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Transcriptional regulation of *Salmonella* Typhimurium Pef fimbriae by H-NS, Hha and YdgT nucleoproteins

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Abstract

Background: Gastroenteritis caused by *Salmonella* Typhimurium is triggered by bacterial adherence to intestinal epithelium cells that leads to invasion and destruction of the mucosal surface. The *pef* operon of *S. Typhimurium* is responsible for the biosynthesis of plasmid-encoded fimbriae (Pef), which mediate adhesion to mouse intestinal epithelium. As most of the 13 fimbriae of *S. Typhimurium*, expression of Pef fimbriae is tightly regulated. *In vitro*, their expression was previously detected only in standing cultures grown in rich acidic medium. Previous work and microarray studies suggested a role of the nucleoproteins H-NS and Hha-YdgT in this negative regulation of *pef* fimbriae expression. In this study, our objective was to demonstrate this repression and to characterize the underlying mechanism. **Methods:** Due to instability of *hns* mutants, strains carrying a deletion of the *hns* gene were freshly constructed by P22 transduction before each experiment. Promoters activities were quantified using plasmid-based transcriptional fusions carried by wild-type, *hns* and/or *hha-ydgT* mutants. Expression of Pef fimbriae was measured by RT-PCR and Western blot by measuring *pefA*/PefA expression, which encodes the major subunit. **Results:** We demonstrate that H-NS and Hha-YdgT negatively regulate *pef* operon transcription by acting on the promoter located upstream of *pefB*, the first gene of the operon. The effect of H-NS was much more pronounced than that of Hha-YdgT. Moreover, we observed that Hha and YdgT can repress *pef* expression independently of H-NS when bacteria were cultivated in acidic medium under standing conditions, but not after culture in neutral pH medium. **Conclusions:** This work demonstrates that the weak expression of Pef fimbriae *in vitro* is partly due to the combined action of H-NS and Hha-YdgT nucleoproteins on the transcriptional activity of the promoter region located upstream of the *pef* operon. A debate still exists in the literature concerning the exact mode of action of these nucleoproteins. Experimental evidence and a mechanistic model recently described indicate that Hha and YdgT act primarily through H-NS to modulate gene expression. On the contrary, few reports show that Hha can bind to specific regulatory sequences independently of H-NS. Our results on *pef* operon transcriptional regulation are in favor of the existence of these two models. Indeed, Hha and YdgT can act through H-NS to modulate *pef* expression. Nevertheless, according to the culture conditions used in our experiments, it appears that Hha and YdgT can also act independently of H-NS.