Master Biologie Moléculaire et Cellulaire 'BMC',
Université de Paris - *UFR Sciences du Vivant*

Parcours : **Biologie et Développement Cellulaires 'BDC'**
http://www.master2bdc.fr/
Fiche de Projet de Stage M2, Année 2021-2022

Unité INSERM ou CNRS ou Université : INSERM U1016, CNRS UMR 8104, Université de Paris **Intitulé Equipe :** Biologie des phagocytes, Infection & Immunité **ED d'appartenance :** BioSPC **Responsable de l'Equipe :** Dr. Florence. NIEDERGANG	**Responsable du Stage :** Dr. Fatah OUAAZ **Contacts** Adresse : Institut Cochin 22, rue Méchain. 75014 Paris Email : fatah.ouaaz@inserm.fr Tel : 01 40 51 64 21

Titre du projet : Role of dendritic cell subsets and extracellular vesicles in antigen transfer and activation of B lymphocytes

Résumé du Projet de Stage (en 300 mots maximum, mots clés en gras)

Dendritic cells (DCs) are professional antigen-presenting cells, which sample **antigens** (Ags) in the periphery and migrate to the lymph node (LN) where they activate T cells and potentially **B cells**. However, the mechanisms of **Ag transfer** and the modalities of **B-cell activation** by DCs remain incompleteley understood. Previousely, we have reported that human monocyte-derived DCs were able to release native Ag internalized by macropinocytosis from the late endosomal endosomes in the extracellular medium by a process that was named "**regurgitation**" Recently, we reported that murine dendritic cells are important peipheral carriers of Ag to the LN B-cell zone and also potent activators of B cells both *in vivo* and *in vitro*. Importantly, we highlight that Ag released by DC regurgitation is sufficient to efficiently induce early B-cell activation through the nuclear accumulation of the transcription factor NF–κB/cRel.

On the basis of these findings, the M2 candidate wll now explore : 1) the respective role of the LN-resident DC subsets (cDC1/CD8α^+, cDC2/CD11b$^+$) in Ag transport, transfer and B-cell activation both *in vivo* and *in vitro*; 2) the molecular mechanisms of the regulation of DC regurgitation and the role of **DC extracellular vesicles** in Ag release and in subsequent B-cell activation.

The candidate will use murine specific anti-HEL B cells (from MD4 transgenic mice) and DCs (Bone marrow-derived DCs; *ex vivo* purified spleen DC subsets) pulsed with Ag (HEL). He/she will explore the Ag distribution, transfer by DCs and B cell activation *in vivo* and in co-culture *in vitro* by multi-color flow cytometry, immuno-histochemistry and confocal microscopy. DCs EVs will be separated by using both immuno-isolation kit (Miltenyi) and double ultracentrifugation (DUC). We expect to provide new insights into Ag transfer and direct B-cell activation modalities by DCs *in vivo* and also new approaches for DC targeting to elicit humoral immuniy.

Publications de l'équipe relatives au projet de stage (max 5)

1- El-Barby H, Capitao M,……., Niedergang F and Ouaaz F (2020). Extracelluar release of antigen by dendritic cell regurgitation promotes B-cell activation through NF-κB/cRel**. J Immunol**, 205, 608-618.
2- Jubrail, J., Africano-Gomez, …….., G., Mootoosamy Cunoosamy, D., Kurian, N., and Niedergang F (2019). Arpin is critical for phagocytosis in macrophages and is targeted by human rhinovirus. **EMBO Rep**. 21, 1.
3- Niedergang F, Grinstein S. (2018). How to build a phagosome: new concepts for an old process. **Curr Opin Cell Biol**. 50:57-.□63.
4- Jubrail J, Africano-Gomez K,……, Kurian N, Niedergang F (2018). HRV16 impairs Macrophages Cytokine Response to a Secondary Bacterial Trigger. **Front Immunol**. 18; 9:2908.
5- Le Roux D, Le Bon A,……., Bismuth G, Niedergang F. (2012). Antigen stored in dendritic cells after macropinocytosis is released unprocessed from late endosomes to target B cells. **Blood**. 19, 95-105.

1 page maximum SVP !