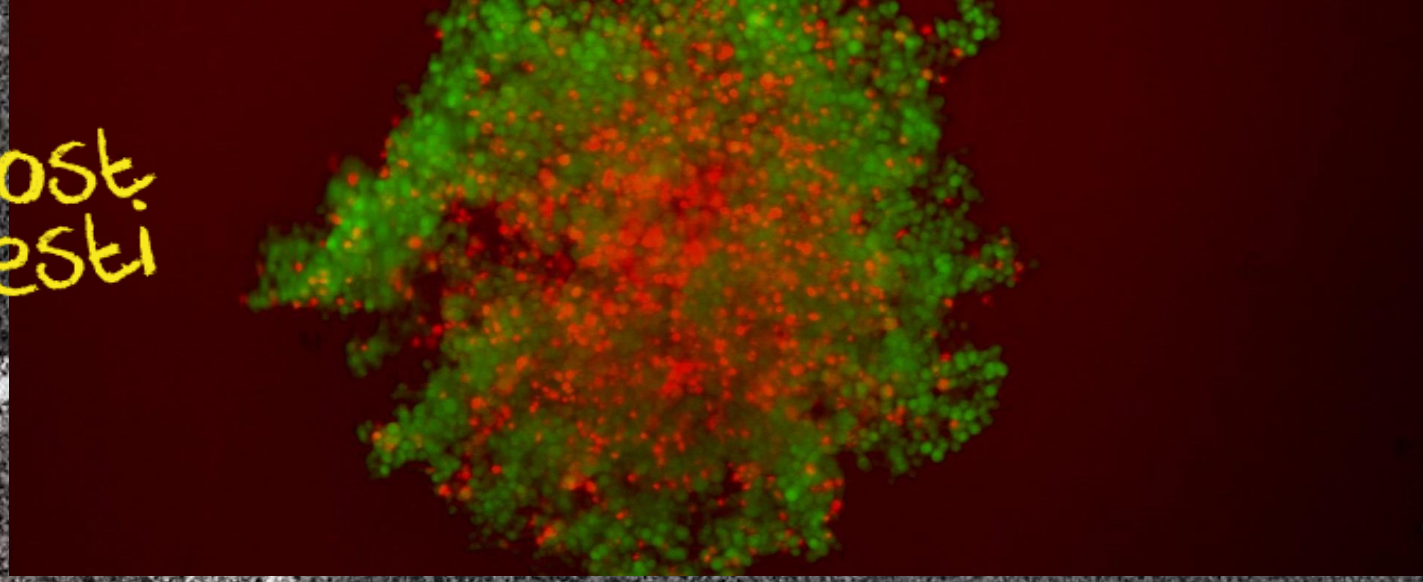
Znanost na cesti



Kavarna Union, Ljubljana

14. junij 2017 ob 19h

Obeti nanotehnologij in varnost nanodelcev

dr. Mojca Pavlin, Skupina za nano in biotehnološke aplikacije
Fakulteta za elektrotehniko, Univerza v Ljubljani

Luka Hvalc, Val 202

SATENA

Javna agencija za raziskovalno dejavnost Republike Slovenije

Institut "Jožef Stefan" Ljubljana, Slovenija

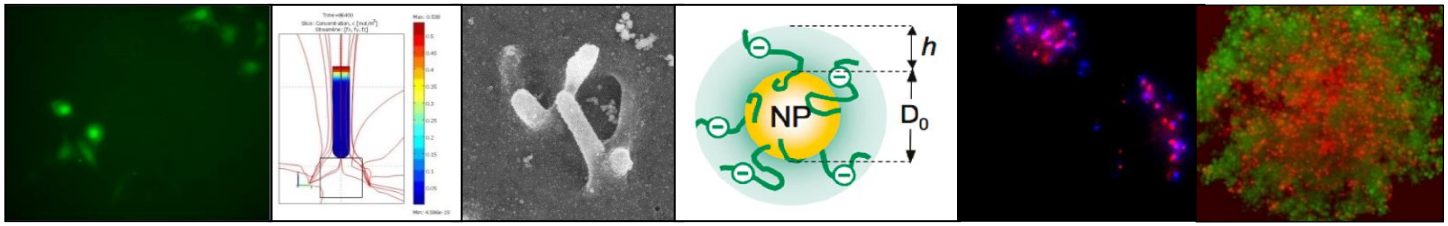
KERAMIKA LIBOJE 1815

VAL 202

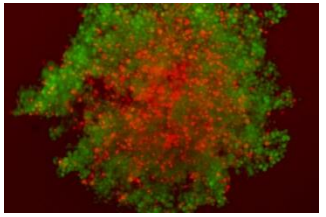
Skupina za nano in biotehnološke aplikacije, FE-UL

Vnos učinkovin, analiza mehanizmov

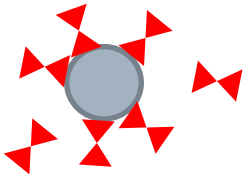
- nanotoksikologija
- razvoj nanodelcev za biotehnološke in biomedicinske aplikacij
- presnovne poti rakavih celic
- genska elektrotransfekcija, utišanje genov

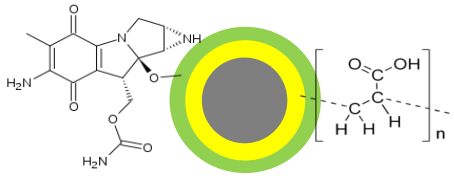


Sinteza, karakterizacija, funkcionalizacija

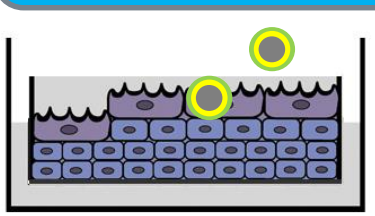


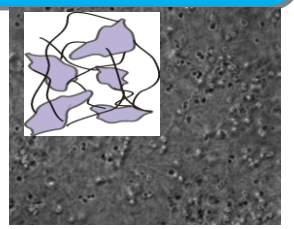
Razvoj nanodelcev za dostavo učinkovin



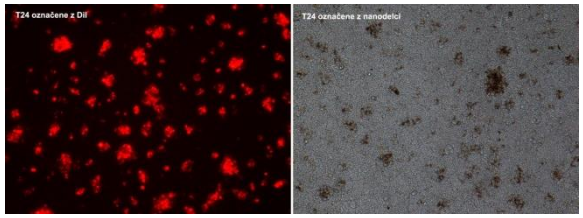


Analiza difuzije v in vitro 3D sistemih

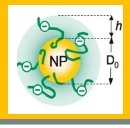




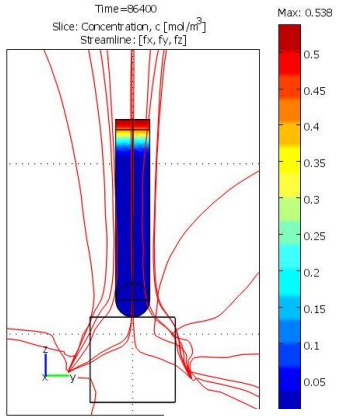
Razvoj nanodelcev za označevanje celic



Razvoj in karakterizacija novih formulacij nosilnih sistemov



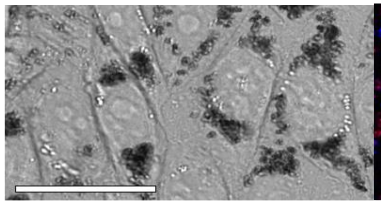
Numerično modeliranje difuzije, magnetoforeze

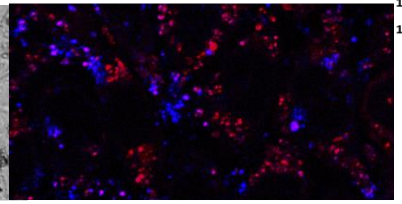


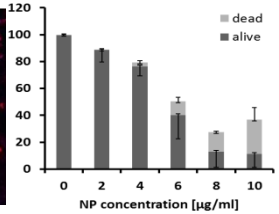
$$v = \frac{\chi_V V_m \nabla B^2}{12\pi\mu_0 R_{hydr}\eta}$$

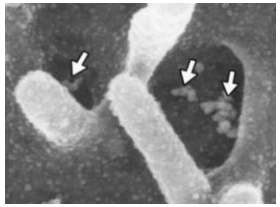
Vizualizacija, znotrajcelična usoda, dinamika, kvantifikacija privzema, analiza toksičnosti

Nanotoksikologija



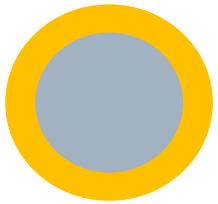


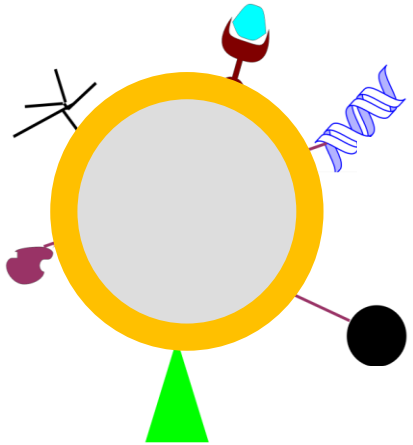




Kaj so nanodelci

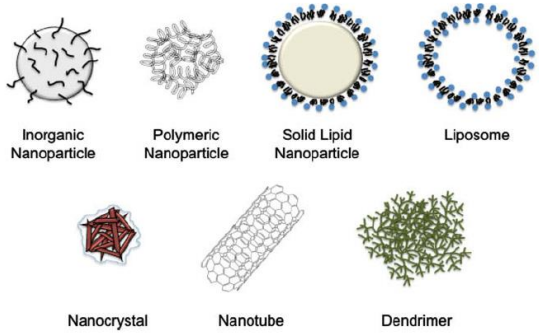
- Velika reaktivna površina (-/+)
- Možnost vezave molekul, tarčna dostava, kontrastna sredstva, katalizatorji, novi materiali...
- (Nekateri) nanodelci prehajajo v notranjost celic
- Koloidno srebro, zlato, železo-oksidni, polimerni, liposomski,...
- Vsak nanodelec svoj material
- Stabilnost!

