MATEMAATIKA-LOODUSTEADUSKOND
KEEMIAINSTITUUT
TEADUS- JA ARENDUSTEGEVUSE AASTAARUANNE 2013

1. Struktuur

Keemiainstituut, Department of Chemistry
Instituudi direktor prof **Mihkel Kaljurand**

- Analüütilise keemia õppetool, Chair of Analytical Chemistry, prof Mihkel Kaljurand
- Anorgaanilise keemia õppetool, Chair of Inorganic Chemistry, prof Toomas Tamm
- Bioorgaanilise keemia õppetool, Chair of Bioorganic Chemistry, prof Nigulas Samel
- Biotehnoloogia õppetool, Chair of Biotechnology, prof Raivo Vilu
- Molekulaartehnoloogia õppetool, Chair of Molecular Technology, prof Mati Karelson
- Orgaanilise keemia õppetool, Chair of Organic Chemistry, prof Margus Lopp
- Keemilise analüüsi teadus- ja katselaboratoorium, Laboratory of Chemical Analysis, v-teadur Maria Kulp

2. Instituudi teadus- ja arendustegevuse iseloomustus (uurimisgruppide kaupa)

T060 - Katalüütilise asümmeetrilise sünteesi ja stereokeemilise analüüsi meetodid ja rakendused, teema juht **Lopp Margus**
Methods and applications of catalytic asymmetric synthesis and stereochemical analysis.

Teadustöö kirjeldus

The goal is to create and develop new methods of asymmetric organic synthesis by designing, preparing and introducing new chiral inducers and by finding new asymmetric transformations. The project is formally divided in three parts: new asymmetric reactions, computational design and analysis and synthesis of bioactive compounds. These parts are tightly linked and support each other. Asymmetric catalytic reactions and stereodirected cascade reactions of compounds with multiple prosterogenic centers creating stereoselectively multiple stereocenters simultaneously will be investigated. Computational modeling develops a robust and user-friendly method for the analysis of VCD spectra and supports understanding of properties and reaction mechanisms of the compounds. The rationally designed nucleoside analogues, dopamine receptor modulators and cucurbiturils will be synthesized and their biological and other properties will be investigated.

Main Scientific Results:

Investigations were carried out in two main directions: new reactions and synthesis of new compounds of interest.
From the first direction

- An asymmetric aminocatalytic approach to alkylidene cyclopropane derivatives via a Michael addition of aldehydes to nitroalkene derivatives was developed. Cyclopropane-containing derivatives were obtained in high enantiomeric purity and moderate to high yields.

- Straightforward cascade reactions for the synthesis of spiro-cyclopropaneoxindoles were described. The target compounds can be obtained in high yields and in good enantio- and diastereoselectivities via hydrogen bonding or iminium catalysis.
- Starting from simple alkylidene oxindoles and nitroketones, a highly stereoselective methodology was developed for the synthesis of spiro-cyclopentaneoxindoles with four consecutive stereogenic centers. Using an organocatalytic cascade of Michael and aldol reactions in the presence of a chiral thiourea catalyst products were obtained in moderate to high yields and excellent enantioselectivities. Nitro, ester, and hydroxyl groups were introduced to the spiro ring, which could be used to facilitate further functionalization of the products.
- A direct cross-coupling reaction to synthesize 3-alkynylsubstituted 1,2-diketones was developed.
- A mechanism of Ti-mediated cyclopropanation was elucidated.
- A convenient method for synthesis of sulphur-containing nucleoside analogues completed successfully

From the second direction

- 21 oxindole derivatives were tested in anti-HIV assay by prof. Merits group at Tartu University. One of the compounds inhibits approximately 2 times at 50mkM concentration.
- Several sulphur-containing nucleoside analogues were synthesized and biologically tested.
- New chiral enantiomeric hemicucurbiturils were synthesized and their some properties were elucidated.

