Joint Annual Meeting of Finnish Synchrotron Radiation User Organisation (FSRUO) and Finnish Structural Biology Network (FinnBoX)

4-5 December 2017, Turku, Finland
Venue: Arc1, Arcanum, University of Turku

✓ Public talks, poster session, networking
✓ Credit points for graduate students

Invited talks:
- Prof. Serguei Molodtsov, European XFEL
- Dr. Harald Reichert, ESRF
- Prof. Derek Logan, MAX IV Laboratory
- Dr. Zoë Fisher, ESS
- Prof. Sarah Butcher, University of Helsinki
- Dr. Tommi Nyrönen, CSC - IT Center for Science
- Dr. Andreas Scherer, Institute for Molecular Medicine Finland
- Dr. Eero Itälä, University of Turku

See the program at www.fsruo.fi
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Organized by:
Finnish Synchrotron Radiation User Organisation (FSRUO) and Finnish Structural Biology Network (FinnBox) in collaboration with University of Turku, Åbo Akademi University, University of Helsinki, University of Oulu and Tampere University of Technology

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Development of Advanced Fe–Cr Alloys for Demanding Applications

Utilizing Synchrotron Light Mediated Electron Spectroscopy

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Abstract

High-temperature corrosion resistance of ferritic stainless steels (Fe–Cr based alloys) is built upon the formation of protective Cr-rich oxide scale. However, Cr vaporization limits the use of Fe–Cr alloys under extreme service conditions; in particular, it has been identified as the most significant failure mechanism in solid-oxide fuel cells (SOFCs). Our study focusses on the initial stages of oxide scale formation on ferritic stainless steels and shows that the Cr vaporization can be controlled via the alloy composition and heat treatments.

In this work, we investigate the influence of heat treatment on the initial stages of oxidation of two Ti–Nb stabilized ferritic stainless steels (EN 1.4509\textsuperscript{1,2} and EN 1.4521\textsuperscript{3,4}) at 650 °C by synchrotron light mediated X-ray photoelectron spectroscopy (XPS) and photoemission electron microscopy (PEEM). The high degree of alloying makes these alloys suitable for high temperature applications, but also renders the alloys prone to microstructural changes that can affect the growth of protective oxide scale. As a demonstration of this, we show that the heat treatment induced precipitation of (FeCrSi)\textsubscript{2}(MoNb)-type Laves phase results in less pronounced surface segregation and oxidation of minor alloying elements (Mo, Mn, Nb, Ti, Si). Most significantly, the diffusion of Mn and the formation of low volatile (MnCr)\textsubscript{3}O\textsubscript{4} spinel oxide at the surface above Cr\textsubscript{2}O\textsubscript{3} are strongly suppressed.

Photodissociation of acetamide clusters by synchrotron radiation in the VUV region

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Large prebiotic molecules, such as acetamide, found in space, are of biological interest. Being the largest interstellar molecule known containing a peptide group, acetamide could be the source of larger peptides and therefore play a potential role in prebiotic chemistry (Ref. 1). The clusterization of acetamide molecules can be considered as a simple model system to explore biologically important chemical processes triggered by the solar radiation.

We studied photofragmentation of small gas-phase acetamide clusters produced by supersonic expansion source using time-of-flight (TOF) ion mass spectroscopy combined with tunable vacuum-ultraviolet synchrotron radiation (Ref. 2). Fragmentation channels of acetamide clusters under vacuum-ultraviolet photoionization resulting in the formation of the protonated and ammoniated cluster ions were identified with the discussion about the preceding intramolecular rearrangements. The influence of the photon energy on the stability of the clusters and their fragmentation channels was addressed. Also, the most stable arrangement of the acetamide dimer was identified. An important new result of this study was tracking the exact proton transfer path in protonation reactions with the help of deuterated sample, identifying that proton transfer from the amino group is a dominant proton transfer mechanism upon near threshold photoionization.

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References
X-ray absorption spectroscopy without a synchrotron for actinide research: example of uranium compounds.

