JOB OFFER: Post-doctoral position for one year

(Possibility of renewal for a second year)

Posted on August 31, 2012

Description:

The successful candidate will have **strong background in molecular biology** (DNA extraction, quantitative PCR…), **immunology** (cell separation based on immune markers…) and/or autoimmunity research. We are looking for an applicant to investigate the **role of natural microchimerism in healthy individuals**.

Due to lack of standardization among measurement and detection techniques, it has often been difficult to assert a clear role for chimerism, natural or not, in autoimmunity or transplantation. Our team developed a large panel of highly sensitive assays, based on Human Leukocyte Antigen (HLA) polymorphisms, to detect and quantify foreign cells. We obtained a grant to create a **platform** dedicated to detection and quantification of foreign cells.

The candidate will run this platform, develop new assays and study the role of fetal and/or maternal microchimerism in health. The candidate will join a research group at the *Institut National de la Santé et de la Recherche Médicale* (INSERM) involved for 20 years in autoimmunity, immunogenetics and clinical research. The group is composed of several scientists and/or medical doctors, a technician, a database manager and students. While knowledge of the French language is not required, English proficiency is mandatory and strong communication and organization skills are expected.

Candidate profile: The applicant must have a PhD in the field of molecular biology, biomedical engineering, immunology or genetics and apply as a post-doc from a non-French laboratory. He/she must be non-French and less than 35 years old.

Workplace: MARSEILLE, Campus de Luminy, France

To apply: please send a CV and a letter of application to **Nathalie C. Lambert (nathalie.lambert@inserm.fr)** with ***"QuantiChimera application"*** in the subject area.

Creation of a platform devoted to foreign cell identification and quantification

Acronym: QuantiChimera

Principal Investigator : Nathalie C. Lambert, CR1 INSERM

Starting date : 29/07/2012

Chimerism is the presence of foreign cells genetically different in an individual. There are two ways to acquire foreign cells: a natural way, through pregnancy and an artificial way through organ or cell transplantation, including blood transfusion **(Figure 1)**. Surprisingly, both chimerism can last for decades following the natural or artificial event. Natural chimerism, present in *small* quantities, called ***Micro*chimerism (Mc)**, has been found in many blood compartments and organ tissues and has focused the attention of several researchers and physicians from multi-disciplinary fields such as **autoimmunity**, **pregnancy**, **cancer**, **infectious diseases**.

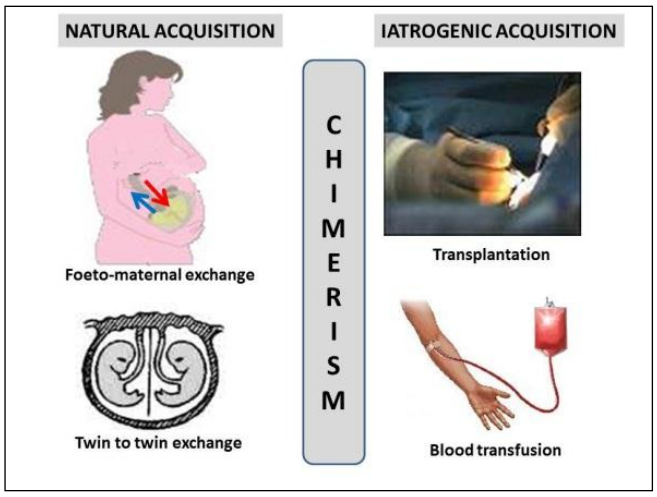


Figure 1- Possible acquisitions of foreign cells in an individual.

Due to lack of standardisation among measurement and detection techniques, it has often been difficult to assert a clear role for chimerism, natural or not, in autoimmunity or transplantation.
We developed highly sensitive tools, based on **Human Leukocyte Antigen (HLA) polymorphisms,** to detect and quantify foreign cells.