Publications 2013

1. Reitel, K.; Lippur, K.; Järving, I.; Kudrjasova, M.; Lopp, M.; Kanger, T. *Synthesis* (**2013**), 45(19), 2679-2683.
2. Noole, A.; Malkov, A.; Kanger, T. *Synthesis* (**2013**), 45(18), 2520-2524.
3. Noole, A.; Ilmarinen, K.; Järving, I.; Lopp, M.; Kanger, T. *Journal of Organic Chemistry* (**2013**), 78(16), 8117-8122.
4. Noole, A.; Ošeka, M.; Pehk, T.; Öeren, M.; Järving, I.; Elsegood, M. R. J.; Malkov, A.; Lopp, M.; Kanger, T. *Advanced Synthesis & Catalysis* (**2013**), 355(5), 829-835.
5. Niidu, A.; Paju, A.; Müürisepp, A.-M.; Järving, I.; Kailas, T.; Pehk, T.; Lopp, M. *Chemistry of Heterocyclic Compounds* (NY, US) (**2013**), 48(12), 1751-1760.
6. Konik, Y.A.; Kananovich, D.G.; Kulinkovivch, O.G. *Tetrahedron* (**2013**), 69, 6673-6678.

T023 - Analüütilised lahutusmeetodid biomeditsiinis ja keskkonnakeemias. teema juht **Kaljurand Mihkel**

Analytical Separation Methods in Biomedicine and Environmental Chemistry

The research aims at developing new methods of analytical separation to the characterization of various samples of biological origin. The research will focus on the following.

1) A search for new buffers for capillary electrophoresis (CE) on analysis of biologically active compounds and biomass components.

2) The investigation of potential extragents and development of methods for the determination of the composition and antioxidative ability of plant extracts.

3) The miniaturization of CE instruments aiming at developing portable analyzers to solve environmental problems in situ.

4) The development of nanoporous materials (organic and carbon aerogels), to be used to separate various analytes by electrochromatography.

Results:

1) Green analytical chemistry methods based on microzone-paper for determining of extractable from plants was developed.
2) Different approaches in CE were developed for glutathione and its analogues, also alkaloids analysis.
3) CE methods for biomass treatment component analysis were developed.
4) Theory of green analytical chemistry was developed.

The most important publications 2013:

1. A.Tamm, A-L. Peikolainen, J.Kozlova, H.Mändar, A.Aidla, R.Rammula, L.Aarik, K.Roosalu, J.Lu, L.Hultman, M.Koel, K.Kukli, J. Aarik, Atomic layer deposition of high-k dielectrics on carbon nanoparticles Thin Solid Films,538 (2013)16-20
2. S.Hyvärinen, J-P. Mikkola, D.Yu.Murzin, M.Vaher, M.Kaljurand, M.Koel, Sugars and sugar derivatives in ionic liquid media obtained from lignocellulosic biomass: Comparison of capillary electrophoresis and chromatographic, Catalysis Today 223 (2013)18-24.
3. Kazarjan, Jana; Vaher, Merike; Mahlapuu, Riina; et al. , Separation of glutathione and its novel analogues and determination of their dissociation constants by capillary electrophoresis, Electrophoresis, 34,12(2013)1820-1827.
4. Kulp, Maria; Bragina, Olga, Capillary electrophoretic study of the synergistic biological effects of alkaloids from Chelidonium majus L. in normal and cancer cells, Analytical and Bioanalytical Chemistry 405,10(2013) 3391-3397.
5. Karmen Kapp, Elina Hakala, Anne Orav, Leena Pohjala, Pia Vuorela, Tõnu Püssa, Heikki Vuorela, Ain Raal , Commercial peppermint (Mentha x piperita L.) teas: Antichlamydial effect and polyphenolic composition, Food Research International 53, 2 (2013) 758-766.
6. M. Vaher, M. Borissova, A. Seiman, T. Aid, H. Kolde, J. Kazarjan, M. Kaljurand. Automatic spot preparation and image processing of paper microzone-based assays for analysis of bioactive compounds in plant extracts, Food Chemistry, 143(2014) 465-471.

T190 - Toidu süsteemibioloogia ja füüsika, teema juht **Vilu Raivo**
Food systems biology and physics

The key subjects of the study were: lactic acid bacteria, probiotics and yeast; single cell model of microorganisms; growth space of microorganisms; peptides and bioactive compounds. The aim of the project was development of systems biology of microorganisms and food production processes using omics methods and mathematical modelling.

Tulemused:

Development of multi-omics approach and single cell model (SCM) for the optimization of biotechnological processes of synthesis of ethanol, lactate etc. using recombinant E. coli cells. Development of models for aerobic growth of lactic acid bacteria. Optimization of anaerobic processes of co-digestion of biodegradable wastes. Evaluation of ecological efficiency of Estonian economy using multisector I/O models to study the growth efficiency and amino acid metabolism of bacteria, development of microcalorimetric methods for the study of lactic acid bacteria in milk and milk gels.

