

Colloque VIIB : Complexes d'intérêt biomédical

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**Structural and Biochemical Analysis of Autoregulation in RalF, a
Legionella Pneumophila ArfGEF**

Legionella pneumophila is an aquatic bacteria that upon inhalation by humans, replicates within alveolar phagocytes by forming a specialized compartment similar to the endoplasmic reticulum of the host. Legionella supports its infectious cycle by utilizing a Type IV secretion apparatus that injects at least 20 bacterial proteins into the host cell. RalF is one component of the Legionella secretion system that was identified as a Guanine nucleotide Exchange Factor for Arf GTPases. RalF possesses at least two distinct domains, an N-terminal catalytic exchange domain that shares 41% homology to the mammalian Sec7 and a C-terminal domain of unknown function that has a similar fold to the "platform domain" present in AP2 and COP adaptor proteins that function in the assembly of clathrin and non-clathrin coated vesicles. Structural and biochemical studies of RalF indicate that it is autoinhibited due to the presence of a "capping helix" that physically occludes the active site of the Sec7 domain, suggesting that a conformational change in RalF is required for optimal substrate recognition. Based on recent findings concerning autoregulation in the Grp1 family of ArfGEFs together with our observations, we present a mode of activation for RalF that involves a GTPase signaling cascade. Additionally, we analyze the kinetic properties of RalF and its specificity for the Arf family which will provide a basis for designing a small molecule inhibitor for this human pathogen.