Profile N° (à remplir par VAS) **FUNDING Planned**
Obtained X

Sheet abstract of thesis 2017 **Disciplinary Fields** **Biologie fondamentale et Bio-informatique**

Thesis Title : (1-2 lines)
Role of dislocations in microtubule dynamics

3 keywords : (1 line) **ACRONYME** MT-DIS
Cytoskeleton / Microtubule / Cryo-electron tomography

Unit/Team of supervising : (1-2 lines)
UMR6290 Institute of Genetics and Development of Rennes / Team Tubulin and Interacting Proteins

Name of the scientific director and co-director : (1 line)

Chrétien Denis (director) and Duchesne Laurence (co-director)

Contact : (1 line)
denis.chretien@univ-rennes1.fr - 02 23 23 67 64; laurence.duchesne@univ-rennes1.fr - 02 23 23 48 82

Socio-economic and scientific context : (10 lines)

Microtubules are constitutive fibers of the cytoskeleton composed of α,β tubulin-heterodimers. They are involved in many cellular processes such as cell division, motility and differentiation. Because of their importance in cell life, they are privileged targets for the development of new therapeutic drugs (e.g. as used in chemotherapy). Tubulin molecules polymerize as outwardly curved 2D sheets that gradually straighten and close into tubes. During this process, tubulin sheets close occasionally onto energetically unfavorable configurations, thereby inducing the formation of local dislocations into their wall. The impact of these dislocations and their potential role in microtubule cytoskeleton functions remain to be determined.

Assumptions and questions (8 lines)

Microtubules assembled *in vitro* or in cell extracts incorporate spontaneously structural defects into their wall (Chrétien et al., 1992). Moreover, recent results by video-fluorescence microscopy reveal incorporation of tubulin into the microtubule lattice *in vitro* and *in cellulo* (Aumeier et al., 2016), suggesting also the presence of local lattice defects. The aim of this project will be to determine the influence of these dislocations on microtubule dynamics: what are their kinetics in *in vitro* reconstituted systems? Are they modulated by the physico-chemical conditions of assembly? Are they effectively presents in cells, and if so, what are their consequences for the dynamics and functions of the microtubule cytoskeleton?

The main steps of the thesis and demarche (10-12 lines)

A significant part of the project will be devoted to the structural analysis of microtubules and of their assembly and disassembly products by cryo-electron microscopy, cryo-electron tomography and 3D reconstructions. The first part of the project will be devoted to the analysis of lattice defect kinetics in *in vitro* reconstituted systems. Microtubules will be self-assembled or nucleated by isolated centrosomes. In parallel, three dimensional reconstructions of these dislocation will be performed by cryo-electron tomography in order to gain a deeper understanding of their structure. In a second part, we will search for the presence of dislocations in cellular microtubules. We will ask whether these dislocations play a role in microtubule dynamics and functions in cells. This project is funded by the ANR for the period 2017-2020.

Methodological and technical approaches considered (4-6 lines)

Methodological approaches will include purification of tubulin and centrosomes, cell culture, sample preparation for cryo-electron tomography, data acquisition (tilt series by electron microscopy), three-dimensional reconstructions and sub-tomogram averaging. Spinning disk microscopy will be used to follow microtubule dynamics.

Scientific and technical skills required by the candidate (2 lines)

Knowledge in biochemistry, molecular biology, structural biology and/or bio-informatics (image analysis). Knowledge of English language required.