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X-ray Absorption Spectroscopy (XAS) is a well-established non-destructive method to determine both oxidation state and local environment of one given element in materials. Its major strength is that no special sample preparation is usually required, and that it is a bulk sensitive method due to the long-range penetration length of X-rays in matter. Thus, XAS allows to perform measurements despite the necessary but highly constraining confinement barrier arising from safety considerations when studying radioactive matter, and especially actinide’s bearing materials such as nuclear fuels. However, XAS experiments need a monochromatic, tunable over a wide range of energy and high flux photon beam. These needs have strongly limited its development to only synchrotron radiation facilities. In addition, the finite and low success rate of synchrotron beamtime access, the high costs of radioactive sample shipment and the low number of dedicated beamline dedicated to radioactive materials have strongly limited the experimental opportunities, and subsequently excluded a large amount of potentially important scientific research to be considered. Thanks to the recent development of spherically bent analyzer crystals, one can develop synchrotron’s alternatives or complement to compensate the currently lacking beamtime availability. One of these an efficient and cost-effective benchtop alternative has been recently built at the University of Helsinki. Its first application on actinide’s bearing materials at the U L₃-edge in transmission mode will be discussed and compared to synchrotron-based studies.
Photo-electrochemical and spectroscopic investigation of ALD grown TiO$_2$: Charge transfer characterization and effect of post annealing at different temperature.

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**Abstract**

Inspired by the photo-electrochemical water oxidation system reported by Fujishima and Honda$^1$, recent work has focused on functionalizing photoactive TiO$_2$ thin films on silicon (Si) semiconductor. Targeting to design an efficient photo-electrochemical device for solar fuel production, finding suitable protection layer material for semiconductors like Si, has recently gained significant attention$^2$.

In this work, TiO$_2$ thin films were deposited on highly doped Si substrate by atomic layer deposition (ALD) technique using tetrakis(dimethylamido) titanium (TDMAT) and water as precursors. In order to understand the influence of ALD parameters on TiO$_2$ film performance in photo-electrochemical cell, ALD growth temperature was varied from 150 °C to 225 °C and film thickness from 20 nm to 50 nm. Further efforts were made to analyze the effect of post-annealing treatment in air on ALD films and its influence on photo-electrochemical water oxidation reaction.

The highest applied bias photon-to-current efficiency for Solar Water Splitting (SWS) was obtained in 30 nm ALD TiO$_2$ film grown at 200 °C after post annealing at 475 °C. Annealing at higher temperatures decreased the photo-activity substantially. X-ray photoelectron spectroscopy analysis of TiO$_2$ (2 nm)/Si samples after annealing in air revealed the onset of interfacial SiO$_2$ formation at 450 °C. SiO$_2$ at the TiO$_2$/Si interface act as a charge transfer barrier with detrimental consequence on SWS on TiO$_2$/Si photo-anode.

New Insights Into the Specificity of Mammalian Copper Amine Oxidases

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Humans have three copper-containing amine oxidases (CAOs): human diamine oxidase (AOC1; hDAO) prefers diamines as substrates, whereas retina-specific CAO (AOC2; hRAO) and human vascular adhesion protein-1 (AOC3; hVAP-1) are monoamine oxidases with preferences for aromatic and aliphatic substrates, respectively. Furthermore, some mammals also contain an additional soluble CAO (AOC4). CAOs are structurally complex and the catalytic reaction can be inhibited in two alternative ways: by targeting the active site topaquinone or by targeting the active site channel. Inhibitors targeted to the channel pose problems for preclinical animal tests, since the channel is evolutionary less conserved and brings about species-specific difference. In this study, an extensive phylogenetic analysis of CAOs comprising 78 mammalian species was complemented with sequence and in silico structural studies to get new insights into the origin of their substrate specificity. The phylogenetic results show a clear division of CAOs into four sub-families, which are in accordance with the known substrate preferences. Additionally, this study helps to derive rules for the classification of new and inadequately characterized CAOs, especially the poorly annotated AOC3 and AOC4 proteins.

Moreover, a detailed analysis of the active site structure of each CAO member was performed and complemented with substrate docking. Thereby, residues, which are conserved within one sub-family but different between sub-families, could be identified. Altogether, the results give new insights into the substrate specificity and preference of each CAO sub-family, which is essential for the design of new and specific inhibitors.
Directed Evolution on FucO – Structural Explanations for Changes in Substrate Scope

