Šifra projekta / Project code **N1-0377**

Dizajn, mehanska stabilnost in uporaba modularnih proteinov na osnovi obvitih vijačnic
Design, mechanical stability and applications of modular proteins based on coiled coils

Vodja projekta:
prof. dr. Roman Jerala

1. VSEBINSKI OPIS PROJEKTA »ERC nova obzorja«:

Projekt ERC AdG MaCChines je bil zelo uspešen, saj je bilo doseženih še več ciljev, kot je bilo sprva predvideno. Dokazali smo nastanek novih vrst proteinskih vzorcev zvitja, ki v naravi niso znani, in njihovo uravnavanje s kemičnimi in biološkimi signali, nedavno smo določili strukture visoke ločljivosti načrtovanega modularnega trikotnika ki je podrobno potrdila, da je bil predlagani model načrtovanja zelo natančen. V projektu MaCChines2, ki je nadaljevanje projekta ERC, predlagamo raziskovanje novih smeri in lastnosti zasnovanih kletk proteinskega origamija z obvitimi vijačnicami, kot sta oblikovanje vozlov in raziskovanje mehanske stabilnosti zasnovanih proteinov, ki z naravnimi proteini ne bi bila mogoča. Poseben poudarek bo na raziskovanju razvitja/ponovnega zvitja posameznih proteinskih molekul z uporabo optične pincete, orodja, ki smo ga pred kratkim uvedli v naš laboratorij, kjer nameravamo razviti več novih eksperimentalnih orodij, ki bi lahko predstavljala temelj prihodnjih projektov za izdelavo odzivnih bioloških sistemov ali novih materialov. Projekt bo prav tako pomagal ohraniti najbolj produktivne člane ekipe.

The ERC AdG project MaCChines has been highly successful, with the achievement of even more goals than initially envisioned. We have demonstrated the formation of new types of protein folds not known in nature, and their regulation by chemical and biological signals and the recent determination of high-resolution structure of a designed modular trigon confirmed in detail that the proposed design model was very accurate. In MaCChines2, as the continuation of the ERC project, we propose to explore new horizons and properties of designed coiled-coil protein origami cages, such as the formation of knots and investigation of the mechanical stability of designed proteins that would not be possible with natural proteins. Emphasis will be on the investigation of single molecule disassembly/reassembly using optical tweezers, a tool that we have recently introduced into our lab where we plan to develop several new experimental tools that could represent the foundation of future projects to make responsive biological systems or new materials. The project will also help maintain the most productive team members.

Sodelujoče organizacije
- **KEMIJSKI INŠTITUT**
- **EN-FIST CENTER ODLIČNOSTI**

a. osnovni podatki glede financiranja

Projekt financira ARRS v okviru cenovne kategorije D za obdobje treh let v obsegu 1996 letnimi urami za obdobje 3 let. Pričetek financiranja je 1. 9. 2024.
The project is co-financed by ARRS with 1996 annual hours of price class D for a period of 3 years. Funding starts on September 1, 2024.

b. sestava projektne skupine s povezavami na SICRIS

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2. faze projekta in njihova realizacija

Projekt sestavljajo štirje sklopi, ki sledijo zastavljenim ciljem:
WP1: Stabilnost CCPO pri mehanskih obremenitvah.
WP2: Raziskovanje nastanka in lastnosti vozličastih struktur
WP3: Priprava proteinov, pri katerih CC par sterično ovira dostop substrata do aktivnega mesta, medtem ko bi premestitev CC ali njihovo razvitje z mehanskim stresom lahko povrnilo njihovo aktivnost.
WP4: Uvedba tehnologije za merjenje interakcij med dvema domenama beljakovin na podlagi genetske fuzije s SpyCatcher/SpyTag

The project consists of four scientific work packages in line with the set specific objectives:
WP1: Stability of CCPO to mechanical stress.
WP2: Investigate the formation and properties of knotted structures
WP3: Prepare proteins, where the pair of CCs sterically hinders the access of the substrate to the active site while the displacement of CCs or their unfolding could regain the activity of those proteins
WP4: Introduction of technology to measure interactions between the two protein domains based on genetic fusion to SpyCatcher/SpyTag

3. bibliografske reference, ki izhajajo neposredno iz izvajanja projekta

SNOJ, Jaka, ZHOU, Weijun, LJUBETIČ, Ajasja, JERALA, Roman. Advances in designed bionanomolecular assemblies for biotechnological and biomedical applications. **Current opinion in biotechnology**. 2025, vol. 92 DOI: 10.1016/j.copbio.2024.103256 [COBISS.SI-ID 223429379]

OREHEK, Sara, ŽELEZNIK RAMUTA, Taja, LAINŠČEK, Duško, MALENŠEK, Špela, ŠALA, Martin, BENČINA, Mojca, JERALA, Roman, HAFNER BRATKOVIČ, Iva. Cytokine-armed pyroptosis induces antitumor immunity against diverse types of tumors. **Nature communications**. 2024, 15, 10801, 1-22. ISSN 2041-1723. DiRROS, DOI: 10.1038/s41467-024-55083-3. [COBISS.SI-ID 221013763]

4. logotip ARIS in drugih sofinancerjev

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