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European Society of Cardiology

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ESC First Contact Initiative Grant Report

Host: Dr. Claude Delcayre, Director of Research at CNRS

UMR 942 INSERM – Univ. Paris- Diderot

“Biomarqueurs et Insuffisance Cardiaque”

As an awardee of the ESC First Contact Initiative Grant, I had the opportunity to visit, from April 2nd to April 30th, the Team headed by Dr. Claude Delcayre at the UMR 942 INSERM, Hôpital Lariboisière in Paris. I am really grateful to the European Society of Cardiology for supporting my visit at this Institution.

In past years I have participated to clinical studies on the pathophysiological implications and on prognostic significance of adrenergic and renin-angiotensin-aldosterone system (RAAS) activation in heart failure patients. In particular, we have recently demonstrated a strong, independent prognostic value for plasma renin activity (PRA) in a

large cohort of systolic heart failure patients receiving optimal pharmacological and non pharmacological treatment (*Vergaro G et al, 2011a; Poletti R et al, 2012*). Furthermore, while investigating the clinical significance of neurohormonal reactivation during RAAS blockade, we have shown that aldosterone elevation occurs frequently in heart failure patients, and that it is associated with high PRA and with the angiotensinogen gene M235T polymorphysm (*Vergaro G et al, 2011b*). Finally, we have found that patients with persistent aldosterone elevation despite complete RAAS antagonism (ACE-inhibitors/angiotensin receptor blockers and aldosterone antagonists) show a higher degree of left ventricular remodelling and higher rate of cardiac death (*Vergaro G et al, 2012*).

The wide experience of Dr. Claude Delcayre and Dr. Jane-Lise Samuel in the field of cardiovascular effects of RAAS has provided me with the opportunity to explore a novel, basic, approach to neurohormonal activation in heart failure, with special regard to aldosterone. I had, indeed, the possibility to complete clinical acquisitions with the knowledge of the experimental models, a step likely representing the basis for translational research.

During the first days of my stay in Unit 942, I had the chance to discuss, together with Dr. Delcayre and Dr. Samuel, about the shared research interests and about the possibility to use animal models of heart failure to clarify the pathophysiological mechanism underlying the resistance to neurohormonal blockade commonly observed in the clinical setting. In particular, we evaluated the appropriateness and feasibility of a rat post-myocardial infarction heart failure model for the assessment of the aldosterone breakthrough phenomenon and for the investigation of potential effects of direct renin inhibitors (aliskiren) in reversing aldosterone elevation during treatment with angiotensin receptor blockers, as well as aldosterone-mediated cardiac remodelling.

I was also invited to present and to discuss with the staff of Unit 942 the background, the details of study design and the expected results of my project. A pilot study was planned

to be started, aimed at testing the occurrence and the timing of aldosterone elevation after long term treatment with irbesartan in a rat model of heart failure (rats submitted to transverse aortic constriction).

I was then presented the ongoing experimental protocols at the time of my visit. In particular, it was of great interest to learn more about transgenic mice with cardiac overexpression of aldosterone synthase (AS mice), which have been extensively studied by the group of Dr. Delcayre. In this mouse model the aldosterone synthase gene is selectively upregulated in cardiomyocytes, under control of the myosin heavy chain- α promoter (*Garnier A et al, 2004*). Interestingly, they found that the cardiac phenotype of AS mice does not differ from that of wild-type mice, except for a coronary dysfunction due to alteration in vascular smooth muscle cells BKCa expression and coronary BKCa-dependent relaxation (*Ambroisine ML et al, 2007*). It was of great interest to discuss the potential mechanism by which an increased cardiac aldosterone content prevents the development of cardiomyopathy and may influence insulin sensitivity in mice with streptozotocin-induced type 1 diabetes and high fat, high sucrose diet-induced type 2 diabetes mice, respectively.

AS mice were also used to clarify the effects of aldosterone in the development of cardiac fibrosis in hypertension. Male AS mice were indeed crossed with female RenTgKC mice (Ren), overexpressing renin in the liver, thus obtaining a double transgenic model of both cardiac hyperaldosteronism and systemic hypertension. Interestingly, AS-Ren mice showed a higher degree of cardiac remodelling and fibrosis when compared to Ren Mice. Cardiac hyperaldosteronism was indeed associated with a mineralocorticoid receptor-mediated reduction of the gene expression of two antifibrotic factors, BNP and BMP4, and with an activation of inflammation/galectin-3-induced fibrosis (*Azibani F et al, 2012*)

Along my stay in Unit 942 I could also learn about the technical details of routinely used protocols in molecular biology, which is fundamental for my future research in basic cardiovascular science. In particular I followed the performance of RT-PCR, Western blot, as well as the techniques of nucleic acids and protein extraction from samples of cardiac tissue. It was also of great interest to make myself familiar with the handling of small animal models, namely rats and mice. I was indeed involved in the non invasive blood pressure measurement with the tail-cuff method and in the performance of echocardiography in both ketamine and isoflurane anesthetized mice and rats (either after transverse aortic constriction or sham operated), for the evaluation of left ventricular morphology and function.

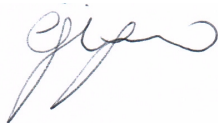
To conclude, this one-month stay at the Unit 942 of INSERM was really useful namely to envision a future collaboration of both laboratories on basic mechanisms of heart failure.

Aknowledgements

I am grateful to Dr. Delcayre and Dr. Samuel for their scientific advice, for their time and for their kind hospitality. I thank Prof. Alain Cohen-Solal for giving me the chance to visit the clinical ward of the Department of Cardiology at Hôpital Lariboisière. I am indebted to Ferial Azibani, Loubina Fazal, Mehdi Maddour, Malha Sadoune, Oksana Boirau, Rebecca Bern, Hélène Ragot, Hugo Breque, Regine Merval and Evelyne Polidano for their patience and kindness in guiding me around the lab.

Yours sincerely,

Giuseppe Vergaro

A handwritten signature in black ink, appearing to read 'G. Vergaro', with a stylized flourish at the end.

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