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Propanediol oxidoreductase from Escherichia coli (FucO) uses NADH/NAD⁺ as cofactors to catalyze the conversion of S-lactaldehyde to S-1,2-propanediol and vice versa. FucO is an attractive enzyme in the search for possible biocatalysts producing α-hydroxy aldehydes, which are important for the synthesis of natural products and synthetic drugs. Enzymes catalyzing these types of reactions are unique in catalytic power and stereoselectivity. The usage of FucO in synthetic industry is limited by the restricted substrate scope, which makes FucO inactive with larger phenyl-substituted alcohols. We used re-engineering and directed evolution to enable FucO to catalyze the regio- and enantioselective oxidation of arylsubstituted vicinal diols, such as phenylpropanediols, into α-hydroxy aldehyde products. We mutated amino acids considered to restrict the entry into the active site, and modeled the mutants that were most active with the substrates phenylacetaldehyde and S-3-phenyl-1,2-propanediol and performed docking studies with them. As expected, our experimental and in silico results show that the mutations enlarge the active site cavity and enable the mutant enzymes to accommodate the new substrates. We also found specific amino acids in the active site, which need to be conserved to allow the substrates to make stabilizing interactions. Interestingly, an asparagine residue makes the mutant enzymes able to discriminate between phenylacetaldehyde and S-3-phenyl-1,2-propanediol. In conclusion, we successfully re-engineered the specialist enzyme FucO to accept also bulkier molecules as substrates, thereby making it more useful for industrial purposes.
The alpha/beta-Hydrolases (ABH) are a structural family of enzymes that share a common fold and a similar acid-base-nucleophile catalytic mechanism, but vary in their sequence similarity and function. The ABH enzymes are prime targets for protein engineering.

In our study, we have chosen 42 representative structures from 40 structural ABH fold families and analyzed the structure of their active sites, focusing on the areas around the catalytic nucleophile residue and the oxyanion hole. We show the occurrence of two conserved geometries: the nucleophile zone and the oxyanion zone, which coordinate each key part of the catalytic mechanism. We present conserved positions of the zones and make use of multiple experimental evidence which confirm that these structural elements in the periphery of the active site are indispensable for the enzymatic activity. We also show the occurrence of an aromatic cluster that lies close to the active site and above the planes of the nucleophile and oxyanion zones.

We propose that the nucleophile zone, the oxyanion zone and the aromatic cluster all compose a three-dimensional construct located near the active site and opposite of the ligand-binding site. We suggest that this assembly outlines the shape of the active site and plays an important role in the enzymatic function by structurally stabilizing the nucleophile and oxyanion hole positions. Finally, we speculate that the aromatic cluster can play a role in the coordination of the catalytic histidine loop and consequently, fix the catalytic histidine next to the catalytic nucleophile.
Observe while it happens: catching a photoreceptor in the act with a free electron laser

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Many biological processes depend on detecting and responding to light. The response is often mediated by a structural change in a protein that begins when absorption of a photon causes isomerization of a chromophore bound to the protein. We have used x-ray pulses emitted by a free electron laser source to conduct time-resolved serial femtosecond crystallography in the time range of 100 fs to 3 ps. This enabled the real-time tracking of the trans-to-cis isomerization of the chromophore in photoactive yellow protein and the associated structural changes in the protein.

Figure 1: a stream of photoreceptor protein crystals is first hit by a blue laser to initiate a chemical (reaction and then by an x-ray laser to probe the dynamics of the atoms in response to the blue light (SLAC National Accelerator Laboratory illustration)

Serial synchrotron crystallography at EMBL PETRA III beamline P14.

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Keywords: serial synchrotron crystallography

Serial synchrotron crystallography (SSX) combines X-ray images taken from randomly oriented crystals, while being passed through the beam using diverse delivery methods, into a single dataset. The method requires a high-brilliance synchrotron source with a beam size similar to the sample size, an appropriate sample delivery method, a detector with sufficient frame rate and a data processing pipeline. SSX can act as a pre-screening method for samples intended for XFEL experiments or a stand-alone experiment to determine the protein structure when crystal growth to larger size cannot be achieved, e.g. in vivo grown crystals. Proof of principle experiments (1) have shown the feasibility of the method.

Our SSX setup at the EMBL beamline P14 on the PETRA III storage ring at DESY (Hamburg, Germany) utilizes the sample delivery systems known from conventional crystallography - thus setup time is negligible and sample consumption is very low. SSX experiments can be done in situ in CrystalDirect™ plates, also in meso, possibly using the same plate the initial crystals grew in. Cryo-samples are mounted in loops or harvested from plates with the CrystalDirect™ Harvester. Generally, the sample size for a feasible experiment is a few microns. Data collection runs as series of helical line scans, typically a dataset is collected in a few minutes, depending on the size of the region of interest, crystal size and the detector used (Pilatus6M, Eiger4M, Eiger16M). Progression of the data collection is monitored throughout the experiment as on-the-fly calculated heat map, displaying the diffraction scores as estimated by program DOZOR (2).