Tähtsamad publikatsioonid 2013

Stulova, I.; Kabanova, N.; Kriščiunaite, T.; Taivosalo, A.; Laht, T.-M.; Vilu, R. (2013). Fermentation of reconstituted milk by Streptococcus thermophilus: Effect of irradiation on skim milk powder. International Dairy Journal, 31(2), 139 - 149

Pitk, P.; Palatsi, J.; Affes, R.; Kaparaju, P.; Vilu, R. (2013). Co-digestion of sewage sludge and sterilized solid slaughterhouse waste: methane production efficiency and process limitations. Bioresource Technology, 134, 227 – 232.

Abner, Kristo; Aaviksaar, Tõnis; Adamberg, Kaarel; Vilu, Raivo (2013). Single-cell model of prokaryotic cell cycle. Journal of Theoretical Biology, 341, 78–87.

Valgepea, Kaspar; Adamberg, Kaarel; Seiman, Andrus; Vilu, Raivo (2013). Escherichia coli achieves faster growth by increasing catalytic and translation rates of proteins. Molecular Biosystems, 2344 - 2358

T133 - Biokatalüütiline stereokeemiline süntees, teema juht **Parve Omar**
Biocatalytic stereochemical synthesis.

A. Teadustöö kirjeldus:

The nucleation studies of 1,2-alkanediols.

The work on developing a scalable lipase-catalytic method for the kinetic resolution of long-chain 1,2-alkanediols and related tetrols, complemented by crystallization of the pure enantiomers from the reaction mixtures has been performed. This work has offered the possibility of a more detailed study of the aggregation of such diols. MD modeling, mass spectrometry, ^{1}H NMR, and DOSY studies provided a novel insight into the nucleation process. The research was carried out with the aim of developing a method for stereo- and chemoselective crystallization of an alcohol enantiomer from the reaction mixture. An efficient protocol for stereo- and chemoselective crystallization of (S)-1,2-dodecanediol and related compounds from the crude bioconversion mixtures was developed.

The experimental and modeling research of the promiscuous catalytic performance of *Thermomyces lanuginosus* lipase (TLL) and related lipases.

The switch of esterification to elimination of 11-acetyl-prostaglandin E_2 catalyzed by *Thermomyces lanuginosus* lipase (TLL), depending on the change of the content of methanol in the reaction medium, is explained by means of MD simulated solvation of the enzyme and docking of the substrate. The structural reason behind the switch is explained by an analysis of the geometry of the solvated TLL structures. We found that TLL with closed lid catalyzes the elimination of acetic acid from the substrate while TLL with open lid catalyzes the esterification of the substrate. The lid of TLL and related lipases opens in hydrophobic solvents. It opens also if the lipase recognizes the interface between water and a fatty lipid substrate in an emulsion. In these conditions the lipase catalyzes normal, acyl transfer reactions. On the other hand, the lid of TLL is closed in water, in some hydrophilic solvents (as was found – in methanol) and in case the lipase is unable to recognize the substrate/water interface. In these cases reactions occurring via proton abstraction and transfer (for instance, elimination) may be catalyzed by the lipase.
The initial rates and selectivities of the alternative reactions catalyzed by different lipases were estimated experimentally, using NMR.
Our findings may extend the scope of use of lipases in catalysis of different reactions of substrates of complex structure in asymmetric synthesis.

Synthesis and quantitative analysis of diastereomeric linked ester conjugates with remote stereocenters using high-field NMR and chiral HPLC.

A synthetic methodology for the preparation of hydroxycarboxylic acid linked ester conjugates has been created.

Several novel hydroxycarboxylic acid linked ester conjugates have been synthesized for being tested as putative agents for regulation of plasma triglyceride level.

Tähtsamad teadustulemused:

1. Several ^{1}H and DOSY spectral indications together with MD simulation results have provided a novel insight into the nucleation process. An efficient protocol for stereo- and chemoselective crystallization of (S)-1,2-dodecanediol and related compounds from the crude bioconversion mixtures was developed.
2. The structural reason behind the switch of esterification to elimination of 11-acetyl-prostaglandin E_2 catalyzed by *Thermomyces lanuginosus* lipase (TLL), depending on the change of the content of methanol in the reaction medium, is explained by means of MD simulated solvation of the enzyme and docking of the substrate: TLL with closed lid catalyzes elimination (an abnormal, promiscuous reaction) while TLL with open lid catalyzes normal, acyl transfer reactions.
3. A stereochemically safe high-yielding procedure for linking unprotected α- and γ-hydroxycarboxylic acids as well as protected hydroxycarboxylic acids to chiral secondary alcohols via glycolic acid linker is proposed. High-field NMR and and chiral HPLC determination of high diastereomeric ratio (>99%) of the products bearing remote stereocenters was explored.