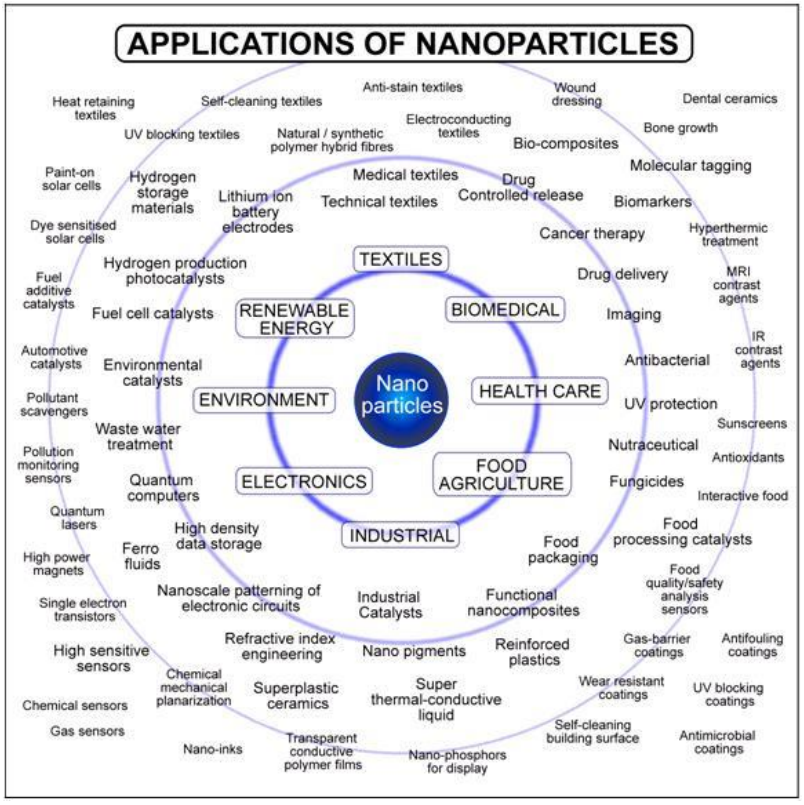




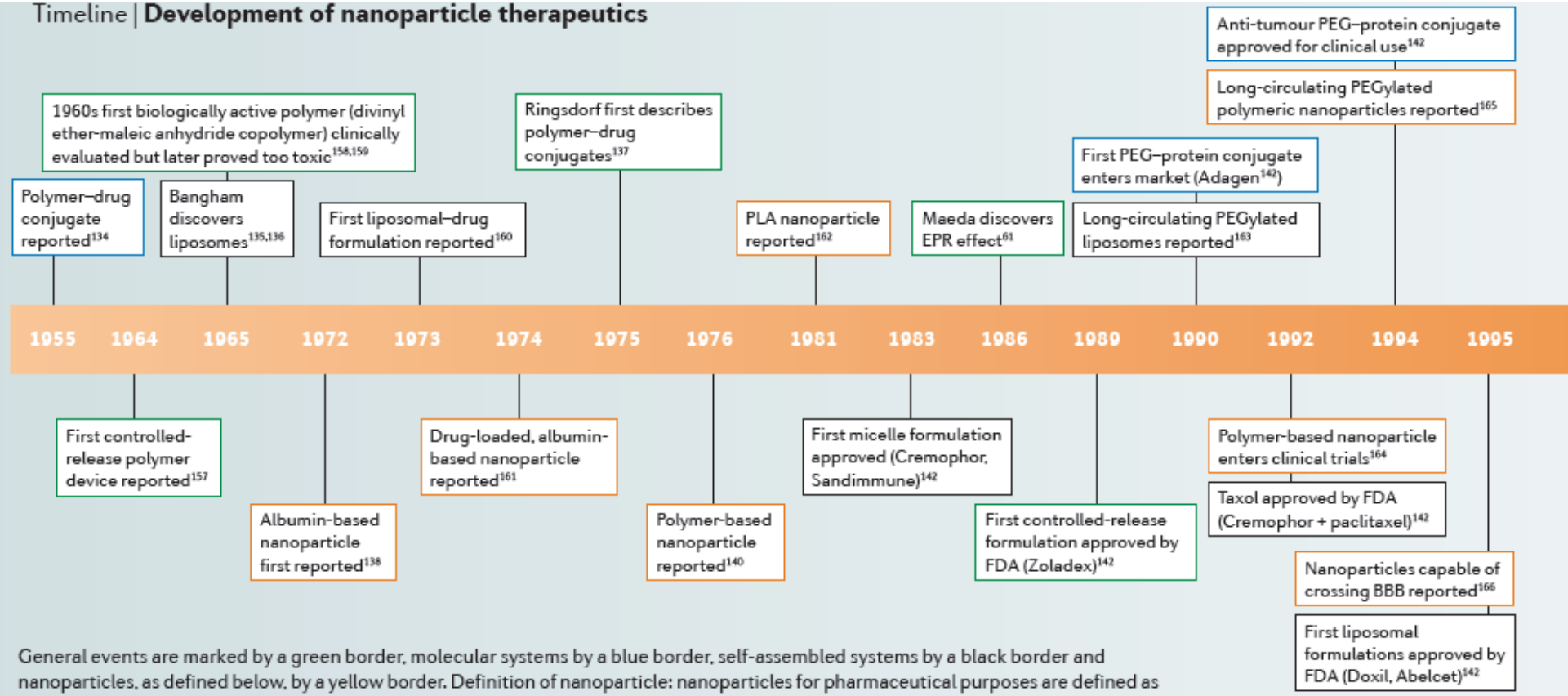
Kje se nahajajo nanodelci

- okoljsko prisotni nanodelci (izpuhi, naravni viri) - saje
- proizvedeni: kozmetika (TiO_2), tekstil (nanosrebro), hrana (E171 food grade TiO_2, SiO_2), premazi...
- specifične aplikacije (elektronika, senzorji, materiali, katalizatorji, biomedicina, biotehnologija)

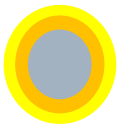




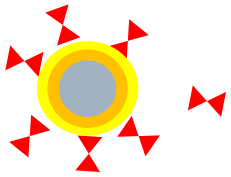
Biomedicinske aplikacije



General events are marked by a green border, molecular systems by a blue border, self-assembled systems by a black border and nanoparticles, as defined below, by a yellow border. Definition of nanoparticle: nanoparticles for pharmaceutical purposes are defined as solid colloidal particles ranging in size from 1 nm to 1,000 nm. They consist of macromolecular materials and can be used therapeutically as drug carriers, in which the active principle (drug or biologically active material) is dissolved, entrapped or encapsulated, or to which the active principle is adsorbed or attached. Abraxane, paclitaxel protein-bound particles for injectable suspension (Abraxis/AstraZeneca); Adagen, PEG–adenosine deaminase (Enzon); BBB, blood–brain barrier; Copaxone, glatiramer acetate for injection (Teva Pharmaceuticals); Cremophor, polyoxyethylated castor oil (BASF); EPR, enhanced permeability and retention; FDA, US Food and Drug Administration; Gliadel, polifeprosan 20 with carmustine implant (Eisai); PEG, polyethylene glycol; PLA, polylactic acid; Sandimmune, cyclosporine injection (Novartis); Zoladex, goserelin acetate implant (AstraZeneca).



DIAGNOSTIKA
NMR
elektronska mikroskopija
patološki preparati



NOSILNA PLATFORMA za MALE MOLEKULE in MAKROMOLEKULE
male molekule, pDNA, miRNA;shRNA,...

Biomedicinske aplikacije

Priložnosti:

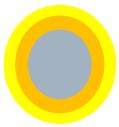
nove nanoformulacije za učinkovine (zmanjšana toksičnost, ciljna dostava)

število razvitih nanoformulacij vsako leto narašča (večina za zdravljenje raka) - poenostavitev postopka FDA

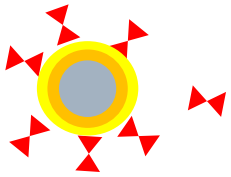
biološka zdravila

Nevarnosti:

imunski odziv, nezadostna učinkovitost - nekatere formulacije umaknjene



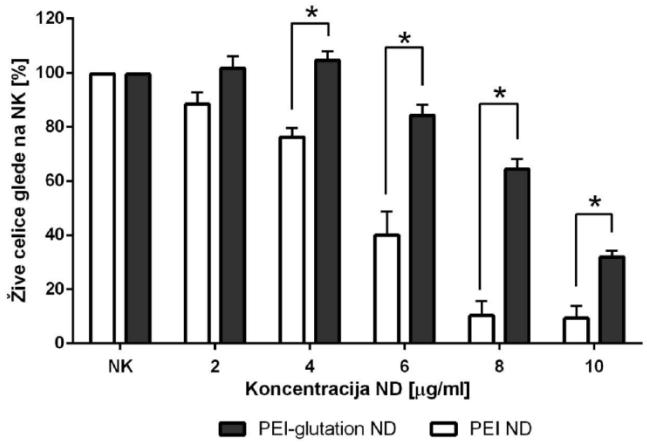
DIAGNOSTIKA
NMR
elektronska mikroskopija
patološki preparati



NOSILNA PLATFORMA za MALE MOLEKULE in MAKROMOLEKULE
male molekule, pDNA, miRNA;shRNA,...

Načini vnosa/izpostavitve tudi določa biodistribucijo, vnos in toksičnost

- Sistemsko (kri: intravenozno, intrarterijsko)
- Lokalno (tumorji)
- Preko sluznic (oralno, pljuča, olfaktorni sistem)

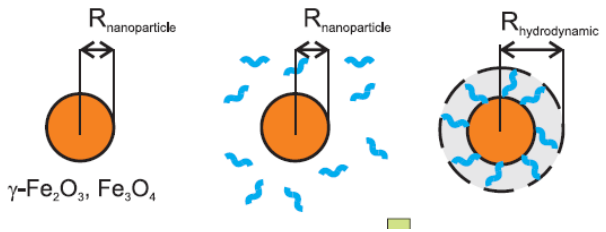


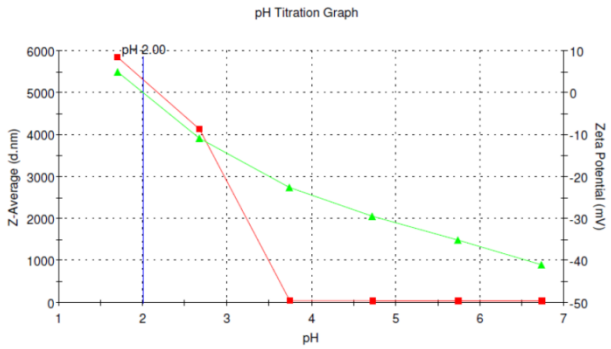
- Mehanizmi (ROS, poškodbe membrane, proliferacija, diferenciacija, transport,...)
- Citotoksičnost (IC50), toksičnost (LD50)
- Akutna toksičnost, toksičnost (dolgotrajna), biodistribucija
- Imunogenost
- Genotoksičnost
- Sproščanje ionov (Ag, ZnO), razgradljivost

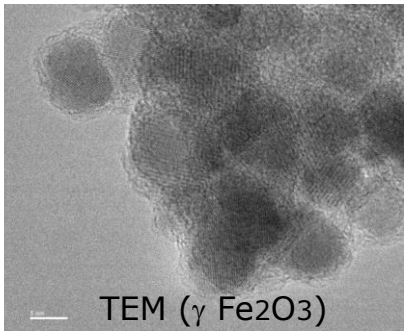
Kdaj so nanodelci res nano ali zakaj karakterizacija ?

