

### UČNI NAČRT PREDMETA / COURSE SYLLABUS

<b>Predmet:</b>	Izbrane metode v biokemiji in molekularni biologiji
<b>Course title:</b>	Selected Methods in Biochemistry and Molecular Biology

Študijski program in stopnja Study programme and level	Študijska smer Study field	Letnik Academic year	Semester Semester
<b>Ekologija z naravovarstvom, 1. stopnja</b>		<b>3</b>	zimski ali letni
<b>Ecology with nature conservation, 1st. degree</b>			

Vrsta predmeta / Course type	izbirni/optional
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Univerzitetna koda predmeta / University course code:	
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Predavanja Lectures	Seminar Seminar	Sem. vaje Tutorial	Lab. vaje Laboratory work	Teren. vaje Field work	Samost. delo Individ. work	ECTS
30			15		135	6

Nosilec predmeta / Lecturer:	Janja TRČEK
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Jeziki / Languages:	Predavanja / Lectures: Vaje / Tutorial:	Slovenski /Slovenian
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**Pogoji za vključitev v delo oz. za opravljanje  
študijskih obveznosti:**

Jih ni	No
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<b>Vsebina:</b>	<b>Content (Syllabus outline):</b>
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Razvoj modernih in učinkovitih biokemijskih in molekularno bioloških metod je v zadnjih desetletjih povzročil pravo revolucijo na številnih področjih naravoslovja. Številne tehnike, ki so se na začetku uporabljale v biokemiji, genetiki in molekularni biologiji, so prodrle tudi v klasične biološke discipline. Začetna predavanja so namenjena ponovitvi strukture genov in genomov. Predstavljene molekularno biološke tehnike so usmerjene v analizo proteinov (elektroforeza izoencimov), kromosomov (molekularna citogenetika) in predvsem nukleinskih kislin. Tehnikam vzorčenja in hranjenja rastlinskih in živalskih tkiv, metodam izolacije, čiščenja in kvantifikacije nukleinskih kislin sledi separacija DNA in RNA z elektroforezo na agaroznih in poliakrilamidnih gelih ter izolacija nukleinskih kislin iz gelov. Poudarek je na metodah encimskih modifikacij DNA kot so rezanje z restriktičnimi nukleazami, lepljenje ali ligacija, fosforilacija in zaznamovanje nukleinskih kislin. Razloženi so principi kloniranja, priprava DNA knjižnic, izolacija pozitivnih klonov, izražanje rekombinantnih proteinov v različnih ekspresijskih sistemih kakor tudi uporaba različnih vektorjev za kloniranje in ekspresijo. Podane so metode hibridizacije nukleinskih kislin, pomembne za prečesavanje po DNA knjižnicah, za metodi Northern blot, Sauthern blot in za kvantitativno ocenjevanje sorodnosti biotskih vrst na osnovi jedrne DNA. Razloženi so tudi principi in možnosti uporabe tehnike polimerazne verižne reakcije (PCR) in principi sekpcioniranja DNA. Nazadnje so podane osnove analize nukleinskih kislin s pomočjo različnih bioinformatskih orodij (kot so programi BLAST, CLUSTAL, EXPASY...), vključno z osnovami različnih pristopov in bioinformatskih orodij pri analizi filogenetskih odnosov s pomočjo molekularno bioloških podatkov. Predavanja so namenjena tudi načrtovanju molekularno bioloških pristopov k problemom v molekularni evoluciji in ekologiji.

The development of modern and powerful methods in biochemistry and molecular biology have been revolutionized in many fields of natural science in the last decades. Many techniques used initially in biochemistry, genetics and molecular biology were adopted in classical biological disciplines as well. The introductory lectures are dedicated to a review in genes and genomes structures. Molecular biology techniques presented are focused on analyses of the proteins (isozyme electrophoresis), chromosomes (molecular cytogenetics) and first of all on nucleic acids analyses. Sampling techniques (collection and storage of plant and animal tissues), nucleic acids isolation, purification and quantification methods are followed with DNA and RNA resolution using electrophoresis methods on agarose and polyacrylamide gels and the resolution and recovery of DNA and RNA fragments from gels. Emphasized are methods for enzymatic manipulation of DNA such as digestion of DNA with restriction nucleases, ligation, phosphorylation and nucleic acids labeling. The principles of cloning DNA, construction of DNA libraries, isolating positive clones (screening), expression of recombinant proteins in different expression systems, the use of different cloning and expression vectors are explained. Nucleic acid hybridization methods, important for screening DNA libraries as well as for Northern and Southern blot methods and for quantitative assessment of relatedness of biotic species on the basis of nuclear DNA are given. The principles and possible applications of polymerase chain reaction (PCR) and the principles of sequencing are also explained. Finally, the fundamentals of nucleic acid analyses using different bioinformatics tools (such as BLAST, CLUSTAL, EXPASY...) are given, including the fundamentals and bioinformatics tools for the analyses of phylogenetic relationships from molecular data. Lectures are also dedicated to plan the molecular biology approaches to problems in molecular evolution and ecology.