The acquired diffraction images are sorted and bunched into sub datasets according to the DOZOR score using a script. The script generates XDS-processing files for each sub dataset and processing of all of these can be launched in parallel. Data is scaled using XSCALE. Data processing takes typically 30 minutes on good quality data. We have demonstrated the feasibility of the pipeline using 5-10 um lysozyme and insulin crystals as test objects. Using an Eiger4M detector, a data set of 65120 images was collected at P14 in 3 minutes as an in situ experiment of 5 micron lysozyme crystals grown and presented to the beam in a CrystalDirect™ plate. About 2000 sub dataset, each containing 5-10 diffraction images, were integrated and scaled to yield complete data to 1.7 Å resolution. The structure could be solved by molecular replacement and electron density maps were of good quality. Beamtime for SSX experiment can be applied for through the EMBL user program at https://smis.embl-hamburg.de.

Biological Sample Preparation at the European XFEL

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The European XFEL is the largest and most powerful X-ray laser in the world and has just hosted the first user experiments. The facility provides ultrashort X-ray flashes on a femtosecond scale, with a peak brilliance orders of magnitude higher than synchrotron radiation. These unique properties provide completely new opportunities for users from different fields of physics, chemistry, material sciences, biosciences, and medicine. For structural biology, the most exciting prospect is the potential to image large biomolecules and complexes - or even entire organelles or cells at high resolution using single-particle approaches. Currently, two of the six planned instruments are operational: SPB/SFX for serial femtosecond crystallography and single particle imaging of biological macromolecules as well as FXE for dynamic studies of chemical reactions. The European XFEL is a joint effort of 12 countries: Denmark, France, Germany, Hungary, Italy, Poland, Russia, Slovakia, Spain, Sweden, Switzerland, and the United Kingdom.

One of the crucial factors for the success of experiments at XFEL is sample quality. The XBI User Consortium has established a biological sample preparation laboratory in direct vicinity of the SPB/SFX instrument. The laboratory offers facilities and expertise for all steps of sample preparation and characterization for a wide range of biological systems. XBI will enable research groups from all over the world to make use of the revolutionary possibilities offered by the European XFEL. The University of Oulu is the current coordinator of the XBI User Consortium. Other members come from the EMBL, Germany, Slovakia, Sweden, and the United States.
HelXAS: a low-cost home laboratory instrument for X-ray absorption spectroscopy

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X-ray absorption spectroscopy (XAS) is an well-established technique for obtaining element-specific information on the local structure and chemical state. Synchrotron beamtime for XAS experiments, however, is in high demand and thus difficult to obtain for e.g. routine characterisation and studies of slow chemical reactions. In addition, utilizing XAS for radiochemistry research is especially difficult as many synchrotrons do not allow radioactive materials on site.

To alleviate these problems, we present HelXAS: a low-cost laboratory XAS instrument utilizing a conventional X-ray tube source and bent Johann-type crystal monochromators. HelXAS is designed for XAS studies in the 5–20 keV range which covers most K edges of 3d transition metals and L edges of 5d transition metals and actinides. The typical energy resolution is around 1–2 eV. Measurements can be performed in transmission and fluorescence detection modes, the latter enabling the study of samples on thick substrate/support. Due to its simple and modular design, HelXAS can modified to accommodate additional equipment and complex sample environments required for e.g. in situ studies. A showcase of various applications is presented.
Synchrotron-based soft x-ray spectromicroscopy for bio-, medical and hard materials characterization

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Various synchrotron based imaging and characterization methods provide novel versatile toolkit, which is opening entries for emerging fields of science and technology as users of synchrotron facilities. In the present talk, an introduction is provided on the existing and planned research collaboration of NANOMO Research Unit benefitting soft x-ray spectromicroscopy known as scanning transmission x-ray microscopy. Few experiments performed at UVSOR-III and SOLEIL facilities on biopolymers, biological and human tissue samples are given. The research involves collaboration with Natural Resources Institute Finland (Luke), Northern Ostrobothnia Hospital District (PPSHP), Center for Advanced Steel Research and EU-MSCA I4Future doctoral programme.
Topic: Structural and functional characterization of neuronal leucine rich repeat adhesion proteins of SALM families

Abstract

Millions of people worldwide are affected by neurological disorders. Growing evidence on genetic components of neurological disorders has been collected and among them are genes encoding for neuronal adhesion proteins like synaptic adhesion-like molecules (SALM) protein families. SALMs has been implicated in severe progressive autism and familial schizophrenia. In my PhD project (2016-2019), I am focusing on structural and functional characterization of SALM proteins, in particular SALM3 an SALM5.