FIZIKALNO - KEMIJSKE LASTNOSTI

- Velikost – hidrodinamski radij, oblika, naboj, nečistoče,kristalna struktura, površinske lastnosti
- Lastnosti delcev determinirajo obnašanje v bioloških sistemih: stabilnost, topnost, mobilnost in možnosti specifične vezave, vnos, toksičnost, imunski odziv
- Karakterizacija nanodelcev je ključna za optimizacijo različnih aplikacij - kontrola procesa
- Meritve pri fizioloških pogojih

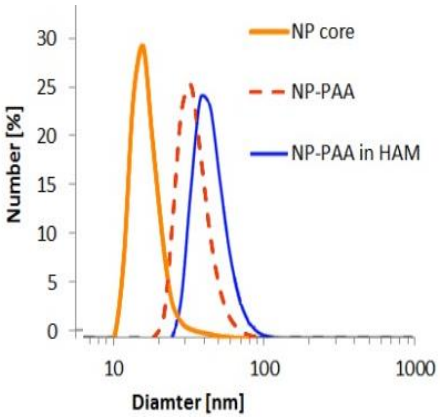


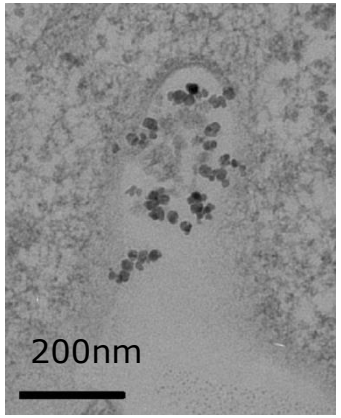


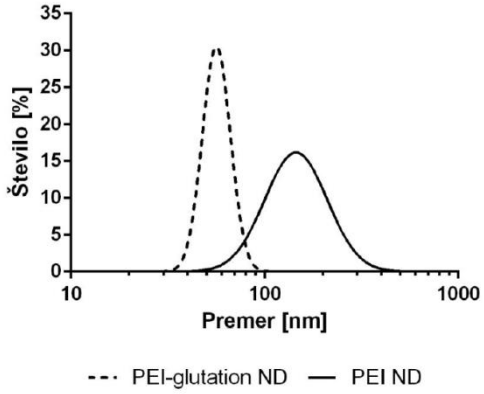


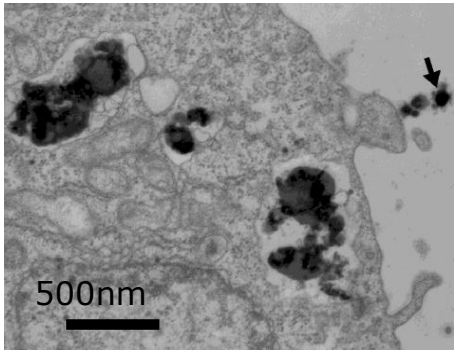
Kaj vidijo celice?

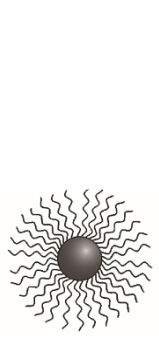
- Ali so nanodelci sploh nano?