1- OBJECTIVES

Based on our expertise on HLA Quantitative PCR assays, we have obtained the 2012 APEX regional grant

1) **To establish a platform devoted to the quantification and detection of chimeric cells in our Inserm unit UMR1097 in Marseille, Luminy.**

2) **To further develop HLA class I -Quantitative PCR assays.**

3) **To better understand mechanisms of acquisition and maintenance of foetal and/or maternal chimerism in healthy individuals.**

2- CONTEXT TO ESTABLISH THE QUANTICHIMERA PLATFORM

3-1 An increasing demand in several research fields:

Increasing interest for detection of foreign, or semi foreign, cells or DNA among physicians and researchers is highlighted, in multi-disciplinary fields, by the emergence of numerous studies on the field. Between 1998 and 2002, a total of 922 studies were published on the topic of chimerism (Medline under the Mesh word

chimerism, not including the 22, 000 studies on transplantation), whereas a 53% increase is noticeable for the following five years. Finally, the creation of a new journal entitled *Chimerism* confirms the tendency (http://www.landesbioscience.com/journals/chimerism).

3-2 An increasing demand to our INSERM team

Aware of our expertise in microchimerism detection, several French and foreign physicians/researchers contacted us to test samples. We then evaluated natural chimerism in a child with juvenile dermatomyositis (Dijon Hospital, France), in a child with DiGeorge anomaly (Angers University and Hospital Center, France), in 55 women with chronic renal diseases (Nice Hospital, France)[1], in a child with biliary atresia who developed graft versus host reaction after liver transplant (La Timone Hospital, Marseille, France), in a man with a scleroderma-like disease for whom foreign female cells were detected by fluorescence in situ in his peripheral blood and maternal/fraternal origin needed to be confirmed (Bruxelles, Belgium) [2]. Investigators were interested in pursuing the analysis on a larger number of patients, but we could not by lack of hands and money.

3.3 A need at the European level

The need for standardization of detection and quantification of chimerism assays has been stressed by several authors in different fields. A Concerted European action, *Eurochimerism*, worked to develop standardized diagnostic methodology for the detection and monitoring of chimerism. It was initiated in 2002 to define a common nomenclature for polymorphic microsatellite (STR) in the field of hematopoietic stem cells transplantations (ESCT) and included leading laboratories from 10 European countries (see below weaknesses of STR methods).
Although lack of common detection and quantification techniques has often been denounced in microchimerism research, such initiative to standardize has never been done. Again, our platform will allow evaluation and comparison of microchimerism/chimerism results between different centers.

The establishment of a platform devoted to the detection and quantification of microchimerism will satisfy the active demand.

3- RATIONALE TO FURTHER DEVELOP HLA- CLASS I –TAQMAN PCR ASSAYS

4.1 In excellent position in the competition of quantification methods

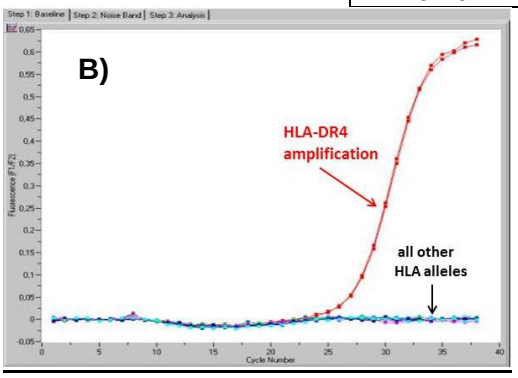
Other investigators have developed quantitative PCR assays based on different polymorphisms to evaluate chimerism and could be in competition with our service. However, up to now the only developed assays available on the market as kits, or as services, are based on Short Tandem Repeat (STR) such as AmpF/STR® Profiler Plus® amplification kit (Applied Biosystem) or BMT CHIMERISM KIT-FL (www. Experteam.it). Such assays have been mainly developed for detection of donor chimerism after peripheral blood stem cell transplantation.
STR strategies have two major limitations: 1) for an accurate assessment of chimerism several polymorphic markers need to be tested when individuals are closely related (mother/child or siblings) which increase the cost; 2) detection sensitivity generally ranges between 1 to 5% which is enough for chimerism detection but *not enough for microchimerism*. These arguments are detailed in § 4.2.
A few investigators have developed HLA specific PCR assays based on the HLA mismatch between the “donor” and the “recipient” [3,4,5]. Artlett et al. had a non-quantitative assay [4]. Garavito et al. and Reed et al. used SYBR Green methods [3,5], which, although very good, are technically less specific than Taqman assays. In SYBR green assays, a small molecule called intercalator: SYBR Green intercalates any double stranded DNA generated at each new cycle of PCR amplification. In our Taqman method a specific fluorogenic probe is added to the classical forward and reverse primers, which makes a higher specificity. Moreover, the sensitivity of our method is 0.005% (see below for details).

