

domain (FlhBc). Homologues of FlhB were found in all bacterial type III secretion systems. The sequence of the protein is highly conserved suggesting that the protein function is also similar.

We have investigated complementation properties of FlhB from *Aquifex aeolicus* to FlhB from *Salmonella typhimurium*. *flhB* gene in *Salmonella* was substituted by *flhB* of *A. aeolicus* or by chimera *flhB* composed of different parts from *A. aeolicus flhB* and *S. typhimurium flhB*. Such mutants were tested for motility. All mutants showed motility although weaker than wild cells. We have found that some mutations in C-terminal part of FlhB resulted in enhanced motility. To explain results obtained we have determined FlhBc structures from both organisms: *S. typhimurium* and *A. aeolicus*. Comparison of the structures gives us new ideas about functional mechanism of FlhB.

**Keywords:** flagella, type III secretion system, FlhB

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#### High-resolution crystal structure of an outer membrane-anchored endolytic peptidoglycan lytic transglycosylase (MltE) from *Escherichia coli*

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The crystal structure of the first endolytic peptidoglycan lytic transglycosylase MltE from *Escherichia coli* is reported herein. The degradative activity of this enzyme initiates the process of cell wall recycling, which is an integral event in the bacterial existence. The structure sheds light on how MltE recognizes its substrate, the cell wall peptidoglycan. It also explains the ability of this endolytic enzyme to cleave in the middle of the peptidoglycan chains. Furthermore, the structure reveals how the enzyme is sequestered on the inner leaflet of the outer membrane.

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#### Structural characterization of the conjugation machinery in G+ bacteria

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Bacterial conjugation is a major mechanism of the horizontal

gene transfer (HGT) in bacteria and thus an important component of bacterial evolution. It provides a route for the rapid acquisition of new genetic information and contributes to the spread of antibiotic resistance [1]. The latter is becoming a primary threat to human health, since currently 70% of all hospital acquired infections are resistant to at least one antibiotic, and antibiotic resistance to at least one drug has been reported for every major strain of pathogenic bacteria [2]. Whilst the plasmid conjugation in Gram-negative (G-) bacteria has been studied in detail, the corresponding process in Gram-positive (G+) bacteria is poorly understood at the molecular level [3]. Among them, the G+ bacterium *Streptococcus pneumoniae* is responsible for nearly 2 000 000 human deaths annually, and this figure represents only 15–20% of the people infected [4]. Thus, understanding HGT in pneumococcus and related bacteria may help in controlling the spread of medically important antibiotic resistance [5]. Progress in the expression, purification, crystallization and structural characterization of key proteins of the conjugative process in *S. pneumoniae* will be presented.

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#### Dimeric and Tetrameric forms of enoyl-ACP reductase

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Most bacteria synthesize fatty acids using a discrete and highly conserved group of enzymes that each carry out a single reaction. Among them is enoyl-ACP reductase (or ENR) which catalyzes the last step in each elongation cycle. It is considered as an attractive target for new antibiotics since it is an essential enzyme and the fatty acid synthesis machinery in human is quite different. Some pathogens such as *Enterococcus faecalis*, *Bacillus subtilis*, *Pseudomonas aeruginosa* have more than one ENR. There have been a number of crystal structures reported from various pathogens: some with an inhibitor and the cofactor (either NADP or NAD), some without either the cofactor or an inhibitor. The overall structures, including the active sites, are quite similar to one another in the case of the ternary complexes, and they are all found as a homo-tetramer in the crystal. The apo structures are also found as a homo-tetramer with the substrate and the cofactor binding domains mostly disordered. However, the recent studies include an apo ENR in a homo-dimeric form, and also in a homo-tetramer but with different arrangement from what was observed earlier. Detailed analysis and its implication will be presented.

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