STRONG NON-COVALENT BINDING OF HYCANTHONE TO DNA

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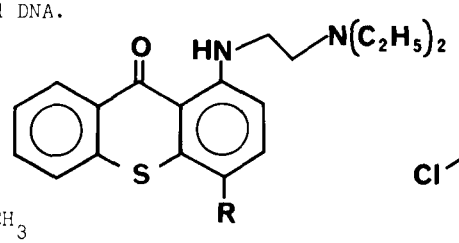
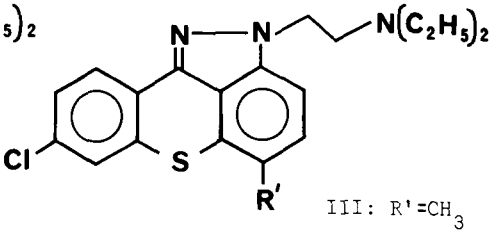
Hycanthone, a drug used in the treatment of schistosomiasis (bilharzia) is mutagenic, carcinogenic and teratogenic (for a review, see Hartman & Hulbert, 1975). Presumably the molecular origin of such undesired genetotoxic effects involves interaction of the drug with DNA. To understand this interaction, and to further our drug-design studies aimed at finding safer antischistosomal compounds (Hulbert, Bueding & Hartman, 1974), we are examining the non-covalent reversible binding of hycanthone and its congeners to DNA.

We report here that lucanthone (I), hycanthone (II) and the benzothiopyranoindazole analogues IA-3(III) and IA-4(IV) bind with very high affinity to double-stranded DNA.



I: R=CH_3

II: R=CH_2OH



III: R'=CH_3

IV: R'=CH_2OH

Addition of low drug concentrations to DNA stabilises the DNA against heat denaturation. The increase in DNA melting temperature measured at a number of drug concentrations allows the dissociation constant (K_d) of the drug-DNA complex to be determined. Different but complimentary experiments, in which changes in the u.v. spectrum of the drug are measured as DNA is added stepwise, also give values of K_d. The table shows values from these latter spectroscopic experiments, carried out in MOPS buffer (10 mM) containing NaCl (100 mM) at pH 7.2 and 20°: 'n' is the number of moles drug bound per mole nucleotide (calf thymus DNA).

Compound	K_d (Molar)	n
I	4.8×10^{-8}	0.14
II	1.8×10^{-8}	0.09
III	1.6×10^{-8}	0.45
IV	8.1×10^{-8}	0.15

No binding is seen when denatured DNA is used (at 95°), and this indicates that binding is by intercalation. Simple monocyclic compounds of the mirisan series show only very weak affinity for DNA.

When taken with the observation that hycanthone is an alkylating agent (Miller & Hulbert, 1976), these results are consistent with the suggestion that chemical carcinogens have the chemical characteristics expected of compounds acting as affinity labels for DNA (Hulbert, 1975).

Hartman, P.E. & Hulbert, P.B. (1975). J. Toxic. Environ. Hlth., 1, 243-270.

Hulbert, P.B. (1975), Nature (London), 256, 146-148.

Hulbert, P.B., Bueding, E. & Hartman, P.E. (1974), Science, 186, 647-648.

Miller, J.L. & Hulbert, P.B. (1976), J. Pharm. Pharmacol., 28, Suppl., p. 18.