**Temeljni literatura in viri / Readings:**

- Berg B., Tymoczko J.L., Stryer L. 2011. Biochemistry, International Edition. 7. izdaja. 1098 str., Palgrave Macmillan.
- Krebs J.E., Goldstein E.S., Kilpatrick S.T. 2012. Lewin's Genes XI. 11. izdaja. 940 str., Jones & Barlett Learning.
- Sambrook J., Russell, D.W. 2001: Molecular cloning – a laboratory manual. Cold Spring Harbor Laboratory press, Cold Spring Harbor. New York

**Cilji in kompetence:**

Namen predmeta je podati osnovne informacije kakor tudi spremljajoč teorijo, oziroma principe na katerih sponzijo zelo številne molekularno biološke tehnike, med katerimi so mnoge aplikirane tudi v klasičnih bioloških disciplinah. Velik poudarek je na praktičnemu laboratorijskemu delu. Študenti naj bi obvladali številne tehnike, kot so:

- Izolacija RNA in DNA iz rastlinskih in živalskih tkiv
- Čiščenje nukleinskih kislin
- Kvantifikacija nukleinskih kislin
- Separacija RNA in DNA na agaroznih gelih
- Separacija proteinov na poliakrilamidnih gelih
- Enzimske modifikacije
- Kloniranje DNA in priprava DNA knjižnic
- Transformacija DNA v različne celične sisteme
- Hibridizacija RNA in DNA
- Prečesavanje pozitivnih klonov
- Northern blot
- Sauthern blot
- Polimerazna verižna reakcija (PCR)
- Sekveniranje DNA

**Objectives and competences:**

The aim of the subject is to give background information as well as underlying theory of the main molecular biology methods currently used, many of them applied also in systematics, evolution and ecology. A great emphasis should be done on practical laboratory work. Students should be able to use many techniques, including:

- RNA and DNA isolation from plant and animal tissues
- Nucleic acid purification
- Nucleic acid quantification
- Separation of RNA and DNA on agarose gels
- Separation of proteins on polyacrylamide gels
- Enzyme modifications
- DNA cloning and DNA library construction
- Transformation of DNA in different cell systems
- Hybridization of RNA and DNA
- Screening of positive clones
- Northern blot
- Sauthern blot
- Polymerase chain reaction (PCR)
- DNA sequencing

**Predvideni študijski rezultati:****Znanje in razumevanje:**

- Študenti dobijo podrobni vpogled v strukturo genov in genomov ter se znanijo z možnostmi in načini uporabe molekularno bioloških tehnik pri študiju biodiverzitete, filogenije, filogeografije, populacijske genetike in ekologije.
- Poleg poznavanja principov številnih

**Intended learning outcomes:****Knowledge and Understanding:**

- Students get a detailed insight in gene and genomes structures, and about possibilities and ways of applications of molecular biology techniques in biodiversity, phylogenetic, phylogeography, population genetic and ecology studies.
- Besides the knowledge of principles of

molekularno bioloških metod in sposobnosti kritičnega ovrednotenja njihove uporabe v praksi dobro obvladajo laboratorijsko delo z različnimi aparaturami ter ravnanje z zdravju škodljivimi kemikalijami.

Prenesljive/ključne spremnosti in drugi atributi:

- Obvladovanje številnih sodobnih metod v molekularni biologiji
- Poznavanje osnovnih pristopov in sposobnost uporabe bioinformatskih orodij pri analizi nukleinskih kislin kakor tudi filogenetskih povezav s pomočjo molekularno bioloških podatkov

numerous molecular biology methods and the capacity of critical evaluations of their applications in practice, they are familiar with various laboratory instruments as well with handling with toxic materials.

Transferable/Key Skills and other attributes:

- Capacity of understanding and handling numerous currently used methods in molecular biology.
- Knowledge of basic approaches and the ability to use current software applications for analyses of nucleic acids as well as phylogenetic relationships using molecular biology data.

#### **Metode poučevanja in učenja:**

- Predavanja
- Laboratorijske vaje
- Individualno delo s študenti

#### **Learning and teaching methods:**

- Lectures
- Laboratory excercises
- Individual work with students

Delež (v %) /

#### **Načini ocenjevanja:**

Weight (in %)      **Assessment:**

<ul style="list-style-type: none"> <li>• Kolokvij</li> <li>• Ustni izpit</li> </ul>	50	<ul style="list-style-type: none"> <li>• Practical examination</li> <li>• Oral examination</li> </ul>
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#### **Reference nosilca / Lecturer's references:**

1. Trček, J., and K. Matsushita. A unique enzyme of acetic acid bacteria, PQQ-dependent alcohol dehydrogenase is also present in *Frateuria aurantia*. Appl. Microbiol. Biotechol., in press.
2. Slapšak, N., Cleenwerck, I., De Vos, P., and J. Trček. *Gluconacetobacter maltaceti*, a novel vinegar producing acetic acid bacterium. 2013. Syst. Appl. Microbiol. 36, 17-21.
3. Trček, J., Oellerich, M., Niedung, K., Ebel, F., Freund, S., and K. Trülsch. 2011. Gut proteases target *Yersinia* invasin *in vivo*. BMC Research Notes 4, 129.
4. Trček, J., Fuchs, T.M., and K. Trülsch. 2010. Analysis of *Yersinia enterocolitica* invasin expression *in vitro* and *in vivo* using a novel luxCDABE reporter system. Microbiology, 156, 2734-2745.
5. Trček, J., Jernejc, K., and K. Matsushita. 2007. The highly tolerant acetic acid bacterium *Gluconacetobacter europaeus* adapts to the presence of acetic acid by changes in lipid composition, morphological properties and PQQ-dependent ADH expression. Extremophiles 11, 627-635.