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XIX CONFERENCE ON ISOPRENOIDS

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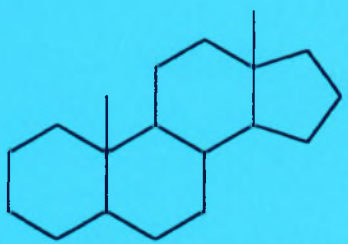

BOOK OF ABSTRACTS

Gdańsk-Jurata, Poland, 8-14 September, 2001

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LIST OF PLENARY LECTURES

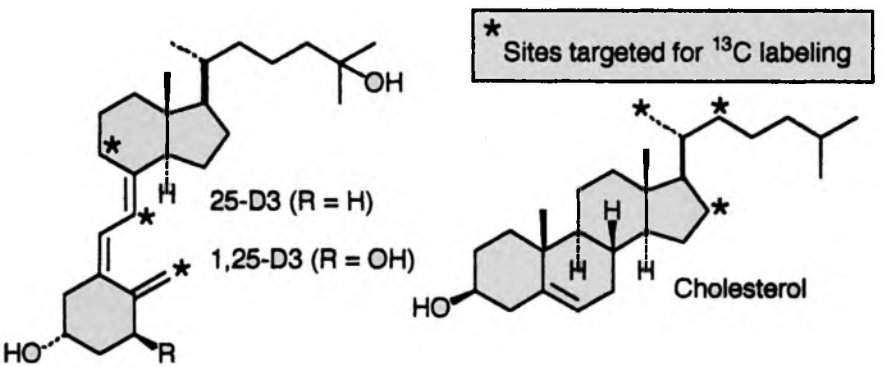
L1	Okamura W. H.	Conformational Probing of Steroids Bound to Proteins: Analogs and Isotopomers of Vitamin D and Cholesterol
L2	Rohmer M.	Isoprenoid Biosynthesis via the Methylerythriol Phosphate Pathway: on the Independent Biosynthesis of Isopentenyl Diphosphate and Dimethylallyl Diphosphate in Higher Plants
L3	Tietze L. F.	Domino and Multiple Heck Reactions for the Synthesis of Monoterpene Alkaloids and Steroids
L4	Kocienski P.	Asymmetric Synthesis Using Planar Chiral π-Allylmolybdenum Complexes
L5	Prestwich G. D.	Proteins that Bind Isoprenoids: Targeting Cell Signaling and Microbial Pathogens
L6	de Groot A.	Clerodanes from Carvone
L7	Rychnovsky S. D.	New Methods for the Synthesis of Highly Oxygenated Natural Products
L8	Kitahara T.	Stereocontrolled Synthesis of an Anticoccidal Sesquiterpene and a Neuroprotecting Amino Acid
L9	LaCour T. G.	Gestalt Effects in Anticancer Steroids
L10	Morzycki J. W.	A New Approach to the Synthesis of a Highly Potent Antitumor Natural Product OSW-1
L11	Fallis A. G.	Taxing Targets: Cycloaddition Routes to Taxoids
L12	Appendino G.	Synthesis of Conformationally Constrained and Conformationally Relaxed Analogues of Taxol. Discovery of Taxoids with Unique Biological Properties
L13	Mori K.	Recent Results in the Synthesis of Bioactive Natural Products
L14	Welzel P.	Forskolin Studies
L15	Macek T.	Approaches in Studies of Brassinosteroide Biological Activity
L16	Tkachev A. V.	New Strategy for Designing Chiral Auxiliary from Natural Terpenes
L17	Neef G.	Carvone as a Starting Material for Achieving Regio- and Stereoselective Intramolecular Attack upon a Quinol bis-Epoxide Moiety

L18	**Mikami K.**	**Design & Asymmetric Catalytic Synthesis of Singly Dehydroxylate 19-nor-1,25$(OH)_2D_3$ Hybrid Analogs for Cancer Cell Differentiatio & Apoptosis**
L19	**Cieplak A. S.**	**Origin of Stereoelectronic Control of Diastereoselection in Nucleophilic Additions to Carbonyls**
L20	**Lee H.-Y.**	**Free Radical Rearrangement Mediated Cyclization Reactions in Total Syntheses of Isoprenoids**
L21	**Daniewski W. M.**	**N-Acylphenylisoserinates of Sesquiterpenoid Alcohols of Lactarius Origin. Synthesis and Biological Properties**
L22	**Connolly J. D.**	**Natural Products from Around the World**

PLENARY LECTURES

CONFORMATIONAL PROBING OF STEROIDS BOUND TO PROTEINS: ANALOGS AND ISOTOPOMERS OF VITAMIN D AND CHOLESTEROL. W. H. Okamura. Department of Chemistry, University of California, Riverside, CA 92521.

Steroids and other small, lipophilic biomolecules (guests) generally mediate their biological actions via association with host molecules or host systems, for example proteins, membranes etc. Drug developments in the vitamin D field have continued to focus on structure-function studies by systematically modifying 1α,25-dihydroxyvitamin D_3 (1,25-D3) or its biosynthetic precursor, 25-hydroxyvitamin D_3 (25-D3). Direct structural information gleaned from crystallographic or NMR studies regarding the ligand-receptor complex and other guest-host systems, also offers potential insight into drug design. Evidence has accrued suggesting that topologically different conformers of 1,25-D3 may bind to proteins in different ways, including the induction of different conformations of protein. This paper will concern our progress on the chemical synthesis of analogs (e.g., provitamins, previtamins, suprasterols, vinylallenes and other analogs) conformationally locked (or at least highly restricted) to mimic higher energy conformers of 25-D3 and 1,25-D3. The synthesis of ^{13}C labeled vitamin D metabolites including initial progress on solution and solid state ligand-NMR applications of labeled steroid bound to host molecules/systems will be described. Progress on studies of D-ring and side chain labeled cholesterol will also be described. Acknowledgement is made to various collaborators (Drs. A. W. Norman, L. J. Mueller, D. C. Borchardt, R. Bouillon, H. v. Baelen) and co-workers as well as to the NIH & NSF and Solvay, Hoffmann-LaRoche, Leo and Cambridge Isotopes.



ISOPRENOID BIOSYNTHESIS VIA THE METHYLERYTHRITOL PHOSPHATE PATHWAY: ON THE INDEPENDENT BIOSYNTHESIS OF ISOPENTENYL DIPHOSPHATE AND DIMETHYLALLYL DIPHOSPHATE IN HIGHER PLANTS

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In plant cells, two pathways are involved in the biosynthesis of isoprenoids: the mevalonic acid (MVA) pathway localized in the cytoplasm and the 2-*C*-methyl-D-erythritol 4-phosphate (MEP) **2b** pathway that occurs in the plastids. First evidence for a branching in the MEP pathway has been found in the bacterium *Escherichia coli*: IPP **3** and DMAPP **4** are separately synthesized from an unidentified MEP derivative (Fig. 1).[1] This feature was pointed out by the incorporation of [4-^{2}H]deoxyxylulose (DX) **1a**, the first C_5 intermediate of the MEP pathway, or of free [3,5,5,5-$^2H_4$]methylerythritol **2a** into the prenyl chains of ubiquinone and menaquinone from *E. coli*. The isoprene units derived from DMAPP quantitatively retained the deuterium, whereas those derived from IPP were characterized by complete deuterium loss. In contrast, no evidence for such a branching was obtained in plant cells after feeding a *Catharanthus roseus* cell culture with [2-^{13}C, 4-^{2}H]DX, which resulted in equivalent ^{13}C labeling and ^{2}H loss for all isoprene units derived from IPP as well as from DMAPP.[2]

Tobacco (*Nicotiana tabacum* L.) cv Bright Yellow-2 (TBY-2) cell cultures proved a powerful biological system for investigation of isoprenoid biosynthesis in plant cells.[3] Feeding TBY-2 cells with [4-^{2}H]DX resulted in the expected labeling of phytoene and the prenyl chain of plastoquinone in the plastids. Significant deuterium retention (ca. 10%) was observed on all carbon atoms corresponding to C2 of IPP and DMAPP in all isoprene units derived from IPP or from DMAPP. This represented the signature for a possible branching in the MEP pathway in plant cells.

IPP and DMAPP have a dual origin in TBY-2 cells. On the one hand, the MEP pathway produces via the branching, as in *E. coli*, DMAPP with deuterium retention from [4-^{2}H]DX and IPP with deuterium loss. On the other hand, a second IPP pool with deuterium retention derives from the isomerisation of the former [^{2}H]DMAPP via the IPP isomerase, and an additional unlabeled DMAPP pool from the isomerization of unlabeled IPP, assuming that the IPP isomerase and the prenyl transferase from plants possess the same enantioselectivity as all known corresponding enzymes from other organisms. The latter IPP and DMAPP pools resulting from an interconversion via the IPP isomerase were not observed in *E. coli*. In this bacterium, the IPP isomerase gene, although present, is not essential,[4] and the corresponding enzyme activity is negligible, preventing thus the interconversion of DMAPP and IPP.

Incorporation of [4-^{2}H]DX into the plastidic isoprenoids of TBY-2 cells was in accordance with the results from studies on the biosynthesis of the monoterpene cineol in eucalyptus twigs, but with a lower DX incorporation yield and no clear labeling of the IPP derived isoprene units.[5]

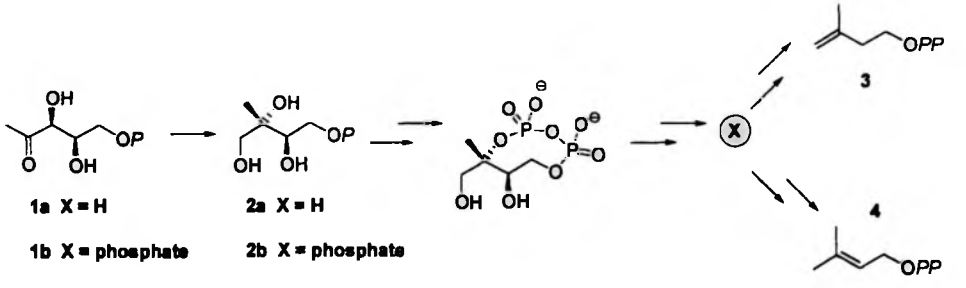

Figure 1. Branching towards IPP **3** and DMAPP **4** in the MEP pathway for isoprenoid biosynthesis

Selected references

1. J.L. Giner *et al.*, *Chem. Commun.* 1857 (1998); L. Charon *et al.*, *Biochem. J.* **346**, 737 (2000); M. Rodriguez-Concepcion *et al.*, *FEBS Lett.* **473**, 328 (2000).
2. D. Arigoni *et al.*, *Proc. Natl. Acad. Sci. USA* **96**, 1309 (1999).
3. A. Disch *et al.*, *Biochem. J.* **331**, 615 (1998).
4. F.H. Hahn *et al.*, *J. Bacteriol.* **181**, 4499 (1999).
5. C. Rieder *et al.*, *Helv. Chim. Acta* **83**, 2504 (2000).

DOMINO AND MULTIPLE HECK REACTIONS FOR THE SYNTHESIS OF MONOTERPENE ALKALOIDS AND STEROIDS

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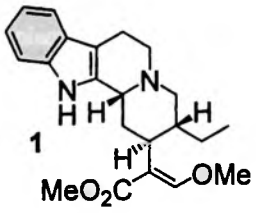
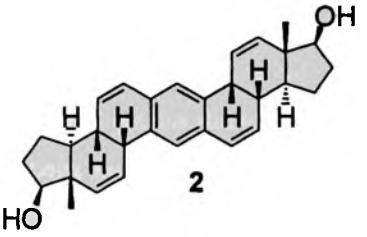
Synthesis of relevant organic compounds such as natural products and analogues, drugs, diagnostics, agrochemicals and any kind of material is a main topic in academic and industrial chemistry. Whereas the race for more selectivity was one of the driving forces in the past - it is still going on - the main goal is now efficiency, the compatibility with our environment, the preservation of our resources and also the economical advantage. This new view is clearly a change of paradigm in synthesis. The proportion of the numbers of steps and the increase of complexity is now an important standard for the quality of a synthesis. Multi-step syntheses with much more than 20 steps are clearly not state of the art since they are neither economically nor ecologically justifiable. In addition, the use of toxic reagents and solvents should be avoided, the amount of waste produced in a process must be reduced and finally one must deal carefully with our resources and our time.

A general way to improve synthetic efficiency and also address the mentioned criteria as well as give access to a multitude of diversified molecules is the development of domino processes and multiple transition metal catalyzed transformations. This methodology allows the formation of complex compounds starting from simple substrates in very few steps. We have defined domino reactions as processes of two or more bond forming transformations under widely identical conditions in which the subsequent reactions take place at the functionalities obtained in the

former transformation and have classified these processes according to the mechanism of the single steps. Domino processes are also suitability in combinatorial chemistry.

In my lecture I shall describe the use of domino processes for the efficient synthesis of monoterpenoid indol alkaloids such as hirsutine **1** which is highly active against the influenza A virus and of simple analogues **2** of the bissteroid cephalostatin as well as of other isoprenoids.





[1] L. F. Tietze, Chem. Rev. 1996, 96, 115–136. [2] L. F. Tietze, F. Haunert in „Stimulating Concepts in Chemistry" (Eds. : M. Shibasaki, J. F. Stoddart and F. Vögtle) Wiley VCH, Weinheim 2000, 39-64. [3] L. F. Tietze, Y. Zhou, Angew. Chem. 1999, 111, 2076–2078, Angew. Chem. Int. Ed. 1999, 38, 2045–2047.

Asymmetric Synthesis Using Planar Chiral π-Allylmolybdenum Complexes

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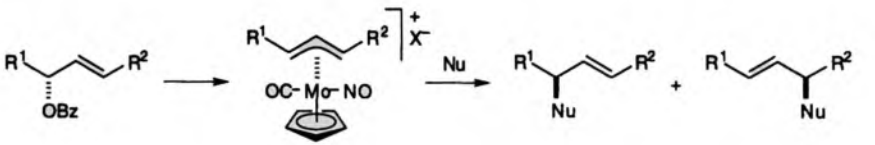
Planar chiral π-allylmolybdenum cationic complexes have been known for more than 20 years but they have been seldom used in organic synthesis. The lecture will focus on the background and recent results from our laboratory in the following areas:

1. New methods for the synthesis of planar chiral π-allylmolybdenum complexes.

2. Factors that influence the stereo- and regiochemistry of their reaction with carbon nucleophiles.

3. Applications of asymmetric allylation to the synthesis of biologically active natural products including the polyether antibiotics salinomycin and ionomycin, the marine antiinflammatory agent pseudopterosin G, and the antitumour agent cryptophycin 4.



Proteins that Bind Isoprenoids: Targeting Cell Signaling and Microbial Pathogens

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Abstract

Much of the understanding the proteins involved in assembly and processing of isoprenoids, as well as those important in the formation and recognition of prenylated proteins has been facilitated with affinity labels. In this presentation, we will explore several topics: (1) cyclization of squalene and oxidosqualene, (2) probes for understanding prenyltransferases, and (3) discovery of a protein important in regulation of polyprenol-mediated apoptosis.

First,, we describe research on determination of the active site of oxidosqualene and squalene cyclases from vertebrate, protozoal, and bacterial sources. These studies employ site-directed mutagenesis, product analysis and peptide mapping with photoaffinity labels to determine important residues for product determination. A library of inhibitors has been prepared and used to identify a OSC inhibitors selective for enzymes from pathogens compared with the vertebrate enzymes.

Second, we designed and stereoselectively synthesized a novel fluorescent probe, (2*E*,6*E*,8*E*,10*E*,12*E*,14*E*)-geranylgeraniol (all *trans*- ΔΔGGOH) and its diphosphate and cysteine methyl ester derivatives. The 8*E* alkene was introduced by Wittig-Horner reaction (> 95% *E*), and the 12*E* double bond was installed using a stereoselective Wittig reaction involving an allylic triethylphosphorane reagent (>99% *E*). The fluorescence spectrum (ΔΔGGOH 10 μM in MeOH) showed λ_{ex} = 310 nm, λ_{em} = 410 nm, quantum yield (QY) = 0.0075, and was quenched fourfold by addition of one equivalent of water. The evaluation of this probe in protein binding and enzymatic recognition will be presented.

Finally, we used photoaffinity labeling techniques to identify a unique isoprenoid binding protein capable of protecting cells from apoptosis. This ubiquitous 50 kDa serum protein has a strong binding affinity for isoprenoids with three or more isoprene units. Initially, the isoprenoid binding was identified using a photoactivatable juvenile hormone I analog. . Both human and bovine forms were shown to be strongly labeled by this analog, and purification and sequencing of the native protein permitted identification, which was subsequently confirmed with commercially available materials. This serum protein also bound the isoprenoid geranylgeraniol (GGOH) and a photoactivatable analog. Because GGOH can induce apoptosis, we used HL-60 cells to test the effects of this serum protein on protection of cells from GGOH-induced apoptosis. Protection was observed in a narrow range, i.e., at approximately 1:1 stoichiometry of protein to ligand. The implications of these observations, the identification of the protein, and the potential of this system for identification of agents that can selectively manipulation of apoptosis will be discussed.

CLERODANES FROM CARVONE

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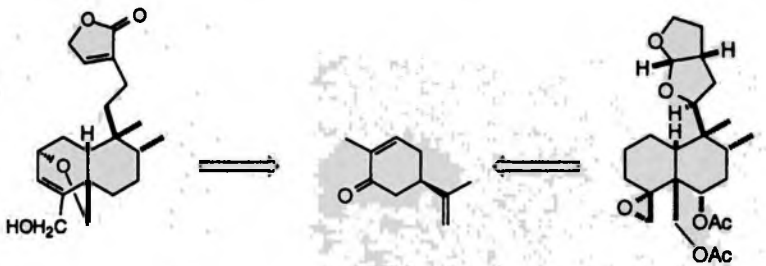
Conjugate addition followed by a Mukayiama reaction in R-(-) carvone leads to highly substituted cyclohexanones that have been studied in several types of annulation reactions. Annulation with MVK and some derivatives leads to highly substituted chiral decalins that can be transformed into clerodanes like brevifloralactone. Other annulations lead to intermediates that are suitable for the enantioselective total synthesis of the insect antifeedant dihydroclerodin.



brevifloralactone R-(-)-carvone dihydroclerodin

NEW METHODS FOR THE SYNTHESIS OF HIGHLY OXYGENATED NATURAL PRODUCTS

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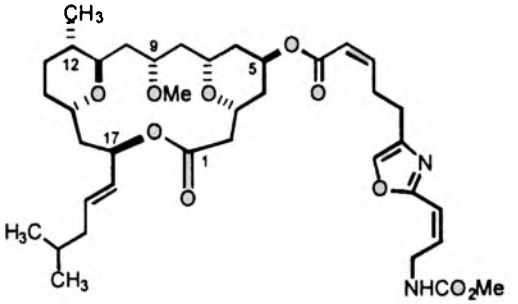
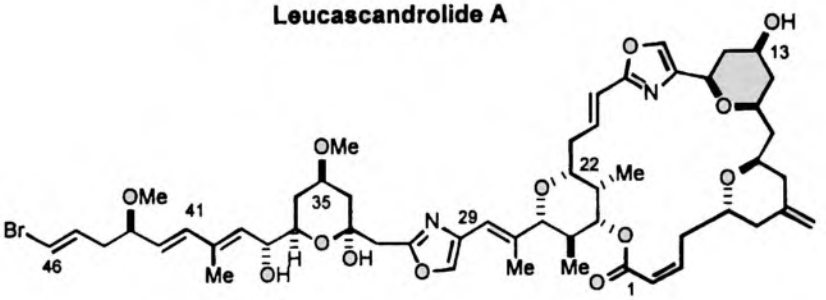
My lab has developed a reductive acetylation reaction of esters that is very useful for preparing α-acetoxy ethers. These α-acetoxy ethers are ideal precursors to oxocarbenium ions. We are investigating the reaction of oxocarbenium ions generated in this manner to prepare new carbon-carbon bonds. I will describe results of our Prins cyclization studies and the development of a new Aldol-Prins tandem reaction. Synthetic approaches to phorboxazole B and to leucascandrolide A will be used to illustrate these new methods.



Leucascandrolide A



Phorboxazole B

STEREOCONTROLLED SYNTHESIS OF AN ANTICOCCIDAL SESQUITERPENE AND A NEUROPROTECTING AMINO ACID

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Stereocontrolled synthesis of the following natural products, Fudecalone **1** and Kaitocephalin **2**, with remarkable biological activities will be presented.

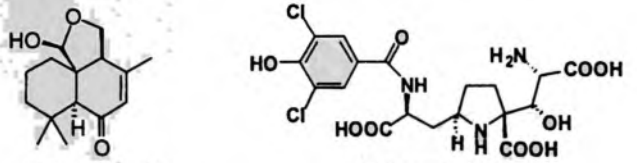

Fig. 1 Structures of Bioactive Heterocycles

Fudecalone, a drimane sesquitepene, was isolated from *Penicillium* sp. FO-2030 as a potent anticoccidal substance.[1] The proposed structure was *cis*-octalone **3** with a non-steroidal conformer as shown in Fig. 2. We succeeded in the synthesis of the proposed structure unambiguously through the retrosynthetic scheme (Fig. 2) and revised the structure to be *trans*-octalone **1**.[2] Details of the story with biological activity will be discussed precisely.

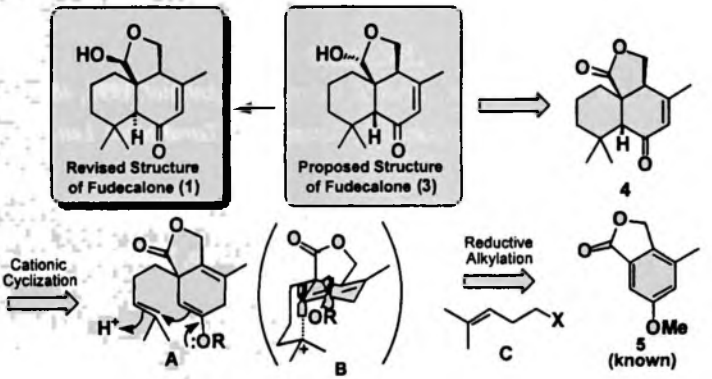

Fig. 2 Retrosynthesis of Fudecalone

Kaitocephalin **2** is an unique amino acid isolated from *Eupenicillium sheari* PF1191 by Seto and co-workers and contains neuroprotecting activity to suppress Glutamate toxicity as an antagonist against the binding of Glutamate to AMPA-KA receptor.[3)] The unique structure and significant activity prompted us to investigate not only the synthesis of itself but of related analogs to understand structure-activity relationship. Through the retrosynthetic scheme shown in Fig. 3, we already obtained the whole skeleton of the proposed structure, but it was not identical, and therefore, we are now investigating the stereochemical assignments carefully. Details will be presented clearly in the lecture.

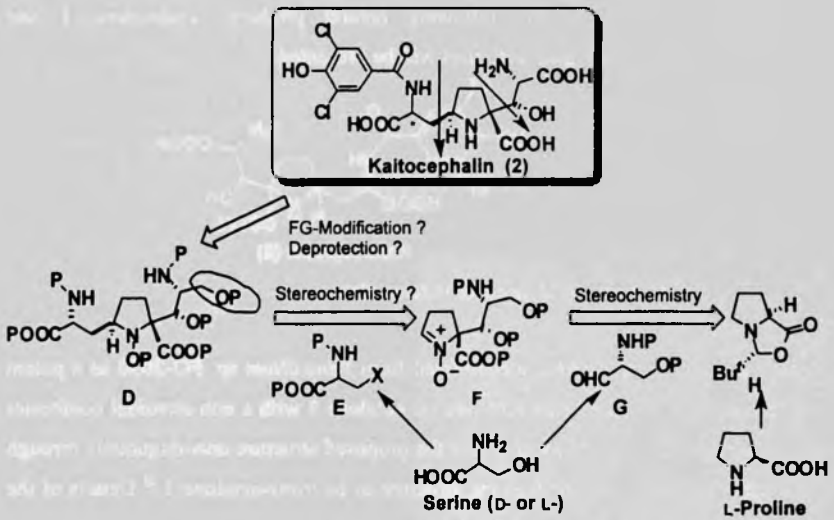

Fig. 3 Retrosynthesis of Kaitocephalin

References

□□ Tabata, N.; Tomoda, H.; Masuma, R.; Iwai, Y.; Omura, S. *J. Antibiot.* **1995**, *48*, 53-58.

□□ Watanabe, H.; Yamaguchi, T.; Furuuchi, T.; Kitahara, T. *Tetrahedron Lett.* **2001**, *42*, 917. *Tetrahedron* **2001**, *57*, in press.

□□ a)Shin-ya, K.; Kim, J.; Furihata, K.; Hayakawa, Y.; Seto, H. *Tetrahedron Lett.* **1997**, *38*, 7079. b) Kobayashi, H.; Shin-ya, K.; Furihata, K.; Hayakawa, Y.; Seto, H. *Tetrahedron Lett.* in press.

GESTALT EFFECTS IN ANTICANCER STEROIDS

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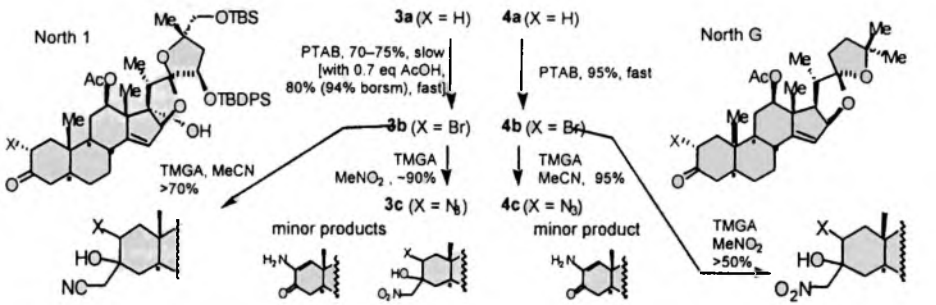
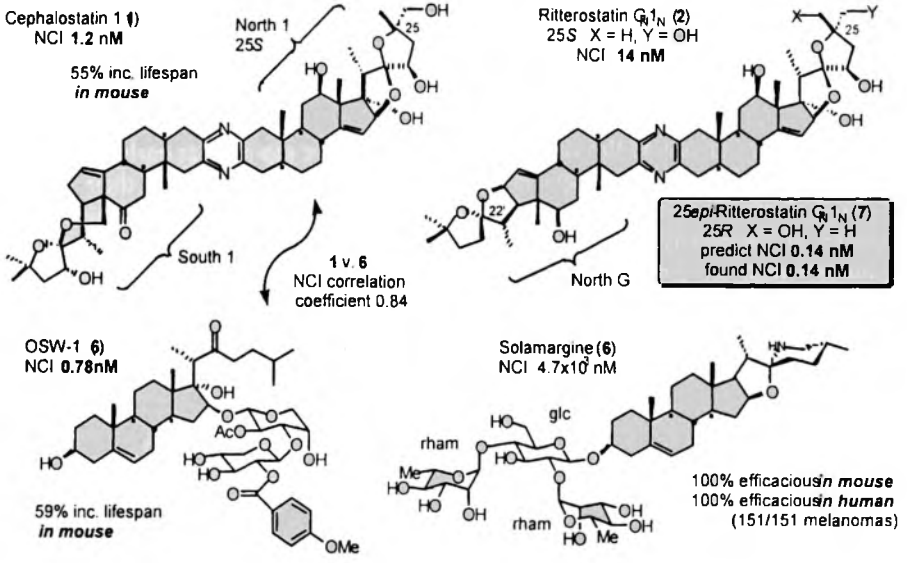
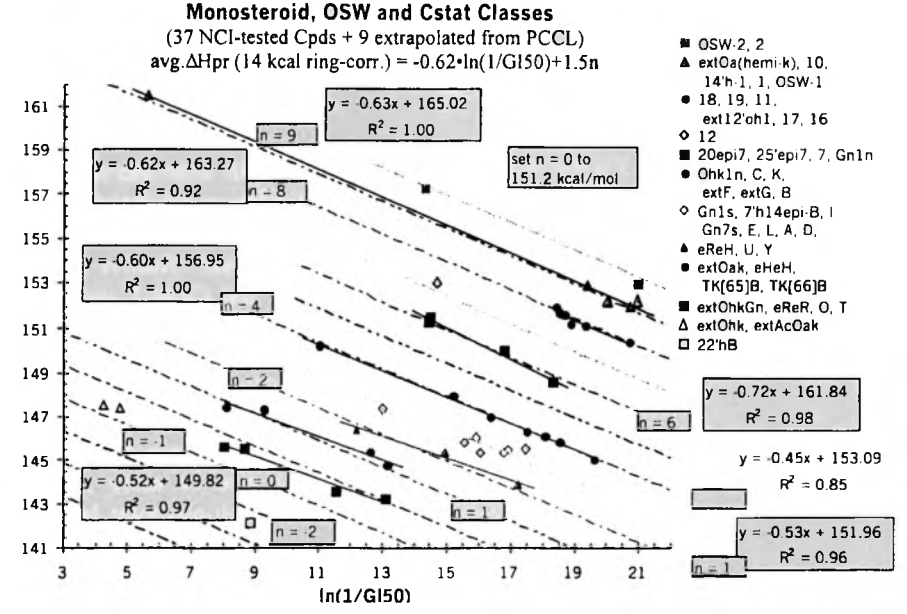
Long-range effects in the chemistry of steroids were reported as early as 1955, and investigations by several groups in the last decade have established their ability to transmit electronic and reactivity information through up to 12 bonds. We have found a number of chemically and biologically interesting differences due to distal structural features which we term the "gestalt effect" in antineoplastic steroids. For instance, during our syntheses of extremely cytotoxic natural cephalostatin 1 (**1**) and analogue **2**, large discrepancies in A-ring reactivity became evident for the North 1 (**3**) subunit v. that for North G (**4**), which differ only in their E/F ring patterns.



These marine bissteroidal pyrazines and "related" glycosidic monosteroids such as terrestrial plant-derived OSW-1 (**5**) and solamargine (**6**) display no functionality typical of antitumor compounds (alkylating sites such as Michael acceptors, radical sources such as quinones, etc.), and yet are among the most potent and effective antitumor compounds ever tested. Interestingly, both cephalostatins and OSW-1 show highly correlated NCI "fingerprint" differential cytotoxicities which are suggestive of a common and possibly novel mechanism of action, and we have found that both are potent inducers of apoptosis with nearly identical behavior. The emerging chemical and biological SAR indicates that all of the most potent steroid-based antitumor agents feature homoallylic oxygen and spiroketal sites, which are sources for stabilized carbenium ions. Further, only conjugated steroids with two or more such pro-carbenium ion sites display high activity, suggesting that multiple-point binding is critical.

We have developed an extensive and precise correlation (R = 0.98) between cytotoxicity and the calculated thermodynamic energy cost to access such ions, which is rendered periodic by the location and extent of polar functionalities (see graph). The utility of the computational method has been verified against oxacarbenium ion mediated reactions of steroid spiroketals, by structure correction of published cephalostatins based on their bioactivity, and by the correct prediction of a new unnatural cephalostatin, 25-*epi*-ritterostatin G_N1_N (**7**), as the most potent member of the family ever tested at the NCI.





It seems that such ions may play a generally unrecognized alkylative or oxidative role in the binding and/or covalent modification of their biological targets. "Unrelated" antineoplastics such as the halichondrins likewise may utilize carbenium ions as active principles.

A NEW APPROACH TO THE SYNTHESIS OF A HIGHLY POTENT ANTITUMOR NATURAL PRODUCT OSW-1

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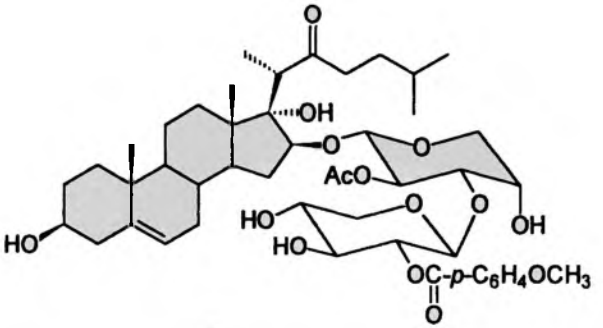
The steroidal saponin OSW-1 belongs to a family of cholestane glycosides that was recently isolated from the bulbs of *Ornithogalum saundersiae*, a perenial grown in southern Africa.[1] Due to its extraordinary antitumor activity,[2] OSW-1 is an attractive synthetic target. During the last two years three different methods of the OSW-1 synthesis (or its aglycone) were reported.[3-5]



OSW-1

Our new strategy for the synthesis of this compound will be presented. The crucial steps were cleavage of the 16α,17α-epoxides with lithium hydroperoxide and glycosylation of the steroid aglycone in its hemiacetal form with disaccharide imidate.

References:

1. S. Kubo, Y. Mimaki, M. Terao, Y. Sashida, T. Nikaido, T. Ohmoto, *Phytochemistry* **31**, 3969 (1992).
2. Y. Mimaki, M. Kuroda, A. Kameyama, Y. Sashida, T. Hirano, K. Oka, R. Maekawa, T. Wada, K. Sugita, J. A. Beutler, *Bioorganic Med. Chem. Lett.* **31**, 633 (1997).
3. C. Guo, P. L. Fuchs, *Tetrahedron Lett.* **39**, 1099 (1998).
4. S. Deng, B. Yu,Y. Lou, Y. Hui, *J. Org. Chem.* **64**, 202 (1999).
5. W. Yu, Z. Jin, *J. Am. Chem. Soc.* **123**, 3369 (2001).

