UPTAKE, ELIMINATION AND SUBCELLULAR DISTRIBUTION OF DAUNORUBICIN IN TRYPANOSOMA RHODESIENSE

J.E. Brown, L.H. Patterson, J. Williamson[1], J.R. Brown[2].
Department of Pharmaceutical Chemistry, School of Pharmacy, Leicester Polytechnic, LE1 9BH, [1]Division of Parasitology, National Institute for Medical Research, London NW7 1AA, [2]Department of Pharmaceutical Chemistry, Faculty of Pharmaceutical Sciences, Sunderland Polytechnic, Sunderland, SR1 3SD.

Trypanosomes are a major source of parasitic diseases in Africa and Central and South America. The clinically useful antileukaemic agent daunorubicin is the most potent trypanocide so far tested *in vitro* but shows complete lack of *in vivo* activity (Williamson and Scott-Finnigan 1978). However, administration of daunorubicin i.p. in mice infected with *T. rhodesiense* has shown that the concentration of drug in plasma is greater than that necessary for *in vitro* trypanocidal activity (Brown et al 1982). Since the nucleus is suggested as the major site of the cytotoxic action of daunorubicin (Brown 1978), we have investigated the uptake, elimination and subcellular distribution of daunorubicin in *T. rhodesiense* to establish whether or not this drug reaches the nucleus.

In all the experiments described, daunorubicin was quantitatively determined by high performance liquid chromatography with fluorimetric detection (Brown et al 1981). *In vitro* uptake of daunorubicin into trypanosomes was shown to increase with time reaching a maximum concentration of drug after 2h. A complementary loss of daunorubicin from the incubation medium was observed. Elimination of daunorubicin from *T. rhodesiense* was investigated by incubating the drug with trypanosomes for 2h, harvesting the drug-loaded trypanosomes and incubating them in fresh medium in the presence and absence of 2-deoxy-D-glucose, a metabolic inhibitor. Daunorubicin content of trypanosomes was generally shown to be greater in the presence of the inhibitor than when it was absent. These results show that daunorubicin is taken into and released from *T. rhodesiense* and suggest a contribution by active transport to the elimination of daunorubicin from trypanosomes.

Subcellular distribution of daunorubicin in trypanosomes was investigated in *T. rhodesiense* isolated from mice that had been i.p. dosed 1h previously with 15mg kg^{-1} daunorubicin. The trypanosomes were subfractionated by ultracentrifugation. Daunorubicin was found in all fractions prepared, typically in the following ratios relative to cytosol: nucleus, 8.75: mitochondrion plus kinetoplast, 3.81: microsomes, 2.44: cytosol, 1 - so showing that the major site of intracellular accumulation of daunorubicin *in vivo* in trypanosomes is the nucleus. Moreover, these results are consistent with the principle of drug-nuclear DNA association, a phenomenon which is believed to be involved in the cytotoxic action of daunorubicin (Henry 1976).

Brown, J.E. et al. (1981) J. Chromatogr. 226 : 521 - 525
Brown, J.E. et al. (1982) J. Pharm. Pharmacol. 31 : 236 - 239
Brown, J.R. (1978) Prog. Med. Chem. 15 : 125 - 164
Henry, D.W. (1976) 'Cancer Chemotherapy' (Ed. A.C. Sartorelli), American Chemical Society, Washington, 15 - 57
Williamson, J., Scott-Finnigan, T.J. (1978) Antimic. Agents. Chemother. 69 : 1 - 2

0022-3573/82/120042P-01$02.50/0