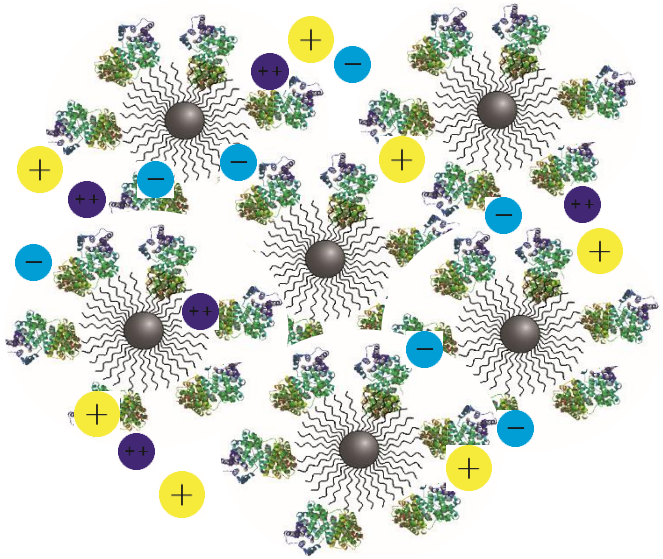


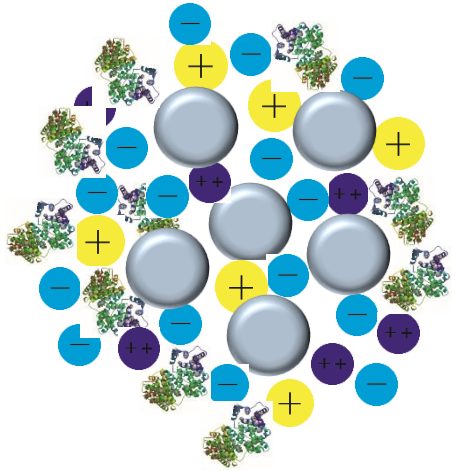








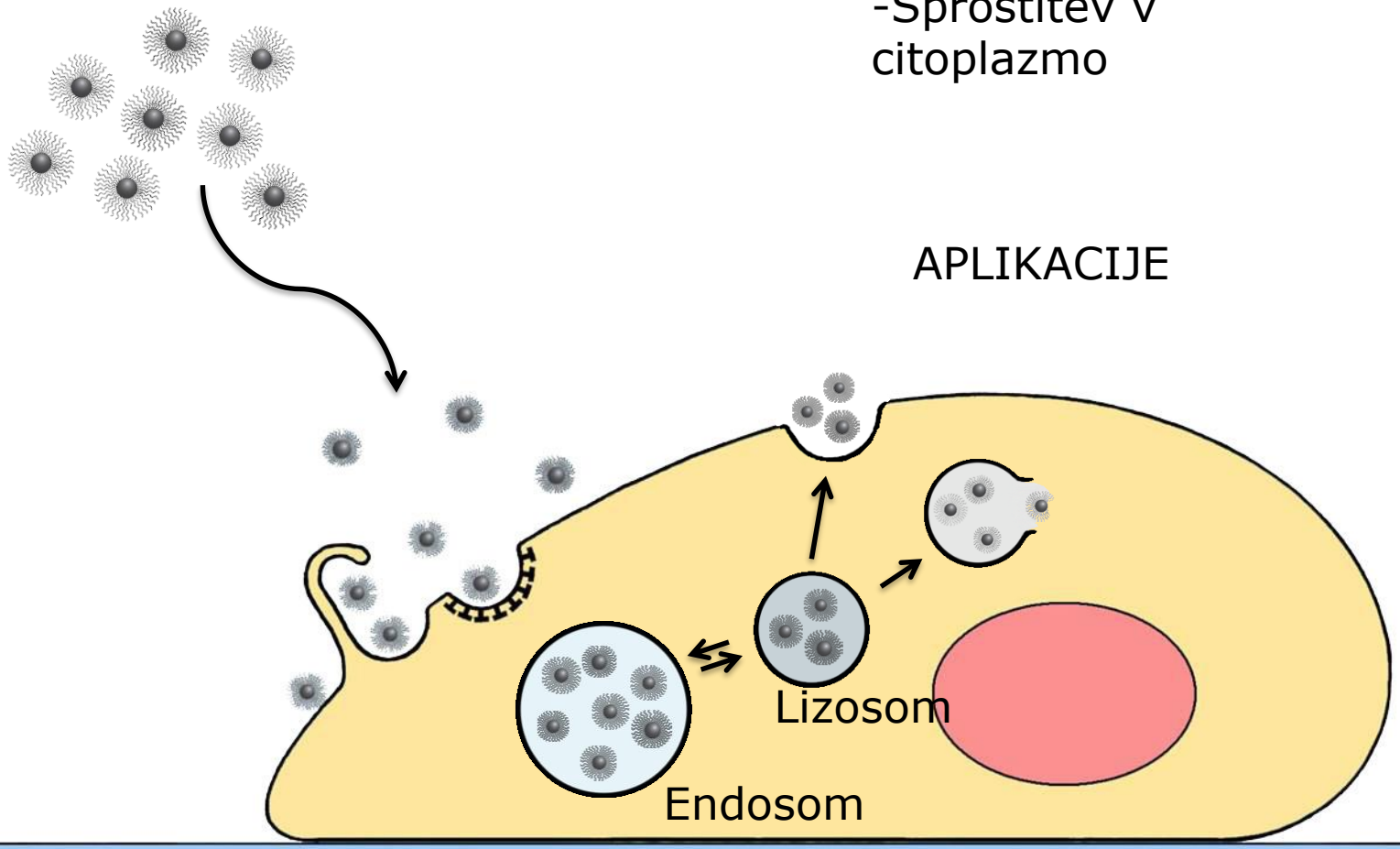




Interakcija nanodelcev s celicami in vitro

Usoda delcev:
-Endocitoza
-Eksocitoza
-Sprostitev v citoplazmo

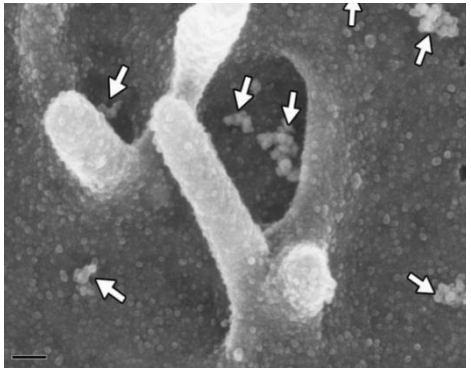
APLIKACIJE



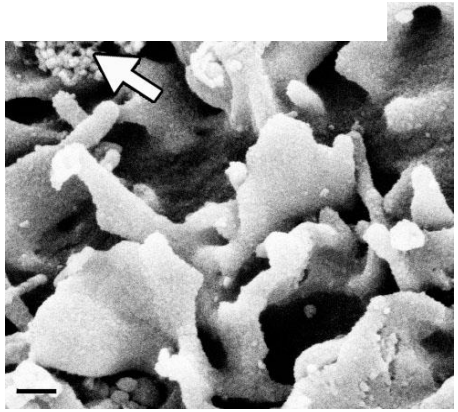
Interakcija, internalizacija

Vrstična in presevna elektronska mikroskopija

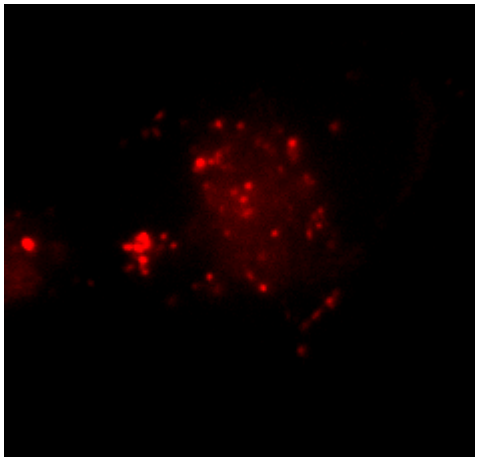
Inštitut za biologijo celice MF -UL

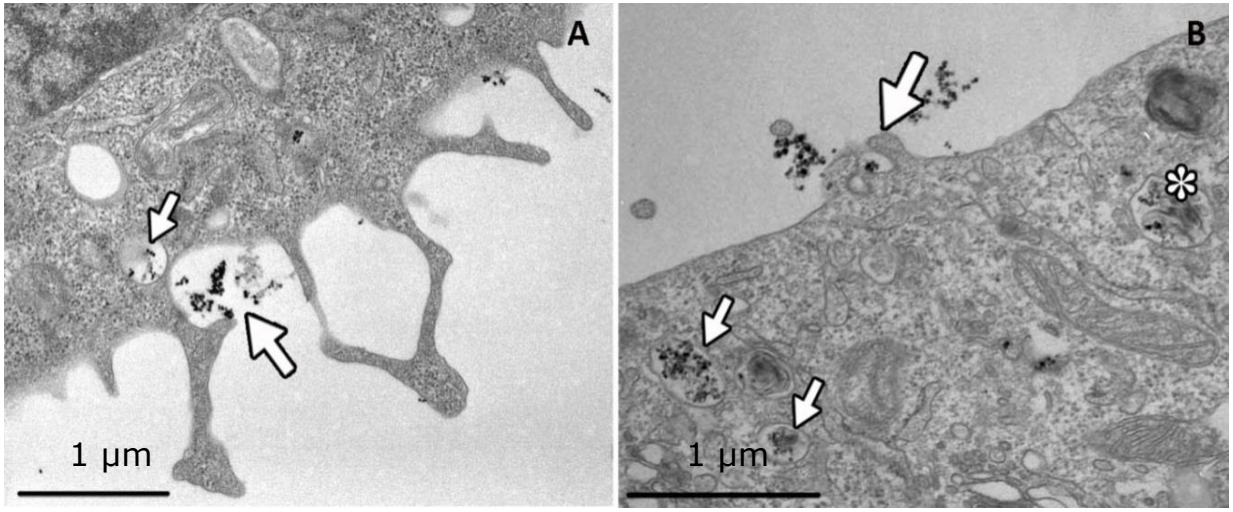


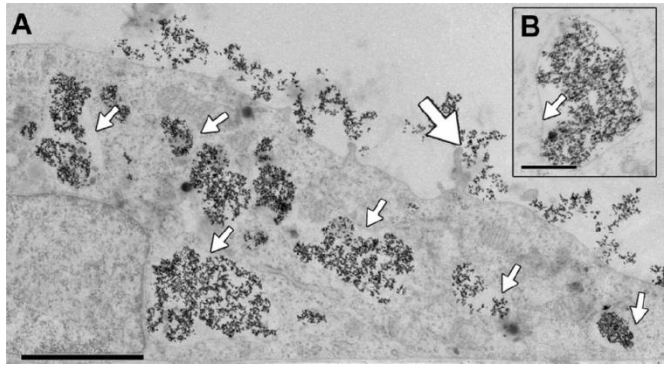
100 nm



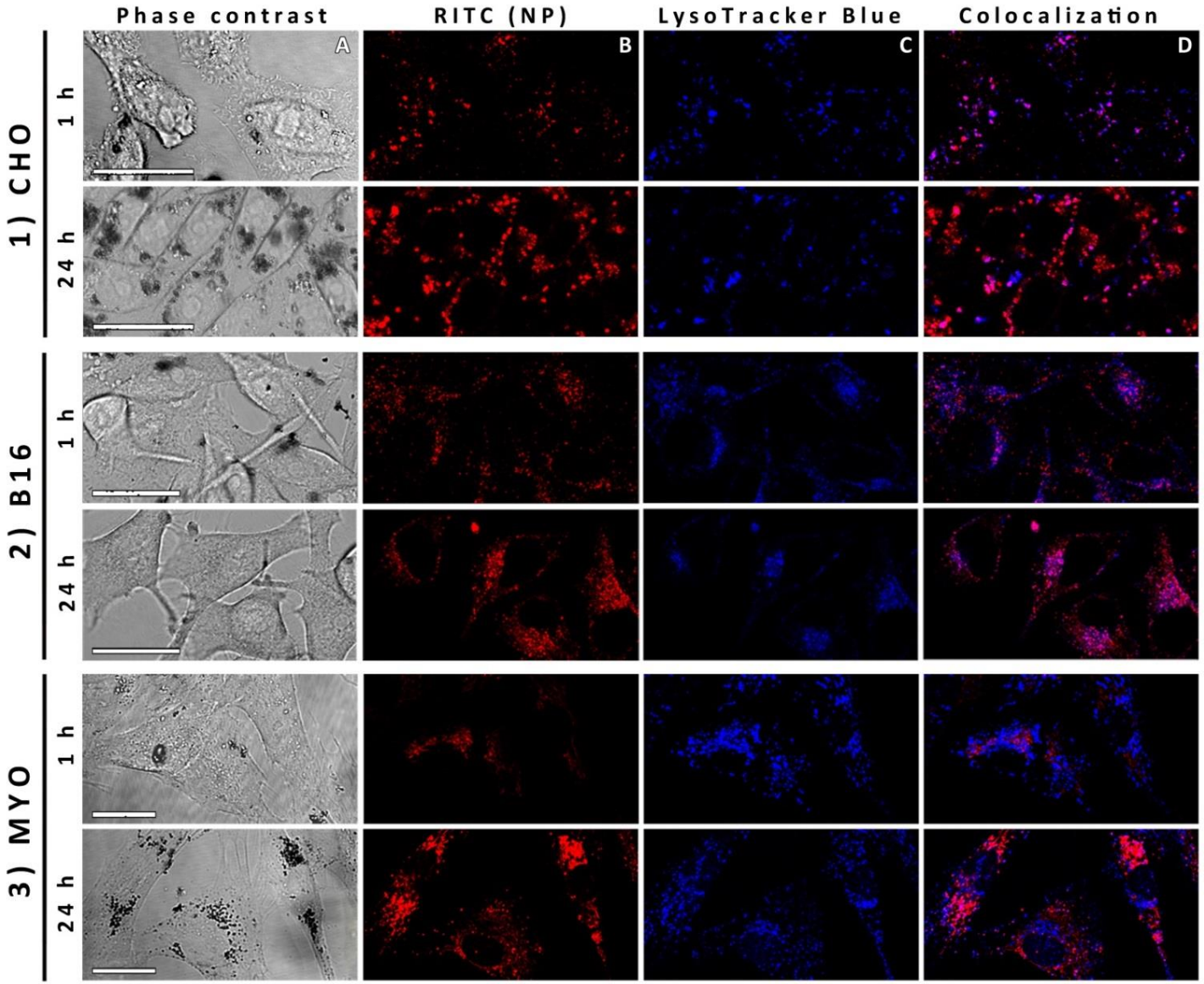
200 nm



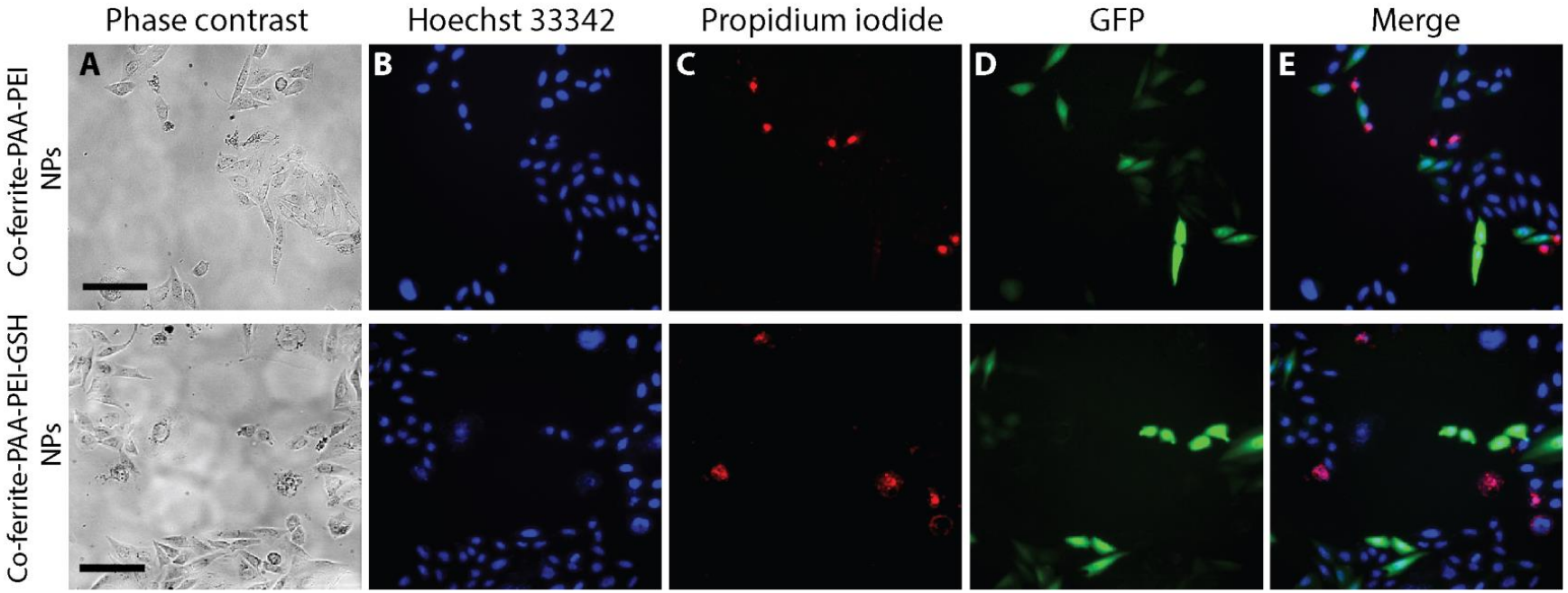




Znotrajcelična usoda

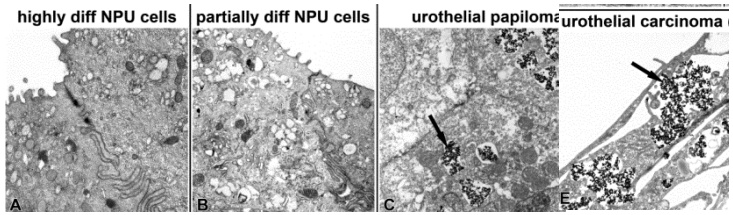


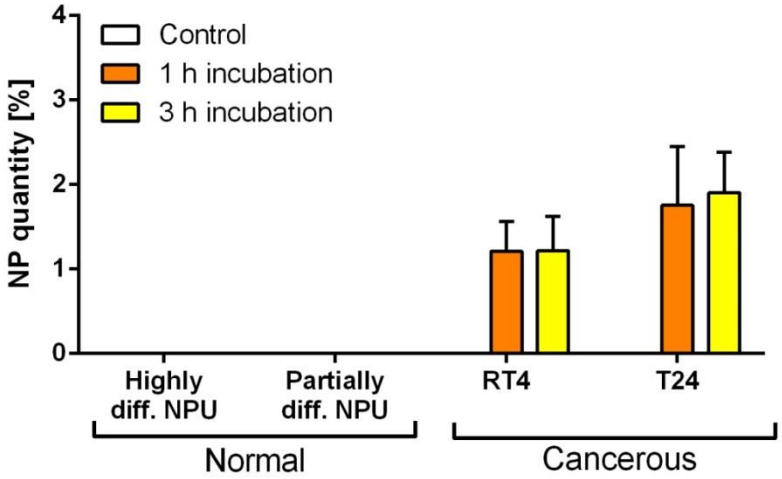
Vnos plazmidne DNA z nanodelci

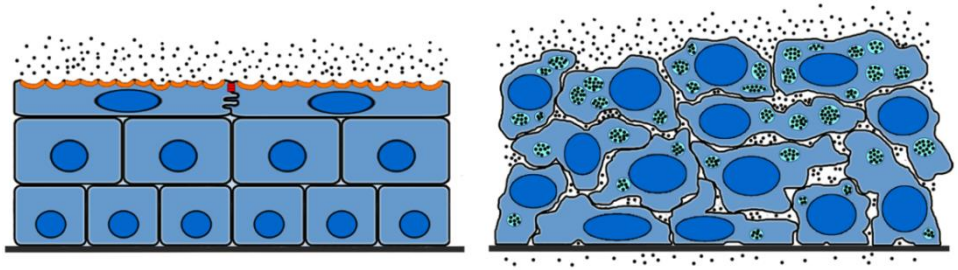


Vnos gena za zeleni flurescirajoči protein (GFP) v celice CHO magnetnih nanodelcev

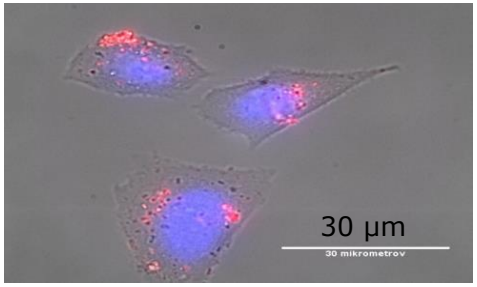
Selektivni vnos v rakave celice urotelija







