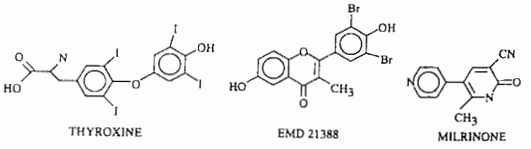
one, its desmethyl 5-amino parent compound, are members of a new class of oral nonglycosidic cardiac positive inotropic agents developed for the treatment of congestive heart failure. Structure activity correlations reveal that milrinone, but not amrinone, stimulates rabbit myocardial Ca^{2+}-ATPase activity as does thyroxine. To further develop this correlation, competive binding studies were carried out which showed that in a similar manner, milrinone, but not amrinone, was a strong competitor for T_4 binding to TTR. Comparison of a series of bipyridine structures revealed homology between the phenolic ring of T_4 and the substituted ring of the bipyridine and a model which defined the conformational features required for activity was developed. The crystal structure of the milrinone-TTR complex was carried out and these data confirmed the model. Structure activity data for a membrane bound T_3 transporter showed that benzodiazepine derivatives were potent inhibitors of T_3 transport. Conformational comparison of their structures revealed homology with T_3 and resulted in a model which incorporates key features of the benzodiazepine subclasses. Thus, these analyses revealed that TTR can be used as a prototypic model to explain the relative potency of these enzyme inhibitors. Based on these results, inhibitors can be designed with selective actions at their respective target sites.



THYROXINE EMD 21388 MILRINONE

Supported in part by DK 494001 and Boots Pharmaceuticals.

MS–04.01.05 Structural Determination of Natural Products. By Angela Y. Lee and Jon Clardy, Department of Chemistry, Cornell University, Ithaca, N.Y. 14853-1301, USA.

Natural products have been a wonderful source of chemically novel and biologically active molecules, but they have also been a severe technical challenge to crystallographers. Most of our research has involved characterizing a variety of biologically active compounds, and more recently, their complexes with cellular receptors. The ultimate goal is to understand not only the structure but also the biochemical function of these compounds. We have worked with marine peptides for many years, and recently have become interested in a novel cyclic peptide from a marine sponge called cyclotheonamide. We never worked on this structure directly because of the inability to grow crystals. Cyclotheonamide is a serine protease inhibitor with K_I ~0.2 nM with trypsin and ~1.0 nM with thrombin. The three-dimensional structure of cyclotheonamide A complexed with bovine β-trypsin has been successfully determined at 2.3 Å resolution. This reveals not only the structure of cyclotheonamide but also its mode of action. The strong electron density and low thermal parameters allowed the conformation of bound cyclotheonamide A to be determined unambiguously—a result that has also clarified the stereochemistry.

Key words: natural product, novel protease inhibitor, mechanistic studies.

MS–04.01.06 POLYMORPHYSM AND BIOINEQUIVALENCE OF 6-METHYLURACIL. By N.B.Leonidov*, S.I.Uspenskaya, Institute "Bioeffect" of Ministry of Science, High School and Technical Policy of Russian Federation. Moscow, Russia; P.M.Zorky & A.E.Masunov, Chemical Department, Moscow State University, Russia.

New perspective trend in drug design is obtaining polymorphic modifications with different molecular conformations. Under consideration has been influence of polymorphism of drugs on their biological activity. Examples of influence of conformational polymorphism of drugs on changes in its biological characteristics in solutions are given. One of the explanations of this phenomenon is the difference between molecular agglomerates which are present in the crystals. The cooperative effect of molecular packing can fasten the details of molecular conformation. After dissolution of a substance such agglomarates or some fragments of them can occur in the solution for relatively long period of time. Specifically two crystal forms of 6-methyluracil differ in the antioxydative and wound-healing effect of their solutions.

X-ray data show that the cyclic dimers formed by a pair of NH...O=C bonds occur in both of them. However, in the form I the dimers are uniteed by single H-bonds and wavy layers arise as a result. In the form II the dimers are joined into ribbons using pairs of H-bonds. It is very likely that hydrated dimers predominate in the solution of the crystals I, but rather long fragments of ribbons occur in the solution of the crystal II. Slightly different molecular conformations are to exist in the agglomerates of these two types. Thus, we obtain a probable explanation of the biological inequivalence of two polymorphs.