Taxing Targets: Cycloaddition Routes to Taxoids

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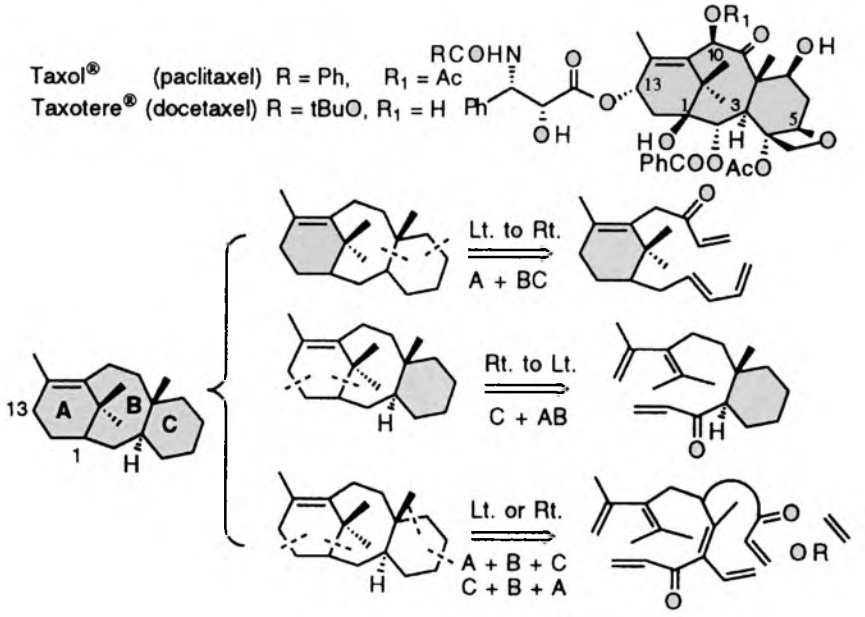
Taxol® (paclitaxel) and Taxotere® (docetaxel) are established therapeutic agents for treating various cancers, particularly ovarian and breast cancer. These drugs operate by a novel mechanism of action that differs from other spindle poisons such as vincristine. Nature has provided us with a novel structure that represents a new chemotherapeutic lead. Thus, as previously demonstrated by other systems (penicillins, tetracyclines, etc.), we should be able to modify these structures to provide new understanding of the mode of action and novel compounds with therapeutic potential. The ultimate objective of this research is to develop a direct, versatile synthesis of new taxoids with a better chemotherapeutic profile and fewer side effects.



Our ongoing investigations to develop general intramolecular cycloaddition strategies to the taxane nucleus will be described. These include the development of tether control groups, magnesium mediated carbometallations and pentadienyl indium reagents.

Related References: Fallis *et. al. Acc. Chem. Research* , **32**, (6), 464-474, (**1999**); *Can. J. Chem.* **77**, (2) 159-177 (**1999**); *Org. Lett.* **2**, (18) 2793-2796, (**2000**). *Synthesis* (7), 921-924, (**2000**).

SYNTHESIS OF CONFORMATIONALLY CONSTRAINED AND COFORMATIONALLY RELAXED ANALOGUES OF TAXOL. DISCOVERY OF TAXOIDS WITH UNIQUE BIOLOGICAL PROPERTIES

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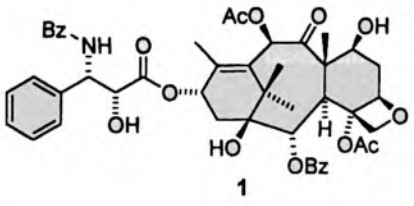
Over the past fifteen years, no natural product has received such a worldwide and multidisciplinary attention as taxol (**1**). The bleak prospects in terms of availability and way of administration which challenged the early investigators were solved in the early 90ties, paving the way to the successful commercialization of the semisynthetic version of the natural product. In just a few years, **1**, under the formulated tradename of Taxol, became the most successful anticancer drug ever, with yearly sells well over the billion dollar level. Within the chemical arena, the focus of taxoid research has gradually moved from the total synthesis of the natural product to the discovery of second-generation analogues which, compared to taxol and taxotere, have more powerful and/or broader activity, a better profile of side-effects, and an easier way of administration. These properties are exemplified by IDN 5109 (**2**), an orally active taxoid active on MDR tumors and currently undergoing clinical development. Furthermore, non-oncological applications of taxoids were also discovered, with clinically useful activity against diseases whose pharmacological treatment is, at present, unsatisfactory (Alzheimer's disease, psoriasis) or non-existing (multiple sclerosis).



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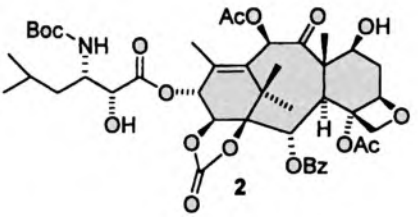

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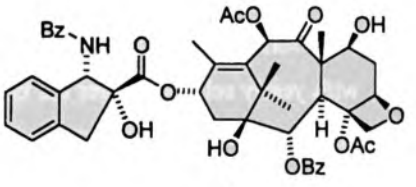
These findings suggest that the pharmacological potential of taxoids is still underexploited, and have served as an inspiration to studies aimed at the discovery of new chemotypes. In this context, we have explored the possibility of obtaining new taxoid leads by suitable carbon-carbon modification of molecular fragments not involved in binding. The conformational hallmark of the natural product is the presence

of a rigid terpenoid core bound to flexible acyl moieties, and modifications of the carbon-carbon framework are expected to change it in a dramatic way.

Results obtained restricting the conformational mobility of the 13-aminoacyl side chain by covalent tethering, and relaxing the taxane core by fragmentation of rings A and C will be presented. These transformations are exemplified by compounds **3-5**. Compounds **3**, **4** and their analogues did not show substantial improvement of activity compared to the taxol, while certain derivatives of **5** were highly active, with a profile of biological activity substantially deviating from that of all known tubulin binders.



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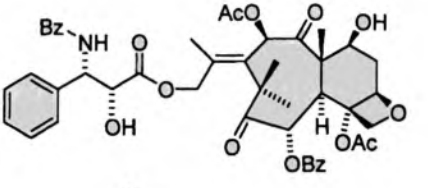

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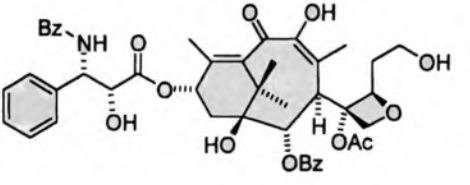

5

During the synthesis of these compounds, several unexpected rearrangements of the diterpenoid core were discovered, and will be discussed along with their possible mechanistic rationale.

RECENT RESULTS IN THE SYNTHESIS OF BIOACTIVE NATURAL PRODUCTS

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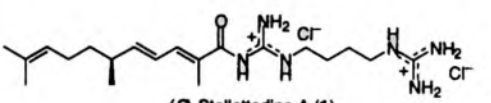
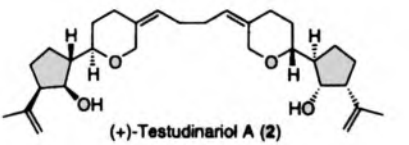
Synthesis of the following marine natural products will be discussed with special emphasis on the determination of their absolute stereochemistry.



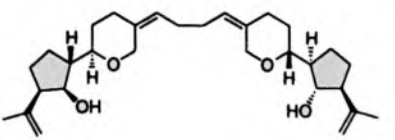
(*S*)-Stellettadine A (1)

Inducer of metamorphosis for ascidian *Halocynthia roretzi*[1]

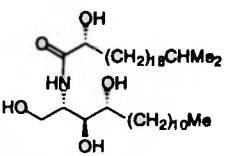


(+)-Testudinariol A (2)

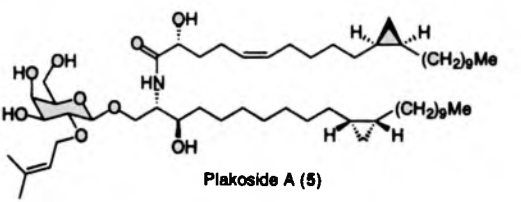
Ichthyotoxic metabolite[2]



(+)-Testudinariol B (3)[2]



Female sex pheromone of the hair crab *Erimacrus isenbeckii* (4)[3]



Plakoside A (5)

Immunosuppressive metabolite[4]

1) H. Takikawa, D. Nozawa, K. Mori, *J. Chem. Soc., Perkin Trans. 1*, **2001**, 657–661.
2) M. Yoshida, H. Takikawa, K. Mori, *J. Chem. Soc., Perkin Trans. 1*, **2001**, 1007–1017.
3) Y. Masuda, M. Yoshida, K. Mori, to be submitted.
4) M. Seki, K. Mori, *Eur. J. Org. Chem.* in press. (*cf. Tetrahedron Lett.*, **2001**, *42*, 2357–2360)

FORSKOLIN STUDIES

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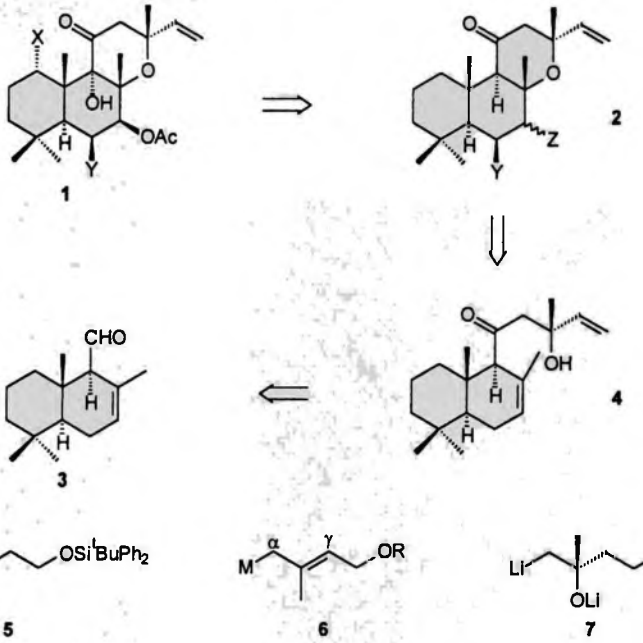
Forskolin (**1**, X = Y = OH) is a labdane diterpene that has been shown to interact with different membrane proteins including adenylyl cyclase.[1] The ability of forskolin to stimulate adenylyl cyclase in intact cells in the absence of hormonal agonists has been exploited by many laboratories for investigation of the role of cyclic AMP in various physiological functions. The mode of binding of forskolin to the adenylyl cyclase has been studied in detail[2,3] and two crystal structures have been published.[4,5] Structure-activity relationships on the basis of these results will be discussed as well as synthetic and mechanistic studies along the lines that follow from the retrosynthetic Scheme.[6]



1 For leading references, see Morris, D.I.; Greenberger, L.M.; Bruggemann, E.P.; Cardarelli, C.; Gottesman, M.M.; Pastan, I.; Seamon, K.B. *Mol. Pharmacol.* **1994**, *46*, 329-337.

2 Yan, S.-Z.; Hahn, D.; Huang, Z.-H.; Tang, W.-J. *J. Biol. Chem.* **1996**, *271*, 10941-10945.

3 Whisnant, R. E.; Gilman, A. G.; Dessauer, C. W. *Proc. Natl. Acad. Sci.* **1996**, *93*, 6621-6625, and references therein.

4 Zhang, G.; Liu, Y.; Ruoho, A. E.; Hurley, J. H. *Nature* **1997**, *386*, 247-253.

5 Tesmer, J. J. G.; Sunahara, R. K.; Gilman, A. G.; Sprang, S. R. *Science* **1997**, *278*, 1907-1916.

6 For leading references, see Behnke, D.; Hennig, L.; Findeisen, M.; Welzel, P.; Müller, D.; Thormann, M.; Hofmann, H.-J. *Tetrahedron* **2000**, *56*, 1081-1095.

APPROACHES IN STUDIES OF BRASSINOSTEROIDE BIOLOGICAL ACTIVITY

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Brassinosteroids (BS) are a group of plant steroids, of which the molecular and biochemical analysis of *Arabidopsis* mutants has furnished conclusive evidence that these compounds are plant growth hormones (1). They are biologically active in the various bioassay systems designed for gibberellins, auxins and cytokinins, eliciting remarkable growth responses. The molecular mechanism of BS action is uncertain, although one might argue from structural considerations that they are likely to work by a mechanism similar to that of animal steroid hormones which generally act *via* a soluble receptor-ligand complex that binds to nuclear sites to regulate the expression of specific genes. Despite many studies on plant steroids there is no report on successful isolation of a receptor.

Our aim was to prepare affinity chromatography carriers for protein receptor isolation from plant extracts (with special respect to *Arabidopsis thaliana*) through BS analogues bound covalently by a proper spacer arm. The ligand must be bound by that part of the molecule which least participates in the biospecific binding. As far as it is not yet known exactly, which parts of the BS molecule are actually necessary for the proper biological activity and specific binding, oriented immobilisation of different ligands to matrix was necessary. First attempts were through carboxyl group introduced into the side chain. Successful binding to MBHA carrier yielded 0.08 mg of steroid per gram of matrix. The use of N-succinimide did not give satisfactory results. GABA proved to be a satisfactory spacer arm for this purpose. Good yield gave agarose modified by adipic acid dihydrazide, too. The amino groups of the polyacrylamide-based carrier PEGA were occupied by BRST with 85% yield.

Among others newly synthesised BRSTs (20S)-2α, 3α-dihydroxy-7-oxa-B-homo-5α-pregnan-6-on-20-carboxylic acid was used for immobilisation. This compound was obtained in eight steps by general synthesis of brassinosteroid skeleton (2, 3) from bisnorcholanic acid, i.e. from 3ß-hydroxy-23,24-dinor-5-cholenic acid. Different newly synthesised BS derivatives (4) were used to obtain chromatography carriers with BS bound through the ring A, ring B or the side chain.

The plant extracts were obtained by grinding frozen plant leaves, salts were removed by gel filtration and the extract applied to the bioaffinity matrix. The proteins bound by non-specific sorption were eluted by the same buffer with increasing gradient of NaCl. After 2 N NaCl also acetic acid was used for elution. All samples were desalted on PD 10 columns, concentrated by lyophilisation and used for electrophoretic estimation of the proteins present.

The prepared new BS derivatives were tested for their biological activities using different tests, among others also their ability to suppress the stress response of tobacco plants (5).

ACKNOWLEDGEMENT: The work was sponsored by grant A4055803 of the Grant Agency of the Czech Academy of Sciences and research project Z4 055 905.

REFERENCES

1. Szekeres M., Nemeth K., Koncz-Kalman Zs., Mathur J., Kauschmann A., Altmann T.,Redei G.P., Nagy F., Schell J., Koncz Cs.: Cell, 85, 171 (1996).
2. Kohout L., Collect. Czech Chem. Comm. 59, 457 (1994).
3. Kohout L., Kasal A., Strnad M.: Collect. Czech. Chem. Commun. 61, 930 (1996).
4. Kohout L., Chodounská H., Macek T. and Strnad M.: Collect. Czech Chem. Comm. 65, 1-7 (2000).
5. Nesnerová P., Svatos A., Kohout L., Macek T.: Plant Physiol. and Biochem., 38, S 08-26, (2000).

NEW STRATEGY FOR DESIGNING CHIRAL AUXILIARY FROM NATURAL TERPENES

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Enantioselective synthesis seems to be one of the most dynamic disciplines of the modern chemistry. Asymmetric synthesis of organic molecules requires the use of chiral auxiliary (catalysts, ligands, modifiers, etc.), but not all sorts of chiral compounds are suitable, the majority of the reported natural and synthetic optically active compounds being ineffective as inductors of chirality. At the same time, it is just the pool of natural products that is the primary source of chirality for preparing all the other chiral chemicals. Among the different types of chiral natural products, terpenes have received the less study (apart from boron derivatives of pinene, natural oxygen-containing derivatives such as menthol, pulegone, carvone and camphor). In spite of extremely high enantiomeric purity of some terpenes and their accessibility, the lack of functional group makes them less attractive as chiral auxiliary. This is the true for natural terpenic hydrocarbons "as is", but what about their synthetic derivatives? Is there a possibility to transform wide spread terpenes to the effective chiral agents? What kind of chemical modification has to be performed to convert unfunctionalized terpenes to the chiral agents?

Addition of simple inorganic reagents with electrophilic nitrogen to unsaturated hydrocarbons is the most important transformation leading to a variety of useful nitrogen-containing derivatives. The detailed study of the addition to natural olefins of different types (C_{10}-acyclic, mono- and bicyclic compounds; C_{15}-mono- and bicyclic compounds with normal and medium-size rings; C_{20}-macrocycles) and study of structures, reactivity and chemical properties of the intermediates resulted in designing general synthetic scheme leading to key nitrogen-containing intermediates and then to different kinds of chiral building blocks and auxiliary.

CARVONE AS A STARTING MATERIAL FOR ACHIEVING REGIO- AND STEREOSELECTIVE INTRAMOLECULAR ATTACK UPON A QUINOL BIS-EPOXIDE MOIETY

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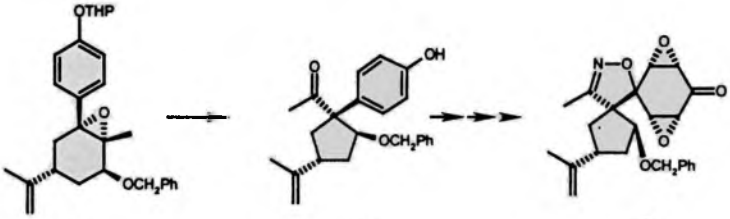
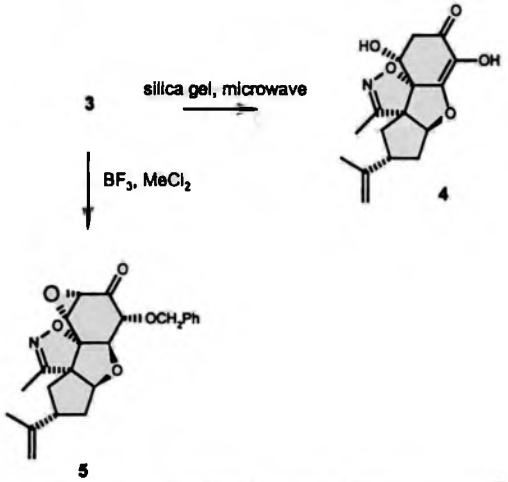
The highly stereoselective rearrangement of a carvone-derived epoxide[1] **1** opens up the possibility of a regio- and stereoselective attack upon a quinol bis-epoxide moiety, which is an essential pharmacophoric portion of several natural products and drug candidates[2].
The challenging situation of discriminating five adjacent electrophilic centers as presented by quinol bis-epoxides such as **3** has not often been dealt with. Previous experience[3] made it rather doubtful whether chemo- and stereocontrol could be achieved otherwise than by synthetic strategies circumventing the bis-epoxy ketone as a central intermediate.



1 **2** **3**

The creation of a chiral environment combined with the appropriate placement of a nucleophilic group made it possible to achieve selective, intramolecular reaction at a defined ß-epoxide position. Surprisingly, the initial attack was followed by a cascade of subsequent transformations leading to a single product **4** that was well prepared for further variation since all the centers of the quinol moiety had changed their functional characteristics in a favorable manner.



Evidence will be presented for an unusual proximity effect that must be interpreted as a direct interaction of a benzyl ether with an epoxide moiety (**3** → **5**). Favorable distance and trajectory for the benzyl ether oxygen enforced by the molecular shape and the high rigidity of educt **3** provoke a transformation where a benzyl ether oxygen can exhibit nucleophilic properties.

References:

1. Neef, G.; Baesler, S.; Depke, G.; Vierhufe H. Tetrahedron Lett *40*, 7969-7973, **1999**.
2. Neef, G.; Baesler, S.; Depke, G.; Vierhufe H. Tetrahedron Lett. *42*, 1011-1015, **2001**.
3. Wipf, P.; Jeger, P.; Kim, Y Bioorg. Med. Chem. Lett. *8*, 351-356, **1998**

DESIGN & ASYMMETRIC CATALYTIC SYNTHESIS OF SINGLY DEHYDROXYLATED 19-NOR-1,25(OH)$_2$D$_3$ HYBRID ANALOGS FOR CANCER CELL DIFFERENTIATION & APOPTOSIS[1]

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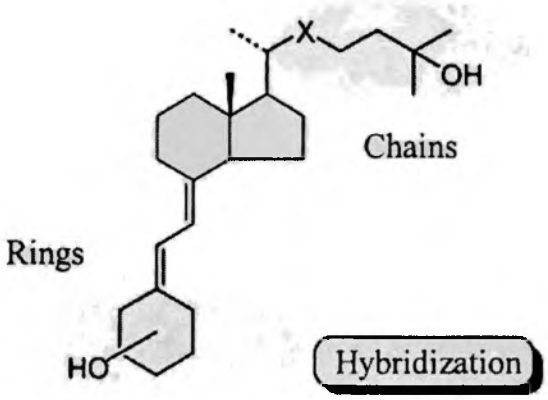
1α,25-Dihydroxyvitamin D$_3$ (1α,25(OH)$_2$D$_3$) has been shown to modulate not only proliferation and differentiation, but also apoptosis in malignant cells, indicating that it could be useful for treatment of cancer and psoriasis. However, little information has been available on the role of the exocyclic methylene group at the C-10 position, a hydroxy group at the C-1 and C-3 positions, and a hetero atom linkage of the side chain. Thus, we designed and synthesized a series of singly dehydroxylated 19-nor[2]-1,25(OH)$_2$D$_3$ hybrid analogs with or without a hetero atom linkage (*e.g.* 22-oxa), via a combinatorial sequence of highly regioselective[3] propiolate-ene reaction[4] and catalytic asymmetric carbonyl-ene cyclization[5]. Significantly, these brave new analogs can be clearly divided into two categories; one group, bearing natural 1α- or 3β-OH, are potent differentiators and the second, with unnatural 1β- or 3α-OH, are potent stimulators of apoptosis.



[1] Mikami, K., et al. *Tetrahedron Lett.* **39**, 3359-3362 (1998); *Biol. Pharm. Bull.* **21**, 1300-1305 (1998); *Synlett* 1899-1902 (1999) ; *Chem. & Biol.* **7**, 173-184 (2000); *Chirality: Nakanishi Issue*, **13**, 366-371 (2001).
[2] DeLuca, H. F., et al. *Tetrahedron Lett.* **31**, 1823-1824 (1990).
[3] Mikami, K., et al. *J. Org. Chem.* **56**, 2952-2953 (1991).
[4] Snider, B. B. *Acc. Chem. Res.* **13**, 426-432 (1989).
[5] Mikami, K., et al. *Chem. Rev.* **92**, 1021-1050 (1992); *J. Am. Chem. Soc.* **111**, 1940-1941 (1989); **112**, 3949-3954 (1990); *Org. Synth.* **71**, 14-21(1992); *J. Am. Chem. Soc.* **116**, 2812-2820 (1994).

ORIGIN OF STEREOELECTRONIC CONTROL OF DIASTEREOSELECTION IN NUCLEOPHILIC ADDITIONS TO CARBONYLS

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The problem of π-face selection was first defined in modern terms half a century ago. In 1952, diastereoselection in organometallic alkylations of chiral aldehydes was shown by Cram to follow a simple rule based on the premise that such reactions are controlled by steric strain. The question of stereoelectronic control of π-face selection was subsequently raised when the Cram rule was found to fail in the case of nucleophilic additions to cyclic ketones. Numerous hypotheses proposed to address this question focused either on the ground-state distortion of the C_{2v} symmetry of the C=O group (leading, for instance, to non-equivalent extension of its LUMO with respect to the carbonyl plane), or on the transition-state interactions (hyperconjugative, coulombic etc.) between the incipient bond and its immediate environment (e.g. vicinal σ bonds). The two hypotheses that presently seem to be most often discussed are the Felkin-Anh and Cieplak models. Both models primarily consider interactions of the incipient bond with the vicinal σ bonds but differ in the assessment of electron demand of the incipient bond, and consequently, in proposing which properties of the stereogenic-center ligands, beside their steric bulk, are important in π-face selection. The Felkin-Anh hypothesis assumes, in accord with the FMO theory, that the incipient bond is electron rich, and the transition state is stabilized by the *anti* oriented electron acceptors; in contrast, the Cieplak hypothesis assumes, in an apparent violation of the common FMO interpretation of nucleophilic addition to carbonyls, that the incipient bond is electron deficient, and the transition state is stabilized by the *anti* oriented electron donors.

To evaluate the two models, the issues of the electron demand at the reaction site and of the properties of the stereogenic center were examined by means of the ab initio calculations. Thus, energy changes, structural variation, and electron density shifts during additions of acetylide ions to cyclohexanone and cyclohexanethione were studied at the HF/6-31G* level. The atomic charge on the carbonyl C was found to become more positive upon approach of the nucleophile; the density deformation maps suggest that the charge polarization occurs to a large extent in the π bond. Since this effect is not compensated for by the charge transfer until in the late stage of the addition, the reaction site is considerably electron deficient (more so than the carbonyl C in the substrate) for most of the reaction path, and its interactions with the ligands are dominated by hyperconjugation with the vicinal C-H and C-C bonds as predicted by the Cieplak hypothesis. Relative importance of different vicinal interactions was also assessed by the natural bond orbital (NBO) analysis of the transition structures. In the framework of the NBO theory, the C-H and C-C hyperconjugation is indeed the major effect, significantly stronger than the alternative interaction proposed by the Anh hypothesis, in all transition structures examined.

To determine relative electron-donor abilities of the common stereogenic-center ligands X, the effect of suitable CH_2-X-substitution on the structure (bond distances), the NBO occupancies, and the energies of the bond-antibond interactions from the second order perturbation theory analysis of the Fock matrix in the NBO basis, were examined in

a series of bicyclic bridgehead cations at the B3LYP/6-31G* level. The obtained order of the σ bond capabilities as mesomeric electron donors is consistent with the previously postulated one: $\sigma_{C\text{-}F} < \sigma_{C\text{-}OCH3} < \sigma_{C\text{-}CH3} < \sigma_{C\text{-}Cl} < \sigma_{C\text{-}Br} < \sigma_{C\text{-}SCH3} \approx \sigma_{C\text{-}H}$. However, extended hyperconjugation in these cations involves not only the $\sigma_{C\text{-}X}$ bond donation into the electron deficient C-C bond ($\sigma_{C\text{-}X} \rightarrow \sigma^*_{C\text{-}C}$ hyperconjugation) but the π_X donation (X $2p_z \rightarrow \sigma^*_{C\text{-}C}$ homoconjugation) as well. Consequently, the order of the total charge transfer is: C-OCH_3 < C-F < C-CH_3 < C-H < C-Cl < C-Br ≈ C-SCH_3. This result suggests that the ranking used to correlate the effect of substituents on π-face selection with their electron-donor properties might depend on the reagent and the molecular embedding of the stereogenic center. Therefore, it is the order of the total charge transfer rather than the σ-bond electron-donor order that should in principle be used to predict the effect of variation in the structure of the stereogenic center on diastereoselectivity.

Free radical rearrangement mediated cyclization reactions in total syntheses of isoprenoids.

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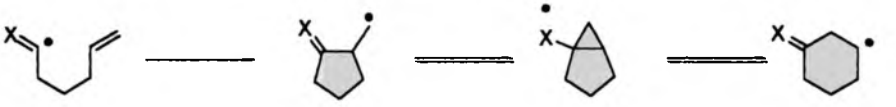
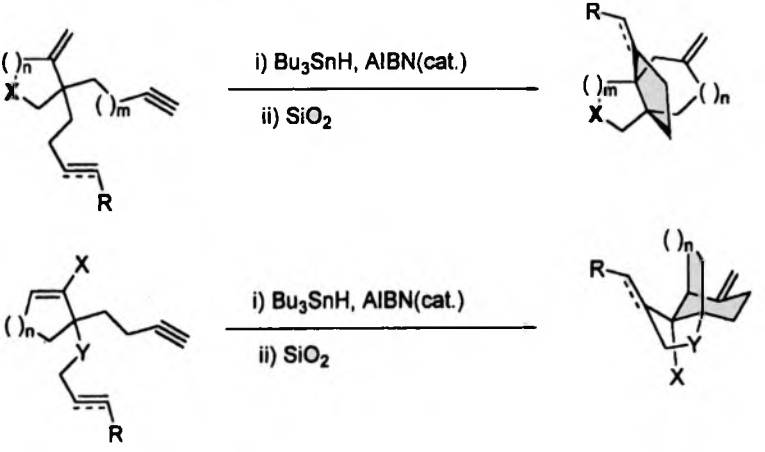
Since the introduction of tin-mediated free radical chemistry, free radical cyclization reaction has become one of the most widely utilized synthetic methodologies in the preparation of various polycyclic skeletons of theoretically or medicinally interesting molecules as well as natural products. As rearrangements of radical intermediates have followed predictable courses, rearrangement during radical cyclization reactions through cyclopropylmethyl radical intermediates (scheme 1) added a unique synthetic versatility to the radical cyclization reaction for the construction of various ring structures.



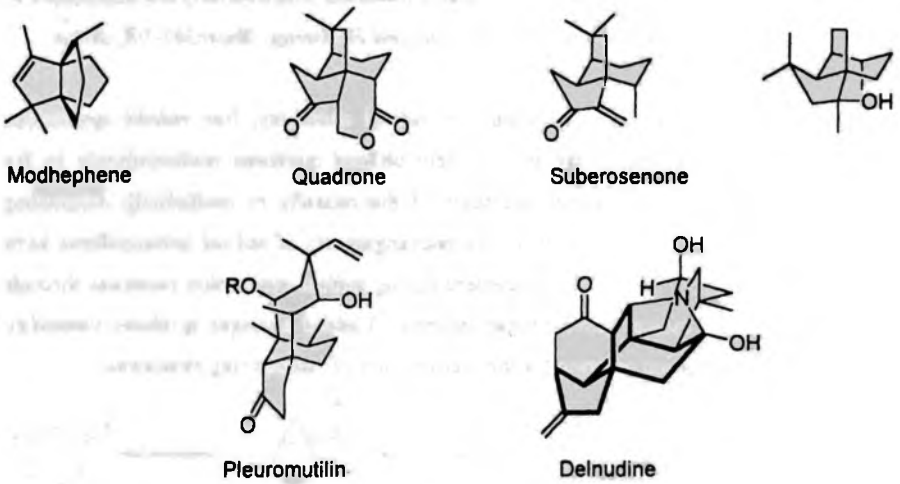
Scheme 1

During the course of the total synthesis of digitoxigenin, one of us observed an unusual cyclopropylmethyl radical mediated rearrangement reaction into the bicyclo[3.3.1]nonane system from 3-butynylcyclohexene system. We became interested in development of new tandem radical cyclization strategies utilizing the radical intermediate obtained from cyclopropylmethyl radical rearrangement. With proper starting materials, construction of propellanes and tricyclo[4.3.n.$1^{1,n}$] system from monocyclic precursors in a single operation was successfully accomplished (scheme 2).



Scheme 2

Since the current synthetic methodologies can introduce core structures of various natural products (scheme 3), total syntheses of natural products based on these synthetic strategies were successfully executed or in progress.



Scheme 3

In this seminar, the scope and limitation of the current synthetic methodologies and application to the total synthesis of natural products will be presented. The current strategies also provide a possibility of asymmetric synthesis of these natural products since a single quarternary carbon center controls all the stereochemistry of cyclization products. Studies on asymmetric synthesis will be discussed.

SYNTHESIS AND BIOLOGICAL PROPERTIES OF N-ACYLPHENYLISOSERINATES OF SESQUITERPENOID ALCOHOLS OF *LACTARIUS* ORIGIN

Rafał Barycki[a], Piotr Kopczacki[a], Maria Gumułka[a], Marek Masnyk[a] Agata Sarosiek[a] Włodzimierz Daniewski[a] Halina Grabarczyk[b] and Gerard Nowak[b] M. Kobus[c] and M. Łuczak[c]

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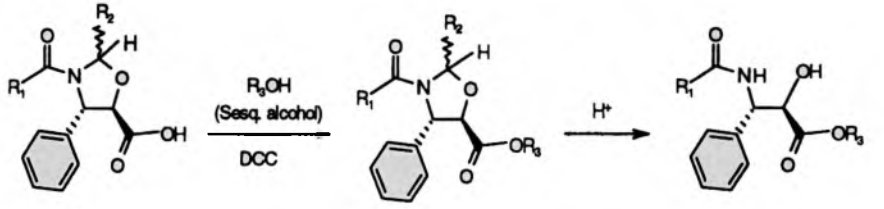
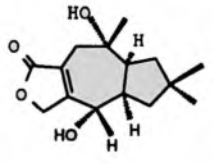
Important biological properties of Taxol® i.e. 13-N-benzoyl phenylisoserinate (2R,3S) of baccatin III prompted us to synthesize and to check biological properties of various N-acyl -(2R,3S)-phenylisoserinates of several sesquiterpenoid alcohols of *Lactarius* origin. Suitably protected N-acylphenylisoserine (**1**) when reacted with sesquiterpenoic alcohols in presence of DCC gave appropriate esters (**2**). These, when hydrolyzed in acidic conditions produced N-acylphenylisoserinates (**3**).