Magnetni nanodelci za označevanje



Katedra za organsko kemijo FKKT UL

Inštitut za biologijo celice MF -UL

Nanotoksikologija

- okoljsko prisotni delci (saje, izpuhi,...)
- industrijsko proizvedeni (TiO2 P25, TiO2 FG, SiO2, železo-oksidni, kolidno srebro, nanozlato, ZnO,...)
- nezadostno poznavanje nanotoksičnosti, problem standardizacije (protokoli, doze, modeli,...)
- vnetje, kancerogeneza,...
- pljuča, možgani,...

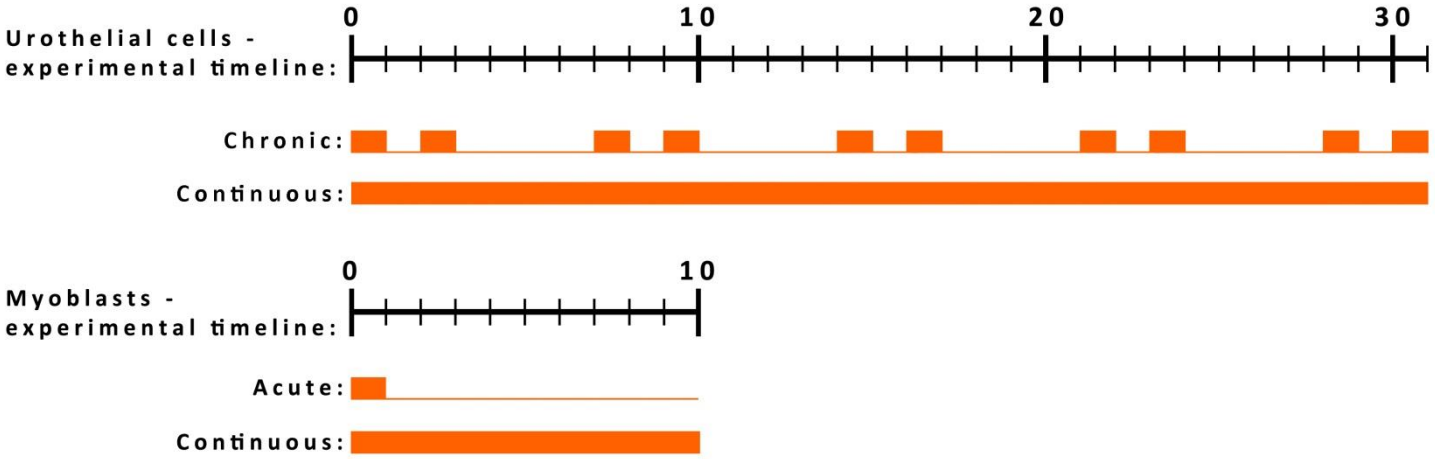
Magnetite pollution nanoparticles in the human brain PNAS 2016 113 (39)

Inhaled Nanoparticles Accumulate at Sites of Vascular Disease ACS Nano, 2017, 11: 4542–4552

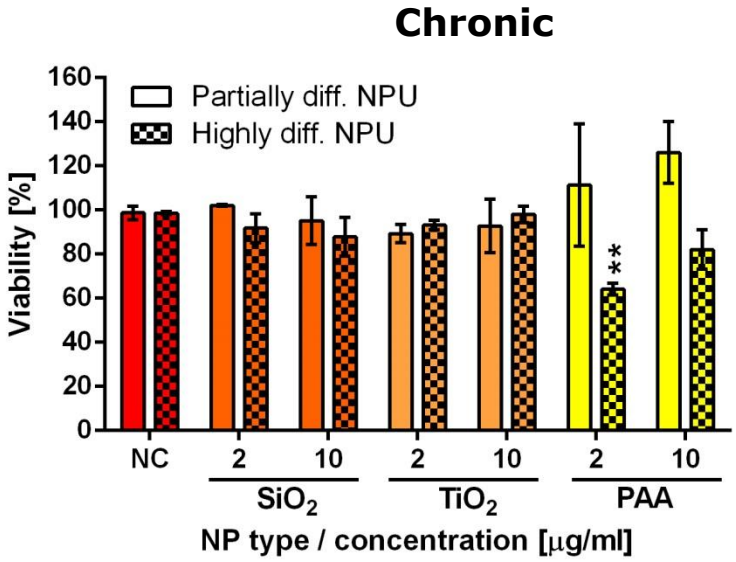
Long-term exposure of A549 cells to titanium dioxide nanoparticles induces DNA damage and sensitizes cells towards genotoxic agents Nanotoxicology 2016, 10:216

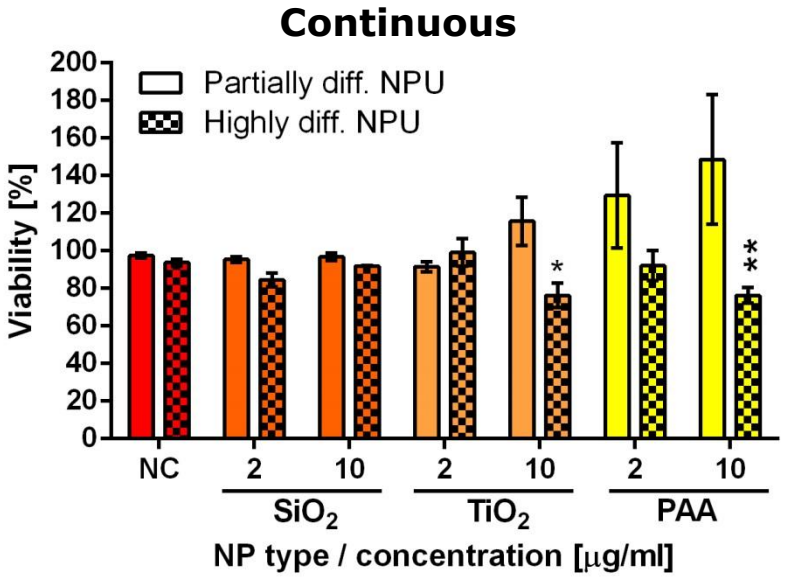
Less Is More: Long-Term in Vitro Exposure to Low Levels of Silver Nanoparticles Provides New Insights for Nanomaterial Evaluation ACSnano 2014, 8: 3260–3271

