Saturday, March 23, Session VII

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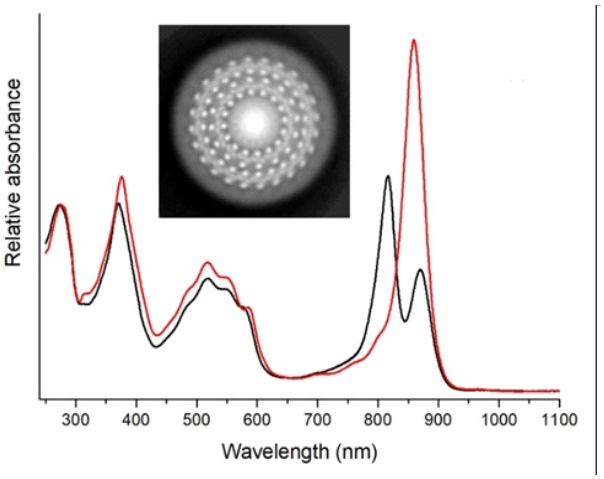
CRYO-EM STRUCTURE OF THE LARGE PHOTOSYNTHETIC COMPLEX FROM TWO SPECIES OF GEMMATIMONADOTA

Alastair T. Gardiner[1], David Bina[2, 3], Pu Qian[4], Zdenko Gardian[2,5], David Kaftan[1,2] & Michal Koblížek[1]*

[1]Institute of Microbiology of the Czech Academy of Sciences, 379 81 Třeboň, Czech Republic
[2]Faculty of Science, University of South Bohemia, 370 05 České Budějovice, Czech Republic
[3]Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Branišovská 1760, 370 05 České Budějovice, Czech Republic
[4]Materials and Structure Analysis, Thermofisher Scientific, Achtseweg Noord 5, 5651 GG Eindhoven, Netherlands
[5]Biology Centre, Czech Academy of Sciences, Institute of Parasitology, Branišovská 1760, 370 05 České Budějovice, Czech Republic

Members of the bacterial phylum Gemmatimonadota (previously called Gemmatimonadetes) inhabit a wide and diverse range of habitats including soils, aquatic environments i.e., marine, fresh and waste water as well as biofilms and sediments. In spite of the prevalence in Nature of this phylum only six cultured species have been described so far. Two of these species, *Gemmatimonas (Gem.) phototrophica* and *Gem. groenlandica* are photoheterotrophic and contain anoxygenic photosynthetic complexes. The photosynthetic pigment-protein complex in *Gem. phototrophica* was found to contain two concentric antenna rings around the central reaction centre (RC), (a projection image is inset in the Figure), and we obtained a high resolution, cryo-EM structure followed revealed many novel features [1] .

Due to the double-ringed antenna in this complex it was termed RC-dLH (d = double Light-Harvesting (rings)). *Gem. groenlandica* was the second phototrophic Gemmatimonadota strain to be characterised [2]. Intriguingly, the absorption spectrum of *Gem. groenlandica* RC-dLH complex (red line) is rather different to that of *Gem. phototrophica* (black line) in the near infra-red (NIR), see Figure. We are currently in the process of final refinement for the RC-dLH complex from *Gem. groenlandica*, therefore, this presentation will compare and contrast the two structures and provide possible functional and physiological reasons for these differences.



1. P. Qian, A.T. Gardiner, I. Šímová, K. Naydenova, T.I. Croll, P.J. Jackson, Nupur, M. Kloz, P. Čubáková, M. Kuzma, Y. Zeng, P. Castro-Hartmann, B.v. Knippenberg, K.N. Goldie, D. Kaftan, P. Hrouzek, J. Hájek, J. Agirre, C.A. Siebert, D. Bína, K. Sader, H. Stahlberg, R. Sobotka, C.J. Russo, T. Polívka, C.N. Hunter, M. Koblížek, 2.4 Å structure of the double-ring Gemmatimonas phototrophica photosystem, Science Advances, 8 (2022) eabk3139.
2. Y. Zeng, Nupur, N. Wu, A.M. Madsen, X. Chen, A.T. Gardiner, M. Koblížek, Gemmatimonas groenlandica sp. nov. Is an Aerobic Anoxygenic Phototroph in the Phylum Gemmatimonadetes, Frontiers in Microbiology, 11 (2021).

