

**TITLE:** TREHALOSE METABOLISM INFLUENCES TYPE 1 FIMBRIA PRODUCTION IN

*Escherichia coli* STRAIN MT78

**AUTHORS:** KLEMBERG, V.S.<sup>a,b</sup> ; PAVANELLO, D.B.<sup>a</sup>; HOULE<sup>b</sup>, S. ; DOZOIS, C. M.<sup>b</sup>; HORN, F.<sup>a</sup>.

**INSTITUTION:**<sup>a</sup> Departamento de Biofísica, Universidade Federal do Rio Grande do Sul, RS, Brazil, <sup>b</sup> INRS- Institute Armand Frappier, Laval, Quebec, Canada

**ABSTRACT:**

Trehalose is present in bacteria, yeasts, and plants. In *Escherichia coli*, trehalose can be metabolized as a carbon source or be accumulated as an osmoprotectant under osmotic stress. Under hypertonic medium, *E. coli* accumulates trehalose internally by synthesizing it from two glucose molecules, catalyzed by OtsA and OtsB enzymes. Trehalose is sent to the periplasm and is degraded by the TreA enzyme into two glucoses, which are transported back to the cytoplasm. Extraintestinal *E. coli* (ExPEC) strain MT78 harbors type 1 fimbria, an adhesin that enables ExPEC strains to adhere and infect eukaryotic cells. In a previous study, we showed that the MT78 $\Delta$ treA mutant displayed impaired type 1 fimbria production, as verified by reduced levels of yeast agglutination, reduced invasion of avian fibroblasts, and reduced bladder colonization in a murine model of urinary tract infection. While absence of the periplasmic TreA likely results in higher internal trehalose concentrations, we wondered if deletion of *otsA* and *otsB* genes, which would lead to decreased internal trehalose concentrations, would instead promote type 1 fimbriae production. Yeast agglutination assays revealed that MT78 $\Delta$ otsBA was also impaired in fimbriae production, and such impairment was even more pronounced in the triple mutant MT78 $\Delta$ otsBA $\Delta$ treA. Thus, the osmoregulated enzymes of trehalose metabolism are somehow required for full production of type 1 fimbriae in ExPEC MT78. Moreover, MT78 $\Delta$ otsBA was unable to grow in minimal medium with glycerol as carbon source in the presence of urea 0.6 M, while MT78 $\Delta$ treA grew even better than the wild type. Urea is known to promote fimbria expression in uropathogenic *E. coli*. Following growth in LB in the presence of 0.3 M urea, a concentration similar to those found in human urine, differences in yeast agglutination among WT and mutants MT78 $\Delta$ otsBA and MT78 $\Delta$ otsBA $\Delta$ treA were significantly decreased. Altogether, our results suggest that trehalose metabolism influences type 1 fimbriae production, even though trehalose does not seem to be the sole or main osmoprotectant against urea in ExPEC MT78.

**Keywords:** Extraintestinal *Escherichia coli*, Trehalose, Type 1 fimbriae

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