4.2- Description of our sensitive and specific HLA Taqman Q-PCR assays

In sex-mismatched pairs, such as women who had given birth to a male, it is easy to use a method of Real Time PCR based on Y chromosome sequence, such as DYS14 [6], a pseudogene of a multigene family [7,8,9]. However it cannot ascertain the foetal origin because of the masculine presence. Moreover to quantify foetal Mc in women who had only given birth to daughters, or to quantify maternal Mc, such sex-mismatched methods cannot be used. We therefore developed and validated a panel of HLA- class II specific quantitative PCR assays (QPCR), designed to target non-inherited, non-shared maternal-specific HLA polymorphisms [10] **(Figure 2 A-C)**.

A)

	HLA- DRB1	
	haplotype 1	haplotype 2
mother	**11**	**04**
child	**11**	**01**



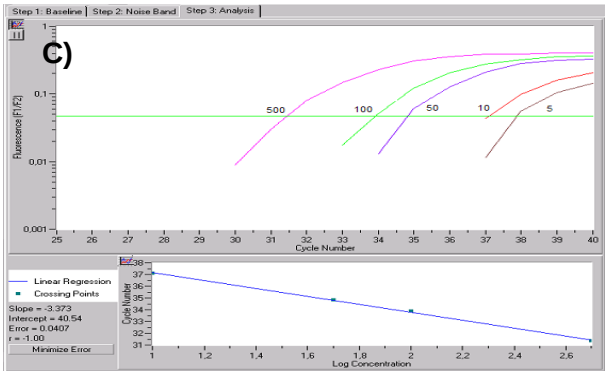


Figure 2- HLA specific Real Time PCR assays

*A) Example of HLA differences between mother and child; the child inherited HLA DRB1*11 allele from his mother and DRB1*01 from his father. Doing a HLA DRB1*04 Q-PCR in child's blood will allow maternal Mc quantification*

*B) Example of specificity of HLA-DRB1*04 assay*

*C) Example of amplification plots from respectively from the left to the right, 500, 100, 50, 10, 5, 1 et 0.5 equivalent DNA of HLA DQA1*03 homozygous cells in a background equivalent DNA of 10,000 non DQA1*03 cells. The x axis indicates the number of cycles and the Y axes the intensity of fluorescence. The lowest quantity of HLA DQA1*03 DNA amplifies at the highest number of cycles.*

A HLA class II panel with ten different assays including HLA-DRB1*01, *15/16, *03, *04, *12, *07, *08 and HLA-DQB1* 02, *03, *06 assays, is already available and the HLA class I panel is currently developed (6 assays) and needs to be further pursued. All our PCR assays have the capacity to detect the equivalent DNA of one cell among 20,000 genome equivalent of host cells (**sensitivity: 0.005%**)

In summary, the platform is unique in France and Europe as our *micro*chimerism quantification methods have 0.005% sensitivity.

4- RATIONALE FOR STUDYING NATURAL MICROCHIMERISM IN HEALTHY INDIVIDUALS

4.1-Quantification of natural Mc needs good methods to avoid wrong interpretations:

It is now well recognized that the placenta is not a strict barrier during pregnancy. Foetal cells and cell-free DNA routinely circulate into the maternal peripheral blood during **normal pregnancy** [11]. Moreover, decades after pregnancy, full term or not, **a woman can still harbour foetal Mc** in her peripheral blood or tissues [12,13].

It is also clearly known that **maternal cells commonly enter the foetal circulation** [14]. First demonstration was done in cord blood samples from male infants, using fluorescence in situ hybridization (FISH) [15]. Female cells were then identified in more than 20% of the samples [15]. Subsequent studies utilizing classical PCR techniques identified maternal DNA with a frequency of 40% [11] to 75% [16] in cord blood samples. As illustrated here, **different techniques conducted to different results**, changing radically the message.

Long term persistence of maternal cells was first described in peripheral blood from infants with severe combined immunodeficiency [17] and more recently in immuno-competent individuals [18,19]. Again, investigation of the biologic significance of maternal Mc has been hampered by limitations in the ability to identify and quantify maternal Mc in progeny.