We have developed robust protein expression constructs and purified proteins of SALM3 and SALM5, also purified the identified ligand; LAR-family receptor tyrosine phosphatase sigma (RPTPs). We have crystallized the extracellular LRR and Ig domains of SALM5 and solved the structure at resolution of 3.0 Å. Biophysical characterization of the SALMs show that they form dimers in solution. Binding experiments show binding affinity for SALM3-RPTPs with mini-exon B (RPTPs-meB) at 2 uM, SALM3-RPTPs at 25 uM, SALM5-RPTPs at 8 uM and SALM3-RPTPs-meB at 6 uM. I am working to solve the structure of protein complex and also map the binding site of the complex using mutational study.

Lack of a high-resolution structure of these synaptic adhesion molecules and their ligand complexes has limited our understanding of how these proteins act in synapse formation. Understanding of the detailed molecular mechanisms underlying the functions of synaptic adhesion pathways will be crucial for the further analysis of the biological function and for potential drug design and development.
Protein production for biophysical and structural characterization in Tampere is essential supplement for Structural biology platform services. Protein expression in several organisms offers suitable system for any recombinant protein that is needed with proper scale-up possibility. Protein quality formulation is defined by SEC-UV/LS/RI analysis. Moreover, service offers protein interaction characterization with wide variety of methods e.g BLI/SPR for high throughput kinetics screening (Octet Red384 and Bionavis SPR Ilves) and calorimetric methods for protein stability, fragment screening (cap-DSC) and complete thermodynamic characterization of the binding process (ITC).
Chaperones from non-classical chaperone-usher pathways preserve folding energy of pilins by providing them an incomplete structural information

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Classical, alternative and archaic chaperone-usher pathways (CUPs) assemble adhesive pili that mediate host or self-recognition of Gram-negative bacteria. The classical CUP has served as a model system to study pilus biogenesis and its inhibition. However, recent structural insights suggested that the mechanism of pilus assembly via the more ubiquitous non-classical CUPs is different. Non-classical chaperones, unlike their classical counterparts, maintain pilus subunits (pilins) in a substantially disordered conformational state, which contradicts with the established steric chaperone concept. To uncover this novel assembly mechanism and gain insight into the architecture of archaic pili, we determined the crystal structure of the self-complemented CsuA/B pilin of Csu biofilm-forming pili from *Acinetobacter baumannii* and characterized conformational dynamics and thermodynamics of CsuA/B before and after assembly. We found that non-classical chaperones use a much shorter donor strand motif than the sequence required for the full complementation of pilins. Thus, non-classical chaperones provide only partial steric information for pilins to fold, preserving their folding energy to drive the pilus assembly. We also discovered that the first β sheet in archaic pilins is disrupted by an insertion of unique hairpins and this structure undergoes a large conformational change that further drives the assembly.
Passivation of surface states on hematite photoelectrode by ALD grown TiO\textsubscript{2} for efficient solar water splitting

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Abstract

Photoelectrochemical water splitting is a method to convert solar energy directly into hydrogen fuel on a semiconductor surface. Unassisted solar water splitting requires a material that absorbs light with photon energies above the thermodynamical energy requirement for H\textsubscript{2}O redox reaction, 1.23 eV. One promising material for large scale solar hydrogen production is abundant and inexpensive hematite, an oxide of iron (\(\alpha\)-Fe\textsubscript{2}O\textsubscript{3}) with a band gap of 2.2 eV [1,2]. The major problem limiting the efficiency of hematite as photocatalyst is recombination of charge carriers in surface states at the semiconductor–electrolyte interface.

