Crystal Structure of Truncated FlgD from the Human Pathogen *Helicobacter pylori*

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Helicobacter pylori is a Gram-negative pathogen able to colonize the human stomach and it is responsible for several gastric pathologies, including gastritis, peptic ulcer, gastric adenocarcinoma and MALT lymphoma.[1] In order to survive in the hard stomach environment and to permanently settle in it, *H. pylori* has to move through the mucous layer and adhere to gastric epithelial cells, in particular during the initial phases of the infection, and in doing so it has to rely on flagella, a rotatory nano-machine. Major sections that define the flagellum are: the filament, the hook and the basal body.[2] In this bacterium FlgD is absolutely needed for the assembly of the flagellar hook, but it has not been detected in the mature flagellum.[3]

In order to clarify the structural and functional properties of this *H. pylori* virulence factor we performed cloning, purification, crystallization and X-ray analysis studies. The HP0907 gene was cloned from two different *H. pylori* strains, 26695 and G27. Single crystals were prepared by the sitting drop vapor diffusion method using an automated crystallization platform (Oryx 8 robot). Two different crystal forms were obtained. Diffraction data of native FlgD_G27 (*Hp*FlgD_m) were measured at the ID14-4 beamline (ESRF, Grenoble, France) and they belong to the monoclinic system, space group *P*2. Crystals of both native and seleno-methionine FlgD_26695 (*Hp*FlgD_t) belong to the tetragonal space group *I*422 and diffraction data of this second crystal form were measured at the PXIII beamline (SLS, Villigen, Switzerland). The quaternary structure in both crystal forms is a tetramer. Four monomers are present in the asymmetric unit of *Hp*FlgD_m, corresponding to a *V*M of 2.63 Å3 Da^{-1} and an approximate solvent content of 53%. In the structure of *Hp*FlgD_t there is one monomer in the asymmetric unit, with a *V*M of 3.26 Å3 Da^{-1} and an approximate solvent content of 62%. The tetramer is generated by the four-fold symmetry axis. Although crystallization of the full length proteins of both strains, FlgD_G27 and FlgD_26695 was attempted (316 and 301 amino acids residues, respectively) the structures revealed the truncated form with the monomers comprising residues Asn127 – Lys272.

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2. Soutourina O.A. and Bertin P.N., *FEMS Microbiol. Rev.,* **2003***.* 27, 505-523.
3. Ohnishi, K., Fan F., Schoenhals G.J., Kihara M. and Macnab R.M., *J. Bacteriol.*, **1997**. 179, 6092-6099.