EDITION DU 19 NOVEMBRE 2014

Bulletin de l'Association pour la Recherche sur les Tumeurs de la Prostate

Editorial

Pour la 23ème fois, l'ARTP se réunit pour échanger sur les nouvelles perspectives des modèles animaux de métastases et sur les traitements spécifiques. Nous aurons aussi l'occasion de remettre plusieurs prix/subventions grâce au soutien de l'industrie pharmaceutique Pierre Fabre Médicament, Astellas, Janssen, Bouchara Recordati, Takeda , Ferring , Sanofi, GSK et l'Association Francaise d'Urologie.

L'ARTP garde sa vocation de se faire rencontrer tous les acteurs travaillant sur les pathologies prostatiques et de subventionner des travaux de recherche. Sa journée scientifique se poursuit cette année encore au sein du congrès de l'Association Francaise d'Urologie ce qui rendra les interactions vers les cliniciens encore plus faciles. L'ouverture vers l'Europe est toujours assurée par le prix ARTP/ESUR.

Très bon congrès.

Pr Alexandre de la Taille

Président de l'ARTP

Palais des Congrès de Paris

2 place de la Porte Maillot

75017 Paris

Comité d'Organisation

Alexandre de la Taille

Olivier Cuvillier

Palma Rocchi

Jocelyn Céraline

Bernard Malavaud

Pascal Rischmann

Programme scientifique de la 23ème journée

Modèles animaux et métastase osseuse du cancer de la prostate

Ce programme 2014 portera sur les principaux aspects de la recherche sur le cancer de la prostate: les modèles animaux et les nouvelles approches pour traiter les métastases osseuses ainsi que les dernières avancées cliniques. Il n'aurait pas été possible sans le dévouement des co-modérateurs et les membres du Comité d'Organisation de la réunion annuelle. Nous tenons à exprimer notre reconnaissance pour les entreprises et les organisations qui ont fourni un soutien financier généreux pour la réunion annuelle 2014 de l'ARTP. Ces contributions importantes ont contribué à faire de ce forum important possible pour l'échange d'idées qui mèneront à des collaborations fructueuses et des progrès significatifs dans le domaine .

8h45 - 9h00

Message de bienvenue

Pr Alexandre de la Taille, Président de l'ARTP

9h00 - 10h15

Session I : Le point sur les modèles animaux

Modérateurs : Dr P. Rocchi, Dr F. Cabon

Intervenants :
Dr Daniel Metzger (IGBMC, Strasbourg)
Pr Laurent Morel (GReD, Clermont-Ferrand)
Dr Vincent Goffin (Institut Necker Enfants Malades, Paris)

10h15 - 10h30 Pause Café

10h30 -11h45

Session II : Cibler les métastases

Modérateurs : Pr S. Oudard, Pr N. Prevarskaya

Intervenants :
Dr François Lamoureux (Inserm U957, Nantes)
Pr Gert De Meerleer (Université de Gand, Belgique)
Dr Eric Gontier (Hôpital du val de Grâce, Paris)

11h45 - 12h15

Les actions de Movember

Intervenant :
Pr Arnauld Villers (CHRU Lille, Lille)

12h15 - 13h30 Pause Déjeuner

13h30 -14h45

Session III : Session clinique

Modérateurs : Dr F. Vacherot, Pr C. Hennequin

Intervenants :
Dr Elena Castro (CNIO, Madrid, Espagne)
Dr Claire Magnon (iRCM, CEA, Fontenay-aux-roses)
Dr Philippe Barthélémy (Institut Jules Bordet, Bruxelles, Belgique)

14h45 - 15h00 Pause Café

15h00 -16h20

Session IV : Auditions Prix ARTP/ESUR

Modérateurs : Dr V. Goffin, Pr L. Morel

Orateurs :
Victor Laurent (Toulouse)
Rafik Boudra (Clermont-Ferrand)
Félicie Cottard (Strasbourg)
Dr Gerald Verhaegh (Nijmegen, Pays-Bas)

16h20 - 17h30

Session V : Session Poster

17h30

Remise des 3 prix Poster de l'ARTP 2014

Résultats de l'AO 2015 des subventions de l'ARTP

17h45 - 18h15

Assemblée Générale de l'ARTP (réservé aux adhérents)

Prix Poster ARTP

Les 3 meilleurs posters recevront un prix Poster de l'ARTP d'un montant de 500 euros et présenteront leurs travaux lors de la 24ème Journée Scientifique l'ARTP

Dr Daniel Metzger (IGBMC, CNRS UMR 7104, Inserm U964, Strasbourg)

Session I :

Le point sur les modèles animaux

New mouse models of prostate cancer generated by conditional mutagenesis

A large number of studies were performed with immortalized human prostate cancer cell lines, to gain insight into (i) the biology of prostate tumor progression and metastasis formation, and (ii) the mechanisms of anti-androgen treatment escape, as well as to test putative chemopreventive compounds. However, as these lines are derived from advanced/metastatic tumors, they do not recapitulate the various stages of the human disease. Moreover, *in vitro* studies do not reproduce interactions of tumor cells with the various cellular compartments present within the prostate gland or the metastatic site, including immune cells. In addition, xenograft models using such transformed human prostate cell lines do not model the progression and heterogeneity of human patient tumors, and require immunodeficient host animals that lack crucial modulators of tumorigenesis present in the tumor microenvironment.

To circumvent these drawbacks, studies were performed using various genetically engineered mice. The transgenic TRAMP mouse line is the most widely used model for pre-clinical testing of chemoprevention strategies and for the identification of pathways involved in prostate cancer progression. These mice express the SV40 oncovirus large T antigen in the prostatic epithelium, and develop autochtonous prostate tumors. However, this mouse model presents a number of flaws that limits its use (e.g. tumour formation driven by viral proteins not involved in human prostate cancers, development of neuroendocrine carcinoma that rarely occur in human, androgen-dependent expression of the transgene…).

In order to better mimic the characteristics of human prostate tumours, several models were established with the Cre/LoxP technology to ablate the most commonly mutated/lost tumour suppressor genes in human tumours (e.g. PTEN and/or p53), specifically in mouse prostate epithelium. New insights into prostate tumour progression gained from studies of these mouse models will be presented.