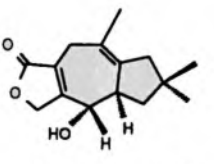


1 2 3

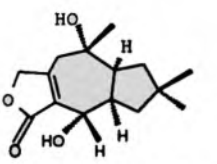
The typical examples of sesquiterpenoic alcohols are shown below:



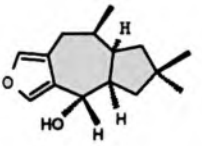
Lactarorufin A



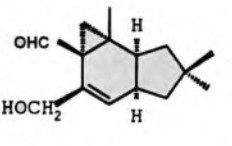
Anhydrolactarorufin A



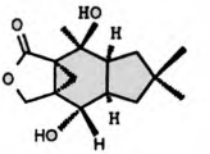
5-deoxy-13-oxo-LRA



Furanol



Isovellerol



Isolactarorufin

Some of the compounds exhibited antifeedant, antiviral and cytostatic properties, these are investigated and a few of them will be presented.

NATURAL PRODUCTS FROM AROUND THE WORLD

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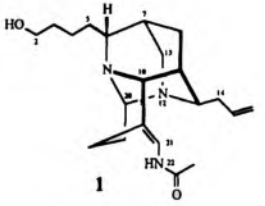
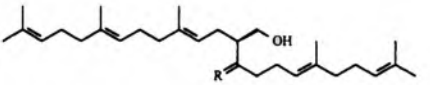
In principle, computer-assisted structure elucidation should facilitate the task of natural product chemists and speed up the process of resolving the structures of the many, often known, compounds which they isolate from natural sources. We have used the Logic for Structure Determination (LSD) program, developed by Dr Jean-Marc Nuzillard of the University of Reims, France, to solve some structures which presented no particular difficulties and could have been readily resolved by normal spectral analysis without recourse to the computer. Recently we were challenged by the structures of three alkaloids, from the seeds of *Acosmium panamense* (Fabaceae), which defied all our efforts to find solutions without using the LSD program[1]. Computer analysis indicated a novel diaza-adamantane skeleton for the alkaloids e.g. acosmine **1**.



1



2 R = CH_3, OH
3 R = CH_2

The formation of bifarnesol *2* seems the obvious first step in the conversion of farnesol into presqualene alcohol. Over a decade ago we isolated an anhydro-derivative of bifarnesol from the latex of *Euphorbia lateriflora*. Structure *3*, proposed for this compound on the basis of its spectroscopic properties, has now been confirmed by synthesis[2]. Recently another isolation of *3* (peplusol) has been reported[3] from *Euphorbia peplus* and the absolute configuration established as in *3*.

Other compounds which will be discussed include a chlorine-containing cyclic peptide from *Baccharis racemosa* and some unusual flavanoids from *Luma chequen*, isolated by Professor Labbé and her colleagues in Chile, a simple isocoumarin from a Nepalese liverwort and several compounds recently isolated from Cameroon medicinal plants by the group of the late Professor Johnson Ayafor.

References

1. J.-M. Nuzillard, J.D. Connolly, C. Delaude, B. Richard, M. Zèches-Hanrot, L. Le Men-Olivier. *Tetrahedron*, 1999, **55**, 11511.
2. S. Faure, J.D. Connolly, C.O. Fakunle, O. Piva. *Tetrahedron*, 2000, **56**,9647.
3. J.-L. Giner, J.D. Berkowitz, T. Andersson. *J. Nat. Prod.*, 2000, *63, 267.*

I am grateful to Professor C. Lavaud, Dr J.-M. Nuzillard and Ms S. Faure (University of Reims), Professor O. Piva (University of Lyon), Professor C.O. Fakunle (Obafeme Awolowo University, Nigeria), Mrs R.Pandey (Kathmandu, Nepal), Professor C. Labbé and her colleagues (University of Chile), Professor J.A. Ayafor and his colleagues(University of Dschang, Cameroon) and Dr D.S. Rycroft and Dr L. Farrugia (University of Glasgow) for their collaboration and friendship.

POSTER COMMUNICATIONS

NEOCLERODANE DITERPENOIDS FROM *TEUCRIUM MONTBRETII* SUBSP. *LIBANOTICUM* AND THEIR ABSOLUTE CONFIGURATION

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It is known that genus *Teucrium*, belonging to the Labiatae (Lamiaceae), is a rich source of neoclerodane diterpenoids[1-6], many of them showing antifeedant[7] activity against certain insect pests. In the frame of our ongoing program of researches on this genus, we report herein on the investigation of *T. montbretii* subsp. *libanoticum* P. H. Davis, growing in Lebanon. From the aerial part of this species ten neoclerodane diterpenoids were isolated. Three of them are new (3β-hydroxy-teubutilin A, 12-*epi*-montanin G, 20-*epi*-3,20-deacetyl-teupyreinidin), whereas the other seven, namely 6-keto-teuscordin, teuscordinon, 6β-hydroxy-teuscordin, montanin D, 3-O-deacetyl-teugracilin A, 3,20-deacetyl-teupyreinidin, montanin G, were already known. The structure elucidation was performed by spectroscopical and chemical means and the absolute neoclerodane configuration of 20-*epi*-3,20-deacetyl-teupyreinidin, 3,20-deacetyl-teupyreinidin, 12-*epi*-montanin G and tafricanin B was determined by chemical correlations.

(1) Piozzi, F. *Heterocycles* **1981**, *15*, 1489-1503.
(2) Piozzi, F.; Savona, G.; Rodriguez, B. *Heterocycles* **1987**, *25*, 807-841
(3) Merritt, A. T.; Ley, S. V. *Nat. Prod. Rep.* **1992**, 243-287.
(4) Piozzi, F. *Heterocycles* **1994**, *37*, 603-626.
(5) Piozzi, F.; Bruno, M.; Rosselli, S. *Heterocycles* **1998**, *48*, 2185-2203.
(6) Ulubelen, A.; Topcu, G.; Sonmez, U. **2000**, *Studies in Natural Products Chemistry*, Atta-ur-Rahman Editor. Vol. *23*, 591-648.
(7) Simmonds, M. S. J.; Blaney, W. M.; Ley, S. V.; Bruno, M.; Savona, G. *Phytochemistry* **1989**, *28*, 1069-1071

VOLATILE COMPONENTS OF *CENTAUREA CINERARIA* L. SUBSP. *UMBROSA* (LACAITA) PIGN. AND *CENTAUREA NAPIFOLIA* L. (ASTERACEAE) GROWING WILD IN SICILY (SOUTHERN ITALY)

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The genus *Centaurea* is a polymorphous genus of the Asteraceae family misses of a satisfactory interpretative model. *Centaurea cineraria* L. subsp. *umbrosa* (Lacaita) Pign. (syn. *C. ucriae* Lacaita ssp. *umbrosa*) is endemic to the western part of Sicily (1) with big flowerheads with red-purplish flowers. *Centaurea napifolia* L. is a annual plant common in Sicily, characterized by numerous flowerheads with purple flowers (1). Up to now no reports are available on the analysis of the volatile components of these plants, although some studies have been previously conducted on the aerial parts (2-4). For this study flowerheads of *C. cineraria* subsp. *umbrosa* and *C. napifolia* were collected at the full flowering from plants grown in Sicily and were subjected to hydrodistillation according to the standard procedure described in the *European Pharmacopoeia* (5). The essential oil were analysed by GC and GC-MS; peak identification was accomplished by comparison of their retention indices with literature values (6) as well as by comparison of their mass spectra with those of NIST 98 and Wiley 5 Libraries and those reported in literature (7, 8).

References

1. Pignatti, S. Flora d'Italia, Vol. III, 173-207, Edagricole, Bologna (1982).
2. Bruno, M. and Werner, H., *Phytochemistry*; **27**, 1873-5 (1988).
3. Bruno, M., Fazio, C., Paternostro, M. P., Diaz, J. and Werner, H., *Planta Med.*; **61**, 374-6 (1995).
4. El-Emary, N. A., Kobayashi, Y. and Ogihara, Y., *Fitoterapia*; **54**, 133-5 (1983).
5. *European Pharmacopoeia*, Vol. III, Maisonneuve SA, Sainte-Ruffine; 69 (1997).
6. Davies, N. W., *J Chromatogr.*, **503**, 1-24 (1990).
7. Adams, R. P. Identification of Essential Oils by Ion Trap Mass Spectrometry, Academy Press; New York (1989).
8. Jennings W. and Shibamoto T. Qualitative Analysis of Flavor and Fragrance Volatiles by Glass Capillary Gas Chromatography, Academy Press, New York (1980).

COMPOSITION OF THE ESSENTIAL OIL OF *PALLENIS SPINOSA* (L.) CASS. (ASTERACEAE)

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The monotypic genus *Pallenis spinosa* (L.) Cass., widespread in the Mediterranean area (1,2), belongs to the Asteraceae family, tribe Inuleae, subtribe Inulinae, Inula group. The plant is a rich source of oxygenated sesquiterpenes, i.e. germacrene D, germacrene A-type epoxides and oplopanones (3). For this study flowerheads of *Pallenis spinosa* were picked from plants grown at Capo Zafferano (Palermo, Sicily), at full flowering, in May 2000. Fresh material was subjected to hydrodistillation according to the standard procedure described in the *European Pharmacopoeia* (4); the oil yield was of 0.04 %. GC and GC-MS analyses were performed as previously described (5). The identification of the oil components was achieved by comparison of their retention times and mass spectra with those reported in literature (6,7) as well as by coinjection, whenever possible, with authentic substances. Results of the analyses show that the essential oil of *P. spinosa* is rich in oxygenated sesquiterpenes. The main components of the oil are germacra-1(10),5-dien-3,4-diol, 3-acetoxygermacra-1(10),5-dien-4-ol, α-cadinol and T-cadinol. The oil shows a weak antibacterial action. This is the first study on the chemical composition of the essential oil of this plant.

References

1 - T. G. Tutin *et al.*, *Flora Europaea*, Vol. IV, p. 139, Cambridge University Press, London (1976).
2 - S. Pignatti, Flora d'Italia, Vol. III, p. 53, Edagricole, Bologna (1982).
3 - G. Appendino, J. Jacupovic and S. Jacupovic - *Phytochemistry*, **46** (6), 1039 (1997).
4 - *European Pharmacopoeia*, Vol. III, p. 69, Maisonneuve SA, Sainte-Ruffine (1997).
5 - Senatore, F. *Flavour Fragrance. J.*, **13**, 263, (1998).
6 - Adams, R. P. Identification of Essential Oils by Ion Trap Mass Spectrometry, Academy Press; New York (1989).
7 - Jennings W. and Shibamoto T. Qualitative Analysis of Flavor and Fragrance Volatiles by Glass Capillary Gas Chromatography, Academy Press, New York (1980).

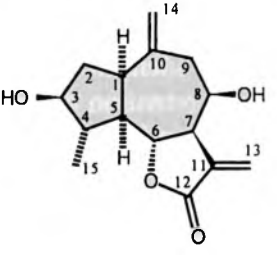
SESQUITERPENOIDS AND PHENOLICS FROM *CREPIS CONYZIFOLIA*

Wanda Kisiel and Klaudia Michalska

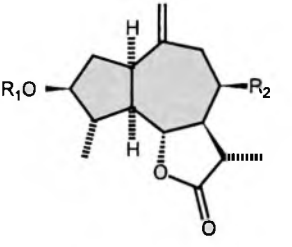
Department of Phytochemistry, Institute of Pharmacology, Polish Academy of Sciences,12 Smętna Str., Pl-31-343 Kraków, Poland

Phytochemical studies of members of the genus *Crepis* (Asteraceae) have revealed that guaiane-type sesquiterpene lactones, accumulated mainly as glycosides, are the most representative secondary metabolites of this taxon. The aim of the present work was to characterize root constituents of *Crepis conyzifolia* (Gouan) Kern. The chemistry of this plant has not been examined so far.

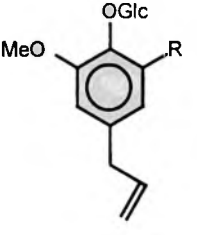
A combination of column and thin layer chromatographies on silica gel followed by semiprep. RP HPLC of the ethanol extract from the plant material yielded two new (**1** and **2**, Glc = β-glucopyranosyl) and two known (**3** and **4**) guaianolides, along with three known phenylpropanoids (**5** - **7**). The known compounds were identified by direct comparison with authentic samples from our collection. Structures of the new compounds were established as 8β-hydroxy-4β(15)-dihydrozaluzanin C (**1**) and 4β(15), 11β(13)-tetrahydrozaluzanin C-3-O-β-glucopyranoside (**2**) by spectral methods, including 2D NMR (NOESY, COSY, HETCOR) experiments.



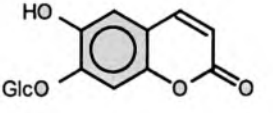
1



2 R_1 = Glc, R_2 = H
3 R_1 = Glc, R_2 = OH
4 R_1 = H, R_2 = OH



5 R = OMe
6 R = H



7

ISOLATION AND TRANSFORMATION OF NATURAL POLYPRENOLS

Alexander V. Kutchin, Irene A. Dvornikova, Alla A. Korolyova,
Ludmila P. Karmanova, Sergey A. Patov

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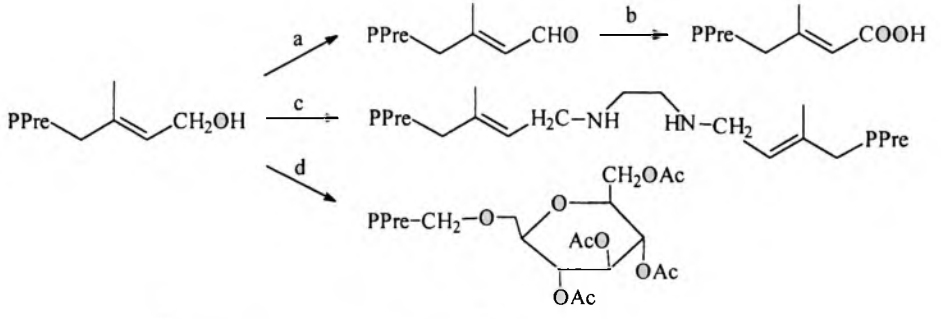
Polyprenols are linear isoprenoid alcohols, containing more than 4 isoprene units and forming in all natural organisms. The most accessible sources of polyprenols are green parts of coniferous species and neutral part of sulphate soap – the side product of cellulose production. Polyprenols are of great interest as biologically active substances, reducing albumeno-carbohydrate metabolism, reproduction of organism function, correlating immune status, possessing of anti-ulcerous effect, exceeding the effect of sea-buckthorn oil. The paper presents the elaborated ways of emulsion isolation of polyprenols C_{70}-C_{80} from the coniferous greenery of *Pinacea* family (the yield is 0,05-0,2 %), as well as polyprenols C_{35}-C_{40} from sulphate soap (the yield is about 1%). Syntheses, leading to medicinal substances with anti-ulcerous and gastro-protective effect and to inhibition of plural medicinal cancerous cell stability have been studied and developed. The corresponding aldehydes, oximes, complex ethers, halogeno-derivatives, amines and other functional derivatives have been synthesized on the base of isolated polyprenols.

Structure and peculiarity of isolated polyprenols and their derivatives has been confirmed by the method of IR-, NMR (H and ^{13}C) spectroscopy and HPLC-chromatography.



a) MnO, hexane; b) Ag_2O;
c) ethylenediamine; d) AcBrGlu, acetone.

FISSISTIGMATINS - NOVEL TYPE PLANT CONSTITUENTS WITH FLAVONOID - SESQUITERPENE HYBRID STRUCTURE

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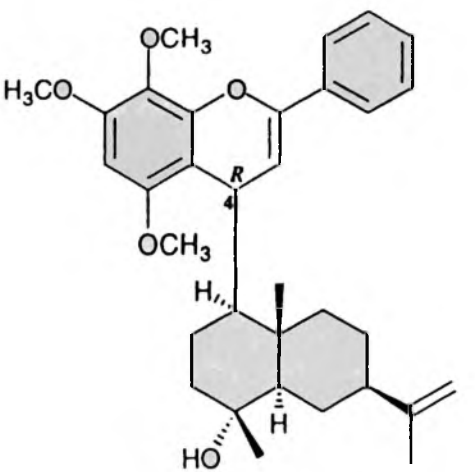
Within a research program on the phytochemical diversity of the flora of Vietnam we have investigated the plant species *Fissistigma bracteolatum* (Annonaceae) used in the folk medicine of Southeast Asia. Besides a series of chalconoids, four new compounds named fissistigmatins A - D were isolated and structurally identified. The complete structural elucidation including the absolute configuration was carried out by combination of extensive NMR studies, MS, CD and molecular modelling calculations. In the case of fissistigmatin A the determined constitution and stereochemistry has been confirmed also by X-ray analysis of its p-bromobenzoate. The fissistigmatins represent the first examples an unprecedented novel type of plant constituents with flavonoid-sesquiterpene hybrid structure. A reaction sequence for their putative biosynthesis will be also discussed.



Fissistigmatin A

References:
T.P. Lien, A.Porzel, J.Schmidt, T.V.Sung and G.Adam, Phytochemistry **53**, 991 (2000)
A.Porzel, T.P.Lien, J.Schmidt, S.Drosihn, Ch.Wagner, K.Merzweiler, T.V.Sung and G.Adam, Tetrahedron **56**, 865 (2000)

ON THE IDENTIFICATION OF THE ACTIVE SITE RESIDUES OF PENTALENENE SYNTHASE BY SITE-DIRECTED MUTAGENESIS.

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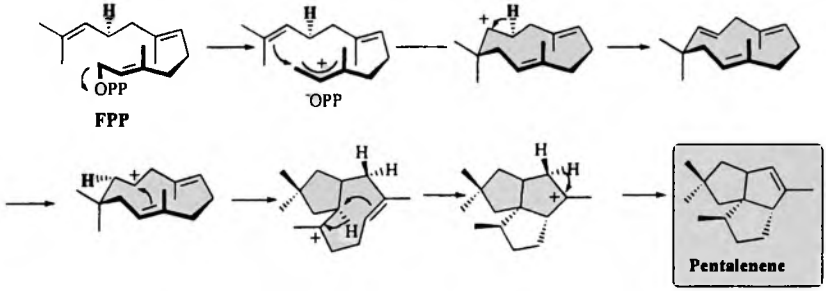
Pentalenene synthase (PS), a sesquiterpene synthase, catalyzes the conversion of FPP to pentalenene, the hydrocarbon precursor of the pentalenolactone family of antibiotics [1]. The cyclase from *Streptomyces* UC 5319 has been cloned and overexpressed in *Escherichia coli* [2]. Extensive studies support the following cyclization mechanism [3].



Based on this mechanism, it was postulated that a single active site base might be responsible for the successive deprotonation-reprotonation-deprotonation steps [2]. The X-ray crystal structure of recombinant pentalenene synthase revealed that histidine-309 (H309) appeared to be ideally situated for abstraction of the relevant protons from FPP and derived intermediates [4]. Further inspection of the PS active site showed that the side chain of asparagine-219 (N219) protrudes into the active site cavity and it was suggested that this residue might influence the cyclization reaction possibly by stabilizing transient cations with the help of phenylalanine-77 (F77).

A set of PS mutants was constructed in which H309, N219 and other amino acids were replaced by other residues. These experiments suggest that histidine is not required for catalytic activity[5], but N219 is essential.

[1] Cane, D. E.; Tillman, A. M. *J. Am. Chem. Soc.* **1983**, *105*, 122-124. [2] Cane, D. E.; Sohng, J. K.; Lamberson, C. R.; Rudnicki, S. M.; Wu, Z.; Lloyd, M. D.; Oliver, J. S. and Hubbard, B. R. *Biochemistry* **1994**, *33*, 5846-5857. [3] Cane, D. E.; Oliver, J. S.; Harrison, P. H. M.; Abell, C.; Hubbard, B. R.; Kane, C. T.; Lattman, R. *J. Am. Chem. Soc.* **1990**, *112*, 4513-4524. [4] Lesburg, C. A.; Zhai, G.; Cane, D. E.; Christianson D. W. *Science* **1997**, *277*, 1820-1824. [5] Seemann, M., Zhai, G., Umezawa, K. and Cane, D. *J. Am. Chem. Soc.*, **1999**, *121*, 591-592.

Studies on the biosynthesis of moenocinol, the lipid part of the moenomycin antibiotics

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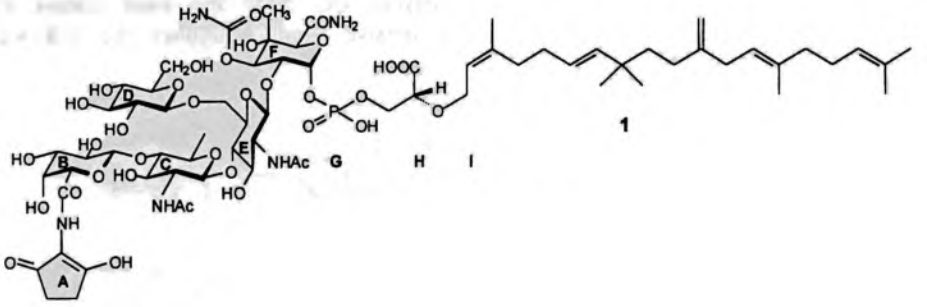
The moenomycin-type antibiotics[1] (see formula **1**) contain a C_{25} lipid unit (the moenocinol part I), that has an interesting structure.



Three isoprenoid C_5 units are easily discernible whereas the central C_{10} part (C-5 through C-11) does not obey the isoprene rule in an obvious way. We have recently shown that the complete moenocinol unit is of isoprenoid origin and that it is formed via the non-mevalonate pathway.[2] This result paved the way to a series of successful feeding experiments starting from differently labeled 1-deoxy-D-xylulose (**1**) preparations. Known routes centered on an asymmetric Sharpless dihydroxylation have been adapted to introduce ^{13}C labels into various positions.[3,4] The results of feeding experiments with [1-^{13}C]-, [2,3-$^{13}C_2$], [4-^{13}C]- and [2,5-$^{13}C_2$]-labeled 1-deoxy-D-xyluloses allowed to postulate that moenocinol originates from a farnesyl precursor and geranyl pyrophosphate.[5,6,7] The poster will show the suggested mechanism and will discuss experiments that are in progress to prove that two allyl starter units are involved in the biosynthesis of moenocinol.

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1 For leading references, see Donnerstag, A.; Marzian, S.; Müller, D.; Welzel, P.; Böttger, D.; Stärk, A.; Fehlhaber, H.-W.; Markus, A.; van Heijenoort, Y.; van Heijenoort, J. *Tetrahedron* **1995**, *51*, 1931-1940.

2 Review: Rohmer, M. *Nat.Prod.Rep.* **1999**, *16*, 565-574. For leading references on new work in this area, see Herz, S.; Wungsintaweekul, J.; Schuhr, C.; Hecht, S.; Lüttgen, H.; Sagner, S.; Fellermeier, M.; Eisenreich, W.; Zenk, M. H.; Bacher, H.; Rohdich, F.; *Proc. Natl. Acad. Sci. USA* **2000**, *97*, 2486-2490, Takagi, M.; Kuzuyama, T.; Kaneda, K.; Watanabe, H.; Dairi, T.; Seto, H. *Tetrahedron Lett.* **2000**, *41*, 3395-3398.

3 Kennedy, I.A.; Hemscheidt, T.; Britten, J.F.; Spenser, I.D. *Can.J.Chem.* **1995**, *73*, 1329-1337.

4 J.-L. Giner, *Tetrahedron Lett.* **1998**, *39*, 2479-2482.

5 Schuricht, U.; Hennig, L.; Findeisen, M.; Welzel, P. *Tetrahedron Lett.* **2000**, *41*, 3047-3051.

6 Schuricht, U.; Endler, K.; Hennig, L.; Findeisen, M.; Welzel, P. *J.Prakt.Chem.* **2000**, *342*, 761-772.

7 Schuricht, U.; Hennig, L.; Findeisen, M.; Welzel, P.; Arigoni, D. *Tetrahedron Lett.* **2001**, *42*, 3835-3837

TRANSFORMATIONS OF α-PINENE USING *Picea abies* SUSPENSION CULTURE

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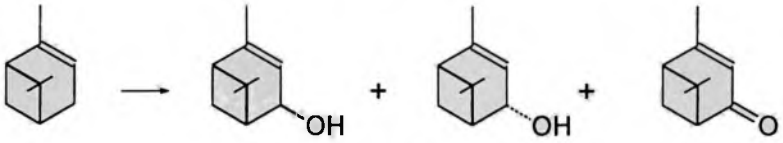
Pure enantiomers of α-pinene were transformed to *cis*- and *trans*-verbenol and verbenone using *Picea abies* plant cells. Longer reaction time led to the formation of verbenone that was not transformed further. Enantiomeric purity of the transformation products corresponded to that of starting α-pinene enantiomers.



α-pinene *cis*-verbenol *trans*-verbenol verbenone

The transformation of α-pinene is very fast. After 3 hours, about 40 % of *trans*-verbenol was present in the reaction mixture. After addition the substrate and immediate workup of the reaction mixture, small amounts of the products were already detected. Furthermore, the filtrate of the cell suspension transformed α-pinene to the same products. The transformation activity of the water phase indicates that it might be possible to isolate an active enzyme from the plant cells.

Financial Support: Grant Agency of the Academy of Sciences of the Czech Republic (grant No. IAA 4055001).

APPLICATION A NEW MICROORGANISM TO OXIDATION-REDUCTION OF STEROID SUBSTRATES

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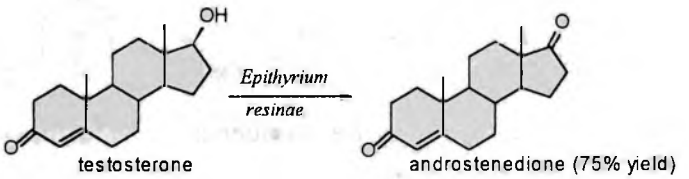
Epithyrium resinae was isolated from wounded trunk of Pinus sylvestris.

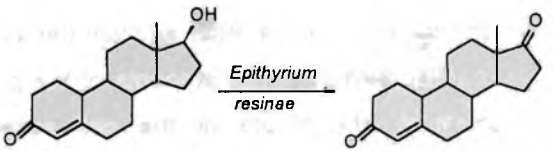
In our previous studies we have found, that *Epithyrium resinae* can easily transform prochiral ketones to optically active alcohols with high yields and high enantiomeric purities. Now this strain was used as a bioreagent so as to investigate its ability to transform steroid compounds. The substrates were: testosterone, 19-nortestosterone and 1-dehydrotestosterone.

Transformations were carried out from 2 to 14 days, depending on the reaction progress. All examined substrates underwent transformation and the reaction products we identified on the basis of TLC and spectra analyses. The yield off all transformations was established on the basis of GLC.

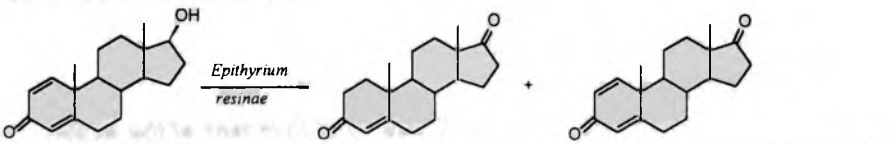
Epithyrium resinae is able to oxidize steroidal 17β hydroxy group to the carbonyl function. Besides 17β alcohol oxidation specific reduction of C1-C2 double bond in 1-dehydrotestosterone was observed.



testosterone androstenedione (75% yield)



19-nortestosterone 19-norandrostenedione (86% yield)



1-dehydrotestosterone androstenedione (68% yield) 1-dehydroandrostenedione (32% yield)

BIOTRANSFORMATIONS OF ISOPRENIC COMPOUNDS BY MEANS OF VEGETABLE ENZYME SYSTEM

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Nowadays food flavour, pharmaceutical and agrochemical industry require syntheses of enantiopure compounds. In the previous papers by the same authors it has been concluded that enzyme systems contained in the mature fragments of high plants are capable of separating racemic mixtures. Recently an optically pure enantiomer S-(-)-1-(2-naphthyl)ethanol (ee=100%) has been obtained at 100% efficiency through the reduction of 2-acetonaphthone by means of a carrot root (*Daucus carota* L.).

On the basis of the conclusions, roots of carrot (*Daucus carota*), celeriac (*Apium graveolens* L. var. *rapaceum*) and horse-radish (*Armoracia lapathifolia* Gilib) have been used as biocatalysts in isoprenic compound transformations.

Substrates used included esters (acetates), alcohols and ketones: androstenolone, testosterone, pregnenolone, cholesterol, FAS, menthol, borneol and their acetates (also testosterone propionate).

In the course of biotransformation ester (acetates and propionate) hydrolysis, reduction of carbonylic group and oxidation of secondary alcohols have been observed Achieved solutions will be the subject of comparison with biotransformations of other living organisms.

The table presents the results obtained in the transformation of steroids by means of specified biocatalysts.

Reaction type	Carbon number	*D. carota*	*A. graveolens*	*A. lapathifolia*
Hydrolysis	C-3	+	+	+
	C-17	+	+	+
	C-21			+
Oxydation	C-3			+
	C-17			
Reduction	C-3	+	+	+
	C-17	+	+	
	C-20			

Lack of „+" indicates that the reaction did not occur.

Menthyl acetate was hydrolyzed by means of all the biocatalysts applied; menthol and bornyl acetate failed to undergo the transformation while menthone was being reduced by *Armoracia lapathifolia* only.

METABOLISM OF STEROID SUBSTRATES IN *FUSARIUM CULMORUM* CULTURE

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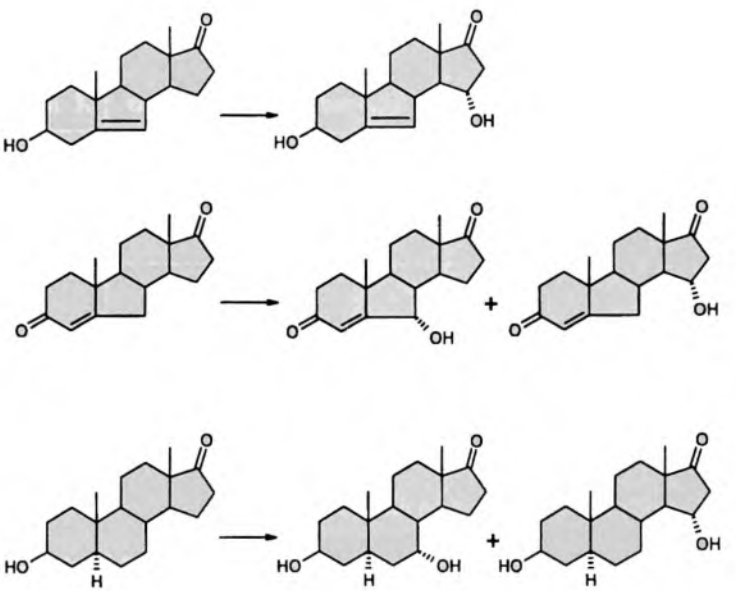
Norwida 25, 50-375 Wrocław, Poland

In our earlier studies we observed that the strain *Fusarium culmorum* was able to carry out regio- and stereoselective hydroxylation of steroid substrates. The directing effect of substrate substituents can be seen in hydroxylation of the same 4-ene-3-oxo steroids with different substituent at C-17. In the presence of 4-ene-3-oxo group the hydroxylation at 12β- and 15α-sites occurred, but only the substrates carrying the oxygen function at C-17 were hydroxylated at 6β-position.

5-ene steroid substrates carrying 3β-hydroxyl group and oxygen function at C-17 were hydroxylated exclusively at 7α- axial, allylic position. The presence of C_5-C_6 double bond seems to have the major impact on the position of 5-ene steroid hydroxylation.

To obtained more information about relationship between the structure of the substrates and course of hydroxylation by *F. culmorum* we decided to study the metabolism of androsterone (DHEA dihydroderivative) and B-nor analogues of: DHEA and androstenedione. The results are presented below.



TRANSFORMATION OF TESTOSTERONE AND ITS DERIVATIVES IN *CLADOSPORIUM CLADOSPORIODES* CULTURE

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The transformation of testosterone and its 1-dehydro-, 17α-methyl- and 19-nor-derivatives was carried out in *Cladosporium cladosporiodes* culture. The purpose of the experiment was to study the influence of methyl group and additional double bond on the course of transformation. The results are presented below:

substrate	products	yield (%)
testosterone	5α-androstan-3β,17β-diol	23
	6β-hydroxyandrostenedione	20
	5α-androstan-3β-ol-17-on	17
	6β-hydroxytestosterone	14
	15α-hydroxytestosterone	14
1-dehydrotestosterone	Androstadienedione	31
	6β-hydroxyandrostadienedione	30
	6β-hydroxy-1-dehydrotestosterone	10
	7β-hydroxyandrostadienedione	10
methyltestosterone	15α-hydroxymethyltestosterone	57
	5α-androstan-17α-methyl-3β,17β-diol	20
	6β-hydroxymethyltestosterone	15
1-dehydromethyl-testosterone	15α-hydroxy-1-dehydromethyltestosterone	48
	6β-hydroxy-1-dehydromethyltestosterone	24
	6β-hydroxyandrostenedione	9
	5α-androstan-3β-ol-17-on	7
	7β-hydroxy-1-dehydromethyltestosterone	5
19-nortestosterone	6β-hydroxy-19-norandrostenedione	37
	6β-hydroxy-19-nortestosterone	17
	15α-hydroxy-19-nortestosterone	13
	10β-hydroxy-19-norandrostenedione	12
	15α-hydroxy-19-norandrostenedione	7
	19-norandrostenedione	5

- the additional C1-C2 double bond inhibited the formation of 3β-ol-5α-products and stimulated hydroxylation at the 7β position,
- the percentage of 15α-hydroxylated products has increased in the presence of 17α-methyl group,
- the lack of C19 methyl group blocked of the reduction in A ring and enabled hydroxylation at the 10β position.

