

INTERNSHIP PROPOSAL

Institute and Group: Institut de Biologie Structurale – Pneumococcus group (T. Vernet)

Supervisor: Cecile Morlot

Email: cecilemorlot@ibs.fr

Phone: 04 57 42 86 55 - 06 24 59 20 37

Research project title: Structural studies of the SpoIIIA-SpoIIQ complex involved in *Bacillus subtilis* sporulation

5 Keywords to describe the project: multi-protein complex – bacterial sporulation – protein-protein interactions – cryo-electron microscopy

Description of the project (aims, experimental techniques, recommended background):

Bacterial sporulation is a differentiation process leading to the development of a highly resistant spore (temperature, antibiotics, irradiation). Spore development requires the assembly of the SpoIIIA-SpoIIQ multiprotein membrane complex, which has been hypothesized to serve as a channel for molecular transport between the mother cell and the developing spore (Morlot and Rodrigues, 2018). In support of this idea, we showed that core components of this complex (SpoIIAG and GerM) contain ring-building motifs required for protein oligomerization in specialized secretion systems, and that SpoIIAG forms an oligomeric ring displaying structural similarities with those found in type III secretion systems, providing direct evidence for a conduit connecting the mother cell and the developing spore (Rodrigues et al., 2016; Trouve et al., 2018). During the M2 internship, the student will purify recombinant membrane forms of SpoIIAF, SpoIIAG, SpoIIAH and SpoIIQ and reconstitute sub-complexes established by these proteins in proteoliposomes or nanodiscs for future structural characterization (possible thesis project). In parallel, the student will set up immunolabeling of those proteins in sporulating *B. subtilis* cells for future localization experiments using super-resolution microscopy (possible thesis project). We are looking for students with experience in recombinant protein purification (mandatory) and bacterial cell imaging (facultative).

Justification that the internship's subject fits with the general theme of GRAL:

This project aims at characterizing the assembly of core components of a large multiprotein membrane complex during bacterial sporulation. The experimental approach includes *in vitro* and *in vivo* methods, which will generate complementary data at the molecular, structural and cellular level.

Relevant publications of the team:

Trouve J, Mohamed A, Leisico F, Contreras-Martel C, Liu B, Mas C, Rudner DZ, Rodrigues CDA, **Morlot C** (2018). Structural characterization of the sporulation protein GerM from *Bacillus subtilis*. *J. Struct. Biol.* pii: S1047-8477(18)30270-3. doi: 10.1016/j.jsb.2018.09.010. [↗](#)

Morlot C, Rodrigues CDA (2018). The new kid on the block: a specialized secretion system during bacterial sporulation. *Trends Microbiol.* pii: S0966-842X(18)30001-5. **Review.** [↗](#)

Rodrigues CDA, Henry X, Neumann E, Kurauskas V, Bellard L, Fichou Y, Schanda P, Schoehn G, Rudner DZ, **Morlot C** (2016). A ring-shaped conduit connects the mother cell and forespore during sporulation in *B. subtilis*. *Proc. Natl. Acad. Sci. USA* 113(41):11585-11590 [↗](#)

[↗](#) **corresponding authorship for C. Morlot**