MS–04.01.07 A STUDY ON STRUCTURE-ACTIVITY RELATIONSHIPS IN 16- AND 17-SUBSTITUTED ESTRANES AND ANDROSTANES. By S. Stankovic*[1], D. Miljkovic[1], R. Kovacevic[1], D. Lazar[1], Lj. Medic-Mijacevic[2], V. Pejanovic[2] and Ch. Courseille[3], [1]Faculty of Sciences, University of Novi Sad, Trg D. Obradovica 4, 21000 Novi Sad, Yugoslavia, [2]ICN Galenika Institute, 29. novembra 111, 11000 Beograd, Yugoslavia, [3]Laboratoire de Cristallographie et de Phisique Cristalline, Faculté de Scienes, Université de Bordeaux I, Talance, France.

A series of synthetic estranes (1-5) and androstanes (6-9) have been prepared and tested for anti-estrogenic and anti-androgenic properties. These steroids have been subjected to X-ray structural analysis to permit structure-activity relationship studies.

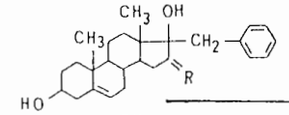
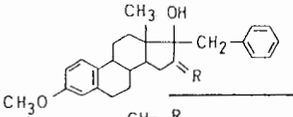
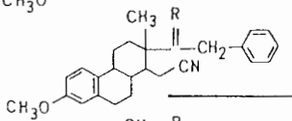
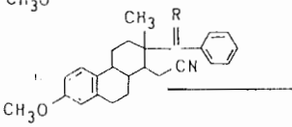
In the first series (1-5) a novel chemical rearrangement during acid-catalized hydrolysis of 16-oximino to 16-carbonyl group has been noticed. Namely, the benzyl moiety at C-17 changes its orientation from α- to β-position. The new β-orientation of the benzyl moiety affects the chemical and biological properties of the C-17 centre.

Among the compounds from the first type (1-3) an expected phenomenon was noticed: β-orientation of the C-17 hydroxyl group produced higher biological activity. The compounds from the second type (7-9) showed differences in biological activity. Namely, the benzyl-derivative 7 possesses an anti-estrogenic activity, while the corresponding phenyl analogue 9 showed no activity at all. The phenyl analogue 8, with 17-OH group, showed in vitro an inhibitory effect on steroidogenesis.

04-Crystallography of Biological Small Molecules

126

Molecular mechanic calculations were performed to provide and to determine minimum energy conformations. Computer aided modeling of the new structures with extended 17-side-chains has been attempted.

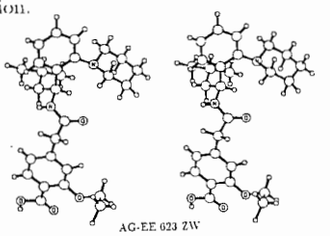
	COMPOUND	R	S.G.	Z	R
	1	<H H	$P2_12_12_1$	8	0.051
	2	=NOH	$P2_12_12_1$	4	0.058
	3	=O	$P2_1$	2	0.071
	4	=NOH	P1	2	0.069
	5	=O	$P2_12_12_1$	4	0.065
	6	=O	$P2_12_12_1$	4	0.048
	7	<H OH	$P2_12_12_1$	4	0.069
	8	=O	$P2_1$	2	0.047
	9	<H OH	$P2_1$	2	0.041

MS-0 .0 .08 X-RAY ANALYSIS OF THE NEW HYPOGLYCEM DRUG REPAGLINIDE: COMMON STRUCTURAL FEA1 RES WITH GLIBENCLAMIDE. By W. Grell, Department of Chemical Research, and M. Mark, Department of Biochemical Research, Dr. K. Thomae GmbH. Biberach, and P. Luger*, Institute for Crystallography, Freie Universität Berlin. Germany.