Publications:

Omar Parve, Indrek Reile, Jaan Parve, Sergo Kasvandik, Marina Kudryašova, Sven Tamp, Andrus Metsala, Ly Villo, Tõnis Pehk, Jüri Jarvet, Lauri Vares. An NMR and MD modeling insight into nucleation of 1,2-alkanediols: selective crystallization of lipase-catalytically resolved enantiomers from the reaction mixtures. *J. Org. Chem.* 2013, *78*, 12795-12801.

Eva Doyle, Jaan Parve, Marina Kudryashova, Sven Tamp, Aleksander-Mati Müürisepp, Ly Villo, Lauri Vares, Tõnis Pehk, Omar Parve. Synthesis and quantitative analysis of diastereomeric linked ester conjugates with remote stereocenters using high-field NMR and chiral HPLC. *CHIRALITY* 25:793-798 (2013).

Hinnang oma teadustulemustele:

Vähemalt „hea“ (4).
Õnnestus kirjeldada terve rida uusi spektraalseid (NMR, MS) efekte, mis ilmnevad dioolide nukleatsioonil sõltuvalt analüüdi stereokeemilisest koostisest (puhas enantiomeer vs. ratsemaat) ning interpreteerida täheldatud efektid kasutades süsteemide molekulaardünaamika simulatsioone. Töötati välja lipaas-katalüütiline meetod 1,2-alkaandioolide ja sarnaste tetroolide stereoisomeeride preparatiivseks eraldamiseks.
Tulemused on avaldatud ajakirjas The Journal of Organic Chemistry.

T010 - Bioaktiivsed lipiidid - metabolism, signaaliülekanne ja regulatsioon , teema juht Samel Nigulas

Bioactive lipids - metabolism, signalling and regulation

Teadustöö kirjeldus

Lipids are essential components of the cell membrane shown to play many dynamic roles in mediating and controlling a wide array of cellular activities including membrane structure, metabolic and gene regulation, protein structure and function, energy production, and signalling pathways. Lipid mediators have been intimately linked to the immune and inflammatory

responses, cell proliferation and apoptosis, as well as shown to be major determinants in many pathologies, including diabetes, cancer, cardiovascular and neurodegenerative disorders. The research has been focused on: (i) elucidation of structural, catalytic, metabolic, regulatory, and evolutionary aspects of enzymes (cyclooxygenases, lipoxygenases and peroxidases) responsible for biosynthesis of lipid mediators, and (ii) study of influence on activity, stability and transport of lipoprotein lipase by apolipoproteins, fatty acids, angiopoietin-like proteins, heparan sulfates, and glycosylphosphatidyl-inositol-anchored high density lipoprotein-binding protein 1.

Aruandeaastal saavutatud tähtsamad teadustulemused

The first non-animal prostaglandin H synthase (PGHS) was cloned, expressed and characterized. The *Gracilaria vermiculophylla* PGHS, which shares only about 20% of the amino acid sequence identity with its animal counterparts, yet catalyzes the conversion of arachidonic acid into prostaglandin-endoperoxides, PGG_2 and PGH_2. Differently from animal PGHSs the *G. vermiculophylla* PGHS easily expresses in *E. coli* as a fully functional enzyme. The recombinant protein was identified as an oligomeric (evidently tetrameric) ferric heme protein. The preferred substrate for the algal PGHS is arachidonic acid and similarly to mammalian PGHS-2, the algal enzyme is capable of metabolizing ester and amide derivatives of arachidonic acid to corresponding prostaglandin products. Algal PGHS is not inhibited by non-steroidal anti-inflammatory drugs. (Varvas et al., 2013)

Our recent results suggest that apolipoprotein C-I and apolipoprotein C-III inhibit lipoprotein lipase activity by a similar mechanism. Both apolipoproteins can dose-dependently displace lipoprotein lipase from the lipid/water interface of lipoproteins and lipid emulsion particles. The presence of increased levels of these apolipoproteins may promote inactivation of lipoproteins by angipoietin-like proteins 4, since patients with renal insufficiency have high plasma levels of both apolipoprotein C-III and angipoietin-like proteins. Thus, our present results can possibly contribute to an explanation for the hypertriglyceridemia associated with kidney disease. (Larsson et al., 2013)

