04-Crystallography of Biological Small Molecules


**PS-04.01.18 STRUCTURE AND CONFORMATION STUDIES OF CRYSTALLINE ANTIBIOTICS: CRYSTAL STRUCTURE AND ABSOLUTE CONFIGURATION OF KANAMYCIN A**

Kanamycin A, an antibiotic complex produced by Streptomyces kanamyceticus from the Japanese soil, was isolated by Oishi, Umezawa and their coworkers as early as 1957. The Kanamycin antibiotic family consists of three components, namely Kanamycin A (the major component), Kanamycin B and C. The disadvantages of Kanamycin A are 4-0-(6-methyl-6-deoxy-d-glucopyranosyl)-6-O-(3-amino-3-deoxy-a-D-glucopyranosyl)-2-deoxystreptamine inhibits the growth of many kinds of bacteria and is widely used in the treatment of gonorrhoea, salmonea, tuberculosis and many other diseases. Kanamycin A consists of a central decamethylene ring linked to two glucopyranosyl rings at the 4 and 6 positions.

The absolute configuration of Kanamycin A is very important in the understanding of its mode of action. An extensive study of the absolute configuration was carried out using chemical methods. As part of our ongoing project on accurate structural investigations of crystalline oligosaccharides of biological importance, we carried out a structural analysis of Kanamycin A from State College, University of California, CA, using CAD-4 diffractometer data. These crystals obtained from water-grown bacteria with x = 7.2254 (3), y = 12.4922 (15), z = 7.1168 (5) Å, α = 94.74 (1), β = 91.16 (1), γ = 51.89 (1)°, V = 640.2 (2) Å³, w(H1) = 18 cm, F.W. 600-6, D = 1.588 g/cm³. The structure was obtained using SHELXS-86 and refined by full-matrix least squares to a final R value of 0.042. Both the D-glucose moieties are attached to the decamethylene ring by linkages. This absolute configuration confirms the earlier determination using optical and photochemical data. The (-)-values for the biphenyl linkages are -101.9, -112.2, -131.5 and -140.4 respectively.

Most aminoglycoside antibiotics including streptomycin and kanamycin do not only inhibit protein synthesis but also cause the amplification of the genetic code. It interacts with the ribosomal 16S protein to stabilize the ribosomal conformation binding between RNA and aminoglycosides and inhibits the elongation of the peptide chain through a series of reactions resulting in prevention of ribosomes from moving along DNA. The specific details of which step of the ribosome cycle, kanamycin blocks the protein synthesis are not yet fully understood. Our attempts to grow crystals of Kanamycin B and C are in progress.

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**Kanamycin antibiotics** (bulldogs)

Kanamycin A: 6
Kanamycin B: 6
Kanamycin C: 6

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**PS-04.01.19 STRUCTURAL AND FUNCTIONAL STUDIES OF TILORONE: A NOUVEAU CRYSTAL STRUCTURE AND CONFORMATION OF 2,7-BIS-(P-PYRIDINYL)PYRIMIDINE ANTIFUNGAL AGENT IN DIFFERENT PRODUCTION STATE**

Tilorone is an orally active pharmacologically agent that protects mice against infections with DNA and RNA viruses. It is also the only known small molecule that is an interferon inducer. It has anti-inflammatory and anti-inflammatory activities and was shown to affect and influence the immune mechanism in humans and animals. Tilorone has been postulated to act by intercalating preferentially between A-T base pairs (Chen, J. Med. Chem. 1986, 29, 1369). Structural and functional studies of these compounds have been undertaken in our laboratories with an objective to correlate the structural features with the immunosuppressive activity. Crystals of the title compound are triclinic, space group P1, with a = 7.058 (2), b = 9.035 (4), c = 23.615 (3) Å, α = 153.91 (2)°, β = 129.5 (2), γ = 93.94 (4)°, V = 1731.9 (4) Å³, D = 1.27 g/cm³, Z = 12 (2). The structure was solved with CAD-4 data (2450 reflections, 1873 > 0.16, using SHELX-86 program and refined to a final R value of 0.063. The propoxy group is in an extended zig-zag conformation and is co-planar with the central fluorine atom. The two terminal propoxy rings are in the preferred chair conformation. The crystal structure lends support to the postulated intercalative mechanism of action for Tilorone. Attempts are being made to crystallize the other Tilorone analogues as well as a complex of Tilorone fragment and DNA, which could afford a detailed interpretation of the intercalation mechanism.

We hypothesize that the stereochemistry and the molecular geometry of the drugs and/or their active metabolite affect both the receptor interaction and the mode of immunosuppression. Lymphocyte proliferation studies were conducted on mice (Cunningham et al., Immunobiol., 1984, 53, 91). The drugs were studied in vitro to assess their effect on immunological function of murine lymphocytes, focusing on cell proliferation assays and on the response to IL-2 and expression of IL-2 receptors. Our preliminary studies showed that Tilorone and its analogs stimulate the IL-2 response in high responder strains (e.g. C3H/HeJ) with a dose of 0.5 mg/kg/day. The studies are ongoing. The findings allow analogs to be tested in vivo to establish their potential for structure-function relationships.

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**PS-04.01.20 CONFORMATION OF 4-SUBSTITUTED 2,6-BIS-(PYRIDINYL)pyrimidines: ANTI-INFECTIVE AGENTS IN DIFFERENT PRODUCTION STATE**

Tilorone is a pyrimidine derivative used in various anti-infective treatments. It is a potent inhibitor of viral replication, and has been shown to have antiviral properties against a wide range of viruses, including herpes simplex virus (HSV), varicella-zoster virus (VZV), and human immunodeficiency virus (HIV).

Our studies on the structure-activity relationships of these compounds have shown that the conformation of the pyrimidine ring is important for their antiviral activity. The crystal structure of 4-substituted 2,6-bis(pyridinyl)pyrimidines was determined using X-ray diffraction techniques, and the conformation of the pyrimidine ring was found to be strongly influenced by the substituent group on the nitrogen atom. These findings suggest that the conformation of the pyrimidine ring is a key factor in determining the antiviral activity of these compounds.