BIOCONVERSION OF PROGESTERONE WITH HALOPHILIC BLACK YEAST *HORTAEA WERNECKII*

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Several black yeasts have recently been isolated from salterns in Slovenia. The dominant species among them was *Hortaea werneckii*, which was found to grow in the media with up to 30% salt concentration. Some adaptations of this microorganism to extreme environments have already been detected; e.g. the changes in the membrane properties and in sterol biosynthesis. In our study we were interested whether the salinity of the medium in any way effects the bioconversion of steroids with the black yeast *Hortaea werneckii*. It was found that the addition of salt to the medium in a concentration up to 17% effects the bioconversion of progesterone only indirectly by slowing the growth of the microorganism. On the other hand two different profiles of progesterone bioconversion dependent on the growth phase of the microorganism were found. In the logarithmic phase of growth several hydroxylated products were detected and the corresponding enzymes were found to be of an inducible nature. At the beginning of the stationary phase the profile changed. The metabolites were found to be the reduced products of progesterone and metabolites, produced by progesterone C_{17} side chain cleavage. The corresponding enzymes were found to be constitutive. In the stationary phase of growth we detected only traces of the hydroxylated products of progesterone which predominated in the logarithmic phase. The same change of profiles was not observed when we followed the bioconversion of progesterone with the mesophilic yeast *Saccharomyces cerevisiae*. In addition since it is known that progesterone has an influence on fungal growth the effect of progesterone as a stressor was tested in the logarithmic as well as stationary phase of growth of *Hortaea werneckii*.

DESIGN OF A NOVEL SYSTEM FOR STRESS RESPONSE OBSERVATION AND ITS USE IN PRACTICE

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Leaves of plants normally release small quantities of volatile chemicals, but when herbivorous insects(1) damage a plant, many more volatiles with different functions are released. The chemical identity of the volatile compounds varies with the plant and insect species and type of wounding. The signalling pathways may vary among plant species. Recent results suggest(2) a general underlying pattern for plant self-defence mechanisms. The key role in induced synthesis of volatiles has the biosynthesis of jasmonic acid (JA) or its methyl ester (MJ). The biosynthesis of JA (from linolenic acid) is induced by recognition of the primary elicitors that activate the plant's signalling system, the derivatives of JA then trigger the specific responses of the plant (gene encoding proteinase inhibitors, antifungal proteins, defensive secondary metabolites etc.). Many of actual bioassay test systems work with wounded plant tissue(2), because it is necessary to wound plant (e.g. to excise leaves) prior to the addition of chemicals (elicitors of defence reactions). It is difficult to interpret results of such experiments, because the high background induced by cutting the leaves may overlap the response induced by MJ. So, we are developing test system where no wounding of plant is required. We use small plantlets of *Nicotiana tabacum* cultivated under *in vitro* conditions. The possibility of using tobacco callus and cell suspension was tested with negative results, because they do not produce volatiles(3). MJ in form of vapours is used as the elicitor of the stress reactions. The production of volatile substances was assayed by a combination of solid phase microextraction (SPME) and gas chromatography (GC), volatiles were identified by mass spectrometry. We tested this system(4) in comparing the antistress activities of some brassinosteroids (24-epiBR, "4075", "4154"). The proposed system proved to be reproducible, sensitive and stable.

Acknowledgement: *The work was supported by grant FRVS 0633/G4, GA AV A-405 5803 and research project Z4 055 905.*

REFERENCES

1. Paré P. W., Tumlinson J. H.: Plant. Physiol. 121, 325 (1999)
2. Schröder F.: Angew. Chem. Int. Ed. 37, 1213 (1998)
3. Nesnerova P., Svatos A. and Macek T.: Chem. Listy 93, S62 (1999)
4. Nesnerova P., Svatos A., Kohout L. and Macek T.: Plant. Physiol. Biochem., 38, p.87 (2000)

CIRCULAR DICHROISM AND CONFORMATION OF 1α,25-DIHYDROXYVITAMIN D_3 AND ITS ANALOGS

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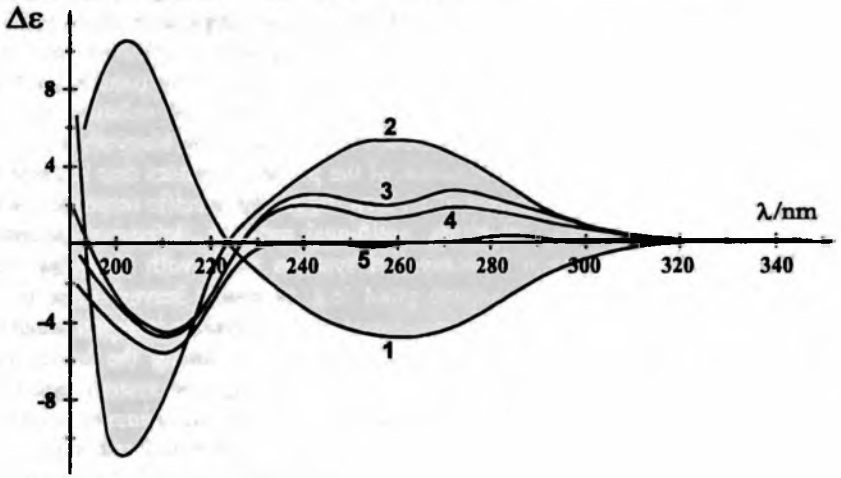
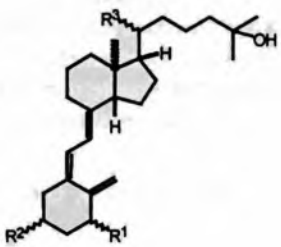
1α,25-Dihydroxyvitamin D_3 **(1)** and its analogs **(2-5)**, synthesized in the laboratory of Professor Wicha, have been analyzed by CD spectroscopy with the purpose to develop a correlation between the conformation and the CD data.





	R¹	R²	R³
2	α-OH	β-OH	β-Me
3	β-OH	α-OH	α-Me
4	β-OH	α-OH	β-Me
5	β-OH	β-OH	β-Me
1		*ent* - **2**	

We used the AM1 method for conformational space search of **1-5**. The results show that the Cotton effect of the chiral triene chromophore present in **1-5** can be rationalized on the basis of the equilibrium between the two chair forms of ring A and taking into account the configuration at C-14. The position of the equilibrium is determined mainly (but not exclusively) by the configuration of the hydroxy groups in positions 1 and 3 of ring A.

STEREOCHEMISTRY OF α-HYDROXY KETONES FROM THEIR CIRCULAR DICHROISM SPECTRA

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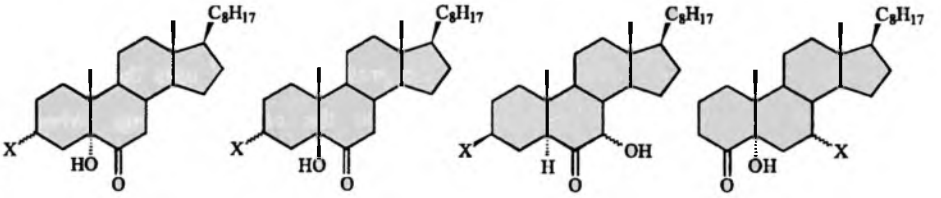
The chiroptical properties of α-substituted ketones have been the subject of many studies for a long time. However, to date there is no simple rule proposed that correlates CD data with the stereochemistry of α-hydroxy ketones. In general, α-hydroxy ketones may be divided into four classes: (1) rigid with axial hydroxyl, (2) rigid with equatorial hydroxyl, (3) rigid with bisectional hydroxyl , and (4) flexible cyclic or open chain α-hydroxy ketones.



Continuing earlier works of W. Klyne,[1] C. Djerassi,[2] G. Snatzke,[3] J.R. Bull,[4] and other authors[5] we have synthesized series of axial and equatorial cholestane α-hydroxy ketones (general structure shown above) and studied their chiroptical properties. Our results as well as previously published demonstrate that we are able to discriminate between axial and equatorial α-hydroxy ketones with help of the following rule: the CD λ_{max} values of the nπ* band occur in the range (a) *ca.* 300 – 310 nm for all α-hydroxy ketones with axially oriented hydroxyl, (b) *ca.* 280 – 290 nm for all α-hydroxy ketones with equatorially oriented hydroxyl, and (c) *ca.* 305 – 325 nm for all α-hydroxy ketones with bisectional hydroxyl. The rule holds independently of the solvent used.

References:

1. W. Klyne, *Tetrahedron*, **1961**, *13*, 29; 2. C. Djerassi et al., *J.Am.Chem.Soc.* **1965**, *87*, 73; 3. G. Snatzke et al., *J.Chem.Soc. (C)*, **1967**, 972; 4. J.R. Bull et al., *Tetrahedron*, **1970**, *26*, 1525; 5. T. Suga et al. *J.Org.Chem* **1967**, *32*, 965.

STEREOSELECTIVE REDUCTION OF VERBENONE TO *cis*-VERBENOL

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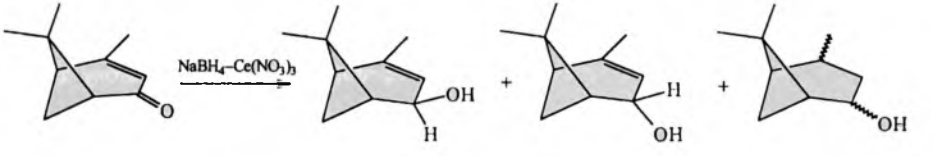
(+)*cis*-Verbenol – a principal component of the sex attractant of the bark beetle may be produced by the reduction of verbenone with $NaBH_4$ [1] $(Pr^i\text{-}O)_3Al$ [2], sodium dihydrobis-(2-methoxyethoxy) aluminate [3], or $LiAlH_4$ [4]. But the physico-chemical characteristics (melting point and $[\alpha]_D$) of *cis*-verbenol, obtained by these authors differ greatly, furthermore, the yields of alcohol is not enough high.

We reduced verbenone by $NaBH_4$ and $NaBH_4$–$Ce(NO_3)_3$. In the first case there was obtained a complex mixture, containing *cis*- and *trans*-verbenol and verbanols. The saturated to unsaturated alcohols ratio was ~50:50, the same one was for *cis*- to *trans*-verbenols – 70:30.

In order to increase the chemo- and stereoselectivity of this reaction we used the reducing system $NaBH_4$–$Ce(NO_3)_3 \cdot 6H_2O$. In the presence of Ce^{+3} ions the reduction generally occurs by 1,2-addition of hydrogen to the carbonyl group, whereas 1,4-addition to conjugated of π–electron double bond system occurs in shade rate.



Verbenone *cis*-Verbenol *trans*-Verbenol Verbanols

In the result of this reaction the mixture with 89-94 % *cis*-verbenol, 2-6 % verbanols and minor amount of *trans*-verbenol are formed. A pure sample of (-)*cis*-verbenol obtained by crystallization from light hydrocarbons has m.p. 62-64 °C, $[\alpha]_D^{29}$ -12.9° (c 0.92, EtOH), $[\alpha]_D^{29}$ +10.1° (c 0.98, $CHCl_3$).

1. Poddubnaya S.S., Cherkaev V.G., Rudolfi T.A., Chem. Abstr., 62, 11855h, 1965.
2. Bain J.P., Pat. US 2,972,632, 1961.
3. Regan A.F., Tetrahedron, 25, 3801, 1969.
4. Kalvoda L., Vrkoc J. Pat. CSSR AO 215485, 1981.

SYNTHESIS OF NEW CHIRAL LIGANDS BASED ON (+)α-PINENE

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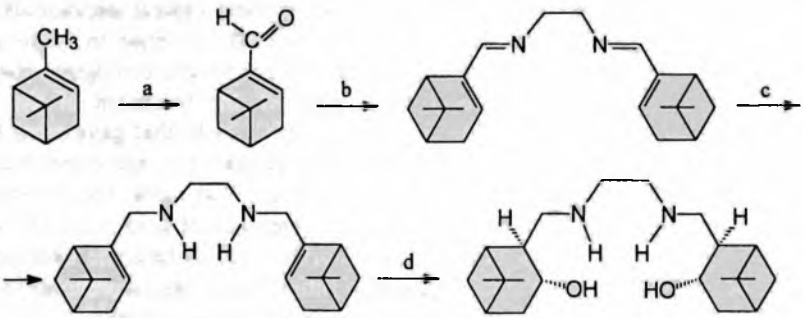
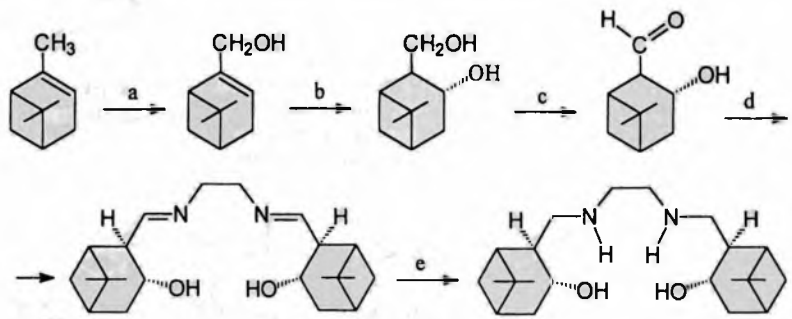
α-Pinene is the most wide spread natural monoterpene. Optically active ligands, based on this terpene are generally interesting for preparing the chiral catalysts, using in organic and bioorganic chemistry. We suggest two ways for synthesis such ligands.



a) SeO_2 / EtOH b) $H_2N–(CH_2)_2–NH_2$ / PhH
c) $NaBH_4$ / EtOH d) B_2H_6 / THF, H_2O_2 / OH^-



a) SeO_2 / EtOH b) $LiBH_4$ / H_2SO_4 / Et_2O; H_2O_2 / OH^-
c) MnO_2 d) $H_2N–(CH_2)_2–NH_2$ / PhH
e) $NaBH_4$ / EtOH

This work is support by INTAS grant 99-1541.

SEMISYNTHETIC SULFUR-CONTAINING ISOPRENOIDS OF *ASTERACEAE* PLANTS AND THEIR SEVERAL BIOLOGICAL ACTIVITIES

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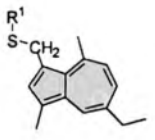
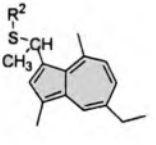
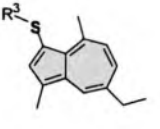
Plants belonging to *Asteraceae* group grown in the Volga-Kama region (Russia) especially *Achillea millefolium l. s. l.* are known to be a rich source of isoprenoids, including new structural types[1]. Two of these, chamazulene (**1**) (7-ethyl-1,4-dimethyl-azulene) and (-)-carvone (**2**) are biologically active, showing anti-inflammatory and anti-ulcer properties. In this connection, it is of interest to synthesize sulfur-containing derivatives on the basis of these natural isoprenoids and investigate their biological activities. Chamazulene is easily obtained in preparative amounts from essential oil of above herb by means of simple and ecological method – hydrodistillation. On the other hand, we suggested[2] the treatment of the raw material with Baker's yeast (BY, 10% aq. sucrose, 30°C, 24 h) that gave more than 30% increase in the yield of (**1**). Chamazulene obtained than way and commercially available (-)-carvone were further used in reactions: 1a) with the aldehydes (formaldehyde, acetaldehyde) plus the thiols in the presence of acid-trace; 1b) with the sulfenylchlorides[4]; 1c) with the disulfenylchlorides; 2a) in catalytic addition of thiols to (-)-carvone. In result, we've got the new sulfur-containing isoprenoids (**3-34**) and tested their several plant-plant, plant-microbe, antioxidant activities[2,3].



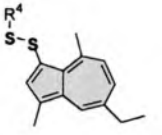
R^1 = Et (**3**), i-Pr (**4**), H_3CO-CO-CH_2- (**5**), HOOC-CH_2- (**6**),Tolyl (**7**), N-phenyl-tetrazolyl (**8**)



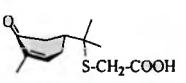
R^2 = Et (**9**), H_3CO-CO-CH_2-(**10**)



R^3 = Me (**11**), Et (**12**), i-Pr(**13**), $H_{33}C_{16}$- (**14**), H_3C-C_6H_4-(**15**),Tolyl (**16**), N-phenyl-tetrazolyl (**17**)



R^4 = Et (**18**), H_3CO-CO-CH_2-(**19**)



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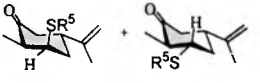
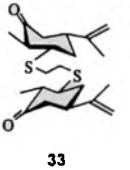


R^5 = i-Pr (**21,27**), -$C_{16}H_{33}$ (**22,28**), -CH_2-CH_2OH (**23,29**), -CH_2-(**24,30**), -CH_2-CH_2SH (**25,31**), -CH_2-CH(NH-$COCH_3$)-COOH (**26,32**)



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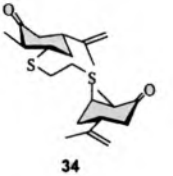

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1. Paley R.V., Plemenkov V.V. et al.//*Rastitelnye resursy* (Rus).1996.V.32.N 4. P. 37-44.
2. Paley R.V., Plemenkov V.V., Ngo Bakopki B. //Abst. of V International Conf. "Bioantioxidant". Moscow. 18-21th November 1998. P. 69.
3. Plemenkov V.V., Yanilkin V.V. et al.//Zhurnal Obshei Khimii (Rus).2001.V.71 N 3.P.494-500.
4. Paley R.V., Plemenkov V.V. et al.//Zhurnal Org. Khimii (Rus). 2000.V.36.N12.P.1772-75.

This research has been supported by INCO-Copernicus under Grant Number IC 15CT98-0150.

CATALYTIC ELECTROPHILIC ADDITION OF DISULFIDES AND THIOLS TO (+)LIMONENE

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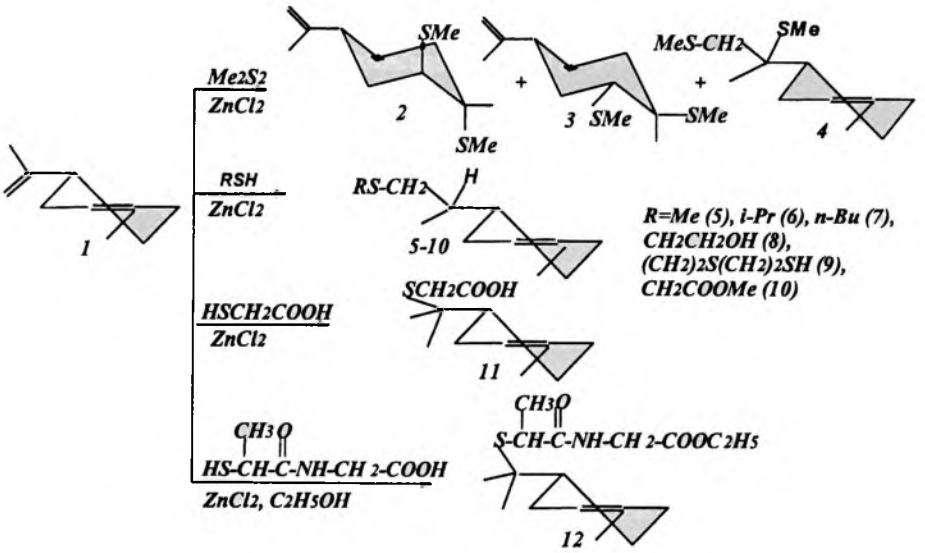
The addition of disulfides to the *exo*- and *endo*-double bonds of (+)limonene ***1*** carried out in the presence of $ZnCl_2$ appears to be equiprobable (the ratio of regioisomers is 1:1 (***2+3***) : ***4***), the ratio of stereoisomers ***2***:***3*** is 9:1) as well as thiols add only onto *exo*-cyclic double bond in an anti-Markovnikov fashion in most cases (***5-10***). The Markovnikov products ***11,12*** were fixed in the reactions with thiols containing carboxylic moiety.



Calculations using the PM3 and B3LYP/6-31G* methods show the high degree of delocalization of the LUMO of $ZnCl_2(RSH)_2$ and $ZnCl_2/(Me_2S_2)_2$ complexes and high sensitivity of the above reactions to steric and electronic factors.

1. Nikitina L.E., Plemenkov V.V. et al., *Zhur. Org. Khimii (Rus),* 1996, Vol. 32, N 7, 1007.
2. Morgunova V.A., Nikitina L.E., Plemenkov V.V. et al., *Zhur. Org. Khimii (Rus)* 2000, Vol. 36, N 4, 512.

THE ALKYLATION OF PHENOLS BY BICYCLIC MONOTERPENS IN THE PRESENCE OF $(PhO)_3Al$

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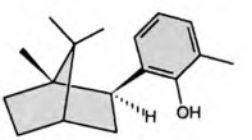
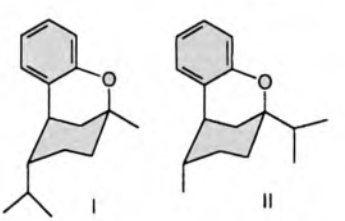
The alkylation of phenols by olefins in the presence of acid catalysts and $(PhO)_3Al$ is the most prevalent method of synthesis of alkyl phenols and terpenic phenols. In the previous paper we have shown, that alkylation of phenol by camphene proceeds selectively in the presence of aluminium phenolate. The main product of the reaction is *ortho*-alkylphenol (80%), in which the terpenic substituent has isobornylic structure.

The condensation of camphene with *ortho*-, *para*-, *meta*-cresols has been carried out on the base of these experiments. Aluminium cresolates were employed as catalysts. Besides the terpenic substituted cresols, ethers are formed, the yield of which is high enough – 40%. The same regularities of the formation of the reaction products and their structure, as in the case with phenol have been established on the base of NMR ^{13}C, ^{1}H spectra of terpenic cresols and X-ray methods of alkylated *ortho*-cresol. Alkylated *ortho*- and *para*-cresols have *exo*-configuration of oxyphenyl and isobornylic structure of alkyl radical.



Furthermore, alkylation of phenol by commercial accessible α-pinene in the presence of $(PhO)_3Al$ has been investigated. It was established, in this case phenylterpenic ethers are formed up to 60% and terpenic phenols – at about 20%. Composition and structure of products of the reaction considerably depend on the reagent ratio. We succeeded in isolating and establishing of two ethers **I** and **II** structure. With the increase of phenol number, the ratio of these ethers changes from 5:1 to 1:2. NMR ^{1}H and ^{13}C spectra of alkylphenylic ethers (**I** and **II**) are in accordance with the literature data[1]. Besides, with the change of reagents ratio, the yield of terpenic phenols and structure of their terpenic radical, also change. *Ortho*-phenols with isobornylic and isocamphenylic structure of terpenic residue have been isolated from the mixture of terpenic phenols obtained.



Thus, alkylation of phenol by α-pinene in the presence of aluminium phenolate proceeds rather selectively, but it is drawn towards the formation of alkyl-phenylic ethers.

[1] John C. Schmidhauser, Garold L. Bryant, Jr., Paul E. Donahue, Mary F. Garbauskas, Elizabeth A. Williams, J.Org.Chem. 1995, 60, 3612-3618.

SYNTHESIS OF OPTICALLY PURE AZAMACROCYCLIC LIGANDS FROM (+)-α-PINENE OF 65% OPTICAL PURITY

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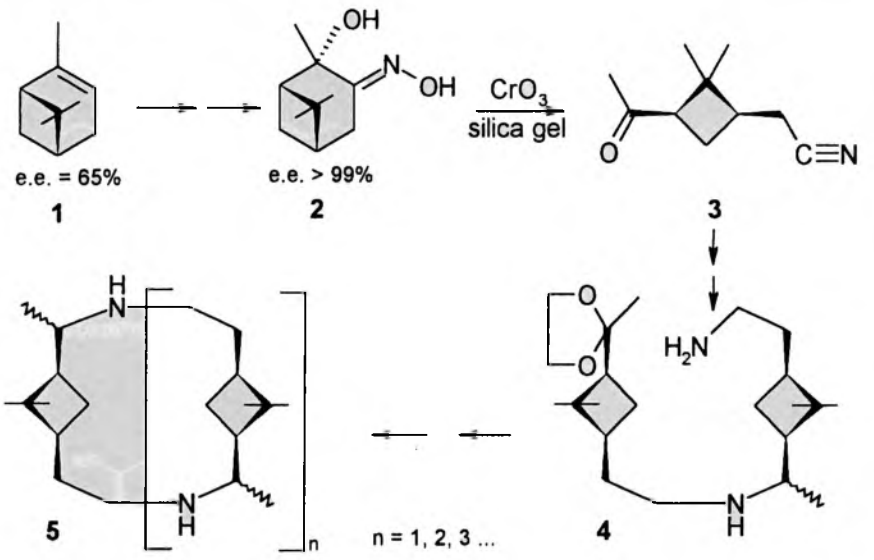
The facile enantiomeric enrichment of 2α-hydroxypinan-3-one oxime **2** makes it a convenient and inexpensive precursor of many pinane-backbone hydroxyamines, and optically pure *gem*-dimethylcyclobutane derivatives. Symmetric C_2, C_3....C_n azamacrocycles **5**, chiral ligands of potential catalysts, have been the compounds of our recent interest.

Optically pure oxime **2** is transformed into ketonitrile **3** by reaction with chromium (VI) oxide on silica gel, using toluene as solvent. Subsequent reactions:carbonyl group protection, reduction of nitrile to the amine, and its condensation with another ketonitrile molecule leads to imine. The reduction of imine (with simultaneous reduction of the nitrile moiety) gives amine **4**. The chain elongation of **4** is possible by further incorporation of next ketonitrile molecule and its reduction. Removal of carbonyl protection, formation of cyclic imine and its reduction, lead to azamacrocycles **5**. Stereocontrolled reduction of imines with LAH affords almost exclusively a single isomer, while traces of the other possible isomer may be removed on purification.



References: Markowicz S. W., Pokrzeptowicz K.; *XVIII Conference of Isoprenoids, Prachatice 1999; Chem. Listy, Symposia* **93**, S33 and references cited therein; Markowicz S. W., et al., in print.

(+)-AROMADENDRENE AS CHIRAL STARTING MATERIAL FOR THE SYNTHESIS OF INSECT PHEROMONES

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Joannes B.P.A. Wijnberg, Aede de Groot*

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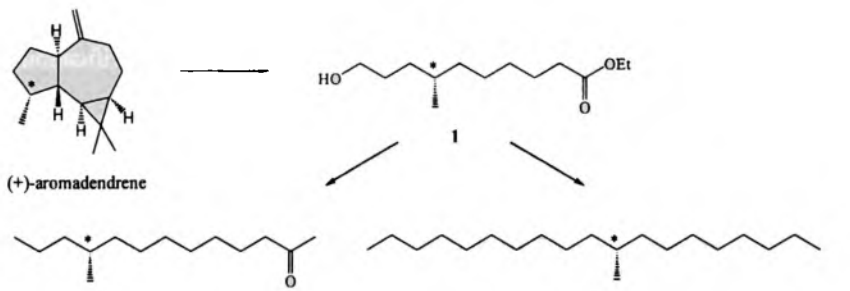
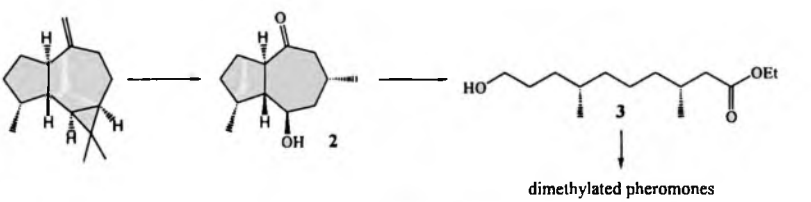
The tricyclic sesquiterpene (+)-aromadendrene is present in the distillation tail of *Eucalyptus globulus* in concentrations of 55-70%. Its five chiral centers and abundant availability make aromadendrene an attractive and cheap starting material for the synthesis of chiral products.
Previous research [1] on aromadendrene has led to the synthesis of various natural products such as (–)-kessane, (+)-maaliol and (–)-cubenol.
In the current research project, aromadendrene is used for the synthesis of insect pheromones. Examples of our target molecules are the pheromones of the southern corn rootworm and the cotton leafworm . The key intermediate in the synthetic route toward these pheromones is the lineair intermediate **1** with different functionalities at both ends of the chain.



Furthermore, aromadendrene has been used for the synthesis of dimethylated pheromones. In this route the extra methyl group is introduced in the aromadendrane skeleton, after which a similar route is followed as mentioned above.



[1] Gijsen, H.J.M.; Wijnberg, J.B.P.A.; De Groot, Ae., *Fortschritte der Chemie Organischer Naturstoffe*, 64, **1995**, 149-193

AMBERGRIS FRAGRANCE COMPOUNDS FROM LARIXOL AND LABDANOLIC ACID

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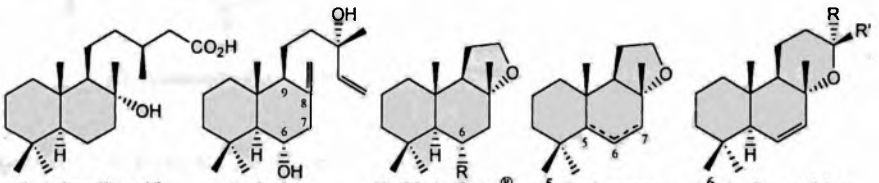
The gum of the *Cistus ladaniferus* consists mainly of labdanolic acid. Larixol can isolated easily from the resin of venice larch turpentine of the *Larix decidua*. It is of industrial importance to investigate which possible compounds can be synthesized from these readily available labdanes. Degradation of the C(9)-side chain will lead to intermediates suitable for the synthesis of interesting odour compounds like Ambrox®, and some of its C(6)-derivatives.



Starting from labdanolic acid a simple synthetic route to Ambrox® has been developed with an iododecarboxylation as key step.[1] A route from larixol to 6α-hydroxy Ambrox® is developed. This derivative, which itself has no fragrance properties, is used for the synthesis of Ambrox®-like compounds. Δ^5- and Δ^6-Ambroxene have been synthesized selectively and show pleasant odour properties.[2]

The combination of the hydroxyl group at C(6) and the exocyclic double bond at C(8) in larixol offers new possibilities for the synthesis of ring B modified odour compounds.[3]

The key transformation in the synthesis of several C(13) substituted Δ^6-tricyclic tetrahydropyranyl ethers (Δ^6-Ambra-oxides), consisted of the easy abstraction of the allylic hydroxyl group at C(6) followed by interception of the resulting carbocation by a nucleophilic hydroxyl group in the side chain.

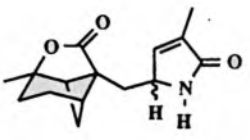
REFERENCES

1. Bolster, M. G.; Jansen, B. J. M.; de Groot, A. *Tetrahedron* **2001**, accepted for publication.
2. Bolster, M. G.; Jansen, B. J. M.; de Groot, A. *Tetrahedron* **2001**, accepted for publication.
3. Bolster, M. G.; Jansen, B. J. M.; de Groot, A. *Tetrahedron* **2001**, submitted.

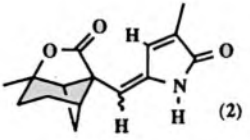
TOTAL SYNTHESIS OF (+)-DIHYDROAMPULLICIN. A FUNGAL METABOLITE WITH GROWTH REGULATING ACTIVITY ISOLATED FROM *Ampulliferina* Sp. No. 27.

Isabel Marcos Robles and Francisco Bermejo*. Departamento de Química Orgánica. Universidad de Salamanca, 37008 Salamanca, Spain.

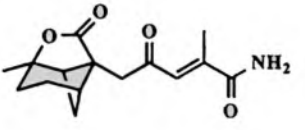
Dihydroampullicin (**1**) (Fig.1) was isolated from a culture filtrate of an *Ampulliferina*-like fungus sp. No. 27. and was biosynthetically related to other sesquiterpenic amides such as (-)-ampullicin (**2a**), (+)-isoampullicin (**2b**) and (+)-pinthunamide (**3**) by Kimura and col.[1, 2] All the representatives of this series of natural products have been claimed to develop growth-regulating properties since they accelerate the root growth of lettuce seedlings by 200% at doses between 30and 300 mg/L over the control.



(+)-Dihydroampullicin (1)



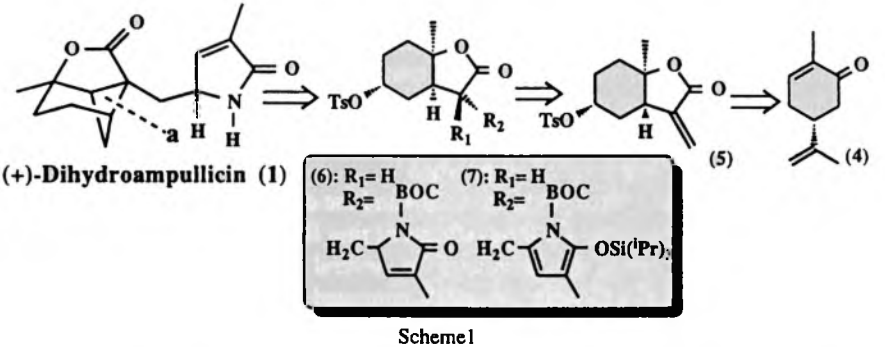
(+)-Ampullicin (2a)= (E)- (2)
(+)-Isoampullicin (2b)= (Z)- (2)



(+)-Pinthunamide (3)

Fig.1

According to our retrosynthetic analysis, the target molecule (+)-**1** was envisaged to be accesssible by internal displacement of tosylates (**6**) and (**7**) (Scheme1)[3].