Pr Laurent Morel (GRED, CNRS 6293, Inserm U1103, Clermont-Ferrand)

Non mammalian models as new tools for cancer research

Model systems allow researchers to examine aspects of disease that may not be easily or ethically achievable in a human patient. Establishing a model system to study how growth factor signaling affects prostate cancer initiation and progression is crucial in understanding how the disease can be treated. Some systems have already been created to better understand the mechanisms by which a prostate becomes cancerous. However, the complexity and the timing of the disease leads to problems with the model systems already in use. Multiple cell line models have been established to study the progression of prostate cancer. Individual cell lines have been created that have different dependencies on androgen, different invasiveness, and different metastatic potential. These lines are able to model some of the hallmarks of prostate cancer, and can easily be used to study the basic mechanisms of cancer progression. Cell line studies are able to closely control environmental conditions and genetic background, helping to reduce some of the complexity associated with the disease. As with other cell culture models, prostate cancer lines have some disadvantages. The in vitro system may only accurately model a single point in the progression of the disease, and interactions with other cell types or molecules may not be represented. These issues require the use of a whole organism model system.

Several transgenic mouse models have been created that allow researchers to study the progression of the disease. Mouse models that generate invasive and metastatic tumors, and allow for tissue specific manipulation of gene expression, have proven useful in the study of prostate cancer progression. Although these animal models have yielded much data on prostate cancer progression, difficulties remain with the systems. The mouse models are costly to maintain, and can be exceedingly time consuming. Prostate cancer is an age dependent disease, and formation of tumors in these animal models can take years. Therefore, efforts have been made for developing in vivo model system that can reproduce the complexity of the disease, almost at cell signalling level, while still maintaining a practical timeframe for study. Drosohila and zebrafish cancer and prostate cancer models will be presented and their contribution to dissection of the pathways and mechanisms underlying tumor progression and to drug discovery will be highlighted.

Dr Vincent Goffin (Institut Necker Enfants Malades, Inserm U1151, Paris)

Animal models to investigate prostate (cancer) stem cells

Determining the cell-of-origin of prostate cancer may have strong basic and clinical (therapeutic) implications. To that end, recent research has focused on the identification of cell populations displaying stem-like features and androgen-independence, two properties that may be required for tumor initiation and/or tumor recurrence. Since human prostate cancer displays luminal characteristics (i.e. loss of classical basal cell markers), it has been assumed for long that it originates from the luminal compartment.

Accordingly, castration-resistant Nkx3.1-positive cells (so-called CARNs) have been identified in the luminal epithelium of the mouse prostate and their involvement in tumor recurrence after androgen deprivation therapy has been suggested. Interestingly, stem cells have also been identified in the basal layer of the human and mouse prostate epithelium, and their ability to generate prostate cancers when transformed by oncogenes has been demonstrated in transplantation experiments.

Although the basal versus luminal origins of prostate cancer are not necessarily mutually exclusive, these studies have stressed that the identification of cell populations displaying tumor-initiating properties may be (highly) dependent on the type of experimental model used. The most classical ones involve in vitro assays (e.g. prostasphere generation and growth in semi-solid conditions), transplantation of transformed cell populations enriched by cell sorting (using cell-specific surface markers), and in vivo lineage tracing using fluorescent proteins expressed specifically in one cell type (e.g. basal versus luminal cells) to address cell fate in physiological or pathological conditions. I will propose a short overview of the main recent studies that have used these approaches. Finally, I will describe in more detail the probasin-Prl transgenic mouse that we currently use in the lab.

This model involves overexpression of the prolactin hormone specifically in the prostate, and we recently showed that prolactin-induced prostate tumors display dramatic expansion of the basal/stem cell compartment and emergence of potential luminal progenitors.

Dr François Lamoureux (Inserm UMR-S 957, Nantes)

Inhibition of BET bromodomain proteins as a therapeutic approach in metastatic castration-resistant prostate cancer

Prostate cancer (PCa) responds initially to anti-androgen therapies; however, after short term remissions, surviving tumor cells recur with castrate-resistant PCa (CRPC). Men who develop metastatic CRPC invariably succumb to the disease. CRPC progression is mainly driven by deregulated androgen receptor (AR) signaling. To significantly improve survival in men with CRPC, new therapeutic strategies are needed. Recently, small-molecule inhibitors (JQ1 or I-BET762) that target the bromodomain and extra-terminal (BET) proteins especially BRD4 have been shown to exhibit anti-proliferative effects in PCa both *in vitro* and *in vivo* by inhibiting AR signaling. Indeed, it was shown that BRD4 physically interacts with AR and can be disrupted by BET inhibitors. JQ1 disrupts AR recruitement to target gene loci, thus inhibiting AR-target genes.

Moreover, CRPC progression is often associated with bone metastasis. In this case, the vicious cycle established between bone associated tumors and bone resorption is the central problem with therapeutic strategies against primary bone tumors and bone metastasis. Recently, we described inhibition of BET bromodomain proteins as an innovative and promising therapeutic strategy that target simultaneously the three partners of the vicious cycle. Inhibition of BET bromodomain proteins using JQ1 inhibits osteoblastic differentiation both *in vitro* and *in vivo*. These effects are associated with transcriptional silencing of *RUNX2,* resulting from the depletion of *BRD4* from the loci. Moreover, JQ1 also inhibits osteoclast differentiation by interfering with BRD4-dependent RANKL activation of *NFATC1* transcription. Collectively, all these recent data demonstrate the therapeutic interest to target BET bromodomain proteins in CRPC associated with bone metastasis.

Session II :

Cibler les métastases

Pr Gert De Meerleer (Department of Radiotherapy and Experimental Cancer Research, Gent University Hospital, Belgium)

Metastasis-directed therapy for low-volume metastatic prostate and kidney cancer: the power of stereotactic body radiotherapy

Metastatic prostate cancer (M1-PC) covers a spectrum from a low (oligometastatic, defined as 1-3 synchronous metastases) to a high (polymetastatic) number of metastases. Metastases mostly appear after curative treatment of the primary tumor and are the leading cause of death in PC.

Treatment-wise, M1-PC is still considered as a homogeneous group of patients. Standard treatment is palliative ADT, irrespective of the number of metastases. ADT, however, induces important toxicity and negatively influences quality-of-life. Ablation of metastatic spots by means of Stereotactic Body Radiotherapy (SBRT) or surgery has been proposed in case of oligometastatic PC, resulting in nearly 100% local control rates and low toxicity rates. For oligometastatic PC, SBRT to a dose of 10x5 Gy or 3x10 Gy postponed the initiation of ADT with 2.5 years. Ideal candidates are those with 1 metastatic spot and a PSA doubling-time of >4 months. These patients should get the opportunity of SBRT before the initiation of systemic therapy.