Repaglinide, (+)-2-ethoxy-α-[[(S)-α-isobutyl-o- piperidinobenzyl]carbamoyl]-p-toluic acid (1), AG-EE 623 ZW, (WHO Drug Inform. 1992, 6(3), List 32), being the most active representative of a series of novel hypoglycemic (B-cytotropic) benzoic acid derivatives, was found to display a 20 times higher blood sugar lowering activity than the sulfonylurea (s.u.) compound glibenclamide (2) in fasted rats. With the aim of obtaining more insight into common or different conformational aspects, the X-ray structure of (1) and of several related compounds was determined. The structure of (1) was computergraphically compared with that of (2) (Byrn,S.R.,McKenzie, A.T., Hassan,M.M.A. & Al-Badr,A.A., J. Pharm. Sci. 1986, 75, 596-600). The superposition was performed in such a way that functional groups which are known to contribute essentially to activity, and which are regarded as being involved in binding to the so called "s.u. receptor" (Kaubisch,N., Hammer,R.,Wollheim,C.,Renold, A.E. & Offord,R.E., Bioch. Pharmacol. 1982, 31, 1171-1174) fit best after having slightly rotated only a few bonds of each of the X-ray structures.

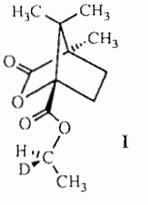
It was found that the acidic (-COOH / -SO2-NH-) and basic (piperidino -N / methoxy-O) functional groups, and the amino hydrogens (-H) are overlapping quite well. In contrast the amido oxo (=O) groups do not, but are yet in positions for enabling hydrogen bonds to the same (presumed) electron accepting binding site of the "s.u. receptor".

It is concluded that the conformations being realized in the superposition are compatible with a common three-point binding model. This, as an extension of the binding hypotheses discussed formerly (Rufer,C. & Losert,W., J. Med. Chem. 1979, 22, 750-752; Brown,G.R. & Foubister,A.J., J. Med. Chem. 1984, 27, 79-81), means that the three main functional groups [the acidic, the basic, and the (ambivalent) amido groups] bind simultaneously to corresponding binding sites of the "s.u. receptor". Additional lipophilic interactions are supposed to occur so that the particular groups of (1) [the (S)-positioned isobutyl group, the (whole) piperidino group, and the ethoxy group], and (2) [the cyclohexyl group], respectively, are binding into (presumed) different corresponding pockets of a given "s.u. receptor". This supports the speculation that "there exists more than one specific binding site for sulfonyl ureas and benzoic acid derivatives" (Verspohl, E.J.; Ammon, H.P.T. & Mark, M., J. Pharm. Pharmocol. 1990, 42, 230-235). Possibly, even the existence of different (sub)types of "s.u. receptors" has to be taken into consideration. - Further investigations are necessary before coming to a decision between the various kinds of binding, and getting conclusive information about the "active conformations" of structurally different stimulators of insulin secretion.



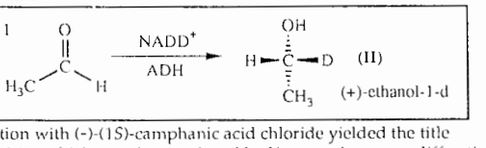
OPS-04.01.09 ABSOLUTE CONFIGURATION OF THE (-)-(1S)-CAMPHANATE ESTER OF (+)-ETHANOL-1-d BY NEUTRON DIFFRACTION by Robert Bau*, Tobias Metzenthin, Department of Chemistry, University of Southern California, Los Angeles, CA 90089, U.S.A., Thomas F. Koetzle, Department of Chemistry, Brookhaven National Laboratory, Upton, NY 11973, U.S.A., and Harry S. Mosher, Department of Chemistry, Stanford University, Stanford, CA 94305, U.S.A.

The absolute configuration of the (-)-(1*S*) camphanate ester (I) of (+)-ethanol-1-*d* has been determined by single-crystal neutron diffraction. (+)-Ethanol-1-*d* (II) was prepared (Scheme 1) via the reduction of acetaldehyde by the enzyme alcohol dehydrogenase (ADH) in the presence of deuterated nicotinamide adenine dinucleotide ($NADD^+$) by the method of Simon[1]:



I

Scheme 1



Esterification with (-)-(1S)-camphanic acid chloride yielded the title compound (I), which was then analyzed by X-ray and neutron diffraction. The neutron diffraction analysis of I, using the known[2] absolute configuration of the (-)-(1S) camphanate group as a reference, showed that the absolute configuration of the chiral CHD group is *R*.

We had earlier analyzed[3] the absolute configuration of (+)-neopentanol-1-*d* (III), prepared by the reduction of deuterated neopentanal by actively fermenting yeast[4] (presumably also involving alcohol dehydrogenase; Scheme 2) and converted to its phthalate half-ester (V):

Scheme 2