In this work, we have fabricated and studied hematite thin films and their surface state passivation using atomic layer deposition (ALD) of TiO\textsubscript{2}. Hematite films were fabricated by anodic electrodeposition of FeOOH from FeCl\textsubscript{2} on indium tin-oxide-coated (ITO) glass. FeOOH was then converted into hematite (\(\alpha\)-Fe\textsubscript{2}O\textsubscript{3}) by annealing the FeOOH-film in air. The effect of film thicknesses and heat treatments were investigated. Hematite films were studied by using spectrophotometry, X-ray photoelectron spectroscopy (XPS), photoelectrochemical (PEC) measurements and electrochemical impedance spectroscopy.

Film thickness was found to depend linearly on the electric charge used to deposit FeOOH. Electrodeposition and heat treatment were optimized so that the hematite films produced the highest possible photocurrent at the H\textsubscript{2}O redox potential. Optimal film thickness was found to be 60 nm. Films annealed to temperatures below 750 °C did not produce photocurrent. The achieved photocurrent increased with annealing time at 750 °C. Annealing times up to 32 hours were tested and photocurrents up to 0.7 mA/cm\textsuperscript{2} were achieved. Tauc-method was used to determine optical band gap of hematite films. The band gap was 2.2 eV regardless of the film thickness. The surface state capacitance was strongly reduced upon deposition of only 0.5 nm TiO\textsubscript{2} layer on hematite photoelectrode.


Tumor suppressor phosphatase PP2A dephosphorylates pro-survival and proliferation factors that drive cancer development. Such diversity in activity is achieved through PP2A’s structural organisation. PP2A exists as a core enzyme composed of catalytic (C) and scaffolding (A) subunit, or as a heterotrimer between the core enzyme and one of the regulatory (B) subunits. As an examples of B-subunit specific biological systems are tumor suppression by B56 family proteins and inhibition of Alzheimer’s disease (AD) relevant Tau protein phosphorylation by B55-PP2A complexes. The most prevalent mechanism of PP2A’s inactivation is overexpression of its endogenous inhibitory proteins, such as Cancerous inhibitor of PP2A, CIP2A. CIP2A is a human oncoprotein, which has emerged as a clinically relevant prognostic marker in cancers. CIP2A also inhibits the Tau and Amyloid Precursor Protein (APP)-related PP2A activities in brain causing to worsening of the Alzheimer-like pathologies. Therefore, reactivation of PP2A is recognised as a valid therapeutic strategy for cancer and AD. CIP2A’s crystal structure published recently showed that the protein forms obligate homodimers and associates with the regulatory B56-PP2A. It is not understood how CIP2A regulates distinct pool of PP2A-B56 and PP2A-B55 complexes, without inducing global PP2A inhibition. Our aim is to elucidate the co-structure of CIP2A with the regulatory B56α subunit of PP2A by chemical cross-linking coupled to mass spectrometry approach (XL-MS/MS), providing the first structure-supported molecular mechanism for CIP2A-mediated inhibition of PP2A activity on B-subunit specific manner. We expect our novel findings to have strong biomedical impact and aid in general understanding of CIP2A's biology.
Prerequisite for the development of new treatments for diseases is the atomic level structural information of the protein(s) involved. The aim of my group is to understand the molecular mechanisms of various diseases by combining structural biology techniques such as X-ray crystallography, SAXS and protein NMR with state of art biochemical and biophysical methods. As an example, I will discuss about our research about X-linked myxomatous valvular dystrophy (XMVD) caused by four different missense mutations in Filamin, a large actin crosslinking protein. Valvular dystrophy is affected ~3% of population and is the main cause of the valvular surgery as no drug-based treatment exists. The underlying molecular mechanism is not understood. We have solved the crystal structure of XMVD causing P637Q mutated Filamin, which revealed that P637Q mutation does not affect the overall folding. This was further confirmed by SAXS experiments. P637Q mutation did not affect Filamin’s interactions with other proteins either. Filamin is known to be a central mechanotransduction element of the cytoskeleton, and endocardial endothelial cell mechanotransduction play a critical role in valve development. Performed steered molecular dynamics simulations revealed that force response of WT and P637Q mutated Filamin is different. Accordingly, the altered mechanotransduction might be the underlying mechanism behind P637Q mutation caused XMVD. For the three other mutations, NMR spectroscopy was applied, which revealed that these mutations partly unfold Filamin. The phenotype of P637Q caused XMVD is different than that of three other mutations, which might be explained by the different molecular level mechanisms.
Catechol oxidases together with tyrosinases belong to a family of coupled binuclear copper (CBC) enzymes. They share a type-3 copper site, where two copper ions bind molecular oxygen. CBC enzymes use dioxygen for oxidizing various phenolic compounds. The copper site exists in four possible forms called oxy-, hydroperoxide-, met-, and deoxy. Despite the similar copper site, tyrosinases are able to oxidize monophenols to diphenols and subsequently to corresponding quinones, whereas catechol oxidases show only the latter activity. Tyrosinases and catechol oxidases are studied over three decades but still no certainty of reaction mechanism exists.