These preliminary data have fuelled our currently running randomized phase II trial (http://clinicaltrials.gov/ct2/show/NCT01558427).

Also for renal cell carcinoma (RCC), SBRT to metastatic spots induces excellent local control rates and is therefore a powerful alternative to metastasectomy. This might seems odd, as RCC is traditionally considered as a radio-resistant tumor. However, when the right radiation schedule is used, other than the traditional pathways of cell death are induced. AS a result, local control rates of around 90% are observed and toxicity is very mild. Even when systemic therapies with TKI's failed, SBRT is able to achieve local control rates of almost 90% leading to a median overall survival of >28 months. Apart from ability to induce excellent local control rates, SBRT might also induce the so-called abscopal effect, which is immune-mediated. The combination of SBRT and first-line TKI in this setting is currently studied in phase I / II trials.

Dr Elena Castro (CNIO, Madrid, Espagne)

Targeted prostate cancer screening in BRCA1 and BRCA2 mutation carriers

Most prostate cancers (PrCa) in the developed countries are diagnosed by prosate specific antigen (PSA) screening. The rationale for PSA screening in the general population is the potential to reduce mortality rates through early detection of the disease. However, many expert groups have concluded that data from existing clinical trials on screening are insufficient to recommend the routine use of this test, and screening for PrCa in the general population is very controversial. But despite all the arguments against screening, PSA is still widely used as a diagnostic tool for PrCa, and the scientific challenge is to differentiate which men will or will not benefit from screening with its related consequences. Over-diagnosis and over-treatment are probably the most important adverse effects of PrCa screening and occur more frequently compared to screening programs for other common cancers such as breast or colorectal. There is therefore an urgent need for novel biomarkers that can stratify patients better, according to their risk of developing clinically significant PrCa, to help us decide who should be recommended for screening. Genetic variants such as *BRCA* germline mutations are an example of such predictive factors that could potentially be used to identify patients for targeted screening. Whilst PrCa screening in unselected men remains controversial, targeted screening aimed at higher risk groups may have a greater impact in men who have a higher risk of aggressive and fatal PrCa. That is the case of BRCa mutation carreiers as deleterious germline mutations in both the *BRCA1* and *BRCA2* genes not only increase the risk of PrCa but have also been associated with more aggressive disease and poor clinical outcomes. Early diagnosis in these patients may be crucial and currently the IMPACT study is evaluating the utility of PSA-based PrCa screening in asymptomatic *BRCA1* and *BRCA2* mutation carriers. Up to date, 791 *BRCA1* carriers, 731 *BRCA2* carriers, and 959 *BRCA1* controls have been enrolled in the study. Preliminary results have shown that the positive predictive value (PPV) for biopsy using a PSA threshold of 3.0 ng/ml in *BRCA2* mutation carriers was 48%—double the PPV reported in population screening studies. A significant difference in detecting intermediate- or high-risk disease has been observed in *BRCA2* carriers. These results support the use of targeted PSA screening based on *BRCA* genotype and show that this screening yields a high proportion of aggressive disease.

Session III :

Session clinique

Dr Claire Magnon (CEA, Institut de radiobiologie cellulaire et moléculaire, Fontenay-aux-roses)

Role of the autonomous nervous system in tumorigenesis and metastasis

Convergence of multiple stromal cell types is required to develop a tumorigenic niche that nurtures the initial development of cancer and its dissemination. While the immune and vascular systems have been described as having robust influences on cancer, a growing body of evidences points to the role of the nervous system in promoting cancer development. Recently, we have uncovered (http://www.ncbi.nlm.nih.gov/pubmed/23846904) that sympathetic and parasympathetic nerve fibers from the autonomic nervous system (ANS) infiltrate prostate tumors and contribute to the early stages of cancer development as well as tumor invasion and metastasis. Chemical or surgical ablation of sympathetic adrenergic nerves prevents the formation of xenogeneic orthotopic or transgenic prostate tumors. Further, genetic depletion of β2- and β3-adrenergic receptors in the tumor microenvironment alters the transmission of adrenergic signals involved in the early phases of tumor development. In addition, in cancer mice models, the parasympathetic cholinergic signaling has been identified as a key regulator of tumor invasion and metastatic spread, by activation of the type 1 cholinergic muscarinic receptor (Chrm1) expressed in the stroma. Pharmacological and genetic approaches recapitulate the mechanism by which parasympathetic nerves might be able to release acetylcholine in tumor tissues that binds Chrm1 expressing cell targets in the stroma. Understanding the specific mechanisms of cancer-nerve interactions is pivotal key to developing novel cancer therapies

Philippe Barthélémy (Institut Jules Bordet, Bruxelles, Belgique)

Cancer de la Prostate métastatique: Résultats des études cliniques de phase III

La prise en charge des cancers de la prostate métastatiques s'est considérablement modifiée ces dernières années grâce à l'arrivée de nouvelles molécules ciblant la voie des androgènes, d'agents ciblant l'os ou encore l'arrivée de nouvelles molécules de chimiothérapie. Les résultats de nombreux essais cliniques de phase 3 ont été rapportés cette dernière année évaluant pour certains l'apport de nouvelles molécules ciblées en monothérapie ou en association avec le docetaxel. D'autres études ont évalué de nouvelles stratégies thérapeutiques dans le but d'optimiser la prise en charge des cancers de la prostate métastatique. La présentation aura comme objectif de faire le point sur les derniers résultats des essais de phase 3 rapportés au cours de l'année écoulée.

Metastatic Prostate cancer : An overview of phase III clinical trial results

The management of metastatic prostate cancer has dramatically changed in recent years with the approval of several new therapies targeting the androgen receptor pathway, targeting bone as well as the approvel of new chemotherapy and immunotherapy agents. Very contrasting results from phase 3 clinical trials have been reported in the last year assessing the impact of new targeted therapies on overall survival and progression free survival. Some studies have assessed new therapeutic strategies in order to optimize the management of metastatic prostate cancer. The aim of the presentation is to review the latest results of Phase 3 trials reported during the past year.