4.2-Natural microchimeric cells might have consequences on human health:

Most of autoimmune diseases have a predilection for females, following childbearing years and share clinical similarities with chronic graft-versus host disease, a known condition of chimerism. ***Foetal Mc*** could then, have a reaction against the host. We and others argued this hypothesis by showing that more frequent and quantitatively greater foetal Mc was present in blood from women with scleroderma (SSc), a rare autoimmune disease, compared to healthy matched women [20]. By contrast, some other studies showed either a marginal difference or no difference for Mc between patients and controls [21,22,23]. Additionally to pregnancy history, ethnics, or clinical distinction between patients, discrepancy in results may come from technical differences in

Mc detection assays [24]. ***The need of standardization for foetal Mc quantification became urgent to clearly understand the role of Mc.***
Maternal Mc has also been investigated as a risk factor in patients with autoimmune diseases. A caveat of most studies is again, that sex-mismatched pairs were selected, providing only presumptive evidence that Mc derived from the expected source. By quantifying maternal DNA with our HLA specific PCR assays, in peripheral blood from children with Type I diabetes, we showed higher quantities of maternal Mc compared to their non-affected siblings and controls [25]. Although tissue' s localization and function of maternal cells are still under investigation, maternal cells have been detected as beta islet cells secreting insulin [25], giving a new role to Mc. Indeed maternal cells would then restore the damaged organ and be then ***beneficial***.
On the opposite, in neonates with lupus congenital heart blocks, maternal cells are seen as the target of host cells and possibly ***detrimental*** to the host [26].
Mc has been studied in many autoimmune diseases (primary biliary cirrhosis, dermatomyositis, Grave's disease, thyroiditis, Sjögren's syndrome, polymorphic eruption of pregnancy, type I diabetes, lupus and rheumatoid arthritis…) and in some cancers, the initial hypothesis of a detrimental role for natural Mc has evolved since [27].

4.3- We all are chimeras:

Apart from its possible implication in diseases, the fact **we all are chimeras** raises questions. By analogy with transplantation studies, one could wonder whether the **HLA relationship between host and donor** could contribute to the acquisition and maintenance of donor cells. This is in this context that we propose to follow fetal Mc during pregnancy and post-partum in healthy women as well as maternal Mc in cord blood samples and adult's blood. Knowing the HLA genotype of future mothers and retrospectively HLA typing of the foetuses, we will be able to distinguish whether i) the host's and/or donor's genotype could influence the passage of cells and/or their maintenance; ii) the HLA relationship between donor and host could influence both .
We, and others, already demonstrated *on a small number* of subjects that the donor's HLA could influence the persistence of Mc cells [28,29] in a particular host's cell sub-population [28]. We also showed that i) women with SSc have more often an HLA compatible child, ii) that they also have more often and in higher quantities fetal Mc, but **we never demonstrated that higher quantities were due to increased foeto-maternal HLA compatibility**.
This is in this context that our QuantiChimera platform has been set up. As **a "proof of concept" a total of 100 healthy pregnant women will be followed to better understand the mechanisms of acquisition and maintenance of foetal and/or maternal microchimerism.**

5- MANAGEMENT AND ORGANIZATION OF THE PROJECT

5.1- Organization

The current project, is managed by Nathalie C. Lambert, INSERM Researcher and a, **to be named Post Doctorant** (see job announcement). They will run the platform for two years following the described organization (**Figure 3**).
At first, like in any technical platform, there is a necessity of maintenance for a certain number of background tools. This is essential for the right development of the whole project. It includes all the necessary for running assays: stocks of control DNA, of cell lines necessary to specificity tests and stocks of standard curves. A good management of this step is the warranty of a running process without delay.
Secondly and in parallel for the first year, new HLA Q-PCR assays will be developed to cover a full class I panel and extend our possibilities of Mc detection. This step called assays development will go from the design of primers/probe sets to the complete validation of a specific standard curve. This will include 4 points: A) primers/probe sets design for a specific HLA-class I assay, B) test of specificity, C) test of sensitivity and finally D) set up of the standard curve. In order to cover 80% of the general population, design will in priority be done for the most common HLA-class I alleles.
Finally, fetal and maternal Mc follow up will be realized on 100 pregnant healthy women and 80 already recruited post-partum women. In collaboration with two Units of gynaecology in Marseille we will follow 100 healthy women during their pregnancy. Blood samples will be collected each trimester and 3 month post-partum. At the first trimester, maternal blood sample will be sent at the HLA laboratory (EFS, Marseille) to be fully HLA typed. Similarly at delivery, the cord blood sample will be HLA typed. Each trimester and post-partum maternal whole blood and PBMC will be frozen for further DNA extraction and fetal Mc analysis. Cells will be separated by RoboSep sytem (StemCell) the third trimester and post-partum, as fetal cells are generally present in higher quantities that trimester compared to the first and second, therefore easier to be