Scheme1

Access to the tosylates (**6**) and (**7**) was based on Michael addition of heterocyclic synthons to α-methylene lactone (**5**), readily accessible from R-(-)-carvone (**4**).[4]

References:

1. Kimura, Y.; Nakajima, H.; Hamasaki, T.; Matsumoto, T.; Matsuda, Y.; Tsuneda, A.; *Agric. Biol. Chem.*, **1990**, *54*, 813-814.
2. Kimura, Y.; Matsumoto, T.; Nakajima, H.; Hamasaki, T.; Matsuda, Y., *Biosci. Biotech. Biochem.*, **1993**, *57*, 687-688.
3. Rico, R.; Bermejo, F., Tetrahedron Lett., **1995**, *36*, 7889-7892.
4. Marcos, I.; Redero, E.; and Bermejo, F., *Tetrahedron Lett.*, **2000**, *41*, 8451-8455.

SYNTHESIS OF SESQUITERPENE ANALOGS OF TAXOTERE®

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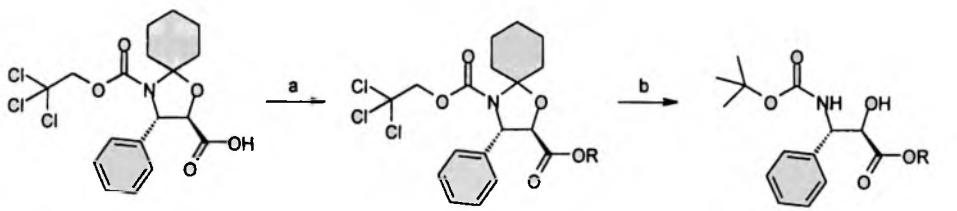
b. Department of Medical Microbiology, Medical University of Warsaw, Chałubińskiego 5, 02-004 Warsaw, Poland

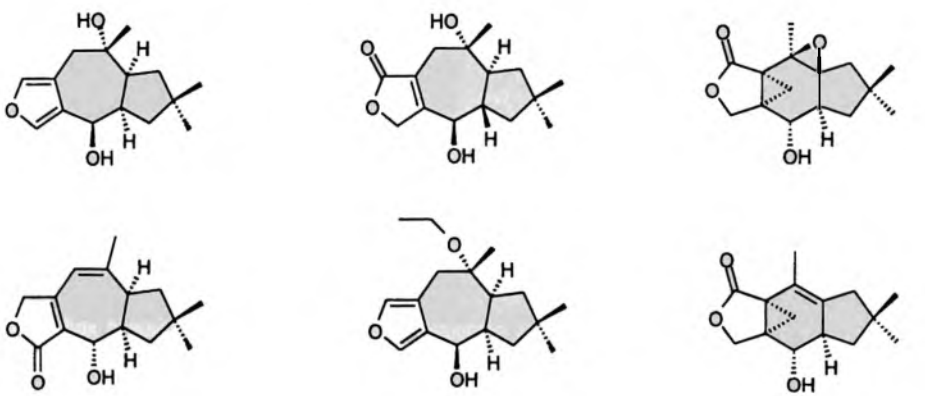
Important biological properties of Taxotere® i.e. 13-N-Boc-phenylisoserinate (2R, 3S) of 10-deacetylbaccatin III prompted us to synthesize N-Boc-phenylisoserinates of several sesquiterpenoic alcohols of *Lactarius* origin. Suitably protected N-acylphenylisoserine (**1**) gave appropriate esters (**2**), when reacted with sesquiterpenoic alcohols in the presence of DCC. Esters **2** were subjected to the reduction with zinc and then acylated with Boc_2O yielding the title N-Boc-phenylisoserinates (**3**).



1 2 3

a- R-OH (sesquiterpenoic alcohol), DCC, DMAP b-Zn, AcOH, MeOH then Boc_2O, Et_2O

The typical examples of sesquiterpenoic alcohols are shown below:



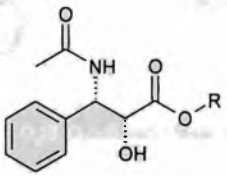
Investigation of the antifeedant and cytostatic properties of the compounds obtained is in progress.

Synthesis and Biological Properties of *N*-Acetylphenylisoserinates of Sesquiterpenoid Alcohols of *Lactarius* Origin

Rafal Barycki[a], Maria Gumułka[a], Marek Masnyk[a], Włodzimierz M. Daniewski[a], Halina Grabarczyk[b], Gerard Nowak[b], Mirosław Kobus[c], Ewa Krawczyk[c] and M. Łuczak[c].

a. Institute of Organic Chemistry, Polish Academy of Sciences, Kasprzaka 44/52, 01-224 Warsaw, Poland; b. Department of Medicinal Plants, Academy of Medicine, Mazowiecka 33, 60-623 Poznań, Poland; c. Department of Medical Microbiology, Medical University of Warsaw, Chałubińskiego 5, 02-004 Warsaw, Poland.

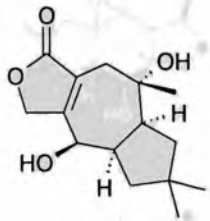
Important biological properties of Taxol® i.e. 13-(2'*R*,3'*S*)-*N*-benzoyl-3'-phenylisoserinate of baccatin III prompted us to synthesize and to check biological properties of (2'*R*,3'*S*)-*N*-acetyl--3'- phenylisoserinates of several sesquiterpenoid alcohols (**1**) of *Lactarius* origin. Phenylisoserinate methyl ester hydrochloride properly N,O-protected was hydrolized to the free acid, which was subsequently attached to the secondary hydroxyl group of the sesquiterpene. After deprotection, the free amine was acetylated.

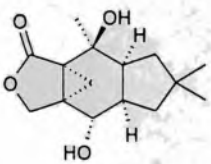


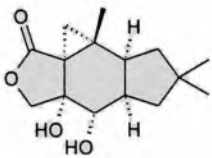
(1)

R-OH means the sesquiterpenoid alcohol

The typical examples of sesquiterpenoid alcohols are shown below:







Antifeedant and cytostatic properties of the compounds thus obtained are investigated and some of them will be presented.

SYNTHESIS OF 4-CARBOXY-*N*-BUTYLCARBAMOYL-SPACERED TAXOIDS AND BINDING TO BOVINE SERUM ALBUMIN

Torsten Blitzke[1)], Alexander Baranovsky[2)], and Bernd Schneider

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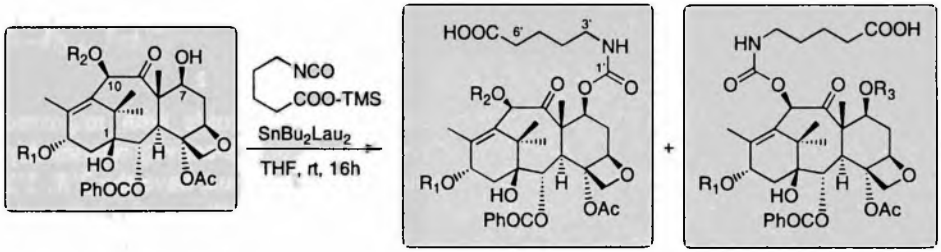
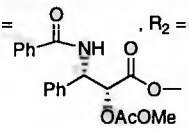
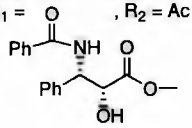
Carbamate side chains have been attached to positions 7 and 10, respectively, of 10-deacetylbaccatin III (DAB) and paclixtaxal (Taxol®) by reaction with trimethylsilyl protected 5-isocyanatopentanoic acid [1]. Conjugation of the carbamate derivatives to bovine-serum albumin was achieved by the carbodiimide method.



10-Deacetylbaccatin III: R_1 = H, R_2 = H;

Paclitaxel: R_1 =  , R_2 = Ac

7-*O*-(4-Carboxybutylcarbamoyl)-10 DAB III: R_1 = H, R_2 = H;

7-*O*-(4-Carboxybutylcarbamoyl)-paclitaxel: R_1 =  , R_2 = Ac

10-*O*-(4-Carboxybutylcarbamoyl)-10 DAB III: R_1 = H, R_3 = H

The approach described here appears to be generally suitable for binding hydroxylated molecules to proteins. Antigens obtained in this manner are useful for preparing antibodies and for use in the development of immunological assays.

[1] T. Blitzke, A. Baranovsky, B. Schneider, Helv. Chim. Acta **84**, in press (2001)

[1)] Present address: Bell, Flavors and Fragrances, Schimmelstr. 1, D-04205 Miltitz, Germany.
[2)] Present address: Institute of Bioorganic Chemistry, National Academy of Sciences of Belarus, Kuprevich Str., Minsk, Belarus.

AN EXPEDITIOUS ENTRY INTO 2'-SUBSTITUTED TAXOL ANALOGS

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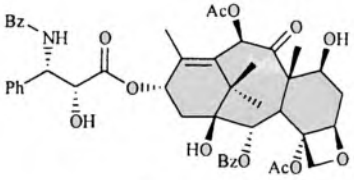
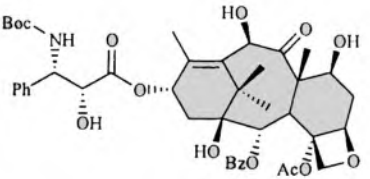
Over the past few years, considerable attention has been given to the synthesis and clinical and pharmacological characterizations of Taxol (**1**) (paclitaxel) and its semi-synthetic analogue Taxotère (**2**) (docetaxel). These two drugs are very active against various cancer diseases, but not devoid of side-effects and shortcomings, including the development of multi-drug resistance.



1



2

For these reasons, the synthesis of newer generations of taxoids with improved pharmacological properties is still a hot field of research . Structure-activity relationship studies have established that the isoserine side-chain of taxanes must have the 2'*R*, 3'*S* configuration for optimal activity. Provided that this 2'R,3'S configuration is maintained, an additional methyl at C-2' is well tolerated, as is its tethering to the 3'-phenyl. Kant[1], Greene[2] and Ojima[3] have independently developed different synthetic protocols for the synthesis of *N,O*-protected-2'-alkylisoserines and their synthetic equivalent *N*-acyl-β-lactams. These procedures reaquire several protection-deprotection sequences and the use of expensive chiral inductors. We have developed an alternative entry to 2'-alkylisoserines which capitalizes on the use of naturally occurring (*S*)-α-hydroxy acids, a cheap starting material. This procedure was applied to the synthesis of various 2'*R*, 3'*S*- 2-substituted isoserines and their synthetic equivalent *N*-acyl-β-lactams, which were then coupled with suitably protected baccatins. The biological activity of the resulting 2'-substituted taxoids will be discussed, highlighting structure-activity relationship within this interesting class of anticancer agents.

[1] Kant, J.; Schwartz, W.S.; Fairchild, C.; Gao, Q.; Huang, S.; Long, B.H.; Kadow, J.F.; Langley, D.R.; Farina, V., Vyas, D. *Tetrahedron Lett.* **1996**, *37*, 6495-6498.
[2] Denis, J.-N.; Fkyerat, A., Gimbert, Y.; Coutterez, C., Mantellier, P.; Jost, S.; Greene A.E. *J. Chem. Soc. Perkin Trans. 1*, **1995**, 1811-1816.
[3] Ojima, I.; Wang, T.; Delaloge, F. *Tetrahedron Lett.* **1998**, *39*, 3663-3666.

Regioselective Norrish Type I Cleavage of a Trimethylsilyl-substituted Estrone Derivative

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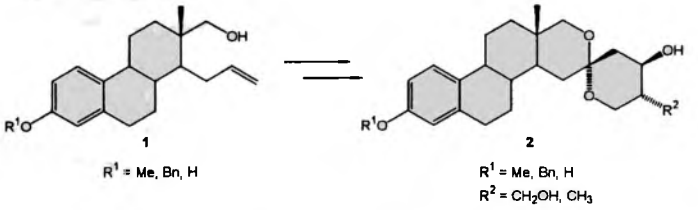
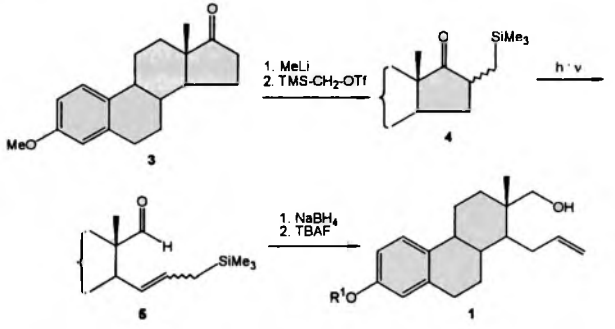
H-6720 Szeged, Dóm t. 8., Hungary

Recently, we reported the synthesis of new estrone-talaromycin natural product hybrids.[1,2] This new class of compounds (**2**) exhibits anticancer activity.



The earlier synthesis method of **1** implies an unselective reaction step.[3] Now we report a new reaction sequence, using a regioselective photochemical cleavage as a key transformation.
After introduction the trimethylsilylmethyl group into position C-16 of estrone-3-methyl ether, a photochemically induced Norrish type I cleavage leads to the allylsilane carbaldehyde **5**. The radical stabilizing ability of the silicium atom in β-position is strong enough to direct the reaction in this way instead of a cleavage between the quaternary center C-16 and C-17. After reduction of the carbonyl group the silicon moiety can be removed in a usual way, producing the required compound **1**.



[1]L. F. Tietze, Gy. Schneider, J. Wölfling, T. Nöbel, C. Wulff, I. Schuberth, A. Rübeling, *Angew. Chem.* **1998**, *110*, 2644-2646, *Angew. Chem. Int. Ed.* **1998**, *37*, 2469-2470.
[2]L. F. Tietze, Gy. Schneider, J. Wölfling, A. Fecher, T. Nöbel, S. Petersen, I. Schuberth, C. Wulff, *Chem. Eur. J.* **2000**, *6*, 3755-3760.
[3]G. Schneider, S. Bottka, L. Hackler, J. Wölfling, P. Sohár, *Liebigs Ann. Chem.* **1989**, 263-267.

SYNTHESIS OF N,N-BIS[2-(2-PYRIDYL)ETHYL]AMINO STEROIDS AS CHIRAL LIGANDS FOR COPPER-MEDIATED O_2-ACTIVATION

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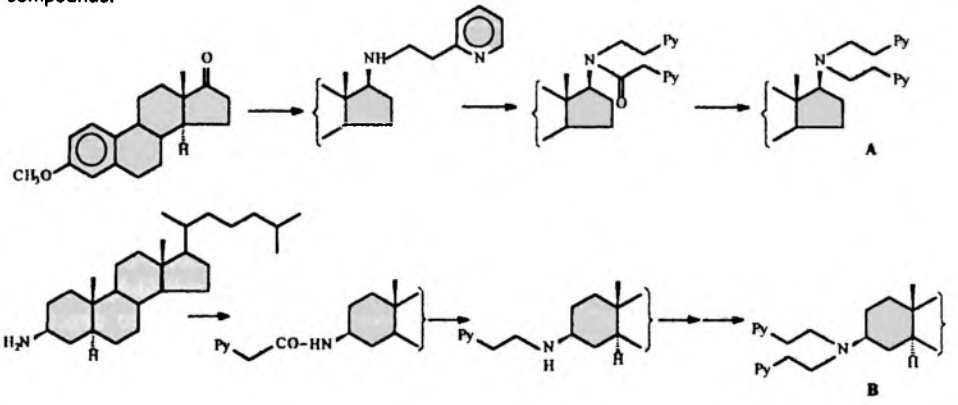
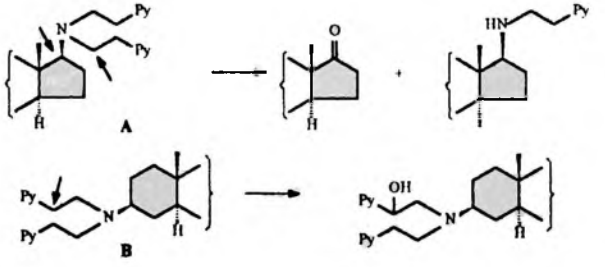
Copper(II) complexes of N,N-bis[2-(2-pyridyl)ethyl]amino compounds (RPY2), which are able to bind and activate O_2, are suitable models for the copper-containing enzyme dopamine β-hydroxylase. Homochiral RPY2 ligands have not yet been synthesized. We have therefore developed routes for the synthesis of such steroidal compounds [1]. The classical synthesis – addition of primary amines to 2-vinyl pyridine – was not very successful using steroidal amines. A new route is the reductive amination of 17-ketones with 2-(2-pyridyl)ethylamine/$NaBH_4$ to the 17β-N-[2-(2-pyridyl)ethyl]amino compounds; acylation of these with 2-pyridylacetic acid/N,N'-carbonyldiimidazole to the corresponding amides and reduction with B_2H_6 to finally yield the N,N-bis[2-(pyridyl)ethyl]amino compounds. Another successful method is the corresponding acylation of primary steroidal amines, reduction with B_2H_6 and repeating this sequence to the desired compounds.



It could be show that the copper(I) complexes of these compounds can activate O_2. Different intramoelcular oxidation reactions take place in dependence of the position: the 17β-compound A was oxidized at the α-position giving 17-ketone and the 17-N-[2-(2-pyridyl)ethyl]amino compound, whereas the 3β-compound B was oxidized at the α-CH_2-group of the pyridineethyl unit.



It seems that the conformation of the ligands is responsible for this different behaviour. Intermolecular oxidation reactions are under way.

[1] M. Gonschior, M. Kötteritzsch, M. Rost, B. Schönecker and R. Wyrwa, *Tetrahedron: Asymmetry*, **2000**, *11*, 2159-2182

ADDITION REACTIONS AT THE 16(17)-DOUBLE BOND OF 13α-3-METHOXY-ESTRA-1,3,5(10),16-TETRAENE

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[a] Institute of Organic Chemistry, University of Szeged, H-6720 Szeged, Hungary

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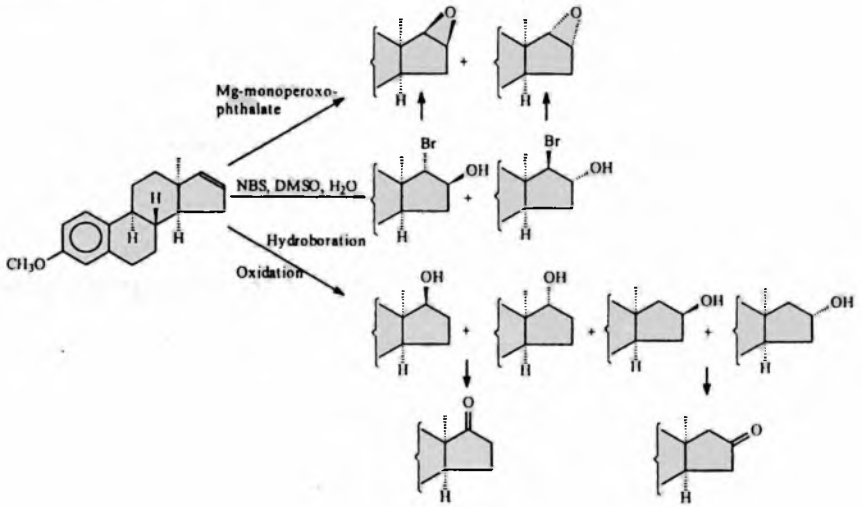
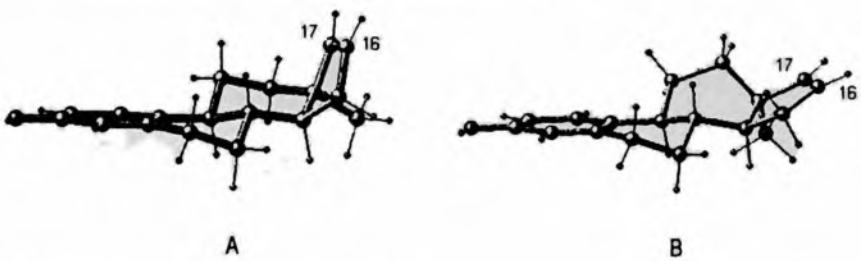
In comparison to the addition at the 16(17)-double bond of 13β-steroids (α-attack), the epoxidation reaction and the addition of HOBr, diborane and 9-BBN to the analogous 13α-compound have been investigated. In all cases α- and β-attack could be observed. In addition, hydroboration/oxidation gave 16- and 17-alcohols.



The ring closure of the 16β,17α-bromohydrine (main product of the HOBr-addition) and the oxidation of the 16-alcohols (main products of the hydroboration/oxidation reaction) open pathways to the 16β,17β-epoxide and the 16-ketone.

In contrast to the addition reactions observed for the normal 13β-series, two conformations (A and B) must be considered either for the olefin or for the reaction intermediates [1].



A B

1 B. Schönecker, C. Lange, M. Kötteritzsch, W. Günther, J. Weston, E. Anders, H. Görls, *J. Org. Chem.*, **2000**, 65, 5487-97

Erzsebet Mernyak thanks the Sigma-Aldrich Kft. (Hungary) for a conference attendance support.

Studies on synthesis of 13,14-seco steroids

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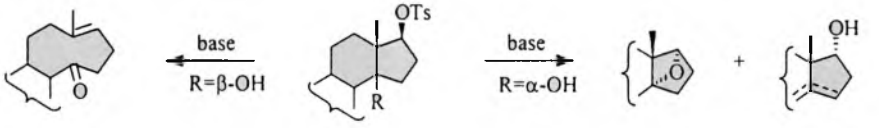
[a]*Institute of Bioorganic Chemistry, National Academy of Sciences of Belarus, Kuprevich str., 5/2, 220141 Minsk, Belarus*

[b]*Department of Medicinal Chemistry, N.V. Organon, P.O. Box 20, 5340 BH Oss, The Netherlands*

[c]*Wageningen University, Laboratory of Organic Chemistry and Research Center, Dreijenplein 8, 6703 HB Wageningen, The Netherlands*

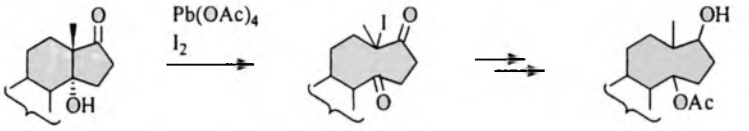
Two synthetic approaches toward 13,14-seco steroids have been investigated. The first one is based on Grob fragmentation of the hydroxy tosylates **1**. The reaction was shown to proceed with formation of the desired seco steroid **2** for 14β-alcohols only.



2 **1** **3** **4**

Compounds with 14α-hydroxy group gave under similar conditions a mixture of the oxetane **3** and alcohols **4**.

The second approach to 13,14-seco steroids included radical oxidation of 14α-hydroxy-17-ketones **5**.



5 **6** **7**

The 13-iodine in **6** could be stereoselectively removed. The functional groups at C_{14} and C_{17} in the obtained diketone were differentiated in a few steps to give the seco steroid **7**.

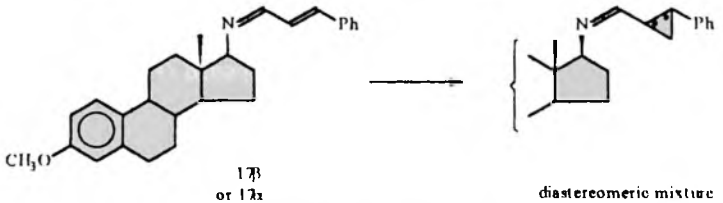
STEROIDS AS CHIRAL AUXILIARIES: HIGHLY DIASTEREOSELECTIVE PALLADIUM-CATALYZED CYCLOPROPANATION OF α,β-UNSATURATED ALDIMINES

B. Schönecker[a], M. Dubs[a], H. Dleks[a], M. Kötteritzsch[a], W. Günther[a], H. Görls[b] and W. Poppitz[b]

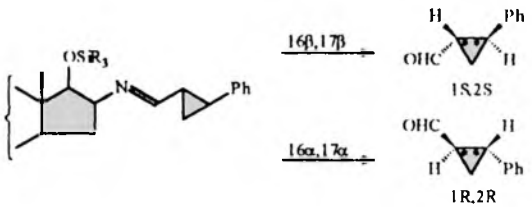
[a] Institut für Organische Chemie und Makromolekulare Chemie, Friedrich-Schiller-Universität Jena, D-07743 Jena, Germany, E-mail: c8scbr@uni-jena.de

[b] Institut für Anorganische und Analytische Chemie, Friedrich-Schiller-Universität Jena, D-07743 Jena, Germany

In connection with investigations on the reactivity of the simple to prepare α,β-unsaturated steroidal aldimines (condensation of primary steroidal amines with α,β-unsaturated aldehydes), we became interested in using these compounds for Pd-catalyzed cyclopropanations with diazomethane. In particular, we employed D-ring substituted 3-methoxy-estra-1,3,5(10)-trienes because of the well-defined and restricted conformation of the anellated cyclopentane ring. The reaction of the α,β-unsaturated imines, obtained from the 17β- and the 17α-amine with (E)-cinnamic aldehyde, $Pd(OAc)_2$ and CH_2N_2 proceeds with high chemoselectivity at the C=C bond to form the desired cyclopropanes, but the diastereoselectivity is low.



Using cis-16-amino-17-silyloxy compounds for condensation and cyclopropanation, high diastereoselectivities can be obtained (de > 90 %). The configuration of the cyclopropanes clearly depends on the configuration of the O-silylated imine. Simple hydrolysis by column chromatography on silica gel furnished the chiral trans-phenyl-cyclopropane aldehydes (ee > 90 %) and the corresponding O-silylated steroidal amine.



A preferred conformation of the unsaturated imines, determined by x-ray analysis, is responsible for the high diastereoselectivity. This method seems to be useful, because a catalytic enantioslective route of cyclopropanation with chiral ligands for cinnamic acid derivatives has been unsuccessful [1].

[1] S. E. Denmark, R. A. Stavenger, A. M. Fauchier and J. P. Edwards, *J. Org. Chem.* **1997**, 62, 3375

LIGANDS FOR AFFINITY CHROMATOGRAPHY: SYNTHESIS OF A BRASSINOSTEROID MODEL

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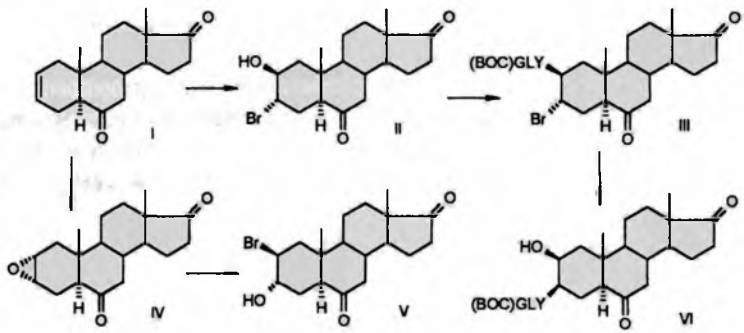
In the course of our studies on structure-activity relationship of brassinosteroid[1-3] there was a need for the synthesis of a compound which would bind to affinity carriers potentially useful for isolation of protein receptors from plant extracts. In this case we planned to bind the brassinosteroid part through an A ring function.

To elaborate the synthesis, we used 6,7-dioxo-5α-androst-2-ene (*I*) as the starting material. An addition of hypobromous acid to a 2,3-double bond afforded 3α-bromo-2β-hydroxy-5α-androstan-6,17-dione *(II)*. To determine the configuration of the adduct, we synthesized an isomeric bromohydrine: 2β-bromo-3α-hydroxy-5α-androstan-6,17-dione (*V*) was formed by a hydrobromic acid cleavage of epoxide *(IV)* obtained on epoxidation of olefin *I*. The structures of these two bromohydrines follow from their NMR spectra before and after treatment with TAI. Bromohydrine *II* was used for synthesis (BOC)glycine-ester *(III)* which on acetolysis of bromine atom afforded compound *(VI)* which will be used for binding experiment.



References

1. Slavíková B., Kasal A., Kohout L.: *Collect. Czech. Chem. Commun.* **63,** 655-661, (1998).
2. Brosa C., Zamora I., Terricabras E., Kohout L.: *Collect. Czech. Chem. Commun.* **63,** 1635-1645, (1998).
3. Kohout L., Chodounská H., Macek T., Strnad M.: *Collect. Czech. Chem. Commun.* **65,** 1754-1761, (2000).

The authors acknowledge financial support by the Grant Agency of the Czech Republic, Grant No. 203/01/0083.

Chiral Nitrogen-Containing Steroid Ligands For Group 4 Metals

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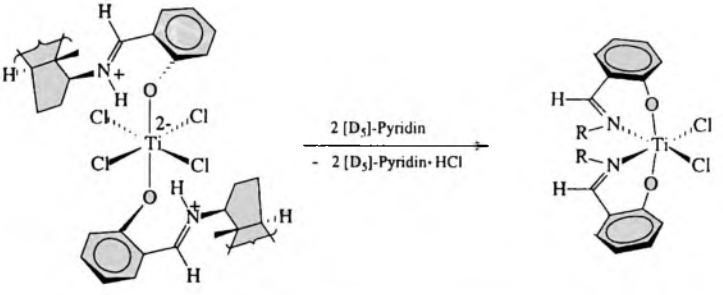
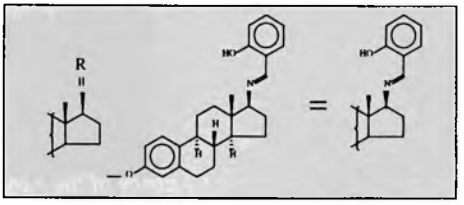
*Westfälische Wilhelms-Universität, Organisch-Chemisches Institut, Corrensstraße 40, D-48149 Münster.
**Friedrich-Schiller-Universität Jena, Institut für Organische und Makromolekulare Chemie, Humboldstraße 10, D-07743 Jena.

In recent years a large number of organometallic complexes with modified salicylaldiminato ligands has been investigated. Current uses of these ligands include the production of syndiotactic polyproylene[1]and the use in ethylene polymerization with neutral catalysts[2], the enantioselective hydrocyanation of imines[3] and the synthesis of syndiotactic polylactic acid.[4] In order to obtain metal complexes of chiral Schiff bases, varios steroid amines of the estrone and cholestane series are prepared diastereoselectively and then condensed with salicyl aldehyde. In the subsequent reaction of two equivalents of the salicylaldiminato ligand with titanium tetrachloride a chiral dibetainic adduct with C_2 symmetry can be isolated.[5]The reaction of the adduct with a Brønsted base leads to HCl elimination and to the formation of a chiral C_2 symmetric titanium complex. It is being investigated if steroid ligands can form similar complexes.





Literature:

[1] J. Tian, G.W. Coates, *Angew. Chem.* **2000**, 112, 3772.
[2] T. R. Younkin, E. F. Connor, J. I. Henderson, S. K. Friedrich, R. H. Grubbs, D. A. Bansleben, *Science* **2000**, 287, 460.
[3] C. A. Krueger, K. W. Kuntz, C. D. Dzierba, W. G. Wirschun, J. D. Gleason, M. L. Snapper, A. H. Hoveyda, *J. Am. Chem. Soc.* **1999**, 121, 4284.
[4] T. M. Ovitt, G. W. Coates, *J. Am. Chem. Soc.* **1999**, 121, 4072.
[5] J. Strauch, T. H. Warren, G. Erker, R. Fröhlich, P. Saarenketo, *Inorg. Chem. Acta* **2000**, 300.

AMINO ANALOGUES OF NEUROSTEROIDS

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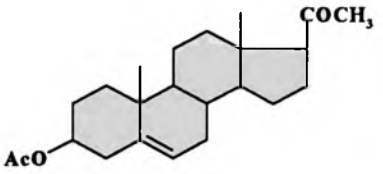
The presence of a 3α hydroxy group was considered essential for neuronal activity involving $GABA_A$ receptors. While substituting fluorine for the 3α hydroxyl in allopregnanolone, we obtained a product significantly reducing aggressivity in mice.[1]

In a search for a new type of neuroactive steroids, we converted pregnenolone acetate (**1**) into 3α-amino-5α-pregnan-20-one (**2**) and 3α-amino-5β-pregnan-20-one (**3**). Their effect on activity of two neurotransmitters will be reported.



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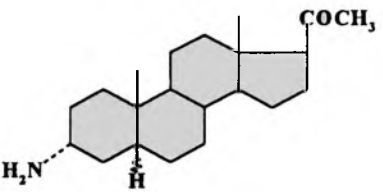

2, R = 5α-H
3, R = 5β-H

References

1. Slavíková B., Kasal A., Uhlířová L., Kršiak M., Chodounská H., Kohout L.: Suppressing Aggressive Behavior with Analogs of Allopregnanolone. *Steroids* **66**, 99-105 (2001).

Acknowledgements

The authors are indebted to the Grant Agency of the Academy of Sciences of the Czech Republic (S5011 07) and the Grant Agency of the Czech Republic (203/01/0084).

SYNTHESIS AND ACTIVITY OF B-NOR-ALLOPREGNANOLONE ANALOGUES

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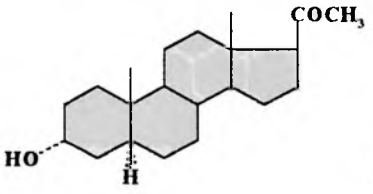
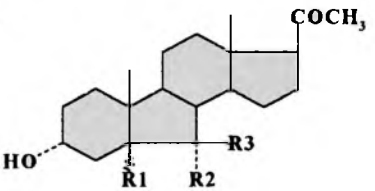
Several B-nor analogues of endogenous neurosteroids - allopregnanolone (I) and pregnanolone (II), compounds III to VI were synthesized from pregnenolone acetate (see ref. [1],[2]). Positive allosteric modulation of γ-aminobutyric acid by these compounds was assessed in two ways:

1. in vitro, using $GABA_A$ receptor membranes: by measurement of the muscimol and TBPS binding before and after application of the compounds;
2. in vitro, using cultured neurons: by measurement of their effect on the flunitrazepam binding and on Cl^- flux across the neuronal membrane.