Dr Victor Laurent

Institut de Pharmacologie et de Biologie Structurale (IPBS), CNRS UMR 5089, Toulouse

Role of periprostatic adipocytes in prostate tumor dissemination: adipocytes-derived fatty acids induce oxidative stress and stimulate tumor invasion

Initially, prostate tumor evolution is slow and remains localized inside prostate. But during tumor progression some cells are able to become aggressive and spread outside the prostate capsule to invade nearby tissues. Prostate is surrounding by periprostatic adipose tissue which is the major component of invasive prostate cancer stroma. Clinically, periprostatic adipose tissue infiltration by tumor cells is a major histological criteria of poor prognosis suggesting that the adipose tissue could be key step in metastatic process. As we have shown for breast cancer, prostate tumor cells are able to induce "cancer-associated adipocytes" delipidation. We have shown that FFA-derived adipocytes have the ability to stimulate pro-oxidant enzymes in prostate tumor cells (particularly NADPH oxidase 5 or NOX5). Oxidative stress generated by coculture with adipocytes induced an increase of tumor invasive potential through a signaling cascade. By different approaches (pharmacological inhibitors, si/shRNA) we have shown that oxidative stress increase induces HIF-1α accumulation which can generate Epithelial-Mesenchymal transition (EMT), MMPs overexpression. Interestingly, in human tumors, the analysis of 10 patients (with pT3 tumors) revealed that NOX5 expression is highly upregulated to the invasive front where the tumor cells are close to adipocytes. These data highlight an important role of periprostatic adipose tissue in prostate cancer dissemination. Among factors increasing prostate cancer aggressiveness, we also find that obesity is associated with the appearance of high-grade tumors with a particular local and distant spread increased. Interestingly preliminary data revealed a high increase of invasion when prostate tumor cells are co-cultivated with adipocytes from obese mice compared lean mice. We thus hypothesize that the signaling cascade involving FFAs/NOX5 could be amplified under conditions of obesity.

Session V :

Auditions Prix ARTP/ESUR

Les lauréats du Prix Poster de l'ARTP 2013

Rafik Boudra

GReD, CNRS UMR6293, INSERM U1103, Clermont-Ferrand

Origin and consequences of NPM1 overexpression in prostate cancer

Nucleophosmin NPM1 is a molecular chaperone involved in many aspect of cellular physiology, eg. ribosomal biogenesis and cell cycle regulation. NPM1 is overexpressed in numerous types of solid tumors, including prostate cancer but the underlying molecular mechanisms are largely unknown. Using both cell lines and transgenic mouse models, we show that NPM1 expression is significantly increased in cells where the PI3K-AKT- mTOR pathway is activated through PTEN deletion. This overexpression is reversed when cells are treated with the pharmacological inhibitor of mTOR rapamycin. In accordance, transfection of small interfering RNA (siRNA) directed against mTOR leads to mTOR and NPM1 downregulation in these cells. In vivo, we found that NPM1 is overexpressed in PTEN knock-out murine prostate, and a one week treatment of these mice with rapamycin leads to NPM1 downregulation. Chromatin Immunoprecipitation (ChIP) assays show that mTOR is localized on NPM1 proximal promoter. mTOR binding on the chromatin seems to be constitutive, since rapamycin treatment did not alter its localization. mTOR could therefore regulates NPM1 expression by phosphorylating others factors involved in NPM1 gene expression, as previously described (Cunningham et al., 2007). We have previously shown that NPM1 overexpression enhance the migration and invasion abilities of prostate cancer cells (Loubeau et al., 2014). Moreover, NPM1 overexpression in mouse prostate epithelium leads to p27 protein downregulation and cyclin E upregulation, two molecular features of aggressive human prostate cancer. We thus propose that NPM1 could be a downstream effector of PI3K- AKT-mTOR pathway, supporting prostate cancer progression by altering expression of a set of genes involved in cellular homeostasis and proliferation.

Félicie Cottard

INSERM U1113, Strasbourg

Constitutively active androgen receptor variants upregulate expression of mesenchymal markers in prostate cancer cells

Androgen receptor (AR) signaling pathway remains the main target of novel therapeutics for castration-resistant prostate cancer (CRPC). However, constitutively active AR variants lacking the carboxy-terminal region in CRPC lead to therapy inefficacy. Moreover, recent studies suggest that AR variants are expressed in primary prostate tumors and may contribute to tumor progression. The aim of this study was to investigate the impact of AR variants on tumor progression.

N-cadherin expression was analyzed in LNCaP cells overexpressing the wild type AR or a constitutively active AR variant by qRT-PCR, Western blot and immunofluorescence. We showed here for the first time that N-cadherin expression was increased in the presence of constitutively active AR variants. Moreover, we have shown that constitutively active AR variants seem to bind Androgen Response Elements (ARE) in the intron 1 of N-cadherin to induce its expression. Furthermore, in addition to the increased expression of N-cadherin, an upregulation of other mesenchymal markers expression such as *VIMENTIN, SNAIL* and *ZEB1* was observed in the presence of constitutively active AR variants.

Nevertheless, these mesenchymal markers up-regulation were not associated with a decrease of E-cadherin expression. This co-expression between epithelial and mesenchymal markers leads us to think that constitutively active AR variants are associated to partial Epithelial Mesenchymal Transition.

Finally, to have a better understanding of mechanisms leading to mesenchymal markers expression in the presence of constitutively active AR variants, we have performed a mi-RNA-seq experiment in LNCaP cells overexpressing the wild type AR or a constitutively active AR variant. At the same time, we have also analyzed RNA profile by RNA-seq in these cells in order to do correlations with mi-RNA-seq profile.

Taken together, our findings will allow us to have a better understanding of the mode of action of AR variants in prostate cancer.

Dr Gerald Verhaegh

Lab. for Experimental Urology, Radboud University Medical Center, Nijmegen, the Netherlands

The functional role of the PCA3 long non-coding RNA in human prostate cancer

PCA3 is a prostate-specific gene that is highly over-expressed in prostate cancer. The *PCA3* gene lacks an extensive open reading frame and no *in vitro* translation products could be identified, suggesting that *PCA3* acts as a long non-coding RNA (lncRNA). To study the phenotypic effects of *PCA3*, prostate cancer cell lines were transfected with *PCA3* expression vectors. Anchorage-independent cell growth was measured by colony formation in soft agar. *PCA3* over-expressing prostate cancer cells displayed an enhanced capacity to form colonies in soft agar (*i.e.* more and bigger colonies were formed). This finding indicates that *PCA3* plays a role in prostate cancer cell proliferation and survival. In UV cross-linking assays it was found that the *PCA3* ncRNA specifically interacts with several low molecular-weight proteins. Furthermore, *in silico* analysis of the *PCA3* transcript revealed a number of energetically favorable secondary RNA structures that may be precursors for microRNA biogenesis. One *PCA3*-derived small RNA could be detected by Northern blot analysis, and this small *PCA3* RNA was found to be in complex with the AGO2 protein. MiRNA qPCR analysis revealed that this *PCA3*-derived miRNA was up-regulated in prostate cancer. Our results demonstrate that *PCA3* contributes to prostate cancer proliferation and survival. The formation of specific *PCA3* RNA-protein complexes and the processing of the *PCA3* transcript into a microRNA may contribute to the observed *PCA3*-mediated growth stimulation. Identification of the *PCA3* RNA-binding proteins and *PCA3* microRNA target genes is ongoing and will be discussed.

