Bacterial Topoisomerases as Target of Peptides Structurally Derived from CcdB Protein

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Bacterial DNA gyrase and topoisomerase IV (Topo IV) are examples of type IIA topoisomerases. DNA gyrase is unique in catalyzing the negative supercoiling of closed circular DNA and Topo IV has the ability to relax positive and negative supercoils, essential processes that control the topological state of DNA in cells. These features make of these enzymes important targets for antibacterial agents including the natural toxins like CcdB protein. The bacterial toxin CcdB is an 11.7 kDa protein that forms with CcdA (8.7 kDa), a toxin-antitoxin (TA) system, which contribute to plasmid stability in *Escherichia coli*. CcdB acts on DNA gyrase forming a stable complex with its catalytic subunit, relieving the supercoiling of the DNA by creating a transient break in the double helix, but no activity on Topo IV has been described for CcdB. In order to the develop inhibitors that might simultaneously target both DNA gyrase and Topo IV, several peptides analogues of CcdB were synthesized by SPPS and tested about inhibition of DNA gyrase and Topo IV activity by agarose gel electrophoresis and by bacterial growth. Peptides analogues with helix structures showed inhibition for both supercoiling and relaxation activity with IC_{100} < 5 µM. However, all peptides were unable to inhibit the bacterial growth, due the poor permeability of the bacterial membrane. To overcome this limitation, the peptides were encapsulated in liposomes (SUV) and other vesicles types. Employing a drug delivery system, the helix peptides were able to inhibit bacterial growth with a MIC very close of the value obtained *in vitro*. Therefore, we developed a new class of topoisomerase inhibitors that, using the appropriate delivery system, may serve as antibacterial agents.

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