I, 5α-H
II, 5β-H



III, R1=R2=R3=H
IV, R1+R2=α,α-O, R3=H
V, R1+R2-β,β-O, R3=H
VI, R1=H, R2+R3=O

References

1. Kasal A.: Epalons. Synthesis of 7-Norallopregnanolone. *Coll. Czech. Chem. Commun.* **64**, 1471-1478 (1999).
2. Kasal A.: Epalons: 6-Substituted Derivatives of 7-Norallopregnanolone. *Tetrahedron* **56**, 3559-3565 (2000).

Acknowledgements

The authors are indebted to the Grant Agency of the Academy of Sciences of the Czech Republic (S5011 07).

FLUORO ANALOGUES OF ALLOPREGNANOLONE, SYNTHESIS AND ACTIVITY

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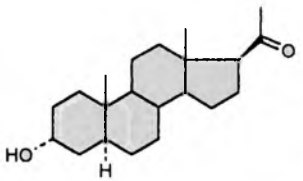
The practical use of neurosteroids (e.g. allopregnanolone, **1**), able to modulate positively effects of γ-aminobutyric acid on neurons, is restricted by its fast clearance and low solubility in body liquids. To increase its metabolic stability, we decided to substitute a fluoro atom for a hydroxy group in position 3α. Striving for increased hydrophilicity of our products, we modified the structure of fluoride **2** in positions 2, 12, 16, and 17, where the newly added substituents were not expected to jeopardize their activity.



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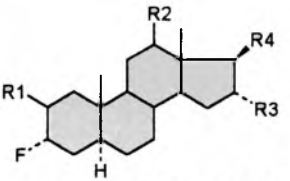

2, R^1, R^2, R^3 = H, R^4 = $COCH_3$

3α-Fluoro compounds were prepared by treatment of 3β-hydroxy derivatives with diethylaminosulfur trifluoride (DAST) at room temperature[1]. Yields varied from 30 - 50%.

Biological activity of all the resulting 3α-fluoro and 3α-hydroxy derivatives, assessed by *in vitro* tests using $GABA_A$ receptors [^{3}H]-muscimol and [^{35}S]-*tert.*butyl-bicyclo[2.2.2]phosporothionate as ligands, will be reported.

References

1. Slavíková B., Kasal A., Uhlířová L., Kršiak M., Chodounská H., Kohout L.: *Steroids* **66**, 99, (2001).

Acknowledgements

The authors are indebted to the Grant Agency of the Academy of Sciences of the Czech Republic (S5011 07).

STEROID MODULATORS OF NMDA RECEPTORS[1]

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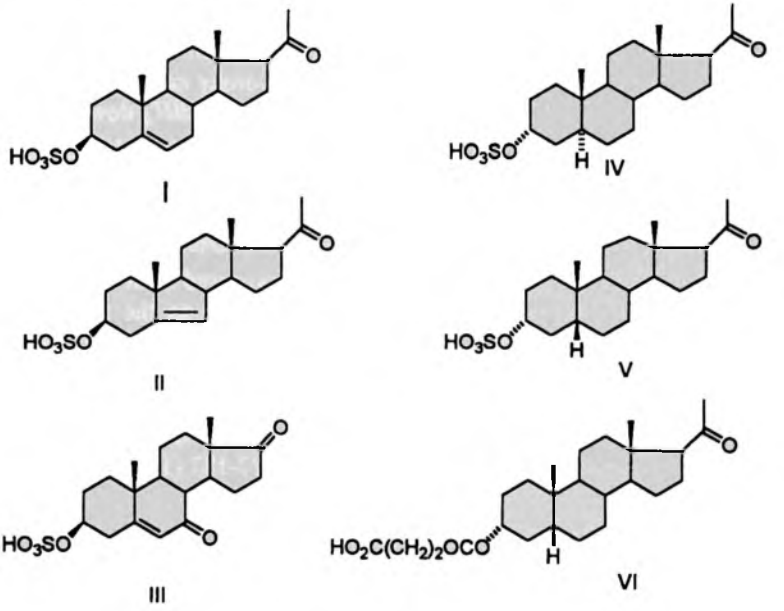
***Institute of Physiology, Academy of Sciences of the Czech Republic, Vídeňská 1083, 142 20 Praha 4, Czech Republic*

Sulphates of pregnane and androstane derivatives **I** - **V** were synthesised by reaction with pyridinium sodium sulphate in methylene chloride[2] or pyridine. Hemisuccinate **VI** was prepared using succinic anhydride.

Electrical recordings from whole-cell and outside-out configurations of the patch-clamp technique were used to characterise effects of the products on neurones in acutely prepared spinal cord slices[3].



References:

1. Compagnone N. A., Mellon, S. H.: *Front. Neuroendocrinol.* **2000,** 1, *21.*
2. Arnoštová L., Pouzar V., Drašar P.: *Steroids* **1992**, 233, 57.
3. Paleček J. I., Abdrachmanova G., Vlachová V., Vyklický L. Jr.: *Eur. J. Neurosci.* **1999**, 827, 11.

The project was supported by the grant of the Grant Agency of the Czech Republic, No. 203/01/0084.

CARDENAMIDES FROM CARDENOLIDES: CARDIAC AND ANTI-CANCER ACTIVITY*

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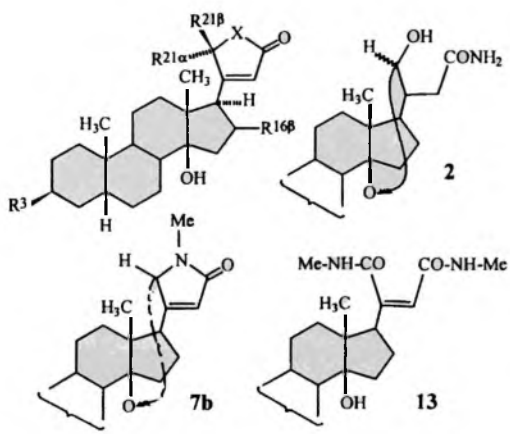
The cardenolide digitoxigenin **1** reacts with *ammonia* in methanol to the halfacetal amide **2** (yield: 53%) with isomerization. The cardenamide **3** can be formed by treatment of **2** with $SbCl_3/SiO_2$ at 120°C (54%) or ethanol/H_2SO_4 under reflux (33%) with loss of 14ß-OH and formation of a Δ^{14} double bond. Reaction of the (21R)-21-bromo cardenolide **4a** (from **4** by bromination with N-bromosuccinimide) with *ammonia* or *methylamine* leads to the (21R)-21-hydroxy cardenamide **5a** (51%) or **5b** (60%) containing 14ß-OH. Treatment of **5a** or **5b** with $NaBH_4$ does not remove the 21-OH group. *SOCl2* and **5b** do not form the 21ξ-Cl derivative **6b** but the (21S)-14,21-epoxi lactame **7b** (71%). **5b** it is reacted with *tosyl chloride* to the (21R)-tosylate **8b** (48%) followed by reduction with *zinc* to the 14ß-hydroxy-cardeneamide **9b** (43%). Otherwise, oxidation of **5a** or **5b** with active MnO_2 leads to the 21-oxo lactame **10a** (72%) or **10b** (70%). In the analogous way **10c** is prepared from **11**. The 21,21-dibromo gitoxigenin 3,16-diacetate **12** reacts with *methyl amine* to the ring-opened 21,23-bis methylamide **13** (23% related to **4**). In the **ATPase test** (guinea pig heart/human heart) **lactames** (except **7b**) show lower activity than the lactone anlogues. Furthermore, the **maleinimides 10a-c** show **-SH group reactivity** and **strong anti-cancer activity** (most active: **10c**). **X-ray structure** of **10a** shows only small changes compared to that of **4.** *For references see: Anti-Cancer Drug Design **10**, 177-187 (1995).

No.	R^3	$R^{16\beta}$	X	$R^{21\alpha}$	$R^{21\beta}$
1	OH	H	O	H	H
3*	OH	H	NH	H	H
4	OAc	OAc	O	H	H
4a	OAc	OAc	O	H	Br
5a	OAc	OAc	NH	H	OH
5b	OAc	OAc	N-Me	H	OH
6b	OAc	OAc	N-Me	H	ξ-Cl
8b	OAc	OAc	N-Me	H	OTs
9b	OAc	OAc	N-Me	H	H
10a	OAc	OAc	NH	=O	
10b	OAc	OAc	N-Me	=O	
10c	TATD	OAc	N-Me	=O	
11	TATD	OAc	O	H	H
12	OAc	OAc	O	Br	Br



* Δ^{14}

TATD = O-3',3'',3''',4'''-tetra-O-acetyl-(digitoxosyl)$_3$

SYNTHESIS OF
(20R)- AND (20S)- METHYL-3-OXO-CHOLA-1,4,22-TRIENE-24-OATE

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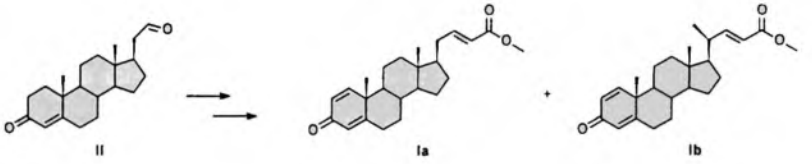
Naturstoffchemie Universität Dortmund, Otto-Hahn-Str. 6, D-44221 Dortmund, Germany

While searching for new antifouling agents, Tomono et al[1] isolated the title compound **I** from an octocoral *Dendronephthya sp.* It shows no antifouling activity against barnacle (Balanus amphitrite) larvae, but is lethal to barnacle larvae at a concentration of 100 µg/ml (LD_{100}). Since the configuration of **I** has not yet been determined we report herein the synthesis of the (20R)- and the (20S)-epimer starting from the same (20S)-aldehyde **II**.



A mixture of the title compounds **Ia/Ib** has been synthesized via epimerization at C-20 of **II** with H_2SO_4/EtOH[2], Horner-Wittig reaction with triethylphosphonoacetate[3], reesterification with p-TsOH in methanol and dehydration with DDQ[4]. Repeated cristallizations from cyclohexane allow for an easy separation and the relative configuration of both compounds has been determined by X-ray analysis[5,6]. Since the analytic data of **Ib** are in good agreement with the published data of the naturally occuring steroid, one can characterize the latter one unequivocally as (20S)-methyl-3-oxo-chola-1,4,22-triene-24-oate.

References:

1 Y. Tomono, H. Hirota, Y. Imahara, N. Fusetani *J.Nat.Prod.* **1999**, 62, 1538-1541.

2 M.E. Herr, F.W. Heyl, *J.Am.Chem.Soc.* **1952**, 74, 3627.

3 E.G. Baggiolini, J.A. Iacobelli, B.M. Hennessy, A.D. Batcho, J.F. Sereno, M.R. Usoković *J.Org. chem*, **1986**,51,3098.

4 J.Fried, J.A.Edwards **1972** *Organic reactions in steroid chemistry* New York Vol. 1, S.317.

5 M. Linker, M. Schürmann, H. Preut, W. Kreiser, *Acta Cryst. E* **2001**, in press.

6 M. Linker, M. Schürmann, H. Preut, W. Kreiser, *Acta Cryst. E* **2001**, in press.

SYNTHESIS OF ALKENES FROM ALDEHYDE ARYLSULFONYLHYDRAZONES AND NITRILES. FACILE TRANSFORMATION CHOLIC ACID INTO CHOLESTANE DERIVATIVES

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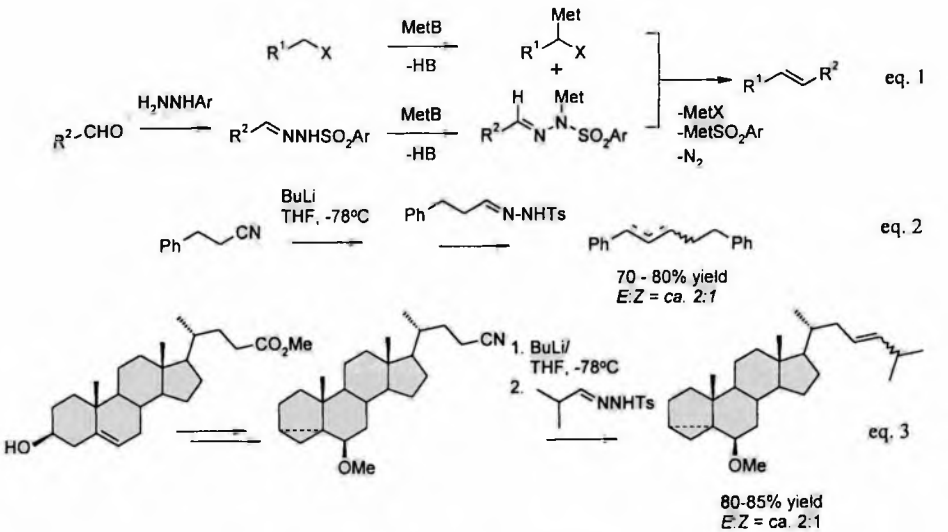
Institute of Organic Chemistry, Polish Academy of Sciences,
Kasprzaka 44/52, 01-224 Warsaw, Poland

Aldehyde arylsulfonylhydrazones react with organometallics bearing in the α-position a leaving group to afford the corresponding olefins,[1] eq. 1. We have recently developed a versatile method for olefin synthesis using α-magnesio sulfones (eq. 1, X=SO_2Ph, Met=MgBu) and aldehyde arylsunfonylhydrazones.[2] The use of nitriles and various metallating agents in this olefination reaction, as illustrated in eq. 2, will be reported. A facile transformation of cholic acid *via* the respective nitrile into cholestane derivatives will be presented (eq. 3).



(1) Vedejs, E.; Dolphin, J. M.; Stolle, W. T. *J. Am. Chem. Soc.* **1979**, *101*, 249-251.

(2) Kurek-Tyrlik, A.; Marczak, S.; Michalak, K.; Wicha, J. *Synlett* **2000**, 547-549.

OLEFINATION VIA REACTION OF ALDEHYDE ARYLSULFONYLHYDRAZONES AND SULFONES. STEROID SIDE CHAIN CONSTRUCTION

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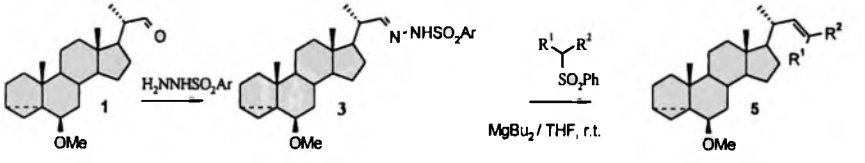
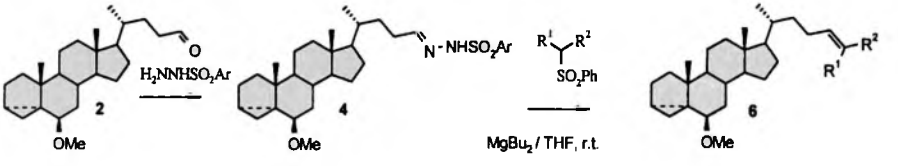
In this communication we present our studies on the new synthesis of alkenes involving coupling of magnesium derivative of aldehyde arylsulfonylhydrazones and α-magnesium derivatives of alkylaryl sulfones.[1] Tosylhydrazones **3** and **4**, prepared from common steroid aldehydes (**1** and **2**), are transformed easily into alkenes **5** and **6** respectively (differ in the position of a double bond in the steroid side chain). The differences in reactivity of **3** and **4** toward various sulfones will be discus. A one pot procedure develop to increase the overall yield of the olefination procedure in case when arylsulfonylhydrazone has a limited stability (e.g. compound **4**) will also be presented.





1. Kurek-Tyrlik, A., Marczak, S., Michalak, K, Wicha, J. *Synthesis of Alkenes by the Reaction of Magnesium Sulfone Derivatives with Arylsulfonylhydrazones of Aldehydes.* Synlett, 2000: p. 547-549.

BIOACTIVITY COMPARISON OF 24-EPIBRASSINOLIDE AND ANDROSTANE ANALOGUE OF CASTASTERONE

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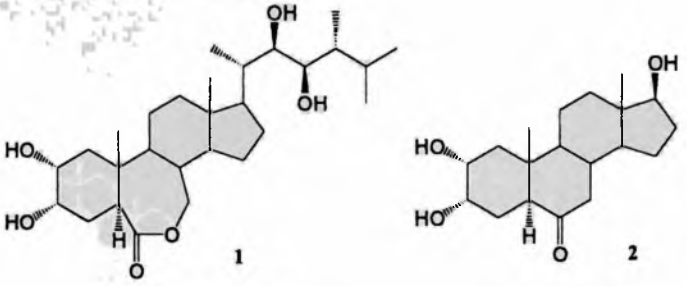
Biological activities of 24-epibrassinolide **1** [(22R,23R,24R)-2α,3α,22,23-tetrahydroxy-24-methyl-B-homo-7-oxa-5α-cholestan-6-one] and an androstane analogue of castasterone **2** [2α,3α,17ß-trihydroxy-5α-androstan-6-one] will be compared.



The activities of these two compounds are different in different bioassays. The biological activity in the following tests will be compared:

Second bean internode bioassay
Growth of roots
Rice lamina inclination test
Growth of *Arabidopsis thaliana* mutant
Activity of Hill reaction in corn
Content of photosynthetic pigments in corn
Activity in field test on wheat
Activity in field test on sugar beat

This work was supported by the Grant Agencies of the Czech Republic (grant No. 203/01/0083) and of Academy of Sciences of the Czech Republic (grant No. 40 55803).

NEW BRASSINOSTEROID DERIVATIVES HAVING AN HYDROXYL IN THE ANDROSTANE ESTER SIDE CHAIN

CARME BROSA[a], LADISLAV KOHOUT[b], MARIA PIQUÉ[a], MARC VILAPLANA[a]

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In the field of brassinosteroids the search for new active brassinosteroid analogs with a good synthetic cost/activity relationship is still a challenge for further application in agriculture of such interesting compounds.

With this aim, we have developed a model based on brassinosteroid-receptor interaction which is useful in explaining the activity of different brassinosteroids from the structural point of view.[1] Following this model, we have found that the electrostatic charges play an important role in explaining the activity and that the hydrogen-bonding could be one of the type of interaction that could take place on binding.

Our QSAR model suggests that the region with high probability of hydrogen-bonding near to the one of the 23R-OH of brassinolide (**1**) is essential for eliciting activity.

The lack of activity of **2** in rice lamina inclination test can be explained if one considers that no interaction is observed in the zone close to the 23R-OH of brassinolide (**1**).[2]

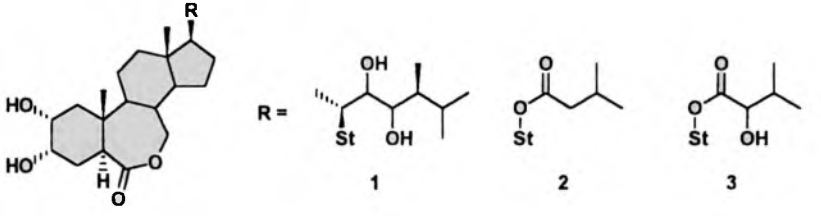
With the aim to assess this, a set of new androstane brassinosteroid analogs having an additional functional group, which fit to the area with high probability of hydrogen-bonding of the 23R-OH group of brassinolide (1), has been evaluated and interpolated in our model to predict its activity.

Through the set of brassinosteroids designed the new analog **3**, differing only to **2** for the presence of an extra hydroxyl group in the side chain, has result to be a good candidate for our purpose.

In this communication, the methodology developed to predict the activity, the synthetic strategy used to obtain **3**, the feasibility of hydrogen-bonding by means of the GRID maps of **3** and some synthetic precursors, as well as their predicted and elicited activity data will be presented and compared with those of **2.**

REFERENCES

1. Brosa C.: Structure-activity relationship in Brassinosterois: Steroidal Plant Hormones (Sakuray A., Yokota T., Clouse S. Eds) Chap 9, Springer-Verlag Tokyo (1999).
2. Brosa C., Zamora I., Terricabras E., Kohout L.: Collect. Czech. Chem. Commun. 63, 1635 (1998).



* This work was done with the help of grants of GA CR No. 203/01/0083 and IQS No. 9430/01

A BRASSINOSTEROID DERIVATIVE USEFUL TO PROVIDE MORE INFORMATION ABOUT HOW DOES THE C2 OH GROUP WORK IN THE BRASSINOSTEROID-RECEPTOR INTERACTION

CARME BROSA, MARC AMORÓS, ESTHER VÁZQUEZ, MARIA PIQUÉ

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In the field of brassinosteroids, which act as potent plant growth regulators, the knowledge of the minimum structural requirements needed for expressing activity is still a challenge. This will help to understand better the mode of action of such interesting compounds.

To achieve this goal in the most efficient way a quantitative structure-activity relationship (QSAR) has been developed in our group based on molecular modeling techniques.[1]

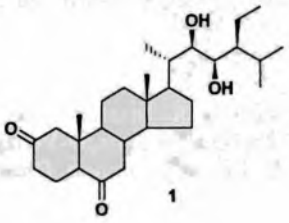
Based on the Grid methodology[1,2] we have found that the hydrogen-bonding could be one of the types of interaction that could take place on binding. Considering this, another interesting point to be determined is if the OH groups present in a brassinosteroid act as an acceptor or a donor in such hydrogen-bonding interaction.

In this sense, we have synthesized a new brassinosteroid analog having only a C2 ketone group in the A ring, compound **1**. The high activity elicited by this compound in the rice lamina inclination test is in agreement with the one predicted in our QSAR model.

In this communication, we will present the synthetic strategy developed to obtain **1** and the activity evaluated and predicted. Also, the feasibility of hydrogen-bonding by means of the GRID maps of **1** and how the functionalities present at C2 may work will be discussed.

REFERENCES

1. Brosa C.: Structure-activity relationship in Brassinosterois: Steroidal Plant Hormones (Sakuray A., Yokota T., Clouse S. Eds) Chap 9, Springer-Verlag Tokyo (1999).
2. Wade R. C., Clark K. J., Goodford P. J.: J. Med. Chem. 36, 140 (1993)



1

B-HOMO BRASSINOSTEROIDS WITH A CHOLESTANE SIDE CHAIN

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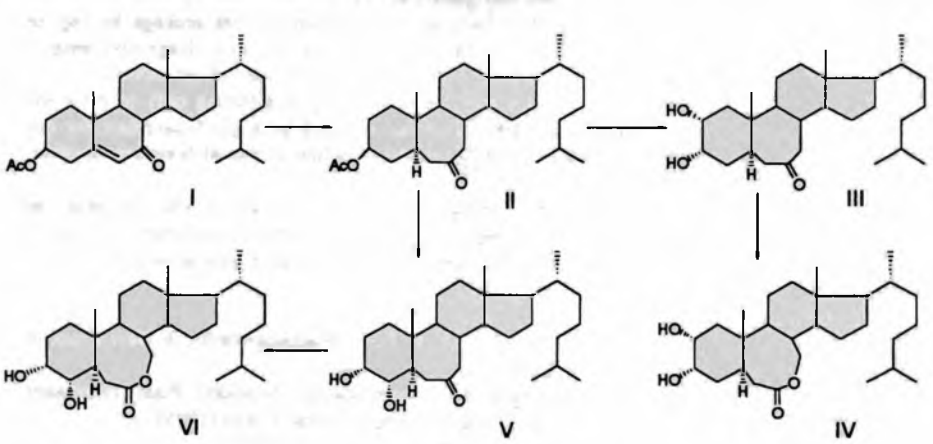
In the course of our studies on structure-activity relationship of brassinosteroid[1] we decided to synthesize B-homo-brassinosteroid analogues.

To elaborate the synthesis, we used 3β-acetoxy-7-oxo-cholest-5-ene (*I*) as the starting material. 3β-Acetoxy-B-homo-5α-cholestan-7a-one (*II*) was prepared from compound (*I*) *via* the corresponding cyanohydrin by the *Demjanoff ring enlargement* as described in literature[2]. Hydrolysis of acetate II afforded 3β-hydroxy-B-homo-5α-cholestan-7a-one which was transformed to the corresponding tosylate.On reaction with collidine, it afforded two isomeric olefins (2,3- and 3,4-olefins). These olefins were hydroxylated with osmium tetroxide[3] to 2α,3α-dihydroxy-B-homo-5α-cholestan-7a-one (*III*) and 3α,4α-dihydroxy-B-homo-5α-cholestan-7a-one (*V*). When treated with trifluoroperacetic acid in dichloromethane, each of compounds *III* and *V* afforded only one product: 2α,3α-dihydroxy-7-oxa-B-homo-5α-cholestan-7a-one (*IV*) and 3α,4α-dihydroxy-7-oxa-B-homo-5α-cholestan-7a-one (*VI*), respectively. Their structures were determined by ^{1}H NMR and ^{13}C NMR spectroscopy.



REFERENCES

1. Kohout L., Chodounská H., Macek T., Strnad M.: *Collect. Czech. Chem. Commun.* **2000**, *65*, 1754-1761.
2. Kohout L., Fajkoš J., Šorm F.: *Collect. Czech. Chem. Commun.* **1967**, *32*, 1210-1228.
3. Kohout L..: *Collect. Czech. Chem. Commun.* **1994**, *59*, 1219-1225.

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NEW BRASSINOSTEROID DERIVATIVES HAVING AN AMINE IN THE ANDROSTANE ESTER SIDE CHAIN

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Brassinosteroids are potent plant growth regulators, which have an exciting potential use in agriculture for improving yield and quality of crops.[1]

The search for new active brassinosteroid analogs with a good synthetic cost/activity relationship is still a challenge for further application in agriculture of such interesting compounds.

To achieve this goal in the most efficient way a quantitative structure activity relationship (QSAR) has been developed in our group based on molecular modeling techniques.[2]

Based on the Grid methodology[2,3] our QSAR model suggest that the region with high probability of hydrogen-bonding near to the one of the 23R-OH of brassinolide (**1**) seems to be more important than the one of the 22R-OH group and essential for eliciting activity. This is in full agreement with the results obtained when the androstane brassinosteroid analog **2** is studied following the same procedure.[4]

The lack of activity of **2** in rice lamina inclination test can be explained if one consider that its ester function presents only an area with high probability of hydrogen-bonding located near to the one of the 22R-OH of brassinolide (**1**), but slightly shifted to the right. No interaction is observed in the zone close to the 23R-OH of brassinolide (**1**).

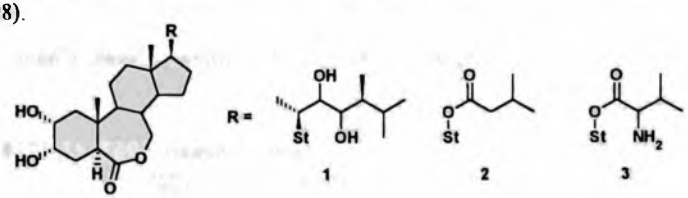
With the aim to assess this a set of new androstane brassinosteroid analogs having an additional functional group, which fit to the area with high probability of hydrogen-bonding of the 23R-OH group of brassinolide (**1**) has been evaluated and interpolated in our model.

Through the set of brassinosteroids designed the new analog **3**, differing only to **2** for the presence of an extra amine group in the side chain, has result to be a good candidate for our purpose. The predicted activity resulting from our model is similar to that of homobrassinolide, a natural potent plant growth regulator.

In this communication, the synthetic strategy developed to obtain **3**, the feasibility of hydrogen-bonding by means of the GRID maps of **3** and some synthetic precursors, as well as their predicted and elicited activity data will be presented and compared with those of **2**.

REFERENCES

1. Khripach V. A., Zhabinskii V. N., de Groot A. E. Eds.: Brassinosteroids; A New Class of Plant Hormones, Academic Press (1999).
2. Brosa C.: Structure-activity relationship in Brassinosterois: Steroidal Plant Hormones (Sakuray A., Yokota T., Clouse S. Eds) Chap 9, Springer-Verlag Tokyo (1999).
3. Wade R. C., Clark K. J., Goodford P. J.: J. Med. Chem. 36, 140 (1993).
4. Brosa C., Zamora I., Terricabras E., Kohout L.: Collect. Czech. Chem. Commun. 63, 1635 (1998).



* This work was done with the help of grants of GA CR No. 203/01/0083 and IQS No. 9430/01

Synthesis of [26-$^2H_3$]-22α,23α-dihydroxy- and [26,27-$^2H_6$]-22α-hydroxy brassinosteroids

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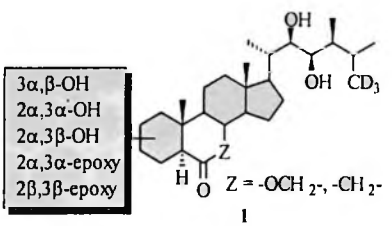
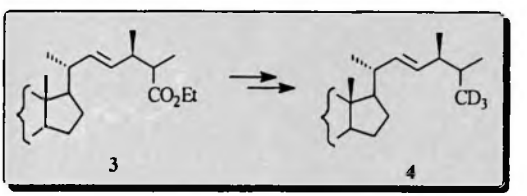
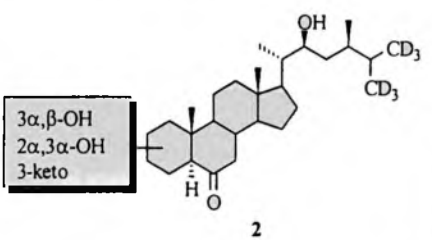
A number of possible biogenetic precursors of brassinosteroids (BS) **1** and **2** deuterated at C_{26} and/or C_{27} has been prepared for biosynthetic studies. Introduction of deuteriums into the target molecules has been done either by reductive deuteration of compound **3** to form triply deuterated derivatives **4**



1



or via addition of the sulfone **6** to the corresponding steroidal aldehyde, yielding brassinosteroids containing six deuterium atoms in the side chain.



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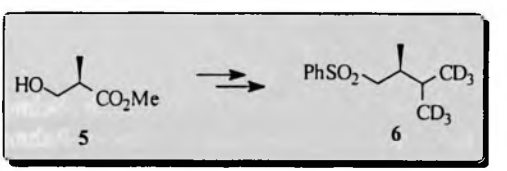

SYNTHESIS OF NEW BRASSINOSTEROID - ECDYSTEROID STRUCTURE RELATED DERIVATIVES FOR BIOCHEMICAL INVESTIGATIONS IN PLANT AND INSECT SYSTEMS

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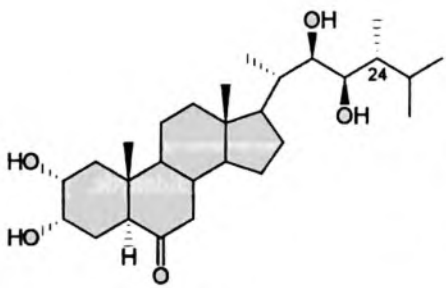
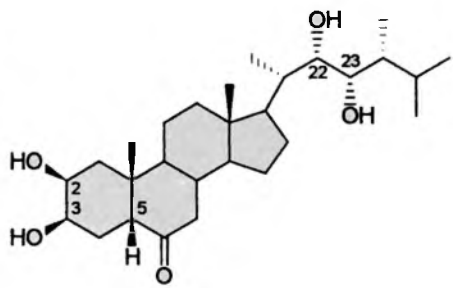
The brassinosteroids represent a new class of steroidal phytohormones of ubiquitous occurrence in the plant kingdom with high growth promoting and antistress activity.[1] The structural similarities of brassinosteroids with the moulting hormones of the ecdysone-type induced us to investigate possible metabolic transformations of the phytohormone 24-epicastasterone in insects. In the course of these studies after feeding of this compound to the cockroach *Periplaneta americana* an organ specific epimerization of the brassinosteroid to 2,24-diepicastasterone could be detected in females. The metabolite being observed only in the ovaries and not in the testes of the insect was identified by GC/MS in comparison with a synthetic sample.[2] These investigations are the first evidence of a metabolic transformation of a brassinosteroid in an insect.

Further investigations required the synthesis of new metabolites as 2,3,24-trisepicastasterone, 2,3,5,24-tetraepicastasterone, 2,3,22,23,24-pentaepicastasterone and 2,3,5,22,23,24-hexaepicastasterone. The structure-activity relationships of the new compounds as phytohormone as well as in an insect moulting hormone assay were investigated.