Olulisemad publikatsioonid aruandeaastal:

1. Kobzar, G.; Mardla, V.; Samel, N. (2013) Lactate is a possible mediator of the glucose effect on platelet inhibition. Platelets, xx - xx. [ilmumas]
2. Varvas, K., Kasvandik, S., Hansen, K., Järving, I., Morell, I., Samel, N. (2013) Structural and catalytic insights into the algal prostaglandin H synthase reveal atypical features of the first non-animal cyclooxygenase. Biochimica et Biophysica Acta - Molecular and Cell Biology of Lipids, 1831, 863 - 871.
3. Larsson, M., Vorrsjö, E., Talmud, P., Lookene, A., Olivecrona, G. (2013) Apolipoproteins C-I and C-III Inhibit Lipoprotein Lipase Activity by Displacement of the Enzyme from Lipid Droplets. . Journal of Biological Chemistry, 288, 33997 - 34008.
4. Mendoza-Barberá, E., Julve, J., Nilsson, S.K., Lookene, A., Martín-Campos, J.M., Roig, R., Lechuga-Sancho, A.M., Sloan, J.H., Fuentes-Prior, P., Blanco-Vaca, F. (2013) Structural and functional analysis of APOA5 mutations identified in patients with severe hypertriglyceridemia. Journal of Lipid Research, 54, 649 - 661.

T031A - Uued arvutusmeetodid keerukate biomolekulide süsteemide kirjeldamiseks, teema juht **Karelson Mati**

Modeling of biomedically and environmentally important systems using computational chemistry

The subject of the research has been the computational study of detailed mechanisms of interactions of chemical compounds with the living organisms and environment. The research has been carried out by the development of new computational methods and the respective software. The novel methodological approaches include development of (1) ab initio quantum-chemical descriptors for molecules in external fields; (2) new quantum molecular dynamics based molecular docking techniques; (3) new algorithms for the search of optimum conformational structure of flexible molecules; and (4) implementation of advanced mathematical methods for the structure-activity relationships. The methodology developed is applicable for the description and prediction of (1) physicochemical properties; (2) pharmacodynamic and pharmacokinetic data; (3) antiviral activity of compounds; (4) activity of mimetics of neurotrophic factors; (5) structure and properties of peptide delivery vectors.

Tähtsamad teadustulemused 2013

A novel computational technology based on modified topoogical molecular descriptors has been used for the fast and efficient prediction of activities of LRRK2 kinase inhibitors. Quantum-chemical methods at various levels were applied for the study of reaction paths of possible synthetic routes for 2,5-diketopyraines. Quantitative structure-activity relationship (QSAR) models of cellular uptake of cell-penetrating peptides were developed using descriptors including hydrogen bonding, peptide charge and positions of nitrogen atoms.

Olulisemad publikatsioonid 2013

1. D.A. Dobchev, I. Tulp, G. Karelson, T. Tamm, K. Tämm, M. Karelson Subchronic Oral and Inhalation Toxicities: a Challenging Attempt for Modeling and Prediction, Mol. Informatics, 32, 793–801 (2013).
2. K.Ha, I. Lebedyeva, Z. Li, K. Martin, B. Williams, E. Faby, A. Nasajpour, G.G. Pillai, A.O. Al-Youbi, A.R. Katritzky, Conformationally Assisted Lactamizations for the Synthesis of Symmetrical and Unsymmetrical Bis-2,5-diketopiperazines, J. Org. Chem.,78, 8510–8523 (2013).
3. J. Regberg, A. Srimanee, M. Erlandsson, R. Sillard, D.A. Dobchev, M. Karelson, Ü. Langel, Rational design of a series of novel amphipathic cell-penetrating peptides, International Journal of Pharmaceutics (in press).

Välisorganisatsioonide liikmed

Mati Karelson – American Chemical Society; International Academy of Mathematical Chemistry

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3. Tunnustus

Eesti Vabariigi aastapreemia kandidaadiks keemia ja molekulaarbioloogia valdkonnas esitati matemaatika-loodusteaduskonna keemiainstituudi juhtivteadur **Aivar Lõokene** uurimistöö „Lipoproteiinide metabolismi regulatsioonimehhanismid” eest.