found in different cell subsets. Retrospectively knowing the foetus HLA typing we will be able to test fetal Mc in the cryopreserved maternal cells and inversely test cord blood samples for their levels of maternal Mc.
To follow up persistence of fetal and maternal Mc in healthy subjects, we propose to test fetal and maternal Mc in women we already have recruited, HLA typed as well as their family members and for whom we know the age and the time post-partum for each child.
Mc frequency will be compared between HLA compatible and HLA incompatible mother/child pairs using a binary outcome (i.e. absence or presence of Mc, measurement >0).
Mc quantities will be compared between the same groups using a logistic regression model to determine if Mc is predictive to HLA compatibility, host's genotype and/or donor's genotype.

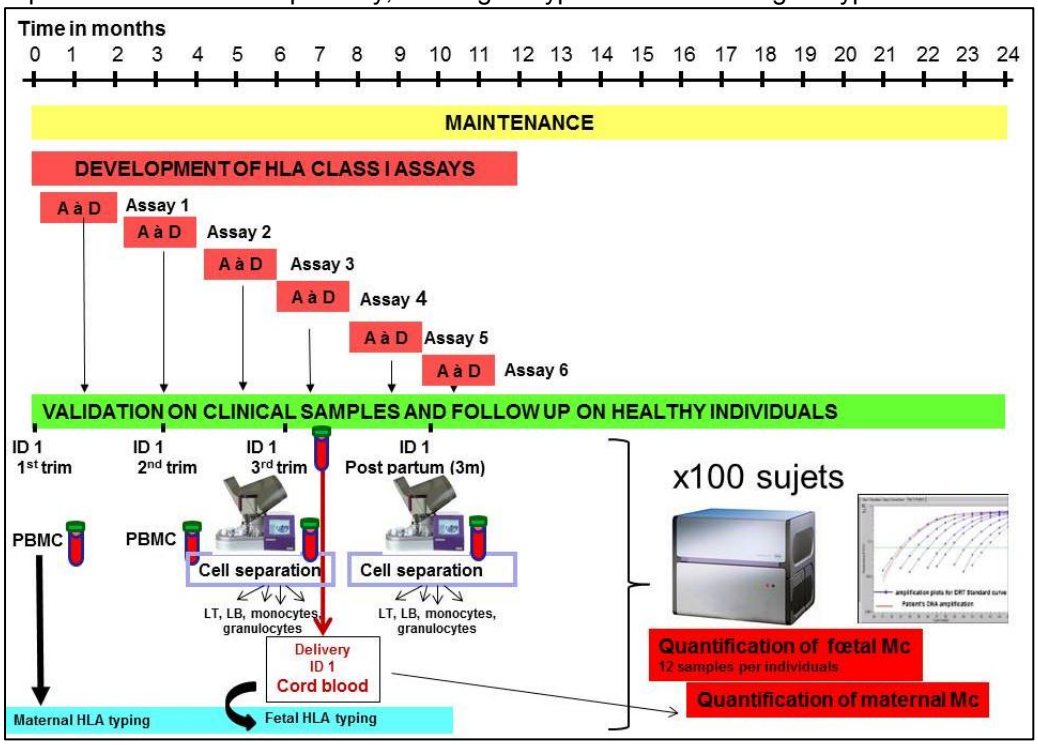


Figure 3- General organization of the platform

5.2- Collaborations:

Teams participating to the project:

- INSERM UMR 1097 directed by Pr Jean Roudier (ex UMR639) **Dr. Nathalie C. Lambert** (CR1 INSERM), 163 avenue de Luminy, Bât TPR2 Entrée A 1er étage 13288 Marseille Cedex 09 ; Tél : 04-91-82-87-68; e-mail: nathalie.lambert@inserm.fr
- Service de Gynécologie du Pr. Gamerre (PU-PH), **Dr Blandine Coubière**, Obstetrician Gynecologist, (MCU-PH), hôpital de la Conception 147, boulevard Baille 13385 Marseille cedex 5, Tél : 04-91-38-37-11; e-mail : blandine.courbiere@ap-hm.fr