L24

COLD-LOVING BACTERIUM FROM A MOUNTAIN LAKE HARVESTS LIGHT ENERGY USING BOTH BACTERIOCHLOROPHYLL-CONTAINING PHOTOSYNTEMS AS WELL AS PROTON-PUMPING RHODOPSINS

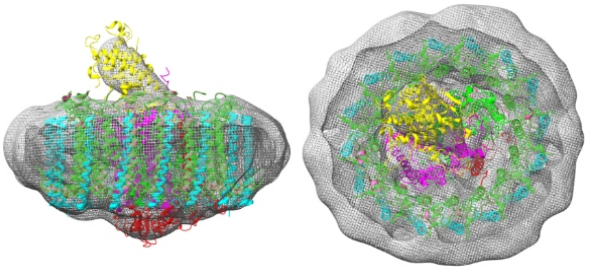
Michal Koblížek[1], Karel Kopejtka[1], Alastair Gardiner[1], David Kaftan[1], David Bína[2], Zdeno Gardian[2]

[1]Centre Algatech, Institute of Microbiology, Czech Academy of Sciences, 37981 Třeboň, Czechia
[2]Faculty of Science, University of South Bohemia, 37005 České Budějovice, Czechia

Bacterium *Sphingomonas glacialis* AAP5 isolated from the alpine lake Gossenköllesee contains genes for anoxygenic phototrophy as well as proton-pumping xanthorhodopsin.

The photosynthetic complexes contain circular light harvesting complex 1 surrounding the type-2 bacterial reaction center. The light harvesting complex is composed from 16 homodimeric subunits. Each subunit binds one bacteriochlorophyll-a pair and one spirilloxanthin molecule. The purified xanthorhodopsin is present as a trimer. It contains carotenoid nostoxanthin serving as an auxiliary antenna and performs the standard photocycle. The xanthorhodopsin-producing cells reduced upon illumination their respiration by 70%. This documents that the harvested light energy was utilized in the metabolism, which can represent a large benefit under carbon-limiting conditions.

The presence of two different photosystems may represent a metabolic advantage in alpine lakes where photoheterotrophic organisms face large changes in irradiance, limited organic substrates and low temperature.



Preliminary cryoEM structure of photosystem in Sphingomonas AAP5

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A FLUORESCENT DEOXYRIBOZYME FOR HIGH-THROUGHPUT SCREENING

M. Volek[1-2], J. Kurfürst[1,3], M. Drexler[1], M. Svoboda[1], P. Srb[1], V. Veverka[1,4], E.A. Curtis[1]

[1]Institute of Organic Chemistry and Biochemistry of the Czech Academy of Sciences, 166 10, Prague, Czech Republic
[2]Department of Genetics and Microbiology, Faculty of Science, Charles University in Prague, 128 44, Prague, Czech Republic
[3]Department of Informatics and Chemistry, University of Chemistry and Technology, 166 28, Prague, Czech Republic
[4]Department of Cell Biology, Faculty of Science, Charles University in Prague, 128 44, Prague, Czech Republic
curtis@uochb.cas.cz

Fluorescence facilitates the detection, visualization, and tracking of molecules with high sensitivity and specificity. A functional DNA molecule that generates a robust fluorescent signal would offer significant advantages for many applications compared to intrinsically fluorescent proteins, which are expensive and labor intensive to synthesize, and fluorescent RNA aptamers, which are unstable under most conditions. Here we describe a novel deoxyriboyzme that rapidly and efficiently generates a stable fluorescent product using a readily available coumarin substrate. An engineered version can detect picomolar concentrations of ribonucleases in a simple homogeneous assay, and was used to rapidly identify novel inhibitors of the SARS-CoV-2 ribonuclease Nsp15 in a high-throughput screen. Our work adds an important new component to the toolkit of functional DNA parts, and also demonstrates how catalytic DNA motifs can be used to solve real-world problems.

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DUAL PERSPECTIVES ON THE EVOLUTION OF SARS-COV-2 RECEPTOR BINDING DOMAIN

Jiří Zahradník[1], Miguel Padilla Blanco[1,2], Prokopios Andrikopoulos[1]

[1]1st Faculty of Medicine, Charles University, BIOCEV, Prague 252 50, Czechia
[2]Departamento de Farmacia, Facultad de Ciencias de la Salud, Universidad Cardenal Herrera-CEU, CEU Universities, Valencia, Spain
jiri.zahradnik@lf1.cuni.cz

The SARS-CoV-2 virus doesn't need a lengthy introduction. We all are aware of it and feel that nothing can surprise us anymore. With more than 16.5 million deposited sequences in the GISAID database and over 4 thousand structures in the PDB, it is by far the most extensively studied virus (20.2.2024). Still, many questions remain unanswered. In my lecture, I will show how the receptor-binding domain is shaped by evolutionary pressures to maintain high binding affinity and escape neutralizing antibodies, and how this interplay continues to surprise us with an ever-changing cooperativity landscape as can be seen from affinities in Table 1. In the second part of the lecture, we will focus on elucidating the change in tropism in SARS-CoV-2 and which structural aspects have contributed to this phenomenon.

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Table 1. The ever-changing cooperativity landscape demonstrated by affinities [nM] and their impact on different SARS-CoV-2 lineages

	R403K	V445H	N450D	L452W	N481K	V483del
WT	0.7	0.8	0.6	0.6	1.0	0.3
BA.2	2.5	0.7	0.9	0.7	0.8	0.4
XBB	1.4	1.0	0.9	1.0	0.8	0.4
XBB.1.5	1.5	0.8	1.0	0.9	0.7	0.5
BA.2.86	0.5	1.0	0.7	1.1	1.6	1.8