Dolgotrajna izpostavitev – in vitro model urotelija

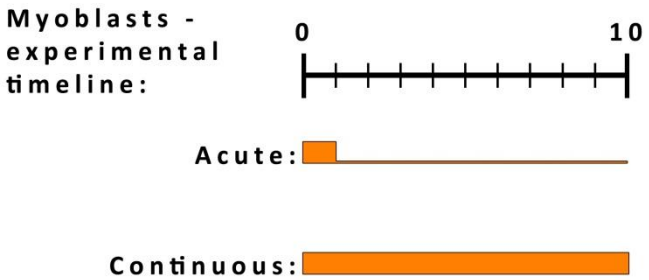


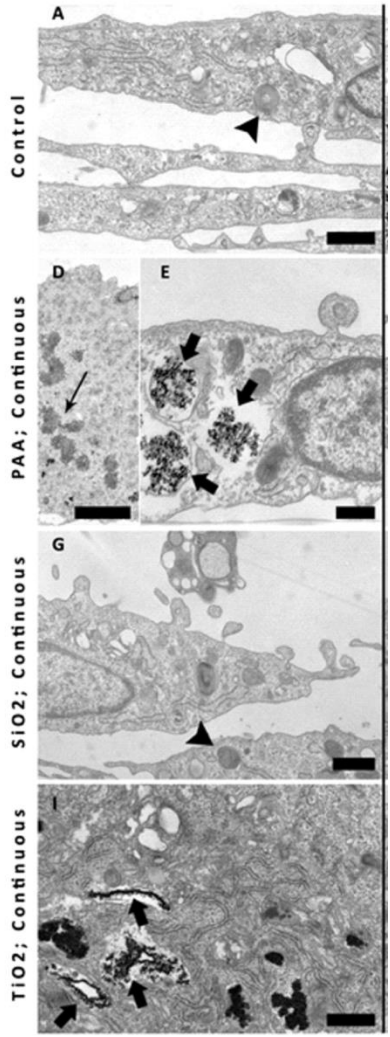
Inštitut za biologijo celice MF -UL



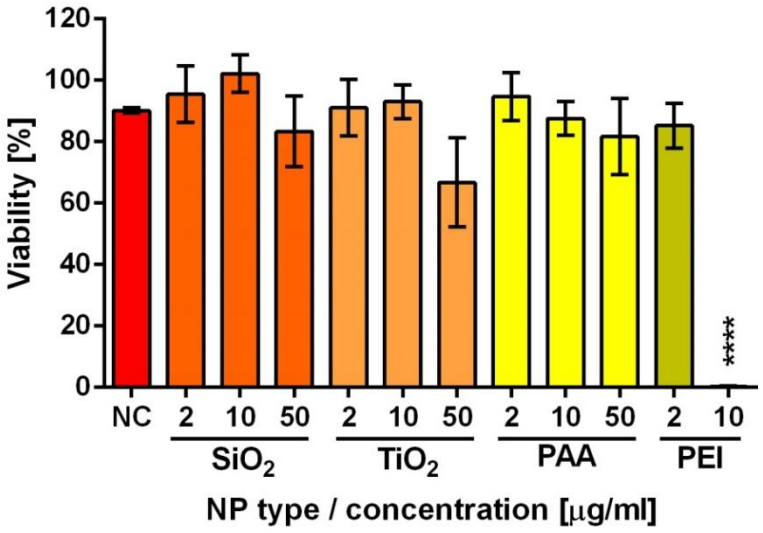


Nanotoksičnost – primarni mioblasti

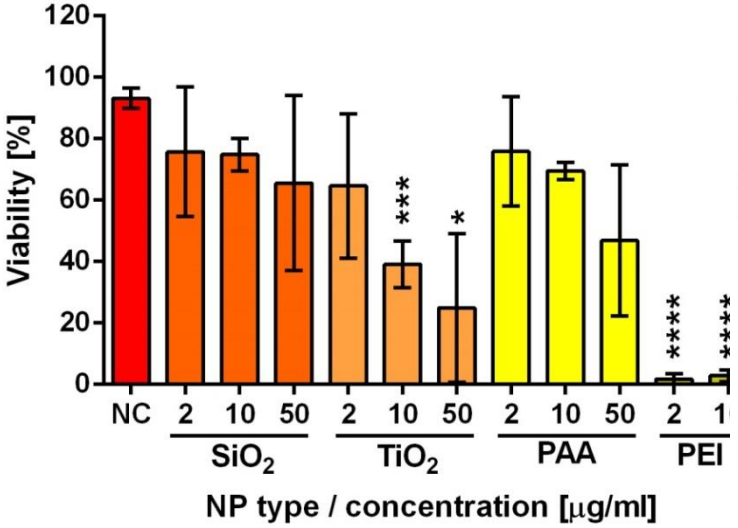




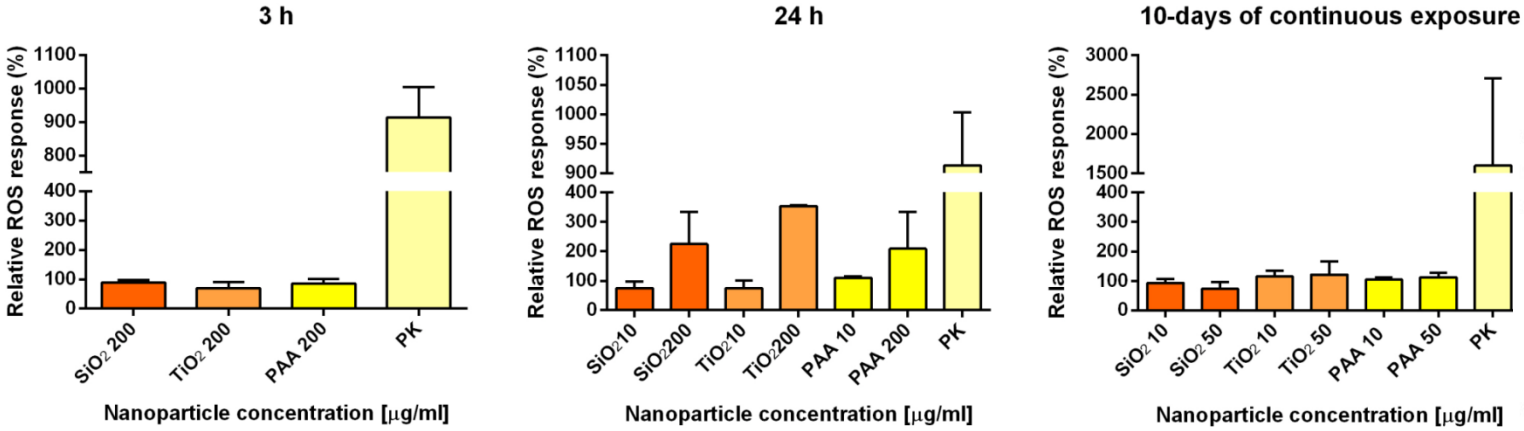
Acute