Adipose tissue acts as a driving force for the local and distant dissemination of prostate cancer in obesity

Victor Laurent[1,3], **Adrien Guérard**[1,3], Catherine Mazerolles[4] ,Odile Schiltz[5], Laurence Nieto[1,3], Philippe Valet[2,3], Bernard Malavaud[3,4], Catherine Muller [1,3]

[1]Team "Microenvironment, Cancer and Adipocytes (MICA)", Institut de Pharmacologie et de Biologie Structurale (IPBS) CNRS UMR 5089, Toulouse, [2]Team "AdipOlab" INSERM U858, I2MR, Toulouse, [3]Toulouse university, [4]University Cancer Institut Toulouse. ,[5]Team " Proteomic and mass spectrometry", Institut de Pharmacologie et de Biologie Structurale (IPBS) CNRS UMR 5089, Toulouse

Abstract cannot be published for confidentiality matter.

Session V : **Posters**

Targeting translationally controlled tumor protein TCTP signaling pathway as a therapeutic strategy in castration-resistant prostate cancer

Sara Karaki[1,2,3,4], Julie Acunzo[1,2,3,4], Philippe Barthelemy[5,6], Khalid Oumzyl[5,6], Palma Rocchi[1,2,3,4]

[1]Inserm, U1068, CRCM, Marseille, F-13009, France ; [2]Institut Paoli-Calmettes, Marseille, F-13009, France ; [3]Aix-Marseille Univ, F-13284, Marseille, France ; [4]CNRS, UMR7258, Marseille, F-13009, France. [5]INSERM U869, Bordeaux, F-33076, France, [6]Université de Bordeaux, Bordeaux, F-33076, France.

Introduction : Castration-resistant prostate cancer (CRCP) correlates with Translationally controlled Tumor protein (TCTP) overexpression and loss of p53. Analysis showed that TCTP's expression was found to be significantly down regulated after androgen ablation to become uniformly highly expressed in 75% of castration-resistant prostate cancer. We decided to develop a TCTP inhibitor that can be used for human therapy and choose the ASO strategy because ASOs inhibit specifically target genes especially whose are difficult to target with small molecules inhibitors or neutralizing antibodies.

We are now developing a second generation ASO by using a lipid-conjugated oligonucleotide modification (LASO) by using lipid-conjugated oligonucleotides via "Click chemistry" in order to improve stability, delivery and biodisponibility of TCTP ASO. Interestingly, LASOs self-assembly give spherical nano-objects of different sizes ranging from few nanometers to 500 nm in diameter. Hence, we will take advantage of the nano-systems obtained from LASOs self-assemblies (i.e. hydrophobic regions resulting from the presence of lipids conjugated to ASOs) to encapsulate docetaxel and promote its delivery.

Methods : In prostatic cell lines resistant to castration, we evaluated TCTP expression with different TCTP-ASO chemically modified by Western Blot analysis. MTT (bromure de 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium) was used to evaluate cell proliferation and flow cytometry to examine the oligonucleotide internalization mode. We finally used confocal microscopy to evaluate TCTP-ASO cellular localization.

Results : The addition of lipid to TCTP ASO (TCTP LASO) allowed faster penetration of the oligonucleotide in the cell via micropinocytosis without any transfecting agent, inducing TCTP's inhibition and significant cell viability decrease.

Conclusion : TCTP's inhibition with specific antisense oligonucleotide lipid-moiety-modified seems to be a promising therapeutic strategy to restore castration and chemo sensitivity for the treatment of therapy resistant prostate cancer.

Prostatic cancer of viral origin: homology of human oncogenic papillomavirus (hpv) l1 with nucleophosmin (NPM1), a controller of androgen receptor transcription

Guy Mong Ky Tran [1], Adrien Caprani [2]

[1] *Retired, Public Health, Hotel Dieu,Clermont-Ferrand, FRANCE; 31 Av du Bois 92290 Chatenay-Malabry FRANCE;* [2] *CNRS UMR, Jussieu, Paris*

Background : We showed previously that HPV contained Prostatic Cancer (PC) related oncogenic proteins: 1°) HPV E2 (51-112) is homologous to an Epidermal Growth Factor (6th HIV Conf, Hamburg, 1997: P474). 2°) E1 to PTEN, E6 and L1 to the c-Myc inhibitor Bin-1, a tumor suppressor deleted in 42% of PC (Bull Cancer 2008, 95: 592). 3°) HPV-18 E2 mimics Osteoprotegerin and ParaTHormone related Protein active site (bone metastasis). Anwar K (1992) found 80% HPV-18 in metastatic PC in Japan.

Our meta-analysis concluded to a frequency of 60% oncogenic HPV (-16, -18, -33) in PC (EuroConf Cancer Pasteur Inst, 2004). Our aim is to link HPV to Androgen Receptor (AR). Another hormonal cancer linked to virus is breast cancer, as the virus integration site is Aromatase, the estrogen synthetizing enzyme (Tekmal RR, 1995).

Methods : Amino Acid (AA) sequence comparison between HPV (Lowe J, 2008) and NPM1 (nucleophosmin), which controls AR transcriptional activity by promoting S-phase entry and hyperproliferation (cyclin switch D1 to E1 and p27kip1 loss) (Boudra R, ARTP 2013). Clinically, high p27kip1 is a correlate of better survival after prostatectomy at 5 years.

Results : HPV L1 chimera (types 16, 18, 31, 33, 44, 56, 66, 115) [type-16, AA 167-219] is homologous to NPM1 chimera (human, duck, alligator, sheep, rhinoceros, turtle,...)[AA 1-48]NPM1 MEDSMDMDSMQPLRPQMFLFGC- - - SGAHWARI SPCSLLGGFFAGCELKSDHPV L1 V EDSMDV – SMDPKQIQMFLI GCKPPTGEHWAR-SPCSPVG---AGDCELKSD

Conclusion : Anti-androgen escape may be explained by AR mutations, but also in a PC subset (about 60%) by a viral infection (oncogenic HPV), as HPV L1 is a viral NPM1 mimetic, enhancing AR transcriptional activity and inducing lethal p27kip1 loss. Japanese mushroom Shiitake is a non toxic and highly efficient anti-HPV (Smith JA, 2014). Anti-cancer drugs discovered by HPV-18 infected KB cells screening (Perdue RE Jr, 1982) may act, by serenpidity, as anti-HPV: Taxol (Paclitaxel, Docetaxel, Cabazitaxel). HPV vaccination of young men could protect against PC.