- Service de Gynécologie du Pr Léon Boubli (PU-PH), Hôpital Nord, chemin des bourrely 13915 Marseille Cedex ; Tél : 15 04 91 96 46 72 / 04 91 96 48 53, e-mail : leon.boubli@ap-hm.fr, **Pr. Florence Bretelle** (PU-PH), e-mail : florence.bretelle@ap-hm.fr, **Pr. Claude D'Ercole** (PU-PH), e-mail : claude.dercole@ap-hm.fr

- Le Centre d'Investigation Clinique de Marseille Nord, **Dr. Nathalie Lesavre**, Hôpital Nord, chemin des bourrely 13915 Marseille Cedex 15, Tél. : 04 91 96 46 14, e-mail : nathalie.lesavre@ap-hm.fr

Recruitment of participants is done by the two OB-GYN units and their associated MDs.

Follow up of participants (convocations, questionnaires…) is under the direction of Dr Lesavre (Centre d'Investigations Cliniques Nord) with respect of ethic and juridic rules.

The general organization described on **Fig. 3** is realized Dr Nathalie C. Lambert, CR1 INSERM UMR 1097, Marseille and a post doc to be named.

6- PERSPECTIVES

The reason for developing such a platform is the importance Chimerism is now taking in the fields of autoimmunity, cancer and transplantation prognosis. We therefore expect the current pilot study on healthy individuals will be the springboard for many clinical applications. Among them we have already identified some, summarized below:

Hematopoietic stem cell transplantation from cord blood (CB) samples can be successful even if donor and recipient are not fully matched for HLA and has become a real therapeutic hope. This mainly would come from the relative "naivety" of fetal stem cells compared to stem cells from adult bone marrow samples. However, as we described above, cord blood samples could host mature maternal cells. We wonder whether high levels of maternal Mc (non-naïve cells) in cord blood samples may correlate with graft versus host disease. In that context, our initial study evaluating maternal Mc levels in cord blood would be usefull. Then if our initial pilot study shows that the highest level of maternal Mc correlates with the highest risk of GVHD on one side and on the other side that the highest level of maternal Mc in cord blood correlate with higher foeto-maternal HLA compatibility; then we would have clues for cord blood selection prior to transplantation.

Biliary atresia (BA) is a rare infantile liver disease in which the intrahepatic and extra-hepatic bile ducts are damaged progressively by an ongoing fibrosing process. If unrecognized, the condition leads to liver failure. The only effective treatments are certain surgeries to reestablish bile flow, or liver transplantation. Several investigators have shown that maternal Mc may play a role in the etio-pathogenesis of BA by causing an alloimmune reaction given the similarities between BA and graft-versus-host disease (GvHD)[30,31,32,33]. We wonder whether HLA identity and/or compatibility between patient's mother and liver's donors could trigger to a rupture of tolerance. Again studies on HLA relationship and Mc acquisistion and maintenance would be helpful.

We previously showed, as detailed in § 4.2, that Mc foetal and /or maternal may have a role in **Scleroderma (SSc)** and that having an HLA-DRB1 compatible child confers an increased risk to develop subsequent SSc in the mother. One could expect that HLA compatibility of fœtus would favour engraftment of its cells into the maternal organism. Only one small human study suggested an association between Mc and maternal compatibility at the class II DRB1 and/or DQB1 HLA loci [34]. The same authors showed in mice that MHC homozygous progeny had higher levels of maternal Mc than MHC heterozygous progeny [35]. Again a reference in healthy individuals would be important as it would reveal whether HLA compatibility triggers to higher Mc levels, which would explain higher levels in SSc women.

Finally we recently showed another possible **function of Mc in rheumatoid arthritis (RA)**: it could contribute to transfer disease-susceptibility alleles in patients who do not carry this susceptibility [36]. Women who do not have the HLA-DRB1*04 susceptibility allele for RA had more often and at higher quantities Mc for HLA-DRB1*04 than healthy controls [36]. However we have no idea of the phenotype of those cells. Cell sorting realized on healthy individuals would serve as reference for this study.

