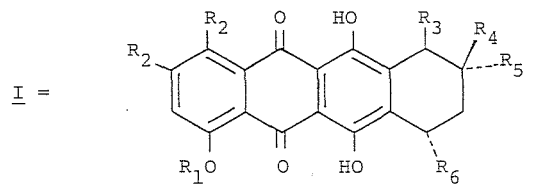
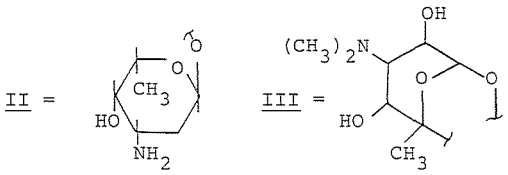
02. X–14 CRYSTAL STRUCTURE ANALYSES OF ANTHRACYCLINES: A CONTRIBUTION TO UNDERSTANDING THEIR STRUCTURE-ACTIVITY RELATIONSHIPS AT THE MOLECULAR LEVEL. Emil Eckle and John J. Stezowski, Institut für Organische Chemie, Biochemie und Isotopenforschung der Universität Stuttgart, D-7000 Stuttgart 80, FRG.

The anthracyclines are a family of organic compounds of general formula I and analogs thereof. Two members, daunomycin, Ia, and adriamycin, Ib, have found extensive clinical use as cancer chemotherapeutic agents. Both produce serious side effects including cumulative cardiotoxicity. Consequently considerable effort has been and is being invested toward the discovery of new analogs that are either active chemotherapeutic agents at much lower dose or are much less cardiotoxic.





Selected Anthracycline Derivatives

	R_1	R_2	R_3	R_4	R_5	R_6
Ia	CH_3	H	H	$COCH_3$	OH	II
Ib	CH_3	H	H	$COCH_2OH$	OH	II
Ic	CH_3	H	H	H	OH	II
Id	H	III	H	OH	H	H

The mechanisms of activity (chemotherapeutic and cardiotoxic) are topics of active current debate. The role that X-ray crystallography will play in helping to understand the mechanisms and in establishing structure-activity relationships at the molecular level remains to be determined.

This report presents the crystal structure determinations for two very different anthracyclines: 9-deacetyldaunomycin, Ic (as the hydrochloride), and 7-deoxynogarol, Id (free base). Their respective space groups and lattice parameters are: $P2_1$, Z=4, a=23.809(3), b=4.879(1), c=25.831(3) β=97.23$\bar{1}$(9) and $C222_1$, $\bar{Z}$=16, a=24.28$\bar{2}$(6), b=9.11$\bar{0}$(2), c=53.00$\bar{7}$(10). The associated R-values are 0.0$\bar{7}$3 (760 parameters, 3407 data) and 0.109 (775 parameters, 4622 data). Both structures contain disordered solvent.

A comparison of these structures with others determined in our laboratory and from the literature will be presented.

02. 1–1 CRYSTALLOGRAPHIC STUDIES OF THE CALMODULIN/TRIFLUOPERAZINE COMPLEX. By L.T.J. Delbaere, L.M.B. Gehrig, and R.A. Hickie. Departments of Biochemistry and Pharmacology, University of Saskatchewan, Saskatoon, Saskatchewan, Canada S7N 0W0.

Calmodulin is a ubiquitous calcium-binding protein which is a key intracellular calcium receptor mediating calcium-regulated enzymes and processes in eukaryotic cells, including those affecting cell replication and proliferation. Mounting evidence suggests that calmodulin levels are abnormally elevated in neoplastic cells, contributing to the unregulated and relatively autonomous growth of cancer cells. Recently, anticalmodulin agents such as trifluoperazine (TFP) have been shown to inhibit the growth of neoplastic cells in a selective dose-dependent manner. Bovine brain calmodulin consists of 148 amino acid residues in a single polypeptide chain with four calcium-binding domains. Anticalmodulin drugs bind to the active form of calmodulin (i.e. the calcium/calmodulin complex), interfering with the activation of calmodulin-dependent enzymes and cellular processes. In the presence of calcium, TFP is proposed to bind to two high affinity sites (K_d=1–1.5μM), with one site located between calcium binding domains II and III and the other site distal to domain IV. The molecular weight of the calcium-calmodulin-TFP complex is estimated to be about 17,500. The complex has been crystallized from 26% PEG 4000 at pH 5.2 with 10mM calcium chloride and 10mM magnesium chloride at 14°C. Characterization of the crystals has provided the following information: space group $P3_121$ or $P3_221$, a=b 40.88Å, c=180.9Å, gamma=120°, Z=6 and a solvent content of about 51% for the crystals. The three dimensional structure of the calmodulin/trifluoperazine complex will provide the tertiary structure of calmodulin itself and the interactions with an inhibitor that inactivates this very interesting protein. (Supported by the Medical Research Council of Canada and the Saskatchewan Cancer Foundation.)

02. 1–2 THE STRUCTURE OF PEA LECTIN AT THREE ANGSTROM RESOLUTION. By Howard Einspahr, K. Suguna, C. E. Bugg, and F. L. Suddath, Department of Biochemistry, Institute of Dental Research and Comprehensive Cancer Center, University of Alabama in Birmingham, University Station, Birmingham, Alabama 35294, U.S.A.

Pea lectin crystallizes in space group $P2_12_12_1$ with a = 50.73(2), b = 61.16(2), and c = 136.59(8) Å . The asymmetric unit contains a dimer of 49,000 daltons, and includes one Mn(II) and one Ca(II) per monomer. The amino-acid sequences of pea lectin and concanavalin A (Con A), the lectin from the jack bean, are very similar, but appear to be circularly permuted, that is, the NH_2- and COOH-termini of pea lectin correspond to a region near the middle of the Con A sequence, and vice versa. The structure of Con A has been refined with data to 1.75 Å (1). Low resolution studies (2, 3) of pea lectin suggest that pea lectin and Con A are also structurally similar. This suggestion is confirmed by comparison of models constructed with the aid of high resolution data. An atomic model of pea lectin has been built with a 3 Å electron density map based on diffractometer data collected to 3 Å from native and derivative crystals and to 6 Å from a PCMBS derivative crystal. Pea lectin shows the same construction of three beta-sheet features as found in Con A, and the architecture of the metal-binding regions in the two lectins is very similar. Differences in the two structures are primarily confined to loop regions. Details of similarities and differences in the two lectin structures will be presented.

This research was supported by grants DE-02670, CA-13148, DE-00106, and GM-30551 from the National Institutes of Health.

(1) Hardman, et al. (1982) J. Mol. Biol. 157, 69.

(2) Meehan, et al. (1982) J. Biol. Chem. 257, 13278.

(3) Riskulov, et al. (1984) F.E.B.S. Letters 165, 97.