Endothelin-1, a gene regulated by TMPRSS2:ERG fusion proteins in prostate cancer bone metastases

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Background : Bone metastases are frequent and severe complications of prostate cancer (PCa). Recently, the *TMPRSS2:ERG* gene fusion, which results in the aberrant androgen-dependent expression of the ERG transcription factor, has been shown to be the most common gene rearrangement in PCa. This study investigates a potential role of the gene fusion in the development and phenotype of PCa bone metastases.

Methods : We previously established three cell clones from a PCa cell line (PC3c), over-expressing different levels of TMPRSS2:ERG. *In vivo*, using intra-tibial injection models, we analysed induced bone lesion phenotype. Furthermore, a transcriptomic study of these clones showed a change of expression in many genes, including *endothelin-1* (*ET-1*). *In vitro*, using RT-qPCR, siRNA, ELISA and chromatin immunoprecipitation (ChIP), we evaluate a direct regulation of *ET-1* by fusion proteins.

Then, using a cohort of carcinoma prostate samples, we study *ET-1* and *ERG* expression in human pathology.

Results : *In vivo* analysis of bone lesions induced by intra-tibial injections of PC3c-TMPRSS2:ERG clones showed an increase of osteoblastic phenotype compared to control cells. Since ET-1 is known to be involved in osteoblast proliferation and in osteoblastic metastasis formation in PCa, we therefore investigated the transcriptional regulation of *ET-1* by fusion proteins. We have shown that this gene was overexpressed in PC3c-TMPRSS2:ERG clones, depending on ERG expression levels, and was inhibited by ERG silencing. *In silico* analysis of the promoter of *ET-1* revealed the presence of several potential binding sites of ERG. ChIP experiments demonstrated a direct binding to one of them. Moreover, in patients, we were able to establish a correlation between the expression of *ET-1* and the expression of the fusion gene *TMPRSS2:ERG*, reinforcing the link between *ET-1* and the fusion.

Conclusion : Taken together, these results strongly suggest that the *TMPRSS2:ERG* gene fusion contributes to the osteoblastic phenotype of PCa bone metastases and that ET-1 is one of the implicated factor, regulated by the transcription factor ERG.

The functional landscape of Hsp27 reveals new cellular processes such as DNA repair and alternative splicing and proposes novel anticancer targets

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Prostate cancer (PC) is a real public health issue in industrialized countries, mainly due to patients' relapse by castration-refractory disease after androgen ablation[1]. Progression to castration-resistant prostate cancer (CRPC) involves variable combinations of clonal selection, ligand-independent androgen receptor activation and alternative growth-factor pathways as well as adaptive up-regulation of anti-apoptotic genes[2]. One strategy to improve current therapies in advanced PC involves targeting genes that are activated by androgen withdrawal, castration-resistant (CR) phenotype[3,4].

Recently, we identified Hsp27 as a highly over-expressed gene in CRPC. Hsp27 knockdown using antisens oligonucleotides (ASO) and small interference RNA (siRNA) increased apoptotic rates and enhanced hormone- and chemo-therapy in PC[5-7]. We developed and patented a second generation ASO targeting Hsp27 that has been licensed (OGX-427) and phase II clinical trials are currently in process in PC[8,9]. Despite OGX-427 efficiency, the functional roles of stress-induced Hsp27 in castration or chemotherapy-induced apoptosis remain undefined. The purpose of this study is to elucidate the pathways leading to Hsp27 action in CRPC in order to increase the pharmacological safety of OGX-427 and obtain the FDA approval and to find new specific therapeutic targets and treatment strategy for CRPC that would have no toxicity for normal tissues. Towards this goal, we undertook an integrated interactomics study aiming at the identification of the Hsp27 protein-protein interaction network by the yeast-two-hybrid method. To comprehend this network we pursued by in-depth primary and comparative bioinformatics analysis for the investigation and classification of the Hsp27 partners according to their cellular functions. Integration of our data to the human large-scale interactome provided an enriched and well-annotated "Hsp27 interactome", underlining the multifunctional character of this heat-shock protein. Interestingly, our screen identified newly described functions in which Hsp27 is involved, providing a complete comprehension of the differential Hsp27 cytoprotective role in normal and cancer cells.

Our present work affords an in-depth analysis of Hsp27 interactome and provides insights into the involvement of Hsp27 in cell functions leading to gene expression regulation, namely telomere maintenance and DNA repair as well as RNA splicing, opening a new promising field of research for multi-target therapeutic approaches. Furthermore, we found that Translationally Controlled Tumor Protein (TCTP) is a new Hsp27 client protein involved in Hsp27 cytoprotection in CRPC. Targeting TCTP with castration and docetaxel therapy seems to be a promising strategy to efficiently treat CRPC progression, with no or little toxicity for normal prostate cells.

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Sensibilisation a la mort de cellules tumorales de prostate chimioresistantes au docetaxel par un donneur de monoxyde d'azote, le glyceryl trinitrate

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Argumentaire : La chimiothérapie au docétaxel est utilisée depuis 2004 pour traiter les cancers de la prostate métastatiques hormono-résistants (CRPC). Néanmoins il est fréquemment observé un échappement à cette chimiothérapie suite à l'acquisition d'une résistance des cellules tumorales. Aussi des combinaisons thérapeutiques consistant à associer le docétaxel à des molécules cytotoxiques agissant par d'autres voies sont recherchées. Dans ce cadre, nous évaluons l'efficacité cytotoxique de l'association du docétaxel au glycéryl trinitrate (GTN), un donneur de monoxyde d'azote (NO) sur des cellules CRCP et l'action du GTN sur la synthèse de clusterine, une cible thérapeutique émergente des CRCP.

Méthodes : Deux modèles cellulaires de cancers CRCP humains résistants au docétaxel, lignées DU-145 DR et PC3-D12 sont utilisés. La survie de ces cellules après traitement au docétaxel et/ou au GTN est évaluée *in vitro* par différents tests (test d'adhérence, culture en milieu semi-solide, mesure de viabilité au MTS, cytométrie en flux). La sécrétion de clusterine est suivie dans le surnageant des cellules traitées par Western Blot et ELISA et la régulation de l'expression du gène par q-RT-PCR. L'action du GTN sur ces lignées cellulaires est aussi recherchée après transfection avec des siRNA clusterine. De plus, la régulation *in vivo* par le GTN de la clusterine dans les cellules DU-145 DR a été évaluée par q-RT-PCR dans un modèle de xénogreffes chez le poisson zèbre.