We have studied catechol oxidase from Aspergillus oryzae (AoCO4)\(^1\). Now, we have solved two new crystal structures of AoCO4 in a new triclinic crystal form at 1.8 and 2.5-Å resolutions. These structures showed different copper site forms (met/deoxy and deoxy) and also differed from the copper site observed in the previously solved structure of AoCO4.

The MAX IV laboratory is a new synchrotron radiation research center located in Lund, Sweden. It was inaugurated in June 2016. Currently, nine beamlines have been constructed at the MAX IV facility.

The FinEstBeAMS is a materials and atmospheric science beamline located at the 1.5 GeV storage ring of the MAX IV facility. The beamline has two branch lines and three permanent end stations. The gas-phase end station and the photoluminescence end station (FinEstLUMI) have been installed and tested with an external light source. The solid state end station is under manufacturing. The FinEstBeAMS is under commissioning and will provide first light for users in 2018.

The beamline will provide photons in the energy range of 4.3–1000 eV with high flux $8 \times 10^{13}$ ph/s – $1 \times 10^{11}$ ph/s and resolving power up to 10000. Dedicated end stations will allow investigations in many different research areas. One can, e.g., study processes occurring in the upper atmosphere, fragmentation pathways of bio- and organic molecules, formation of nanoparticles, luminescent materials, nanomolecular layers on alloy surfaces, and electrochemical double layer capacitors \textit{in situ}.

Main funding for the FinEstBeAMS beamline has been received from the Academy of Finland through the Finnish Research Infrastructure funding projects [1-3] and from the European Union through the European Regional Development Fund [4].

Crystallisation and crystal structure of *Caulobacter crescentus* xylonolactonase

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*Caulobacter crescentus* xylonolactonase (XylC) that catalyses the lactonisation of L-xylonic acid has been successfully crystallised, and its crystal structure has been preliminarily determined with molecular replacement using X-ray diffraction data. Small crystals obtained in optimised conditions have been measured in synchrotrons at ESRF in Grenoble, France and at Diamond in Oxfordshire, England. The best crystal structure thus far has a resolution of 2.8 Å, which gives a good overview of the general structure, but for determining important structural details at the active site higher resolution is still desired.
Molecular mechanisms of Charcot-Marie-Tooth neuropathy linked to mutations in human myelin protein P2

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Charcot-Marie-Tooth (CMT) disease is one of the most common inherited neuropathies. Recently, three CMT1-associated point mutations (I43N, T51P, and I52T) were discovered in the abundant peripheral myelin protein P2. These mutations trigger abnormal myelin structure, leading to reduced nerve conduction velocity, muscle weakness, and distal limb atrophy. P2 is a myelin-specific protein expressed by Schwann cells that binds to fatty acids and membranes, contributing to peripheral myelin lipid homeostasis. We studied the molecular basis of the P2 patient mutations. None of the CMT1-associated mutations alter the overall folding of P2 in the crystal state. P2 disease variants show increased aggregation tendency and remarkably reduced stability, T51P being most severe. In addition, P2 disease mutations affect protein dynamics. Both fatty acid binding by P2 and the kinetics of its membrane interactions are affected by the mutations. Experiments and simulations suggest opening of the β barrel in T51P, possibly representing a general mechanism in fatty acid-binding proteins. Our findings demonstrate that altered biophysical properties and functional dynamics of P2 may cause myelin defects in CMT1 patients. At the molecular level, a few malformed hydrogen bonds lead to structural instability and misregulation of conformational changes related to ligand exchange and membrane binding.
Role of Oxide Defects in ALD grown TiO$_2$ Coatings on Performance as Photoanode Protection Layer

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Abstract

Photoelectrochemical (PEC) water splitting is one of the potential methods of utilizing solar energy. A major issue for the method and for renewable energy production is the development of an efficient, chemically stable and cost-effective semiconductor photoanode. Recently, titanium dioxide (TiO$_2$) coatings grown by atomic layer deposition (ALD) have appeared to be a promising approach to stabilize semiconductor photoanodes under PEC conditions. In particular, amorphous ALD grown TiO$_2$ has shown exceptional charge transfer properties compared to its crystalline form that are not properly understood yet [1]. Therefore, we target to gain better understanding on the defect structure of ALD grown TiO$_2$ and utilize the information in the development of optimal photoanode protection layer for efficient solar water splitting.