7- CONCLUSIONS

Our QuantiChimera platform supported by the 2012 Apex regional grant will allow starting a first pilot study on natural microchimerism in healthy individuals and helping for future clinical projects to be developed.

8- REFERENCES

1. Albano L, Rak JM, Azzouz DF, Cassuto-Viguier E, Gugenheim J, et al. (2012) Male microchimerism at high levels in peripheral blood mononuclear cells from women with end stage renal disease before kidney transplantation. PLoS ONE in press.
2. de Bellefon LM, Heiman P, Kanaan SB, Azzouz DF, Rak JM, et al. (2010) Cells from a vanished twin as a source of microchimerism 40 years later. Chimerism 1: 56-60.
3. Reed W, Kong DZ, Lee TH, Cowan MJ, Busch MP, et al. (2002) Non-invasive determination of the paternal HLA haplotype of a fetus using kinetic PCR to detect fetal microchimerism in maternal plasma. Bone Marrow Transplant 29: 527-529.
4. Artlett CM, Cox LA, Jimenez SA (2000) Detection of cellular microchimerism of male or female origin in systemic sclerosis patients by polymerase chain reaction analysis of HLA-Cw antigens. Arthritis Rheum 43: 1062-1067.
5. Garavito G, Klein D, Denis M, Pugliese A, Ricordi C, et al. (2002) Real-time sequence-specific primer polymerase chain reaction amplification of HLA class II alleles: a novel approach to analyze microchimerism. Transplantation 73: 822-825.
6. Lambert NC, Lo YM, Erickson TD, Tylee TS, Guthrie KA, et al. (2002) Male microchimerism in healthy women and women with scleroderma: cells or circulating DNA? A quantitative answer. Blood 100: 2845-2851.
7. Arnemann J, Epplen JT, Cooke HJ, Sauermann U, Engel W, et al. (1987) A human Y-chromosomal DNA sequence expressed in testicular tissue. Nucleic Acids Res 15: 8713-8724.
8. Arnemann J, Jakubiczka S, Thuring S, Schmidtke J (1991) Cloning and sequence analysis of a human Y-chromosome-derived, testicular cDNA, TSPY. Genomics 11: 108-114.
9. Vogel T, Schmidtke J (1998) Structure and function of TSPY, the Y-chromosome gene coding for the "testis-specific protein". Cytogenet Cell Genet 80: 209-213.
10. Lambert NC, Erickson TD, Yan Z, Pang JM, Guthrie KA, et al. (2004) Quantification of maternal microchimerism by HLA-specific real-time polymerase chain reaction: studies of healthy women and women with scleroderma. Arthritis Rheum 50: 906-914.
11. Lo YM, Lo ES, Watson N, Noakes L, Sargent IL, et al. (1996) Two-way cell traffic between mother and fetus: biologic and clinical implications. Blood 88: 4390-4395.
12. Yan Z, Lambert NC, Guthrie KA, Porter AJ, Loubiere LS, et al. (2005) Male microchimerism in women without sons: Quantitative assessment and correlation with pregnancy history. American Journal of Medicine 118: 899-906.
13. Lambert NC, Pang JM, Yan Z, Erickson TD, Stevens AM, et al. (2005) Male microchimerism in women with systemic sclerosis and healthy women who have never given birth to a son. Ann Rheum Dis 64: 845-848.
14. Lo YM, Lau TK, Chan LY, Leung TN, Chang AM (2000) Quantitative analysis of the bidirectional fetomaternal transfer of nucleated cells and plasma DNA. Clin Chem 46: 1301-1309.
15. Hall JM, Lingenfelter P, Adams SL, Lasser D, Hansen JA, et al. (1995) Detection of maternal cells in human umbilical cord blood using fluorescence in situ hybridization. Blood 86: 2829-2832.
16. Bauer M, Orescovic I, Schoell WM, Bianchi DW, Pertl B (2002) Detection of maternal deoxyribonucleic acid in umbilical cord plasma by using fluorescent polymerase chain reaction amplification of short tandem repeat sequences. Am J Obstet Gynecol 186: 117-120.
17. Pollack MS, Kapoor N, Sorell M, Kim SJ, Christiansen FT, et al. (1980) DR-positive maternal engrafted T cells in a severe combined immunodeficiency patient without graft-versus-host disease. Transplantation 30: 331-334.
18. Maloney S, Smith A, Furst DE, Myerson D, Rupert K, et al. (1999) Microchimerism of maternal origin persists into adult life. J Clin Invest 104: 41-47.
19. Lambert NC, Erickson TD, Yan Z, Pang JM, Guthrie KA, et al. (2004) Quantification of maternal microchimerism by HLA-specific real-time polymerase chain reaction - Studies of healthy women and women with scleroderma. Arthritis and Rheumatism 50: 906-914.
20. Lambert NC (2010) [Microchimerism in scleroderma: ten years later]. Rev Med Interne 31: 523-529.

