Thermodynamic and kinetic study of Ti(IV) interaction with transferrin

Arthur D. Tinoco, Ann M. Valentine

Department of Chemistry, Yale University, New Haven, CT 06520

Contrary to popular belief, evidence exists implicating titanium bioactivity. A compelling finding is the anticancer activity exhibited by titanium(IV) compounds. Budotitane (diethoxybis(1-phenylbutane-1,3-dionato) titanium(IV)) and titanocene dichloride (Cp_2TiCl_2) were two promising Ti(IV) leads. The drugs accumulate Ti(IV) in the chromatin of cancer cells but cause minor DNA crosslinking suggesting a different mode of action from cisplatin.[1] Although DNA is thought to be the target of Ti(IV) anticancer drugs, the instability of the drugs due to metal hydrolysis has made it difficult to characterize DNA interaction with them.

Determining the mechanism of action of Ti(IV) anticancer drugs is crucial for synthetic efforts to improve medicinal activity. In 2000 Sadler *et al.* proposed that the anticancer activity of Ti(IV) drugs probably derives from complexation of the metal by the 80 kDa serum protein transferrin.[2] Transferrin, present at 30 μM in serum, is the major Fe(III) transport into cells and is only 30 % Fe(III) saturated. It binds and circulates other hard metal ions. It is plausible that transferrin serves as an effective carrier of Ti(IV) into cells.

To shed light on the cancericidal mechanism of Ti(IV) drugs isothermal titration calorimetric and kinetic experiments were performed to investigate the interaction between Ti(IV) and human transferrin. Extensive work was done with Ti(IV) citrate. ITC data revealed that at blood plasma conditions (pH 7.4, $[HCO_3^-]$ = 27 mM) Ti(IV) has a greater affinity (log K = 27.0 and 25.9) than Fe(III) (log K = 22.5 and 21.4) for the C and N binding sites of transferrin. The result implies that Ti_2-Transferrin would effectively block Fe(III) from entering cancer cells, which are known to have a larger population of transferrin receptors at the plasma membrane. Kinetics with Ti(IV) citrate, a very water soluble and stable complex, show a multiple step metal binding process. The complex delivers the metal to transferrin at slower rates than Cp_2TiCl_2. This suggests that more promising Ti(IV) drug leads would be ones that display comparable solubility and stability to Ti(IV) citrate but deliver the metal at rates comparable to Cp_2TiCl_2.

1. Christodoulou, C.V.; Eliopoulos, A.G.; Young, L.S.; Hodgkins, L.; Ferry, D.R.; Kerr, D.J. *Br. J. Cancer* **1998**, *77*, 2088-2097.

2. Guo, M.; Sun, H.; McArdle, H.J.; Gambling, L.; Sadler, P.J. *Biochemistry* **2000**, *39*, 10023-10033.