24-Epicastasterone



2,3,5,22,23,24-Hexaepicastasterone

References
1. Adam, A. G. Porzel, J. Schmidt, B. Schneider, B. Voigt: New Developments in Brassinosteroid Research. In: Atta-ur-Rahman (Ed.) Studies in Natural Products Chemistry, **1996**, Elsevier, Amsterdam, London, New York, Tokyo, Vol. 18, 495-549.
2. J. Schmidt, K. Richter, B. Voigt, G. Adam: Metabolic transformation of the brassinosteroid 24-epicastasterone by the cockroach *Periplaneta americana*. Z. Naturforsch. 55c, 233-239 **(2000).**

SYNTHESIS OF NOVEL STEROIDAL $\Delta^{1,4,6}$-3-KETONES

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AEDE DE GROOT[b], and VLADIMIR KHRIPACH[a]

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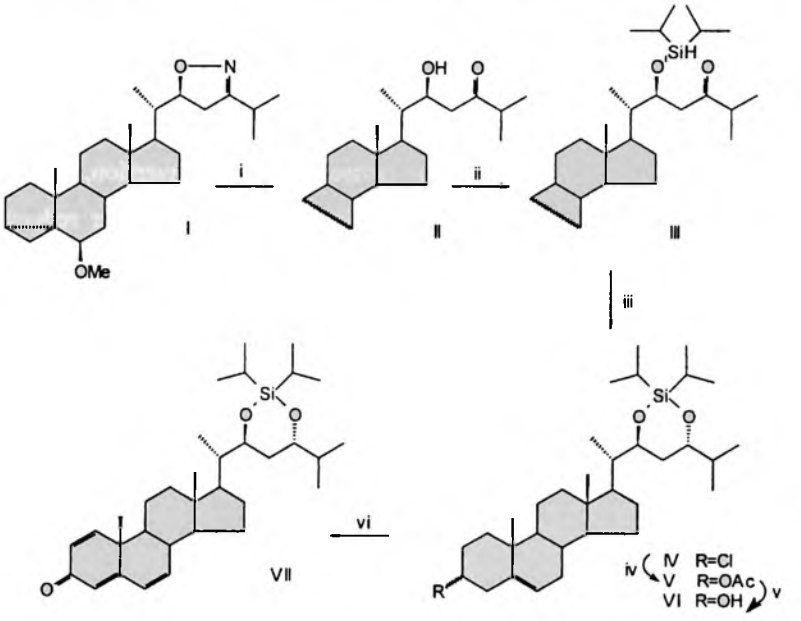
Major synthetic routes utilized to introduce the 1α-hydroxy group into steroidal molecules, *e.g.* vitamin D precursors, are methods using easily available $\Delta^{1,4,6}$-3-ketones as key intermediates.

Herein we wish to report on the synthesis of steroidal $\Delta^{1,4,6}$-3-ketone *VII* starting from 20-isoxazolin-5′-yl-steroid *I* obtained from stigmasterol[1].



i) Ni-Ra, $AlCl_3$, MeOH-H_2O, r.t.; ii) iPr_2SiHCl, Et_3N, DMAP, petroleum ether, reflux
iii) $SnCl_4$, CH_2Cl_2, -78°C; iv) KOAc, AcOH, reflux; v) KOH, EtOH, reflux;
vi) DDQ, dioxane, reflux

The chiral side-chain centers at C-22 and C-24 were stereoselectively generated *via* cleavage of the isoxazoline ring followed by intramolecular carbonyl hydrosilylation[2].

The reaction conditions as well as the spectral data of the obtained compounds will be discussed.

REFERENCES

1. Khripach V.A., Litvinovskaya R.P., Baranovskii A.V., Akhrem A.A.: Dokl. Akad. Nauk SSSR. 3, 597 (1991).
2. Anwar S., Davis A.P.: J. Chem. Soc., Chem. Commun. 831 (1986).

IMPROVED SYNTHESIS OF ZYMOSTEROL

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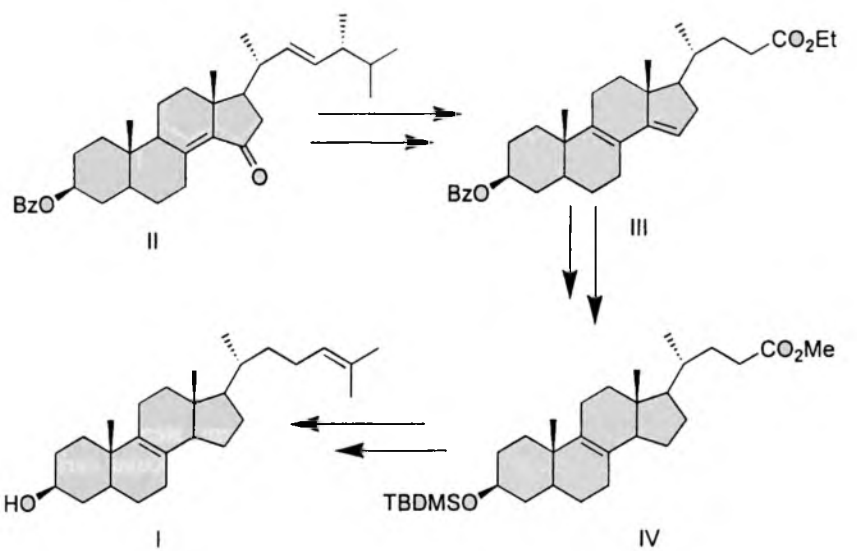
Zymosterol (I) is known metabolite of biosynthesis of cholesterol and ergosterol. However, the natural sources of this sterol is rather scarce. Dolle[1] and coworkers had developed an effective large scale synthesis starting from ergosterol. Here we propose an alternative synthesis of side chain which reduces steps and cost of reagents.

The synthesis starts from enone (II) which in a few steps is converted in diene ester (III). The key step is the Wittig-Horner reaction with C^{22}-aldehyde. Further, ester (III) is transformed in compound (IV) via the protection group conversion, hydroxylation and the Barton xanthate reduction. The last steps include the ester reduction, the Wittig reaction and deprotection.



REFERENCE
[1]Dolle R.E., Schmidt S.J., Erhard K.F., Kruse L.I.: J.Am.Chem.Soc.. *111*, 278 (1989).

CLEAVAGE OF THE STEROID 16α,17α-EPOXIDES WITH LITHIUM HYDROPEROXIDE

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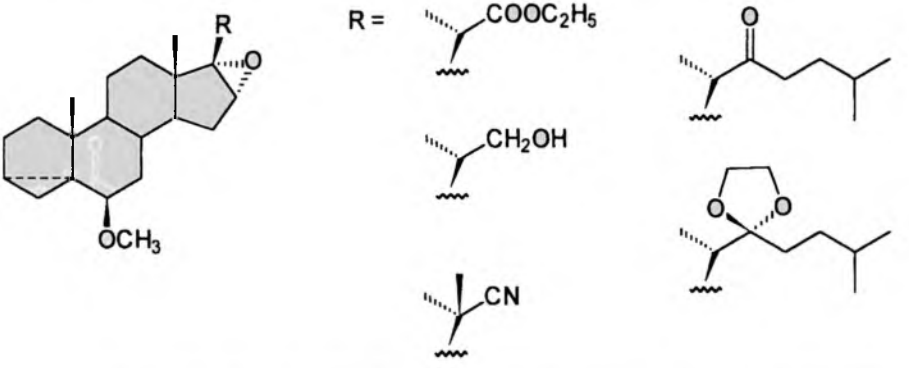
Five 16α,17α-oxidosteroids were subjected to acids, bases and lithium hydroperoxide.[1,2] Acids caused Wagner-Meerwein type rearrangement irrespective of the side-chain structure. The 16α,17α-epoxides proved resistant to bases unless a C(22)=O group was present; in the case of 22-esters or 22-ketones the oxirane rings were cleaved with base and the corresponding allylic alcohols were formed.



The reactions of 16α,17α-oxido-22-carbonyl compounds with lithium hydroperoxide resulted in the epoxide cleavage to the desired 16β,17α-diols which underwent further transformations.

References:

1. J. W. Morzycki, A. Gryszkiewicz, I. Jastrzębska, *Tetrahedron Lett.* **41**, 3751 (2000).
2. J. W. Morzycki, A. Gryszkiewicz, I. Jastrzębska, *Tetrahedron* **57**, 2185 (2001).

A REARRANGEMENT OF 23-SUBSTITUTED SPIROSTANES TO THE BISFURANS

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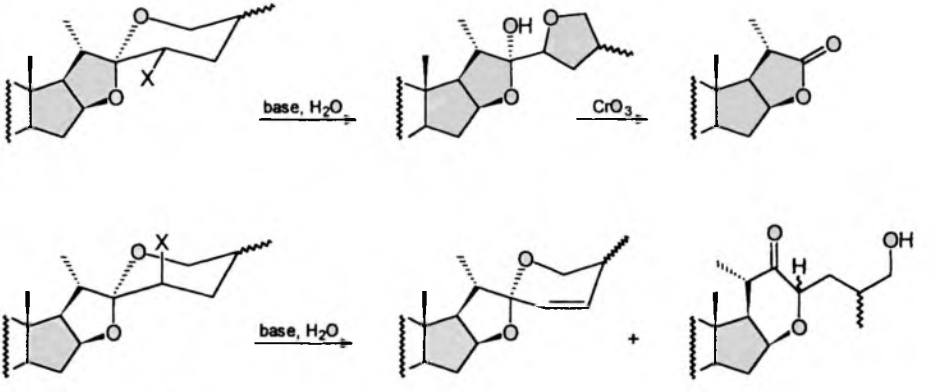
Solvolytic reactions of spirostane 23-bromides and 23-tosylates were studied.[1] In the 23*S* series a new rearrangement to the bisfurans was observed irrespective of the configuration at C-25. The optimal conditions for the reaction were found. An efficient degradation procedure of sarsapogenin via the corresponding bisfuran to the C_{22} lactone was elaborated.[2]



X = Br or OTs

23*R*-Spirostane derivatives proved more resistant to solvolysis and form the elimination products when treated with base at elevated temperature. However, the solvolysis of 23*R*-tosylate with 25*S* configuration yielded the rearranged E-homo product in addition to the 23-olefine. The decomposition of 23-tosylhydrazones under basic conditions was also examined.

References:

1. I. V. Torgov, *Chemistry of spirostanols* (in Russian); Nauka: Moscow, 1986.
2. J. W. Morzycki, I. Jastrzębska, *Tetrahedron Lett.*, in press

SYNTHESIS OF A-RING PRECURSORS TO 1α-HYDROXY-19-NORVITAMIN D COMPOUNDS STARTING FROM PHLOROGLUCINOL

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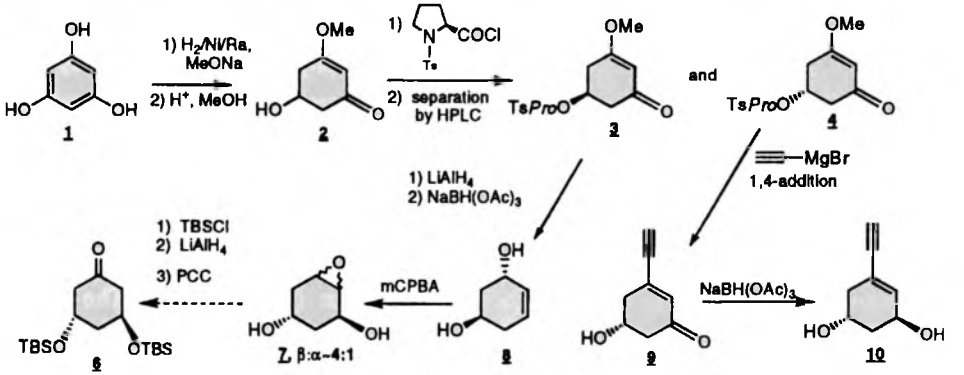
The known[1,2] A-ring synthons **6**, **10** for 1α,25(OH)$_2$-19-norvitamin D$_3$ and 1α,25(OH)$_2$-19-norprevitamin D$_3$ are accessible from phloroglucinol **1**. The absolute stereochemistry of intermediary (S,S)- and (R,S)-esters **3**, **4** was assigned from their Cotton effect in Circular Dichroism spectra (taking into account the predominant conformation in solution with an axial ester group: ^{1}H NMR in $CDCl_3$: $J_{5Heq,6Hax}$ = 5.5 Hz), on the base of comparison with the Cotton effect of regioisomeric 6-substituted esters **11**[3]. This absolute configuration was proved by conversion of (R,S)-ester **4** into (S)-5-hydroxy-2-cyclohexenone **5**.



The addition of ethynylmagnesium bromide was shown to proceed regiospecifically in 1,4-fashion, on the modelling non-symmetric 3-methoxy-6-tosylprolinoxy-2-cyclohexenones (R,S)- and (S,S)-**11**.[3]

References

1. K.L. Perlman, R.E. Swenson, H.E. Paaren, H.K. Schnoes, H.F. DeLuca. Tetrahedron. Lett. 1991, **32**, 7663.
2. L.A. Sarandeses, J.L. Mascarenas, L. Castedo, A. Mourino. Tetrahedron Lett. 1992, **33**, 5445.
3. a) V.G. Zaitsev, D.G. Sachava, G.S. Yankovskaya,. N.I. Garbuz. Chirality 2000, **12**, 287; b) V.G. Zaitsev, A.L. Mikhal'chuk. ibid. 2001, **13**, (8), 000.

SYNTHESIS AND BIOLOGICAL ACTIVITY OF 2-METHYL AND 2-METHYLENE-1α-HYDROXY-19-NORVITAMIN D ANALOGS WITH ALKYL SIDE CHAINS

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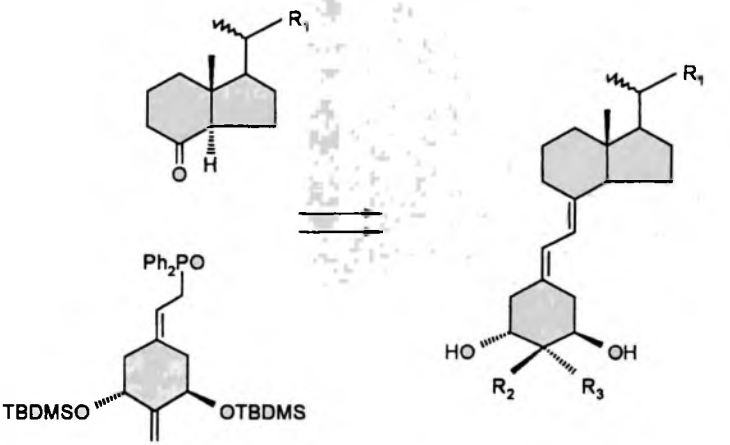
It is well known that the natural hormone, 1α,25-dihydroxyvitamin D_3, in addition to its classical role as a highly potent regulator of calcium homeostasis in animals and humans, also regulates cellular differentiation and immunology. Recently, its analogs characterized by the absence of the 19-methylene group were synthesized and tested. These 19-norvitamins have exhibited similar binding affinity to the vitamin D receptor as 1α,25-dihydroxyvitamin D_3 and some of them are also characterized by high calcemic and cell differentiation activities. In continuing efforts towards the synthesis of biologically active vitamin D compounds of potential therapeutic value, some new analogs have been synthesized and will be presented. The key synthetic step involves the Wittig-Horner coupling of Windaus-Grundmann type ketones, possessing different 17β-alkyl substituents, with the phosphine oxide prepared from (-)-quinic acid. The A-ring conformations of the synthesized vitamins and their biological activity will also be discussed.



Active Metabolites of Vitamin D_3 and Analogs of Vitamin D_2 - Synthesis and Biological Evaluation

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Andrzej Kutner[a]
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A series of highly hydroxylated metabolites of vitamin D_3 was obtained with the additional chiral center in the aliphatic side-chain and a series of homologated and branched analogs of an active form of vitamin D_2. Vitamin D C-22 synthon was used as the common advanced intermediate in these syntheses. Vitamin D_3 metabolites were obtained by Davis enantioselective

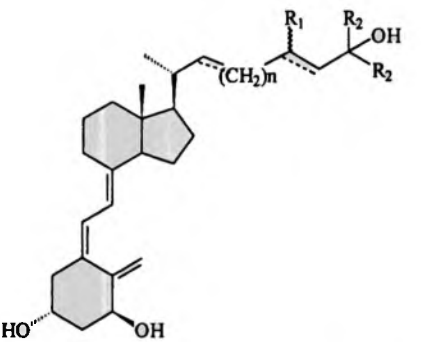

Figure 1. General structure of vitamin D analogs.

hydroxylation of vitamin D enolate with chiral oxaziridine. Vitamin D_3 analogs were synthesized by the ring opening of the side-chain epoxide with the sulfone stabilized C-22 carboanion. Vitamin D_2 analogs were obtained by Julia olefination of C-22 sulfone with a series of side-chain aldehydes. Some of the analogs showed statistically significant synergistic activity in suppressing the growth of murine breast tumor in the combined treatment with cytostatics like doxorubicin and cisplatin.

[1]Chodyński, M.; Odrzywolska, M.; Zorgdrager, J.; Velde van de, J-P., Kutner, A. *Chirality*, **1999**, *11*, 701-706;

[2]Siwińska, A.; Opolski, A.; Chrobak, A.; Wietrzyk, J.; Wojdat, E.; Kutner, A.; Szelejewski, W.; Radzikowski, C. *Anticancer Res.* **2001**, 21, in press.

Efficient Total Synthesis of Vitamin D Analogs

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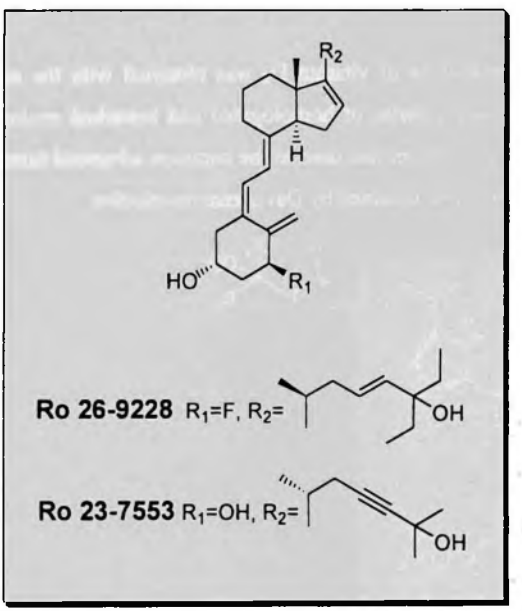
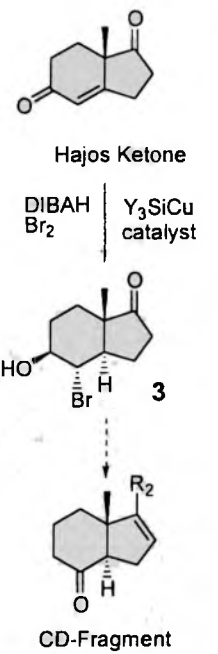
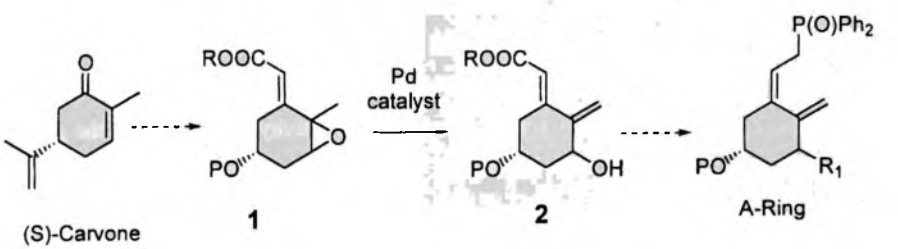
The total synthesis of Ro 26-9228 and Ro 23-7553 was developed using a Lythgoe type strategy. The key step for the synthesis of A-rings, was the palladium-catalyzed rearrangement of allylic epoxide **1** into allyl alcohol **2**. For the construction of CD-fragment, the intermediate **3** was obtained by the reductive bromination of Hajos Ketone using silylcopper catalyst. Most of the steps do not require chromatography and the methods described in this poster can be adopted for the large scale synthesis of many vitamin D analogs.







X-RAY DIFFRACTION STUDY OF STRUCTURES OF OLEANOLIC ACID OXIDATION PRODUCTS

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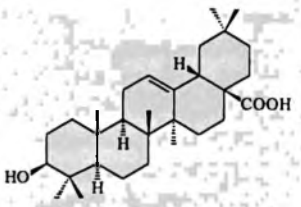
The X-ray diffraction studies involving products of oxidation of oleanolic acid and its 18α-diastereoisomer has been performed. These products used as starting materials in the synthesis of azatriterpenoids of supposedly interesting biological properties are the oleanolic acid derivatives including additional oxygen functions,.



oleanolic acid

The analysed compounds are mainly of carbonyl character or are lactons. The former compounds are products of mild oxidation, while the latter ones are obtained as results of large structural changes in the carbocyclic skeleton of oleanolic acid and its 18α-diastereoisomer. The results, that are presented at the conference, comprise the X-ray diffraction studies of eight carbonyl derivatives and seven triterpenoid lactones, namely:

3-oxo-18β-olean-12-en-28-oic acid methyl ester, 2-oxo-18β-olean-12-en-28-oic acid methyl ester, 2-formyl-3-hydroxy-18Hβ-olean-2,12-dien-28-oic acid methyl ester, 3β-acetoxy-12-oxo-18β-oleanan-28-oic acid methyl ester, dimethyl 1,3,11-trioxo-1,2-seco-18b-olean-12-ene-2,28-dioate, 3,11-dioxo-18α-olean-12-en-28-oic acid methyl ester, 3β-acetoxy-12,19-dioxo-olean-9(11),13(18)-dien-28-oic acid methyl ester, 3β-acetoxy-12,19-dioxoolean-9(11),13(18)-dien-28-oic acid methyl ester, 3β,11-dihydroxy-12-oxoolean-9(11),13(18)-dien-28-oic acid methyl ester methanol solvate, 11α-hydroxy-28-methoxy-28-oxo-2,3-dinor-18β-olean-12-en-1,4-olide hydrate, 3,28-dimethoxy-3,11,28-trioxo-2,3-seco-18β-olean-12-en-2,9α-olide, 3β-hydroxy-28-methoxy-11,28-dioxo-2-oxaolean-12-ene-3,1-carbolactone, 9α-hydroxy-28-methoxy-11,28-dioxo-1,2,-dinor-10α,18α-olean-12-en-3,10β-olide, 9β-hydroxy-28-methoxy-11,28-dioxo-1,2,-dinor-10α,18α-olean-12-en-3,10β-olide, 9β-hydroxy-28-methoxy-11,28-dioxo-1,2,-dinor-10α,18β-olean-12-en-3,10β-olide, 28-methoxy-28-oxo-1,2-dinor-10α,18β-olean-9,12-dien-3,10β-olide.

AZATRITERPENOIDS AS TRANSDERMAL PENETRATION ENHANCERS

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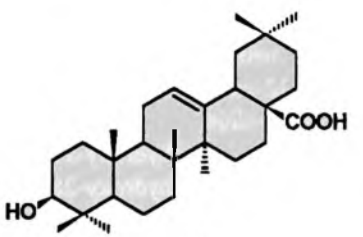
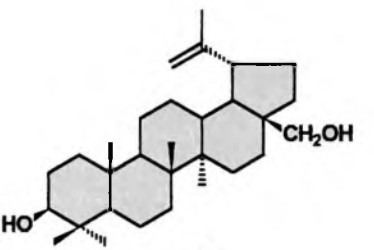
A series of nitrogen derivatives of triterpenoids from the group of oleanolic acid (**1**) and betuline (**2**) was obtained. In these compounds the nitrogen atom was either included in the skeleton of the molecule (compounds with the lactam system) or in the functional group (amide, hydroxyimine, nitrile). On synthesis of lactams (e.g. **3a**, **4a**), different reactivity of similar ketone and hydroxyimine systems occurring in ring A or ring C of the derivatives of oleanolic acid was observed and a very low susceptibility of the lactam systems formed to chemical modifications was established. One of the few reactions in which lactams were involved to a satisfactory degree was the conversion into thiolactams.

All the azatriterpenoids obtained were tested as to the ability of activation of transdermal penetration of drug, with the use of progesterone as the model compound. Similar tests were performed also for the parent oleanolic acid and betuline as well as some of their nitrogen-free derivatives. It has been established that only azatriterpenoids are good promoters of transdermal transport. The compounds with the lactam structure have been shown be more effective than the other derivatives, while thiolactams (e.g. **3b**, **4b**) not only enhance the amount of the absorbed substance but also increase the depth of its penetration. The strength of the promoting effect of the majority of the compounds studied was the same or higher than that of N-dodecylcaprolactame (Azone) used as the reference compound.



1



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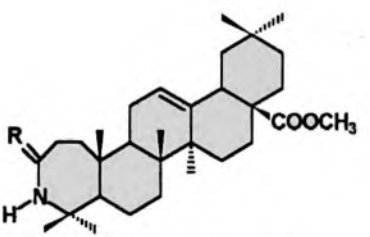
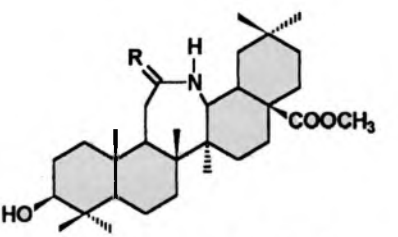


3a R = O **3b** R = S



4a R = O **4b** R = S

ALKALOIDS FROM THE POTATO AS STARTING MATERIAL FOR THE SYNTHESIS OF STEROID HORMONES

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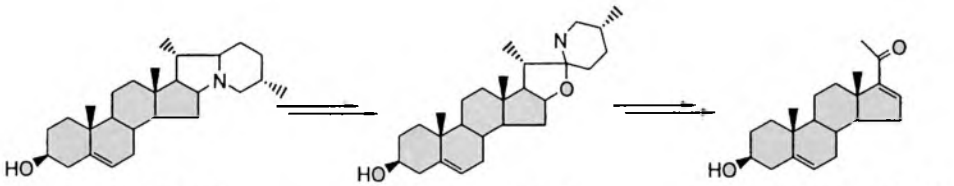
Large quantities of steroid alkaloids present in potatoes are produced in the starch industry as by-products. During the processing of starch, the proteins together with the glycoalkaloids are separated from the starch. The protein fraction is then further processed to isolate the proteins, which are used as nutriciants in cattle fodder. The waste from the protein fraction contains a high amount of glycoalkaloids, which can be isolated from the waste stream. Today these glycoalkaloids are considered as waste products without any value. However, after hydrolysis of the glycoside bond, the nitrogen-containing compound solanidine **1** is obtained which can serve as a starting material for the synthesis of steroid hormones.



solanidine **1** solasodine **2** 16-dehydropregnenolon **3**

For this purpose solanidine **1** has to be transformed into 16-dehydropregnenolon **3**. Starting from this compound **3** several classes of steroid hormones are accessible. At the moment dehydropregnenolon is obtained through degradation of diosgenine.

Gasi *et al.*[1] have already shown that it is possible to transform solanidine into dehydropregnenolon but because of low yields and expensive reagents this method is not suitable in industry.

The Polonovski reaction and the Cope reaction were investigated in order to open up the five-membered ring with the tertiary nitrogen. The Von Braun reaction was re-investigated to cleave the five-membered ring. This reaction gave access to structures like solasodine **2**. This compound is then used for accomplishing the degradation of solanidine to dehydropregnenolon.

REFERENCES

1. Gasi K.T.M., Djurendic E.A., Colic D.R., Sakac M.N., Arcson O.N., Mejacevic L.M., Miljkovic D.A.: *J. Serb. Chem. Soc. 62*, 451 (1997).

Efficient Biomimetic Synthesis of Monoterpenoid Alkaloids by a Three Component Domino Process

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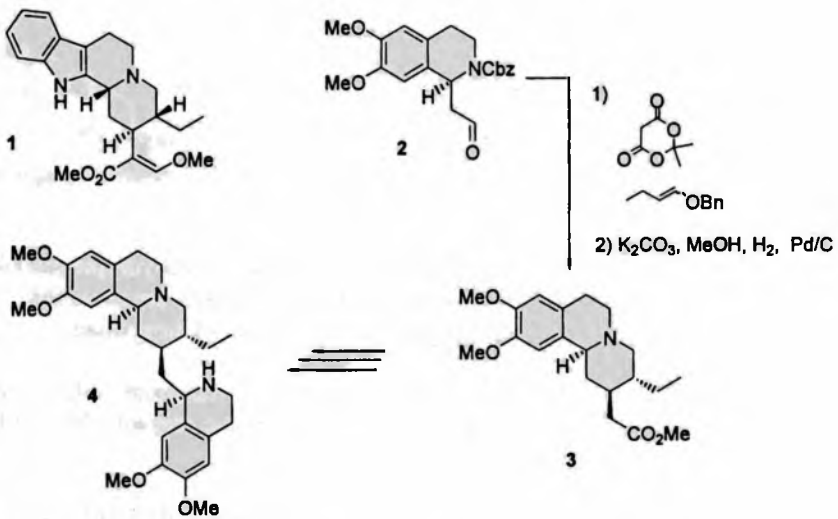
The monoterpenoid indole and isochinoquinoline alkaloids hirsutine **1** and emetine **4** are formed in Nature by condensation of tryptamine and dopamine, respectively with secologanin. Both compounds exhibit strong physiological activities, e.g. hirsutine **1** is highly effective against the Influenza A virus.[1] Recently we have developed an efficient synthesis of hirsutine **1** by a three component domino-Knoevenagel-hetero-Diels-Alder-reaction followed by a biomimetic solvolysis-hydrogenation process.[2,3]
Here we describe the use of this process for the synthesis of Ipecacuanha alkaloids of the emetine type as **4** via **3**. For this transformation the isoquinoline **2** was treated with Meldrum's acid and an enol ether followed by treatment with methanol in the presence of K_2CO_3 and hydrogenation.



[1] H. Takayama, Y. Iimura, M. Kitajima, N. Aimi, K. Konno, H. Inoue, M. Fujiwara, T. Mizula, T.Yokola, S. Shigeta, K. Tokuhisa, Y. Hanasaki, K. Katsuura, *Bioorg. Med. Chem. Lett.* **1997**, *7*, 3145–3148. [2] L. F. Tietze, *Chem. Rev.* **1996**, *96*, 115–136. [3] L. F. Tietze, Y. Zhou, *Angew. Chem.* **1999**, *111*, 2076–2078, *Angew. Chem. Int. Ed.* **1999**, *38*, 2045–2047; L. F. Tietze, F. Haunert in „Stimulating Concepts in Chemistry" (Eds. : M. Shibasaki, J. F. Stoddart and F. Vögtle) Wiley VCH, Weinheim **2000**, 39-64.

BINDING PROPERTIES OF STEROID SUBSTITUTED CALIXPRROLES

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Synthetic protocol is based on reaction of ketones derived from cholic acids and pyrrole under acid catalysis (Lewis acid) and was described earlier.[1,2]

We report hereby the further investigations leading towards the utilization of the calixpyrroles, namely as tools in analytical and bio-analytical chemistry. Also we report their NMR spectra prediction and spatial arrangement calculations.

Both calix[4]pyrroles derived from cholic and lithocholic acids were separated by column chromatography on silica gel as a mixture of isomers and final separation was performed by HPLC, silica column, in ethyl acetate – dichloromethane mixture.

α,β,α,β and α,α,α,α isomers were tested for binding efficiency. As our novel receptors offer 12 well-oriented OH groups as well as numerous opportunities for lipophilic interactions we tried to find complexation substrates first among different organic acids. Hence, we tested the formation of non-covalent complexes by MS (FAB, MALDI) and by IR methods. The binding of malic acid, tartaric acid (tartarate) and squaric acid was observed. Enantioselective binding studies are in progress.

A structure of calixpyrrole used is given on a Fig. 1.:

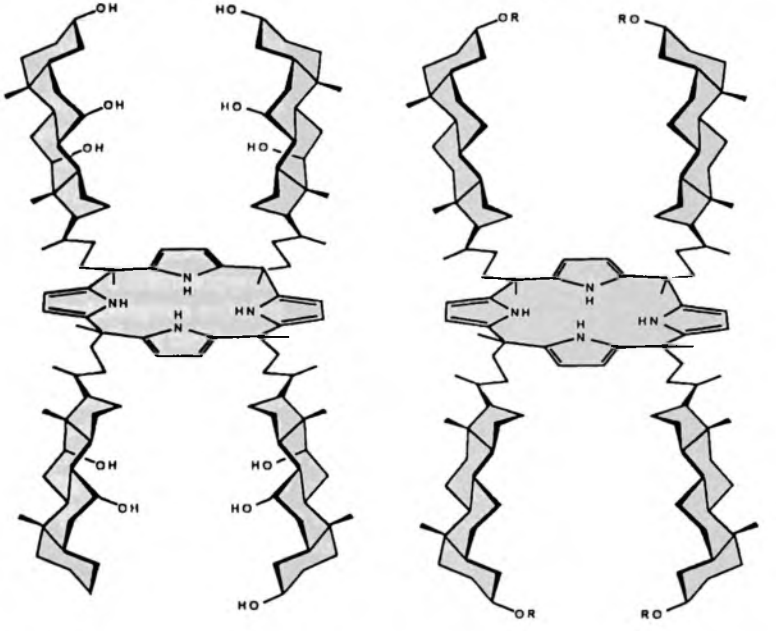

Fig. 1

LITERATURE:

[1] Dukh, M.; Černý, I.; Urbanský M.; Pouzar, V.; Král, V.; Drašar P.: Chem. Listy, 93, Symposia s63 (1999).

[2] Dukh, M.; Černý, I.; Urbanský, M.; Pouzar, V.; Král, V.; Drašar P.: Czech Pat. Appl. 1999, PV3098-99 (31. 08. 1999).

