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Binding of the Marine Sponge Pigment Fascaplysin to DNA: Comparison of Calorimetric and Spectroscopic Titrations

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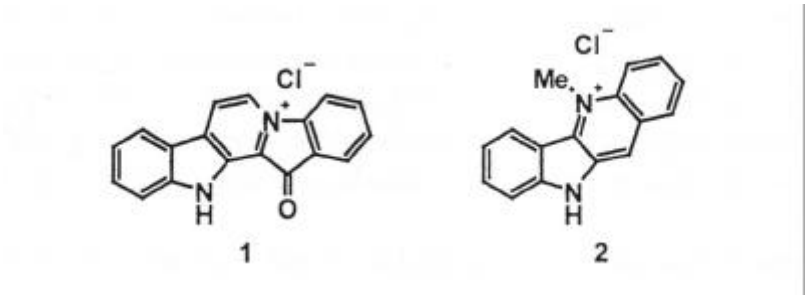
Fascaplysin (1) is a red pigment isolated in 1988 from the Fijian sponge *Fascaplysinopsis* Bergquist sp., and was reported to inhibit the growth of several microbes, and to suppress proliferation of mouse leukemia cells at low concentrations [1]. In order to investigate the possibility of interference of fascaplysin with the genetic material as an explanation for the biological activity, we initiated a study on the binding of fascaplysin with DNA.



We report the results on titrations of calf thymus (CT) DNA with fascaplysin (1) monitored by isothermal titration calorimetry (ITC), which is a general method for measuring binding affinities and stoichiometries of binding phenomena [2], by ultraviolet absorption spectroscopy (UV), and by circular dichroism (CD). While the thermal method directly addresses the thermodynamics of the binding process, the Spectroscopic methods probe the change of the electronic properties of the drug molecule as it binds to DNA. Thus, the thermal and Spectroscopic methods have different point of views complimenting each other and revealing the complete picture of the binding process. Results are compared with those of the cytotoxic agent cryptolepine (2), which has been demonstrated to efficiently intercalate into DNA and to act as a potent topoisomerase II inhibitor [3].

The UV Spectroscopic data could be well interpreted in terms of a two-site model for the binding of fascaplysin to DNA revealing affinity constants of $K_1 = 2.5 \times 10^6$ M^{-1} and $K_2 = 7.5 \times 10^4$ M^{-1} (base pairs of DNA). Based on the typical change observed in both the absorption and circular dichroism spectra, intercalation of fascaplysin is regarded as the major binding mode. The calorimetric titration curves showed an exothermic reaction which was exhausted at a 2:1 base pair/drug ratio. This finding is in agreement with an intercalation model comprising nearest neighbor exclusion. In addition, significantly weaker non-intercalative DNA interactions can be observed at high drug concentration. By comparison of all the data with the binding behavior of known intercalating agents, it is concluded that, besides weaker binding modes, fascaplysin intercalates into DNA. Thus, some of the biological activity can be attributed to interference with the genetic material.

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