Résultats : Nous montrons *in vitro* que le traitement au GTN sensibilise les cellules DU-145 DR et PC3-D12 à la mort et diminue fortement l'expression et la sécrétion de clusterine, cette inhibition de la synthèse de clusterine étant impliquée dans l'activité sensibilisatrice à la mort du GTN. De plus, nous vérifions *in vivo* dans un modèle de xénogreffes de cellules DU145 DR chez la larve de poisson zèbre que le GTN inhibe la synthèse de clusterine et réduit la croissance des cellules tumorales.

Conclusions : Nos résultats montrent l'intérêt d'un traitement combiné GTN/docétaxel capable de réduire l'expression et la sécrétion de la clusterine et de diminuer la résistance des tumeurs à la chimiothérapie.

Emergence of progenitor/stem cell populations at preneoplastic stages of prostate cancer in a mouse model of constitutive prl/stat5 signaling

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Prolactin (PRL) signaling via signal transducer and activator of transcription (Stat5) has recently emerged as a putative alternative target for prostate cancer treatment. PRL expression and Stat5 activation correlate with disease progression and recurrence. Our previous work has shown that transgenic mice expressing PRL in the prostate epithelium (Pb-PRL mice) develop prostate tumors displaying an amplification of epithelial basal/stem cells. This cell population has been reported to participate in cancer initiation and recurrence. Thus, we have further analyzed the amplification of basal/stem cells in Pb-PRL mice. For this, we have compared prostates from Pb-PRL and WT mice using immunostaining, and cell sorting followed by cell culture.

Immunohistological analysis of basal cell clusters and the immediately adjacent luminal cells found higher Stat5 activation, and distinct differentiation status in Pb-PRL compared to wild-type prostates: higher expression of stem cell antigen 1 (Sca-1, a stem/progenitor cell marker) and higher abundance of intermediate cells (which display both basal and luminal cytokeratins). In addition, using FACS we discovered the existence of a new subpopulation of Sca-1-positive luminal cells that also was amplified in Pb-PRL compared to wild-type prostates. In vitro evaluations showed that these cells were able to become Sca-1-negative (mature) luminal cells upon androgen stimulation, and that they had a weak but detectable sphere-generating capacity. We postulate that Sca-1-positive luminal cells represent luminal progenitors that are amplified in response to increased local PRL/Stat5 signaling in the prostate. These luminal progenitors could originate from basal/stem cells through differentiation into intermediate cells which later can commit to the luminal lineage and thus may be relevant for the initial steps of PRL-induced prostate tumorigenesis.

Targeting Hsp27-Eif4e interaction as a new therapeutic strategy for castration resistant prostate cancer

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Background: we shown that silencing Hsp27 expression induce tumor regression and restore castration and chemotherapy sensitivity in castration-resistant prostate cancer (CRPC).We developed and patented a 2nd generation antisens (OGX-427) targeting Hsp27 that has been licensed and phase II clinical trials are in process in patients with PC. Despite the promising results of these trials, long-term inhibition of survival protein Hsp27 might be accompanied with cytotoxic effects due to the role of this protein in several physiological processes. To avoid this problem, an alternative therapeutic strategy is to use compounds that disrupt specific protein-protein interactions between Hsp27 and partners that mediates its cytoprotective effects.

We have shown that Hsp27 interacts with eIF4E and protects it from degradation by ubiquitin-proteasome pathway. The Hsp27-Eif4E interaction leads to the protection of protein synthesis initiation process and enhances cell survival during stress induced by castration or chemotherapy.

Our aim is to understand Hsp27-eIF4e interaction mechanism and found chemical compounds that disrupt this interaction.

Methods: Towards this goals, we have proceeded to the identification of the interaction site of Hsp27-eIF4E by using docking experiments and co-IP with mutants of Hsp27.In the same time, we started to make a high- throughtput screening of compounds that potentially inhibit this interaction by Bioluminescence Resonance Energy Transfer (BRET).We are now considering the effect of chemical compounds that inhibit Hsp27-eIF4E interaction on cell proliferation and apoptosis of CRPC models.

Results: Our results show that eIF4E interacts preferentially with the C-ter part of the phosphorylated Hsp27 and the loss of this interaction increases sensitivity to castration and docetaxel. Preliminary screening using BRET system allowed us to identify one phenazine compound (#14) that inhibits Hsp27-eIF4E interaction. We have shown that treatment of CRPC models with these compounds significantly decrease cell proliferation and increase apoptosis.

Conclusion: Our data allowed us to better define the interaction between Hsp27 and eIF4E. Furthermore, we identified one phenazine compound that specifically inhibits Hsp27-eIF4E interaction and increase castration and docetaxel sensitivity, representing a novel therapeutic strategy for CRPC.

TRP Channel-Associated Factors Are a Novel Protein Family That Regulates TRPM8 Trafficking and Activity

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Background: TRPM8 is a cold sensor that is highly expressed in the prostate as well as in other non-temperature sensing organs and that is regulated by downstream receptor-activated signaling pathways. However, little is known about the intracellular proteins necessary for channel function.

Methods: Here, we identify by GST pull-down followed by mass spectrometry, two previously unknown proteins that we have named "TRP Channel-Associated Factors" (TCAFs) as new TRPM8 partner proteins. We studied the functional interaction of TCAFs with TRPM8 by using binding assays such as co-immunoprecipitation, GST pull-down and FRET, as well as functional assays such as patch-clamp experiments, calcium imaging, and time lapse videomicroscopy migration assays. Expression of all genes was analyzed in different healthy tissues as well as in prostate cancer specimens.

Results: TCAF1 and TCAF2 both bind to the TRPM8 and are necessary to the channel function. However, they exert opposing effects on TRPM8 cell surface trafficking and differentially affect its gating properties. Functional interaction of TCAF1/TRPM8 also leads to a reduction in both the speed and directionality of migration of prostate cancer cells, consistent with an observed loss of expression of TCAF1 in metastatic human specimens, whereas TCAF2 promotes migration.

Conclusions: The identification of TCAFs introduces a novel mechanism for modulation of TRPM8 channel activity and also has potential clinical implication, as this regulation affects directional cell motility in prostate cancer cells, which is a critical process for cancer metastasis.

