M.Sc. Project

TITLE: Studies on the role of DsrD in dissimilatory sulfite reduction

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Duration: 1 year

Background

Comprehensive studies of the human gastrointestinal microbiota have revealed the presence of sulfate reducing bacteria (SRB) along with levels of sulfate in the gut that can sustain proliferation [1]. These anaerobes are common members of the human gut microbiota [2,3] and have been implicated in inflammatory bowel diseases (e.g., ulcerative colitis and Crohn's disease), due to the toxic and corrosive effects of sulfide produced from sulfate reduction, which lead to cell inflammation [2,4,5]. Other illnesses of the human gut tract have also been linked to SRB activity, such as cholocystitis, abdominal abscesses, spondylitis or even colorectal cancer (where sulfide is considered to be an initiator of the disease by activating a number of biochemical pathways) [6]. The enzyme responsible for the production of sulfide by SRB and other sulfidogenic gut bacteria is the dissimilatory sulfite reductase DsrAB together with the small protein DsrC and the DsrMKJOP membrane complex, as discovered in our laboratory [7]. Immediately upstream of the *dsrAB* genes is the *dsrD* gene encoding a protein with a still unidentified function. The *dsrD* gene is present in all organisms that have DsrAB, i.e. in all SRB, sulfite-, thiosulfate- and organosulfonate-reducing organisms, strongly suggesting an important role of DsrD in the reduction of sulfite.

DsrD is a small protein containing several conserved Lys residues that may be involved in the binding of anions. The crystal structure of DsrD showed the presence of a conserved winged helix-turn-helix (wHTH) motif, characteristic of DNA binding proteins or protein-protein interactions [8]. Curiously, in the sulfite-reducer gut pathogen *Bilophila wadsworthia,* the DsrB and DsrD proteins are fused [9], raising the possibility that DsrD may have a functional role in the DsrAB-catalysed sulfite reduction.

In our lab we have preliminary data suggesting that DsrD plays an important role in the dissimilative metabolism of SRB, at the catalysis level, but in what specific role is not yet clear. Thus, the objective of this project is to study the effect of several conserved amino acid residues of DsrD in the catalysis of sulfite reduction using *in vitro* functional studies. This work will allow a better insight into the mechanism of the toxic production of sulfide in the gut, and how to inhibit it.

Objectives

The main goal of this project is to elucidate the role of DsrD in the sulfite reduction pathway by studying the role of several conserved residues of this protein. To achieve this purpose, the main objectives are:

- To produce several DsrD variants containing point mutations in conserved residues;

- To study the impact of DsrD variants in sulfite reductase enzymatic activities;

- To investigate the properties of DsrD variants in protein-protein interaction studies with DsrAB.

References

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