21. Murata H, Nakauchi H, Sumida T (1999) Microchimerism in Japanese women patients with systemic sclerosis. Lancet 354: 220.
22. Selva-O'Callaghan A, Mijares-Boeckh-Behrens T, Prades EB, Solans-Laque R, Simeon-Aznar CP, et al. (2003) Lack of evidence of foetal microchimerism in female Spanish patients with systemic sclerosis. Lupus 12: 15-20.
23. Gannage M, Amoura Z, Lantz O, Piette JC, Caillat-Zucman S (2002) Feto-maternal microchimerism in connective tissue diseases. Eur J Immunol 32: 3405-3413.
24. Rak JM, Pagni PP, Tiev K, Allanore Y, Farge D, et al. (2009) Male microchimerism and HLA compatibility in French women with sclerodema: a different profile in limited and diffuse subset. Rheumatology (Oxford) 48: 363-366.
25. Nelson JL, Gillespie KM, Lambert NC, Stevens AM, Loubiere LS, et al. (2007) Maternal microchimerism in peripheral blood in type 1 diabetes and pancreatic islet beta cell microchimerism. Proc Natl Acad Sci U S A 104: 1637-1642.
26. Stevens AM, Hermes HM, Rutledge JC, Buyon JP, Nelson JL (2003) Myocardial-tissue-specific phenotype of maternal microchimerism in neonatal lupus congenital heart block. Lancet 362: 1617-1623.
27. Lambert NC, Reed AM (2005) Maternal-Fetal Aspects of Autoimmune Diseases. In: Lockshin M, Branch W, editors. Reproductive and hormonal aspect of systemic autoimmune disease: Elsevier.
28. Lambert NC, Evans PC, Hashizumi TL, Maloney S, Gooley T, et al. (2000) Cutting edge: persistent fetal microchimerism in T lymphocytes is associated with HLA-DQA1*0501: implications in autoimmunity. J Immunol 164: 5545-5548.
29. Reed AM, McNallan K, Wettstein P, Vehe R, Ober C (2004) Does HLA-dependent chimerism underlie the pathogenesis of juvenile dermatomyositis? J Immunol 172: 5041-5046.
30. Suskind DL, Rosenthal P, Heyman MB, Kong D, Magrane G, et al. (2004) Maternal microchimerism in the livers of patients with biliary atresia. BMC Gastroenterol 4: 14.
31. Kobayashi H, Tamatani T, Tamura T, Kusafuka J, Yamataka A, et al. (2007) Maternal microchimerism in biliary atresia. J Pediatr Surg 42: 987-991; discussion 991.
32. Hayashida M, Nishimoto Y, Matsuura T, Takahashi Y, Kohashi K, et al. (2007) The evidence of maternal microchimerism in biliary atresia using fluorescent in situ hybridization. J Pediatr Surg 42: 2097-2101.
33. Muraji T, Hosaka N, Irie N, Yoshida M, Imai Y, et al. (2008) Maternal microchimerism in underlying pathogenesis of biliary atresia: quantification and phenotypes of maternal cells in the liver. Pediatrics 121: 517-521.
34. Berry SM, Hassan SS, Russell E, Kukuruga D, Land S, et al. (2004) Association of maternal histocompatibility at class II HLA loci with maternal microchimerism in the fetus. Pediatr Res 56: 73-78.
35. Kaplan J, Land S (2005) Influence of maternal-fetal histocompatibility and MHC zygosity on maternal microchimerism. J Immunol 174: 7123-7128.
36. Rak JM, Maestroni L, Balandraud N, Guis S, Boudinet H, et al. (2008) Transfer of the shared epitope through microchimerism in women with rheumatoid arthritis. Arthritis Rheum 60: 73-80.