Prostate cancer and cadmium : a silent inodore toxic penetrating by inhalation

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Background: Studies on identical twins show that prostate cancer (PK) is essentially of environmental origin: There is agreement 16 times, and discordance 135 times (Morganti G, 1956). Cadmium (Cd) is a toxic environmental candidate because there is a rat model of PK caused by Cd (Waalkes MP).

Methods: Literature review.

Results: Cd level, when sought, is statistically higher in the case of PK that in normal tissue (Ogunlewe JO, 1989, in Nigeria) (West, 1991, in USA: OR=1.35) (Brys M, 1997) (Julin, 2012, in Sweden: RR=1.13 - 1.55). Vinceti M (2007) in Italy found an OR = 4.7 by studying toenail Cd levels. Red meat (relatively high Cd level) consumption is correlated with PK (Ekman P, 1999). Cd soil levels are higher in areas with high PK incidence as Salamanca in Spain (Sanchez Garcia A, 1992). Cd accumulates in the prostate with time, which corresponds to an increased PK risk with age. Cd vapours (even in the solid state) penetrate insidiously, are odorless and tasteless. Mussels, oysters and mushrooms are rich in Cd. Tobacco contains Cd. Studies in the workplace show a correlation with the work in contact with Cd (Kjellstrom T, 1979, in Sweden). Cd increases the oncogene c-Myc (Tang N, 1991) and stimulates telomerase (high levels in PK). Subjects with loss of heterozygosity (LOH) of Bin-1, an anti-Myc tumor suppressor (in 50% of PK) (Sakamuro D, 1996) are likely to be

Conclusion: Cd is a risk factor in a PK subgroup; selenium (Yoshizawa K, 1998) and vitamin E synergistically decreased the PK frequency by 63%; selenium binds to prostate metallothionein and releases Cd; α-lipoic acid is a Cd chelator. Some soils are rich in Cd: discharges, gold mines, dams funds; professionally, workers at risk are those of Cd-Ni battery (Sahmoun AE, 2005), anti-corrosion coating, plastic paints, luminescent materials, metalworking (INRS toxicological card, 1992).

Targeted delivering siRNA therapeutics for treating castration-resistant prostate cancer via amphiphilic dendrimer based nanoassemblies

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Background: Prostate cancer (PCa) is the second most common cause of cancer death for men in the Western world. Current chemotherapy effects only a modest improvement in median overall survival for castration-resistant prostate cancer (CRPC). Therefore, novel therapeutic modalities are highly demanding. Anticancer treatment based on molecular targeting of survival genes using small interference RNA (siRNA) constitutes an innovative and promising therapeutic approach. Nevertheless, successful clinical translation of siRNA therapeutics requires essentially safe and efficient delivery systems.

Here, we present targeted transportation of siRNA therapeutics attacking heat shock protein 27 (Hsp27) for anti-PCa study using a novel amphiphilic dendrimer (**AD**) based nanovector.

Methods: siRNA targeting Hsp27 (Hsp27 siRNA) was firstly encapuslated into the dendrimer **AD** based nanoassemblies. Then this system was further decorated with a targeted moiety of $E_{16}G_6$RGDK peptide. The RGDK peptide has recently been shown to target the tumor endothelium through the interaction of RGD (Arg-Gly-Asp) sequence with $\alpha_v\beta_3$ and $\alpha_v\beta_5$ integrins overexpressed on many cancer cells including prostate cancer, and also enhance cell penetration via the binding of RGDK, a CendR motif, towards neuropilin-1 receptor. The gene silencing and anticancer effect were evaluated in vitro and in vivo in PCa model.

Results: The $E_{16}G_6$RGDK peptide decorated **AD**/Hsp27 siRNA delivery system yielded improved gene silencing and inhibition of tumor growth in PCa xenografted nude mice in comparison with non decorated delivery system, confirming the targeting ability of the peptide $E_{16}G_6$RGDK.

Conclusions: This targeted delivery of siRNA therapeutics via amphiphilic dendrimer based nanoassemblies successfully magnified therapeutic potential of RNAi with minimal toxicity profile in PCa model. Hence this target approach may constitute a promising strategy for further translational research in siRNA-based gene therapy for treating CRPC as well as various other diseases, advancing the current therapeutic modality in the view to achieving higher efficacy, less systemic toxicity and better targeting.

Involvement of the ion channel trpa1 in epithelial-stromal interactions in human prostate cancer

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Background : Many data show the impact of the tumor microenvironment and the role it can play in prostate carcinogenesis. Indeed, there exists a close contact between the epithelial tumor cells and tumor stroma which is favorable to the development of cancer in spite of the absence of androgens.

In this context, laboratory's recent works have shown that TRPA1 ion channel is exclusively expressed in stromal cells of prostate cancers. The activation of this channel may allow an increase in intracellular free calcium leading to the secretion of factors by stromal cells and may participate in epithelial- stromal interactions of prostate cancer. It is therefore important to study the role of this channel in the epithelial-stromal interactions and carcinogenesis of the prostate.

Methods : RT-PCR, Western Blot, Immunofluorescence, ELISA, wound healing assay, calcium imaging.

Results : First of all, we studied the role of TRPA1 channel in stromal cells showing in a first time by calcium imaging that TRPA1 channel is functional in stromal cells and secondly, dosing conditioned medium by ELISA, that it is involved in the secretion of growth factors like HGF (Hepatocyte Growth Factor). After, we have study how this channel can be activated and have shown that it is involved in calcium capacitive entry induced by Thapsigargin, and also, in the calcium entry of transduction pathways induced by epithelial agonists like ET-1 (Endothelin-1). Finally, we have studied HGF's effects on epithelial cancer cell lines, and have observed that this factor is able to induce the migration and the epithelial-mesenchymal transition (EMT) of the human prostate cancer cell line androgen-independent DU145 as our results suggest it.

Conclusion : Our work shows the importance of the TRPA1 channel in epithelial-stromal interactions in human prostate cancers. Indeed, this channel, specifically expressed in cancer-associated stromal cells, saw its activity and its expression directly modulated by factors secreted by prostate epithelial tumor cells as it is the case for ET-1. In response to these cancer cells agonists, the TRPA1 channel seems to be involved in the growth of stromal cells and the secretion of growth factors such as HGF, a factor able to induce the migration and the epithelial-mesenchymal transition, suitable process for the metastasis of tumor epithelial cells. Thus, TRPA1 channel could be a particularly attractive therapeutic target in the targeting of epithelial-stromal interactions in the treatment of prostate cancers.

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