In this work [2], structural, optical and photoelectrochemical properties of the ALD grown TiO$_2$ films were studied in as-deposited condition and after annealing in air at 500 °C. TiO$_2$ films were grown on n-type phosphorus-doped silicon and fused quartz by ALD at 200 °C using tetrakis(dimethylamido)titanium (TDMAT) and deionized water as precursors. The properties of TiO$_2$ were investigated by X-ray photoelectron spectroscopy (XPS), ellipsometry and UV/Vis/NIR spectrophotometry. In addition, results from X-ray diffraction (XRD), Raman spectroscopy and photoelectrochemical (PEC) cell are discussed.

Based on the results, as-deposited TiO$_2$ is amorphous and absorbs visible light as "black" TiO$_2$. After annealing in air at 500 °C TiO$_2$ crystallizes as rutile and becomes "white" TiO$_2$ that absorbs light only in the UV region. As-deposited TiO$_2$ contains significant amount of Ti$^{3+}/2+$ oxygen vacancies that are oxidized as Ti$^{4+}$ upon annealing in air. In addition, nitrogen is found only in as-deposited titanium dioxide. As-deposited TiO$_2$ is not chemically stable under PEC conditions. In contrast, the annealed TiO$_2$ is chemically stable and showed 0.20 % ABPE efficiency for water splitting reaction.

ALD-grown cathode material for LIBs: XANES and EXAFS analysis of Li$_x$Mn$_2$O$_4$

Owing to their light weight and high energy density, lithium-ion batteries (LIBs) have become the technology of choice for energy storage in a wide range of applications such as hybrid and electric vehicles, portable consumer electronics and microelectronics. Among the available cathode materials for this type of batteries, lithium manganese oxide spinel (Li$_x$Mn$_2$O$_4$) is a promising candidate due to its low cost, low toxicity, high specific capacity, and minimal structural distortion during charge and discharge cycles.

In this project, thin-films samples of Li$_x$Mn$_2$O$_4$ were produced by means of ALD. Different lithiation cycles were used to transform ALD-MnO$_2$ into Li$_x$Mn$_2$O$_4$ spinel. To gain a fundamental understanding of this process, the local atomic and electronic structure of the samples were investigated by means of X-ray absorption spectroscopy. Experimental measurements were collected at the CLÆSS beamline, from ALBA synchrotron radiation facility, at the Mn K-edge in fluorescence and total electron yield mode. A XANES (X-ray absorption near edge structure) analysis was performed to qualitatively determine the changes in the Mn average oxidation state (AOS) upon increasing lithiation cycles. Likewise, an EXAFS (extended X-ray absorption fine structure) analysis was done to obtain information of the geometrical changes near the Mn atom by fitting the EXAFS region of the spectra with theoretical models. It was demonstrated that upon increasing the number of lithiation cycles, there is a decrease in the Mn AOS and an increase of the local distortion around the Mn atom.
Structural basis for *Acinetobacter baumannii* biofilm formation

One sentence summary: Hydrophobic loops exposed at tips of Csu pili enable *Acinetobacter baumannii* attachment to abiotic surfaces

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Abstract

*Acinetobacter baumannii*—a leading cause of nosocomial infections—has a remarkable capacity to persist in hospital environments and medical devices due to its ability to form biofilms. Biofilm formation is mediated by Csu pili, assembled via the ‘archaic’ chaperone-usher pathway. The X-ray structure of the CsuC-CsuE chaperone-adhesin pre-assembly complex of Csu pili reveals the basis for attachment of the bacterium to abiotic surfaces. CsuE exposes highly hydrophobic finger-like loops at the tip of the pilus. Decreasing hydrophobicity of the loops abolishes bacterial attachment, suggesting that the pilus tip detects and binds to hydrophobic regions of the substrate surface. Anti-CsuE antibody completely blocks biofilm formation, presenting a means to prevent the spread of the pathogen. The use of hydrophilic materials instead of hydrophobic plastics in medical devices may represent another simple and cheap solution to reduce pathogen spread.