SUGAR COMPLEXING RECEPTOR BUILT AS NOVEL STEROID-PORPHYRIN CONJUGATE FOR UTILISATION IN WATER

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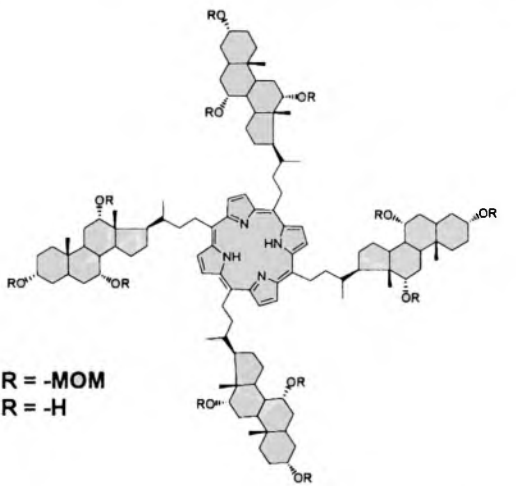
We present a synthesis of *meso*-substituted porphyrin **2** with sufficient solubility in water with twelwe of hydroxyls. The synthetic protocol starts with methyl cholate, which was prepared using altered published method[1], Then, it was transformed into its tris-MOM ether by condensation with chloromethyl methyl ether and resulting blocked ester was transformed into the respective alcohol by reduction with LAH/THF. The resulting alcohol was oxidized to its aldehyde counterpart by chromium(VI)oxide in pyridine. Formyl compound was condensed with pyrrole in dichloromethane with the catalysis of boron trifluoride etherate to give the protected tetrasteroidyl porphyrin **1**, which was in final step deprotected giving receptor **2**. We value steroidal moiety as a building block, which brings chiral discrimination potential into the molecule. New receptors were tested for binding of biologically important substrates, namely mono and oligo saccharides in natural environment. Receptor **2** exhibits very effective saccharide binding in aqueous media, with preferential complexation of oligosaccharides (K_a $10^4[M^{-1}]$ up to 13.0 for maltotriose). Mechanism of binding was studied by ^1H NMR, IR and Raman spectroscopy on complex of **2** with model alkylglucopyranoside. In conclusion, the new type of water-soluble porphyrin receptor was synthesized and fully characterized. Dodecahydroxyporphyrin **2** displayed effective complexation with unmodified saccharides, including monosaccharides as pentoses and hexoses, disaccharides and even trisaccharides in natural environment. The different affinity to several substrates could be explained by the overall spatial fit of saccharide and size of porphyrin host cavity with highest binding affinity for trisaccharides, while higher oligosaccharides are bound much weaker. The study of membrane saccharide transport system based on receptor **2** is in progress.



1 R = -MOM
2 R = -H

Acknowledgements. The project was supported by the grants of the Grant Agency of the Czech Republic No. 203/97/0695, of IOCB (No. 04/91), of HHMI #75195 and of Ministry of Education of the Czech Republic (No. VS 97135) and of COST No. OCD12.20.

(1) Rubottom, G.M.; Chong-wan, K. J. Org. Chem. 1983, 48, 1550.

SYNTHESIS AND STRUCTURE OF LINEAR OLIGOMERS OF ETIENIC ACID CONNECTED WITH AMIDE BOND.

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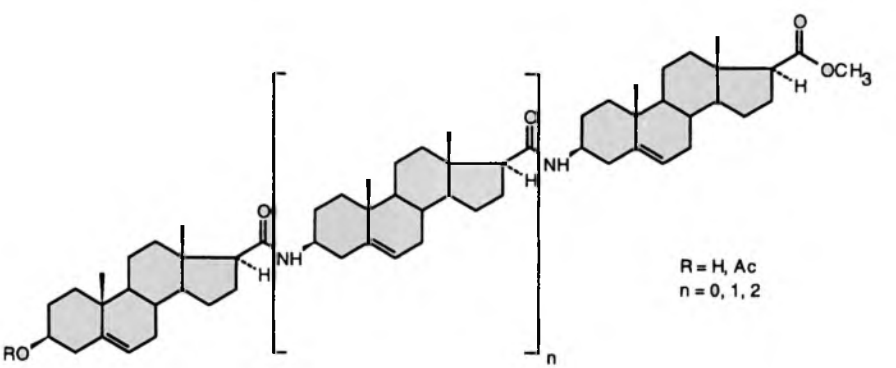
Inst. Org. Chem. Biochem., Flemingovo 2, 166 10 Prague 6, Czech Republic

Continuing our effort[1] in construction of larger rod shaped molecules using a steroid skeleton as a rigid building block, we developed a method for chaining etienic acid derivatives through the amide bond[2]. Methyl ester of etienic acid was transformed into corresponding 3β-azido derivative, and both etienic acid and this derivative were activated in form of N-hydroxysuccinimide ester. Succesive azide reduction and condensation with the active ester gave higher oligomer. By the repeating of this procedure we were able to prepare up to four membered oligomers.

Detailed NMR study has resulted in nearly complete structural assignment of carbon and hydrogen signals in NMR spectra. Information about conformation behavior of the selected dimer and trimer has been obtained from interproton NOEs, relaxation times T_1 of individual carbon atoms and molecular modeling.



1. Černý I., et al.: Collect. Czech. Chem. Commun. **2000**, *65*, 1597.
2. Černý I., et al.: Collect. Czech. Chem. Commun. **2001**, *66*, Accepted.

Supported by COST OC D12.20 and performed as a part of CAS research project Z4 055 905.

EFFICIENT SYNTHESIS OF 2-C-METHYL-D-ERYTHRITOL BY EPOXY ESTER - ORTHOESTER REARRANGEMENT

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We describe a new synthesis of 2-C-methyl-D-erythritol, the dephosphorylated form of a biosynthetic intermediate in the 1-deoxyxylulose pathway to isoprenoids. The key step in the preparation of this five carbon tetrol is the acid-catalyzed rearrangement of an epoxy ester, readily available via Sharpless asymmetric epoxidation. The mechanism of the epoxy ester-orthoester rearrangement has been studied using O-18 labeling and C-13 NMR analysis. The data show that 5-*exo* cyclization leads to a [2.2.1]-bicyclic orthoester with inversion at the proximal epoxy center. Hydrolysis of the orthoester and deprotection leads to the desired branched sugar in good yield.

CYCLIC ODORANTS BY RING CLOSING METATHESIS

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Firmenich SA R & D Division POBox 239 CH-1211 Geneva 8

Ring closing metathesis [1] of suitably substituted dienes provides a convenient entry into 5-, 6-membered ring [2] or macrocyclic odorants [3] and other economically important perfume ingredients [4].

The olfactory nuances of the macrocyclic products depend, in a subtle way, on the configuration of the double bond in their backbones.

Fürstner has demonstrated recently that ring closing alkyne metathesis followed by *Lindlar* reduction of the resulting cycloalkyne products may serve to control the Z double bond of civetone [5].

Diverse applications and complementary strategies shall be discussed [6].

[1] Recent reviews: R.H. Grubbs, S. Chang, *Tetrahedron* **1998**, *54*, 4413; A. Fürstner, *Top. Catal.* **1997**, *4*, 285; M. Schuster, S. Blechert, *Angew. Chem. Int. Ed Engl*, **1997**, *36*, 2036; A. Fürstner, *Top. Organomet. Chem.* **1998**, *1*, 37.

[2] A. H. Hoveyda, R. R. Schrock, *Chem. Eur. J.* **2001**, *7*, 945; D. S. La, J. B. Alexander, D. R. Cefalo, D. D. Graf, A. H. Hoveyda, R. R. Schrock, *J. Am. Chem. Soc.* **1998**, *120*, 9720; J. B. Alexander, D. S. La, D. R. Cefalo, A. H. Hoveyda, R. R. Schrock, *J. Am. Chem. Soc.* **1998**, *120*, 4041.

[3] R. Hamasaki, S. Funakoshi, T. Misaki, Y. Tanabe, *Tetrahedron* **2000**, *56*, 7423; H. Hagiwara, T. Katsumi, V.P. Kamat, T. Hoshi, T. Suzuki, M. Ando, *J. Org. Chem.* **2000**, *65*, 7231; V. P. Kamat, H. Hagiwara, T. Katsumi, T. Hoshi, T. Suzuki, M. Ando, *Tetrahedron* **2000**, *56*, 4397; A. Fürstner, K. Langemann, *Synthesis*, **1997**, 792; Y. M. Choo, K. E. Ooi, I. H. Ooi, *J. Am. Oil Chem. Soc.* **1994**, *71*, 911; J. Tsuji, S. Hashiguchi, *J. Organomet. Chem.* **1981**, *218*, 69; J. Tsuji, S. Hashiguchi, *Tetrahedron Letters*. **1980**, 2955.

[4] A. Fürstner, K. Langemann, J. *Org. Chem.* **1996**, *61*, 3942.

[5] A. Fürstner, G. Seidel, *J. Organometal. Chem.* **2000**, *606*, 75.

[6] A. Fürstner, O. R. Thiel, L. Ackermann, *Org. Letters* **2001**, *3*, 449.

REACTION OF 2-HETEROSUBSTITUTED ALDEHYDE TOSYLHYDRAZONES WITH α-MAGNESIO SULFONES

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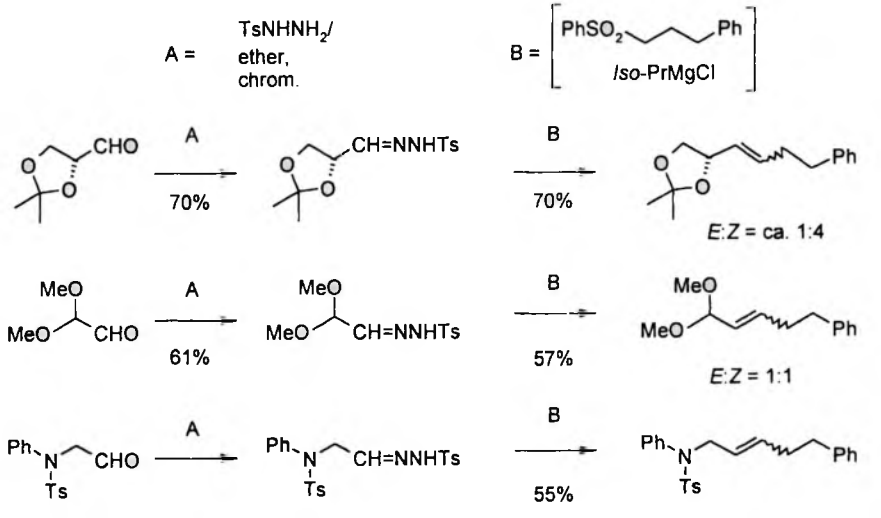
We have recently described a new method for aldehyde olefination, involving transformation of an aldehyde into the corresponding tosylhydrazone and reaction of the latter with an α-magnesio sulfones.[1] Now, we show that this method may be used for synthesis of allylic alcohols or allylic amines derivatives, starting from the corresponding α-hydroxy or α-amino aldehyde derivatives. Typical examples are included in the scheme.



(1) Kurek-Tyrlik, A.; Marczak, S.; Michalak, K.; Wicha, J. *Synlett* **2000**, 547-549, cf. Vedejs, E.; Dolphin, J. M.; Stolle, W. T. *J. Am. Chem. Soc.* **1979**, *101*, 249-251.

Synthesis of polyhydroxyindolizidines from 5,6-dihydro-2H-pyran-2-one

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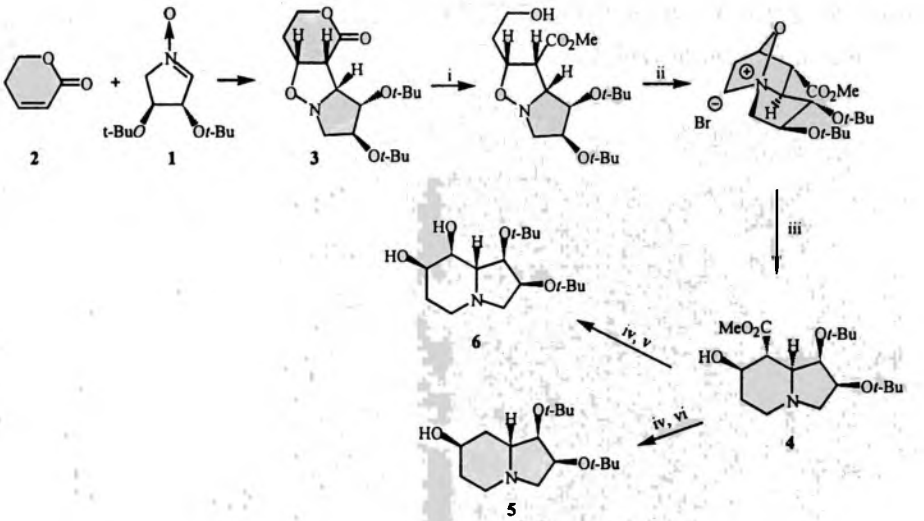
Institute of Organic Chemistry of the Polish Academy of Sciences, 01-224 Warsaw, Poland

The 1,3-dipolar cycloaddition reaction of nitrone **1** to commercially available lactone **2** proceeds with high stereoselectivity. The adduct **3** can be easily transformed into indolizidine **4** *via* Brandi's methodology (Scheme). The carboxylic function can be removed or converted into hydroxy group to provide derivatives of 7-hydroxy-lentiginosine (**5**) or 7,8-dihydroxy-lentiginosine (**6**), respectively.



i: K_2CO_3/MeOH or H^+/MeOH; ii: CBr_4, TPP; iii: H_2-$Pd(OH)_2$/C, MeOH; iv: LiOH$\times H_2O$; v: 1) N-hydroxy-pyridine-2-thione, DCC, DMAP, 50°C, 2) *t*-BuSH, 50°C; vi: 1) N-hydroxypyridine-2-thione, DCC, DMAP, argon, 50°C, 2) *t*-BuSH, argon, 50°C.

Stereocontrol formation of oxa-cecphams

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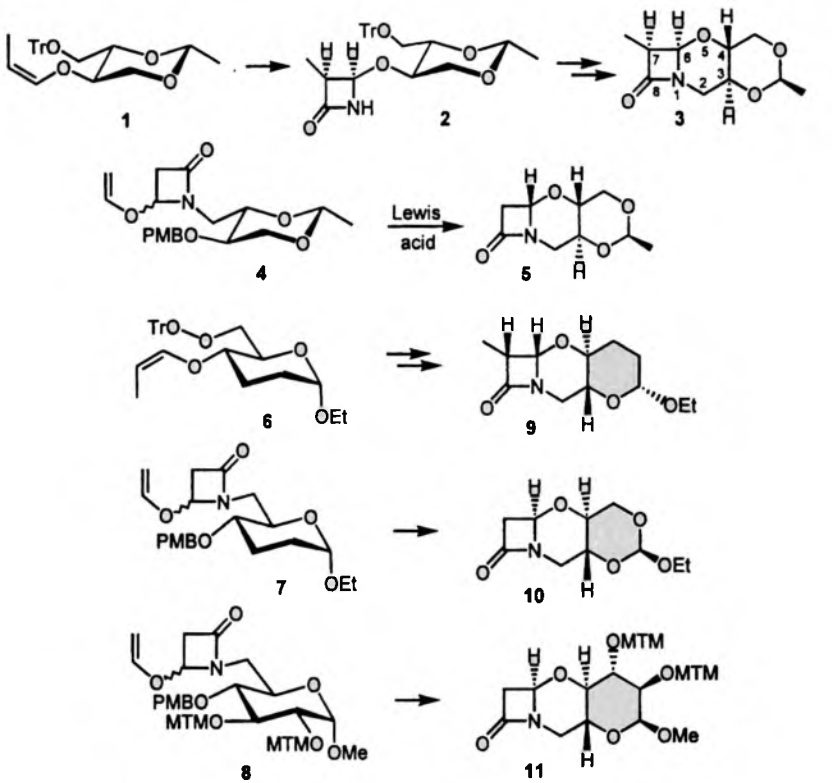
[2+2]Cycloaddition of chlorosulfonyl isocyanate (CSI) to (*Z*)-1,3-*O*-ethylidene-2-*O*-propenyl-4-*O*-trityl-L-erythritol (**1**) proceeds with excellent stereoselectivity to afford corresponding (*R*)-4-alkoxy-azetidin-2-one **2** which can be transformed into 5-xacepham **3** by intramolecular alkylation of the β-lactam nitrogen atom. Cepham having the alternative (*S*) configuration at the bridgehead carbon atom **5** can be achieved by another methodology based on intramolecular displacement of vinyloxy group in β-lactam **4** [1]. Both approaches have been successfully applied to compounds **6**-**8** to obtain corresponding cephams **9**-11 with full control of the absolute configuration at C-6 carbon atom.



PMB = *p*-methoxybenzyl; MTM = methylthiomethyl

[1] Z. Kałuża, R. Łysek, *Tetrahedron: Asymmetry*, **1997**, *8*, 2553; Z. Kałuża, *Tetrahedron Lett.*, **1998**, *40*, 8349; ibid, **1999**, *40*, 1025.

Stereocontrolled approach to 4-methyl-5-oxacephems from ethyl lactate and 4-vinyloxy-azetidin-2-one

A. Kazimierski,[a] K. Lewandowski,[b] M. Chmielewski [a] and Z. Kałuża [a]

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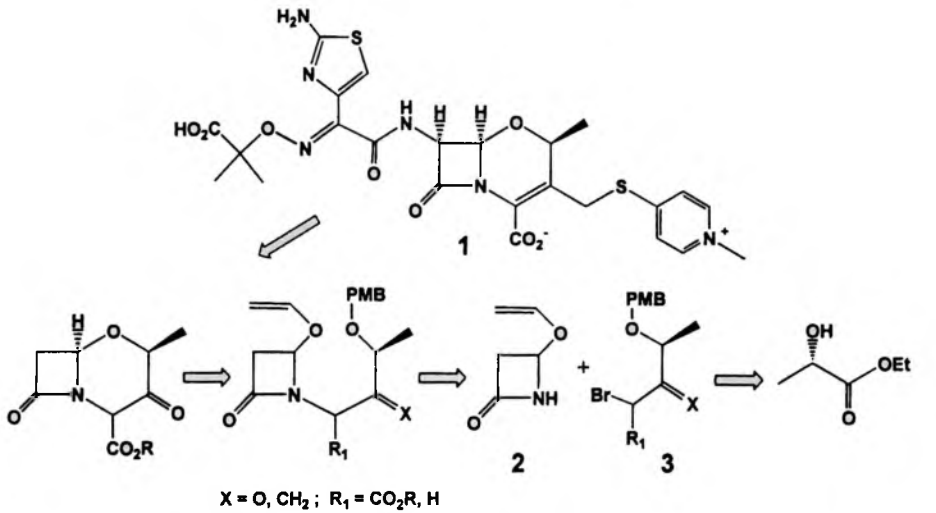
[b] *Institute of Chemistry, University of Podlasie, 08-110 Siedlce, Poland*

Synthetic oxa- analogues of cephalosporines, exhibit high antibiotic activity and are resistant to β-lactamases. In 1988 the Merck and Meiji groups reported a new oxacephem OCP-9-176 **1** having a 4β-methyl substiuent [1]. The introduction of a 4β-methyl group to the oxacepham skeleton increases stability to β-lactamases without changing its activity.

In this report we present studies toward the stereocontrolled synthesis of 4-substituted 5-dethia-5-oxacephems employing our new methodology [2].

The retrosynthesis of oxacephem **1** leads to simple starting materials: 4-vinyloxy azetidin-2-one **2** and readily available from ethyl lactate chiral bromide **3** (Scheme 1).



Scheme 1

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[1] S. Shibahara, T. Muray, K. Kuodo, T. Yoshida, S. Kondo and B. G. Christiansen, *J. Antibiot.*, *41*, **1988**,1154.

[2] Z. Kałuża, S-H. Park. *Synlett*, **1996**, 895. Z. Kałuża, R. Łysek, *Tetrahedron: Asymmetry*, *8*, **1997**, 2553. Z. Kałuża, *Tetrahedron Lett.*, *39*, **1998**, 8349. B. Furman, R. Thürmer, Z. Kałuża, R. Łysek, W. Voelter, and M. Chmielewski, *Angew. Chem. Int. Ed. Engl.*, *38*, **1999**, 1121.
Z. Kałuża, *Tetrahedron Lett.*, *40*, **1999**, 1025.

ACTIVATION OF CARBOXYLIC AND THIOCARBOXYLIC ACIDS BY LAWESSON REAGENT.

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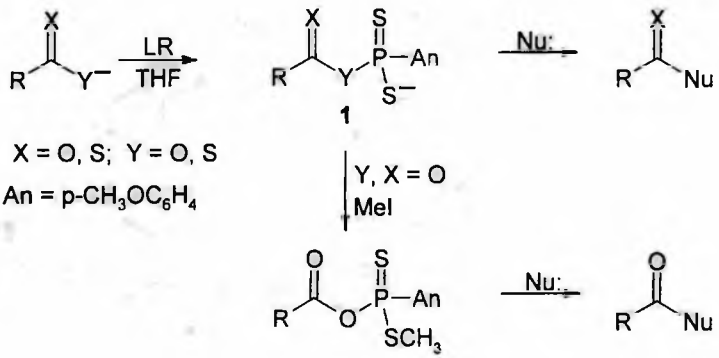
Taking into consideration a limited number of thioacylation methods we focus on the reaction of carboxylic and thiocarboxylic acids with Lawesson reagent. We found that the respective adducts / mixed anhydrides triethylammonium salts as well as their *S*-methyl esters are good acylating / thioacylating agents for preparation of amides / thioamides and peptides (Scheme 1).



Scheme 1

A reaction of N-nucleophiles with an activated form of thiocarboxylic acid (**1**) is thermodynamically controlled, i. e. it gives higher yields of thioamides at lower temperatures.

The scope and limitations of the above mentioned method of activation will be discussed.

THIONATION OF MIXED *N* – HYDROXYAMIDE – AMIDE SYSTEM

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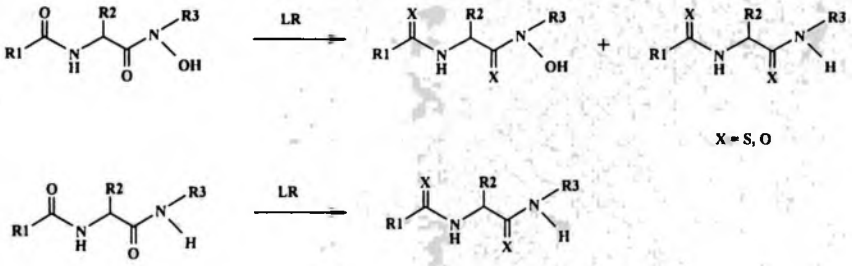
Structural modification of biologically active organic compounds uses the carbonyl - thiocarbonyl transformation due to the use of such a compounds for research in medicine and biochemistry.

The direct thionation seems to be the simplest method of *N*-hydroxythiopeptides synthesis. There is no any of convenient synthetic method,[1] which allows acces to *N*-hydroxythioamide in high yield and purity. We synthesized some diamides containing *N*-hydroxamide function and studied possibilities of their thionation with Lawesson reagent. As model compounds we choose the corresponding diamides (Bz-Gly-N(OH)iPr, Bz-Ala-N(OH)iPr, Ac-Gly-N(OH)Bzl). This work also provided several monothiodiamides by conversion of the (Bz-Gly-NHiPr, Bz-Ala-NHiPr, Ac-Gly-NHBzl). The observed chemoselectivity of thionation will be discussed.



[1] L. Wang, O. Phanistiel IV J. Org. Chem. **2000**, 65, 1442-1447

HIGHLY SELECTIVE CROSS-METATHESIS WITH VINYL SULPHONES AND SULPHOXIDES USING THE "SECOND GENERATION" RUTHENIUM CATALYSTS

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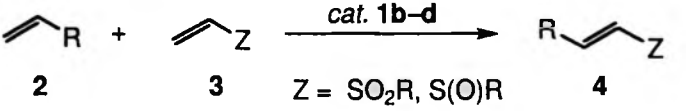
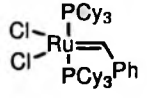
Keywords: carbene complex; olefin metathesis; cross-metathesis; sulphone; sulphoxide; ruthenium; homogenous catalysis

Substituted α,β-unsaturated sulphones are generally well accepted as useful intermediates in organic synthesis. Thus, vinyl sulphones serve efficiently, *e.g.* as Michael acceptors and as 2π partners in cycloaddition reactions. In addition, the stability and easy further transformations of sulfonyl group *via* elimination or either reductive or alkylative desulphonylation render further advantages of vinyl sulphones as intermediates in total synthesis.[2]

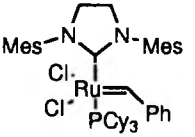
In this communication, we report the single-step synthesis of functionalized α,β-unsaturated sulphones and sulphoxides **4** *via* highly selective cross-metathesis reaction catalyzed by the "second generation" ruthenium alkylidenes **1b–c**.



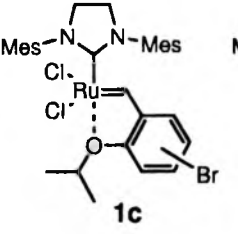
2 **3** **4**



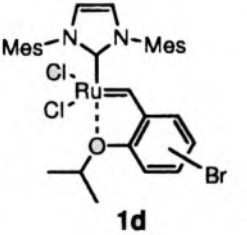
1a



1b



1c



1d

[1]Tel.: +48-22-6323221 ext. 2108; fax: +48-22-6326681; e-mail: grela@technologist.com.

[2](a) Procter, D. J. *J. Chem. Soc. Perkin Trans. 1* **1999**, 641-667; (b) Simpkins, N. S. *Tetrahedron* **1990**, 46, 6951-6984; (c) Patai, S.; Rappaport, Z.; Stirling, C., Eds. *The Chemistry of Sulphones and Sulphoxides*, John Wiley and Sons Ltd., 1988.

GENERATION OF CARBANIONS IN TWO-PHASE SYSTEMS IN THE PRESENCE OF AQUEOUS SODIUM HYDROXIDE. BLOCKING OF THE INTERFACE BY HIGHLY LIPOPHILIC I^- ANIONS.

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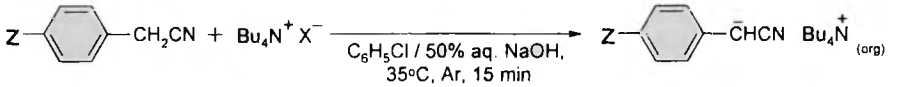
Hydroxide ion initiated reactions of CH-acids carried out under phase transfer catalysis conditions (concentrated aqueous sodium or potassium hydroxide / organic substrates; PTC/OH systems) are very important processes of great practical value.[1,2] Among these PTC/OH promoted reactions alkylation of CH-acids with alkyl halides is the most common process which results in the formation of a new C-C bond (Scheme 1; X=Cl, Br, I).

$$\geq C\text{-}H_{(org)} + R\text{-}X_{(org)} \xrightarrow[50\%\ NaOH_{(aq)}]{(cat.)\ Q^+X^-_{(org.)}} \geq C\text{-}R_{(org)} + Na^+X^-_{(aq)} + H_2O$$

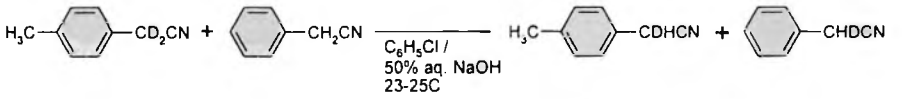
Scheme 1

Although alkyl iodides RI are the most reactive halides the produced I^- anions inhibit PTC alkylation of carbanions.[1,2] We have found that in such PTC systems the equilibrium concentrations of typical carbanions in the organic phase decrease when going from X^-=Cl^-, Br^- to I^- (Scheme 2; Z=MeO; Me; H; Cl).



Scheme 2

We have also found that in the absence of quaternary ammonium cations Q^+ the interfacial generation of carbanions is retarded when the aqueous phase is saturated with NaI. This kinetic effect was detected using deuterium exchange measurements in chlorobenzene/50% aq.NaOH two-phase system (Scheme 3).



Scheme 3

The reasons for such inhibitory effect and its experimental verification will be discussed.

1. M. Mąkosza, M. Fedoryński *Advances in Catalysis*, Vol.35, Acad. Press, **1987**, 375.
2. E.V. Dehmlow, S.S. Dehmlow *Phase-Transfer Catalysis*, Verlag-Chemie: Weinheim, 3-rd Ed. **1993**.

NOVEL SYNTHESIS METHOD FOR THE PREPARATION OF 2,3 SUBSTITUTED TETRAHYDROFURANS

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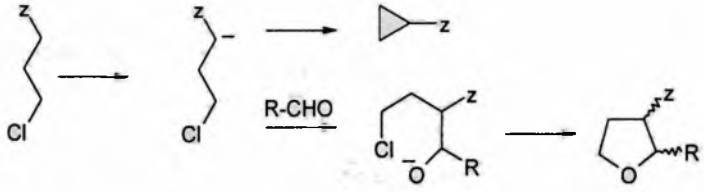
Treatment of γ-chlorobutyronitryle, γ-chlorobutyrophenone, γ-chloropropylphenyl sulfon etc. with strong base results in formation of the corresponding γ-chlorocarbanions which undergo rapid cyclization to produce substituted cyclopropanes. We have found that when these reactions are carried in the presence of active electrophiles such as aldehydes the intermediate carbanion enter fast intermolecular addition to produce aldol anions which cyclize to substituted tetrahydrofurans.



Z= CN, SO_2Ph, COOBu, C(O)Ph

R= Ph, p-MePh, p-ClPh, PhCH=CH

Usually trans isomer is the major product. This new method of synthesis of tetrahydrofurans can be of substantial practical value because starting materials are readily available and yields of the products are high. Scope and limitation of the new process will be discussed.

SYNTHESIS OF AMINO SUGARS FROM N-PROTECTED α-AMINOALDEHYDES

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The synthesis of enantiomerically pure organic compounds from chiral substrates is very advantageous, as it enables precise programming and efficient realization of synthetic pathways. During recent years, this approach to organic synthesis greatly contributed to progress in directed introduction of various functionalities, and in closely controlled formation of new stereogenic centers.

α-Amino acids are the important natural source of chiral starting materials, useful in stereocontrolled organic synthesis. On account of the increasing interest in α-amino acids and their suitably protected derivatives, we resolved to gather and present the actual knowledge concerning the use of α-amino aldehydes in stereocontrolled syntheses of amino sugars.

Several examples of the application of N-protected α-amino aldehydes to the asymmetric synthesis of useful chiral intermediates are given. Furthermore, these optically pure compounds are shown to be versatile synthons for preparation of many amino sugars and their derivatives of biological importance, e. g., cis-3-hydroxyproline and 1,3-dideoxynojirimycin.

DIASTEREOSELECTIVE NITROALDOL REACTION WITH CHIRAL DERIVATIVES OF GLYOXYLIC ACID

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The nitroaldol addition is one of the basic method for the construction of a carbon-carbon bond, 2-nitroalcohols formed in this process offer an easy access to a variety of numerous biologically important compounds.[1] There is currently substantial interest in development of stereocontrolled versions of the Henry reaction, however, only one example of application of chiral auxiliary has been reported.[2] In our studies we decided to examine the diastereoselectivity of nitroaldol addition of simple nitrocompounds to glyoxyimides and glyoxylates bearing four different types of chiral inducers, proved to be very efficient in many other reactions.

1. Oshida, J.; Okamoto, M.; Azuma, S.; Tanaka, T. *Tetrahedron: Asymmetry* **1997**, *15*, 2579 and references cited therein.
2. a) Solladié-Cavallo, A.; Khiar, N. *Tetrahedron Lett.* **1988**, *29*, 2189, b) Solladié-Cavallo, A.; Khiar, N. *J. Org. Chem.* **1990**, *55*, 4750.

ENANTIOSELECTIVE CATALYTIC HYDROGENATION OF KETO ESTERS

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Stereoselective hydrogenation of ketones possessing a heteroatom in the vicinity of the carbonyl group has been extensively studied, due to the synthetic significance of chiral alcohols.[1] In recent years, an enantioselective version of this process, using chiral phosphine-rhodium and -ruthenium complexes as catalysts, has been developed mainly by Noyori and coworkers.[2] This new methodology offers a simple and highly efficient route to many valuable building blocks, useful in total synthesis of natural products.
In this communication, we would like to present a short survey of enantioselective hydrogenation reactions of various keto esters. Special attention will be paid to 3-oxo-, 4-oxo-, and 3,5-dioxoesters as substrates, and to BINAP-Ru complexes as catalysts, both extensively studied in our laboratory.

1. Ohkuma, T.; Noyori, R. *Carbonyl Hydrogenation,* in T*ransition Metals for Organic Synthesis,* Beller, M.; Bolm, C. (EDS), Wiley, Weinheim 1998, Vol. 2, p. 25.
2. Ohkuma, T.; Noyori, R. *Hydrogenation of Carbonyl Groups,* in *Comprehensive Asymmetric Catalysis,* Jacobsen, E. N.; Pfaltz, A.; Yamamoto, H. (EDS), Springer, Berlin 1999, Vol. 1, p. 199.

THE ENANTIOSELECTIVE HETERO-DIELS-ALDER REACTION CATALYZED BY A CHIRAL (SALEN)CHROMIUM (III) COMPLEX

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The Diels-Alder reaction of 1,3-dienes with alkyl glyoxylates leads to formation of 3,6-dihydro-2*H*-pyran derivatives which are convenient substrates for further modifications. Several research groups have reported an enantioselective catalytic version of this reaction.[1] We applied chiral chromium(III) complex as a catalyst and found that for 1,3-butadiene the reaction proceeds only under high-pressure conditions. For cyclic dienes, like 1,3-cyclohexadiene, however, the reaction under atmospheric pressure was successfully carried out. Moreover, the stereochemical course of this reaction was much more selective. The hetero Diels-Alder reaction between 1,3-dienes and nonactivated dienophiles, such as alkoxyacetaldehydes, will be also presented.

1. For a review see: Jorgensen K. A. *Angew. Chem. Int. Ed.* **2000**, *39*, 3558-3588.

AUTHOR INDEX