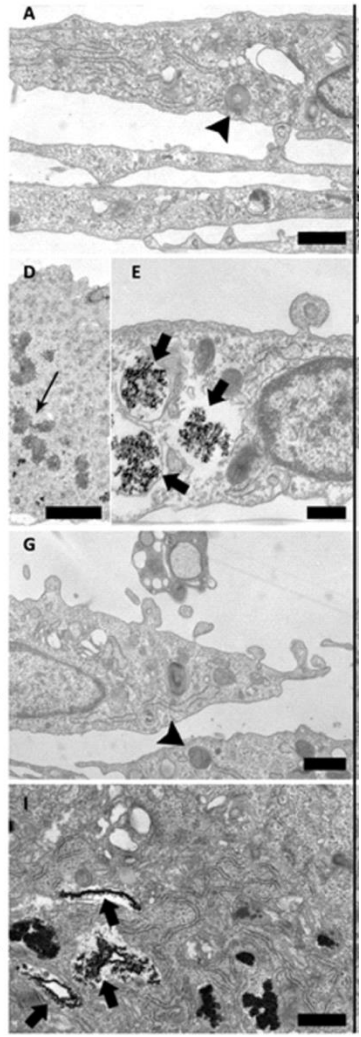


10 days Continuous

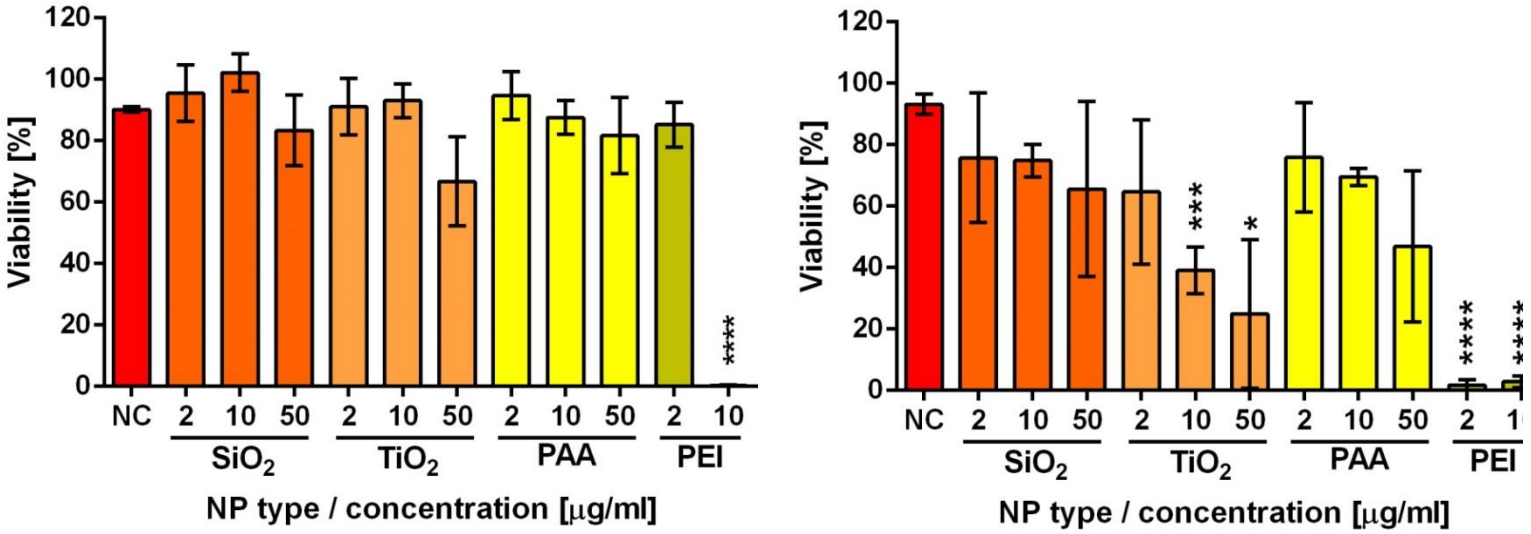


Nanotoksičnost – primarni mioblasti



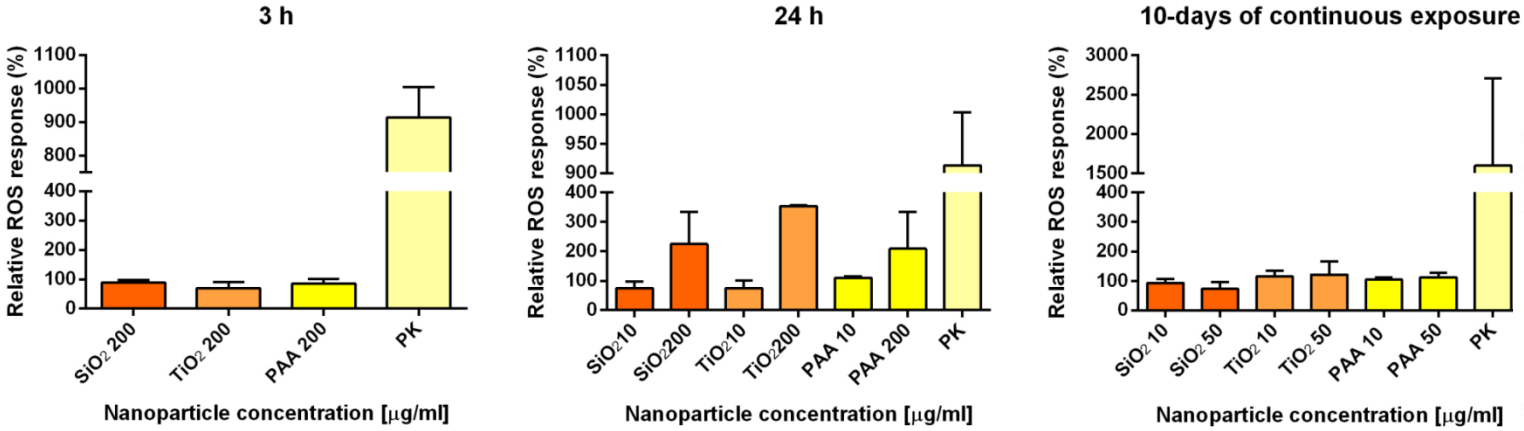


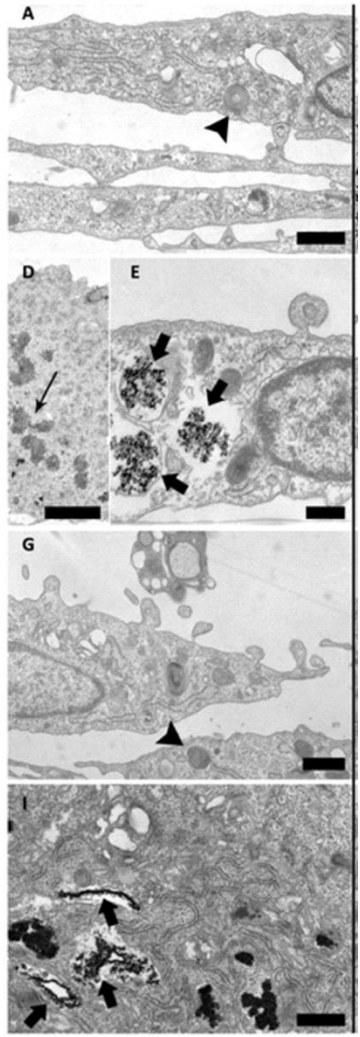
Acute **10 days Continuous**



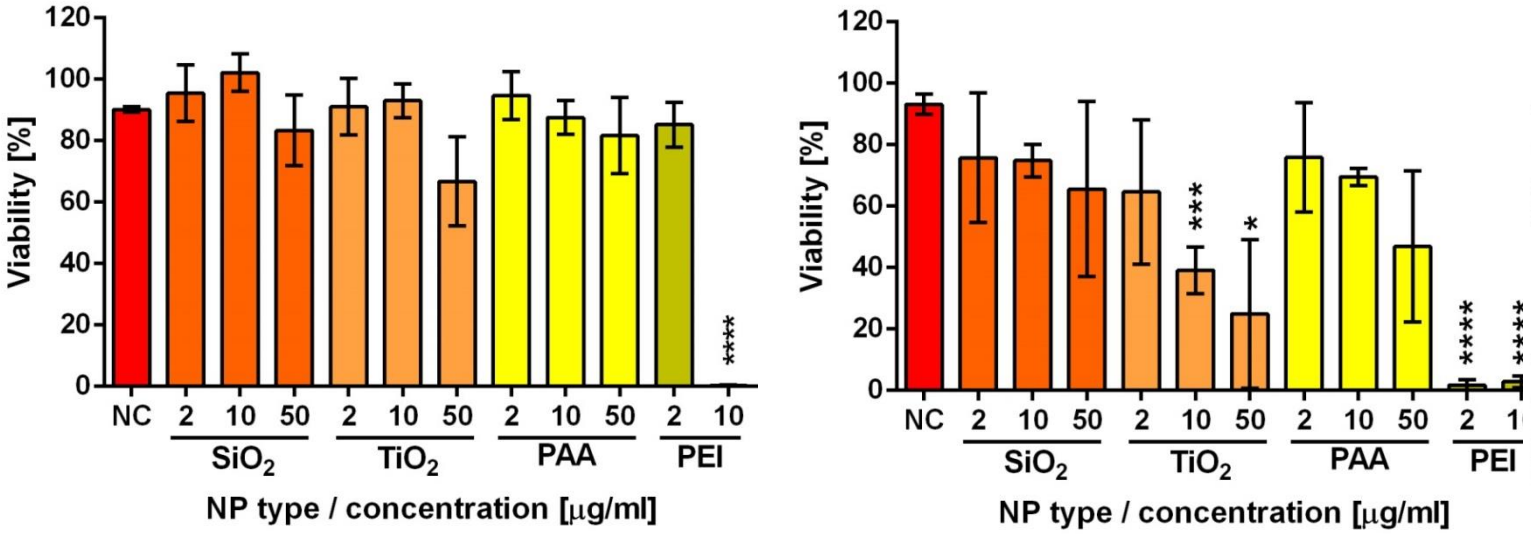
Laboratorij za molekularno nevrobiologijo MF-UL

Nanotoksičnost – primarni mioblasti

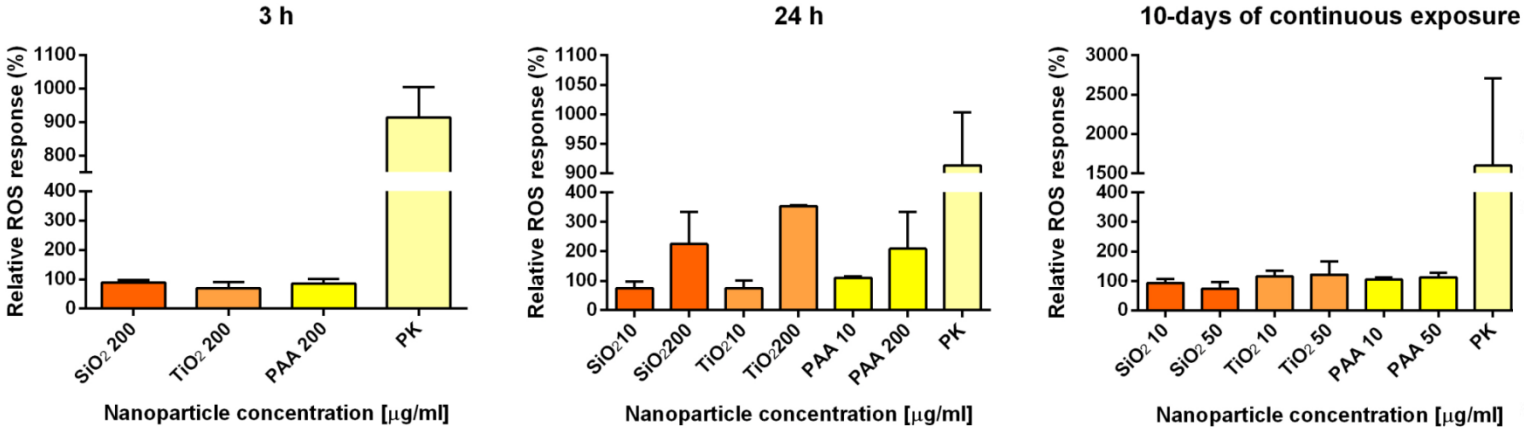




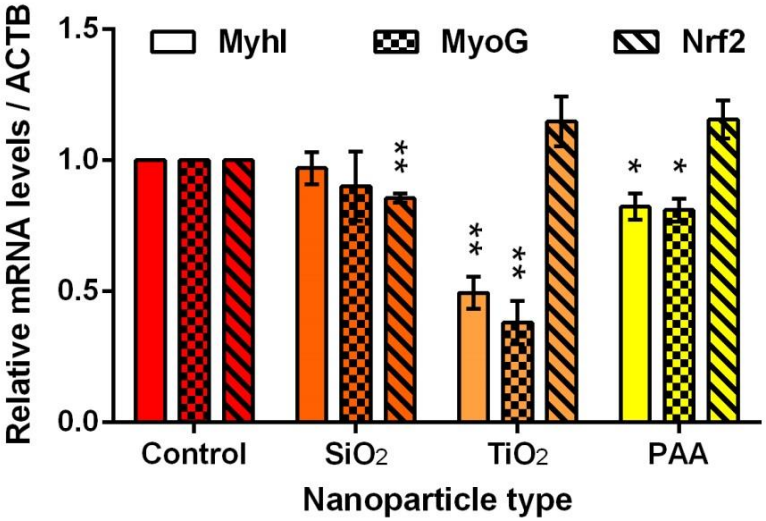
Acute **10 days Continuous**

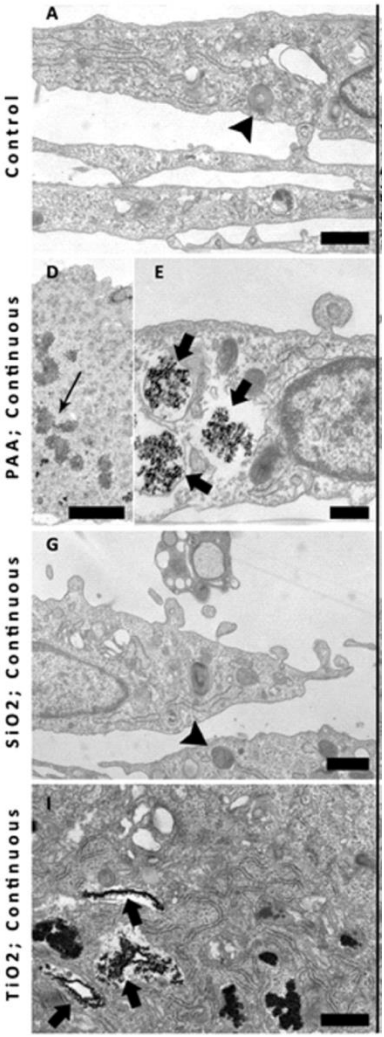


Nanotoksičnost – primarni mioblasti



Acute **10 days Continuous**

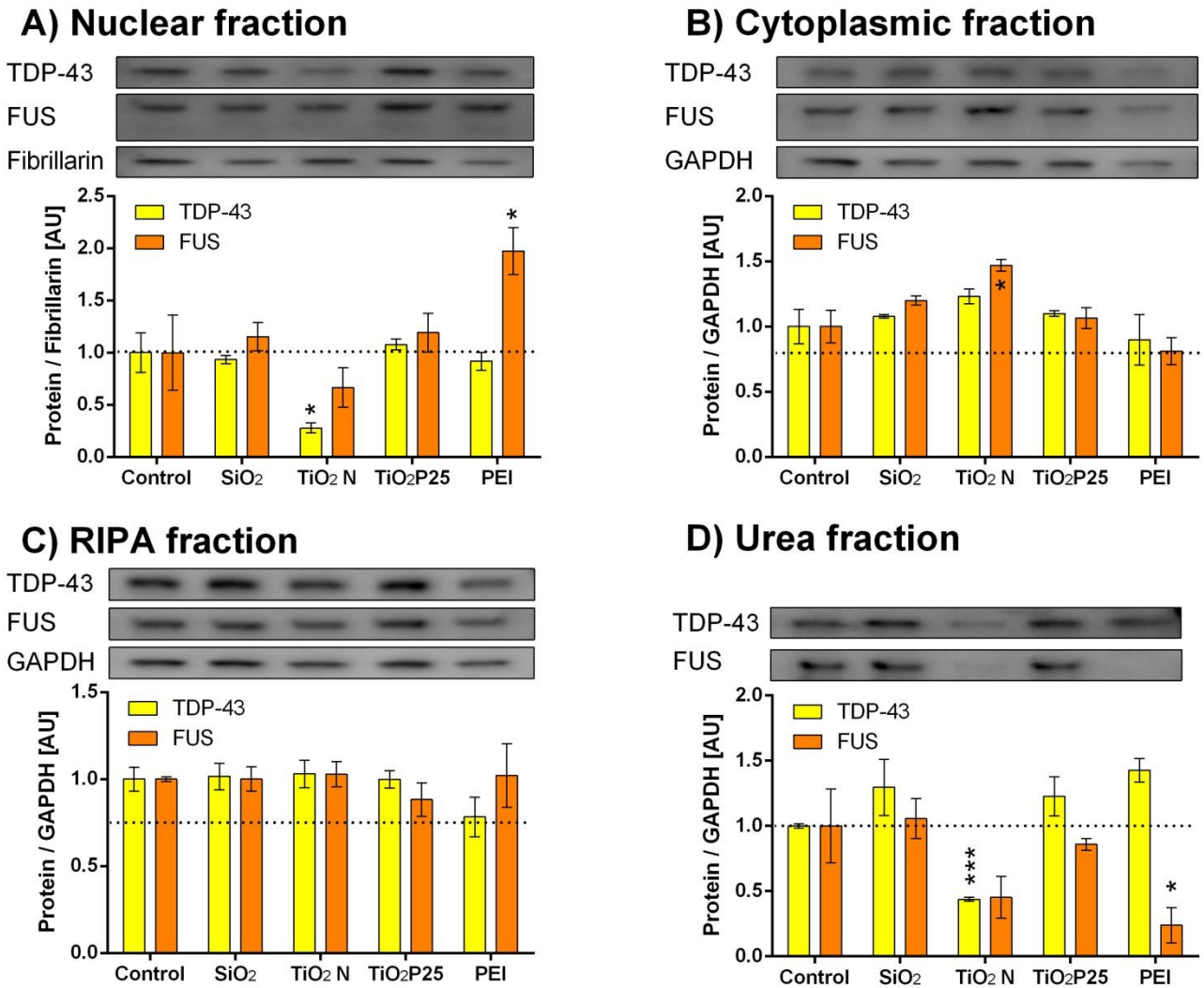




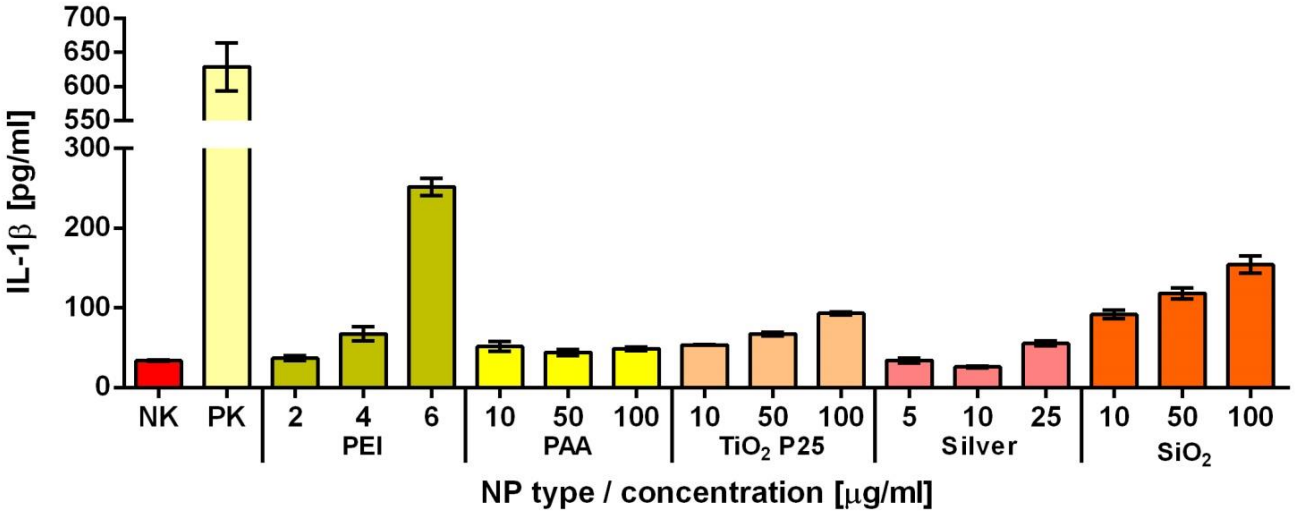
Laboratorij za molekularno nevrobiologijo MF-UL

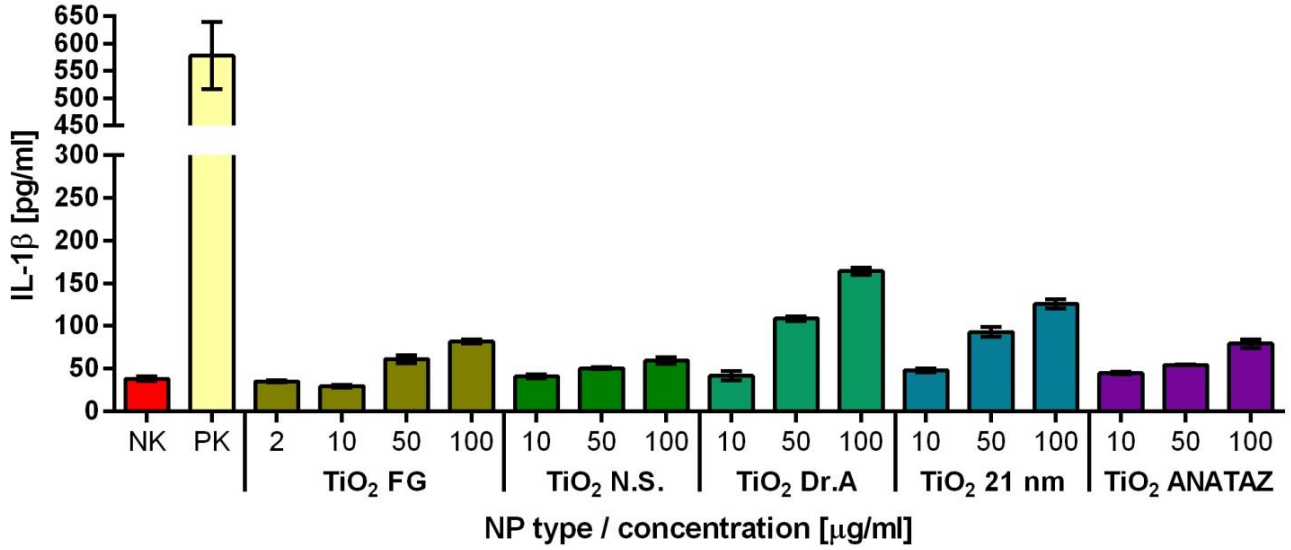
Neurotoksičnost

Odsek za biotehnologijo IJS



Imunski odziv



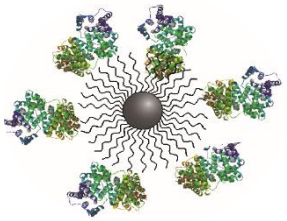


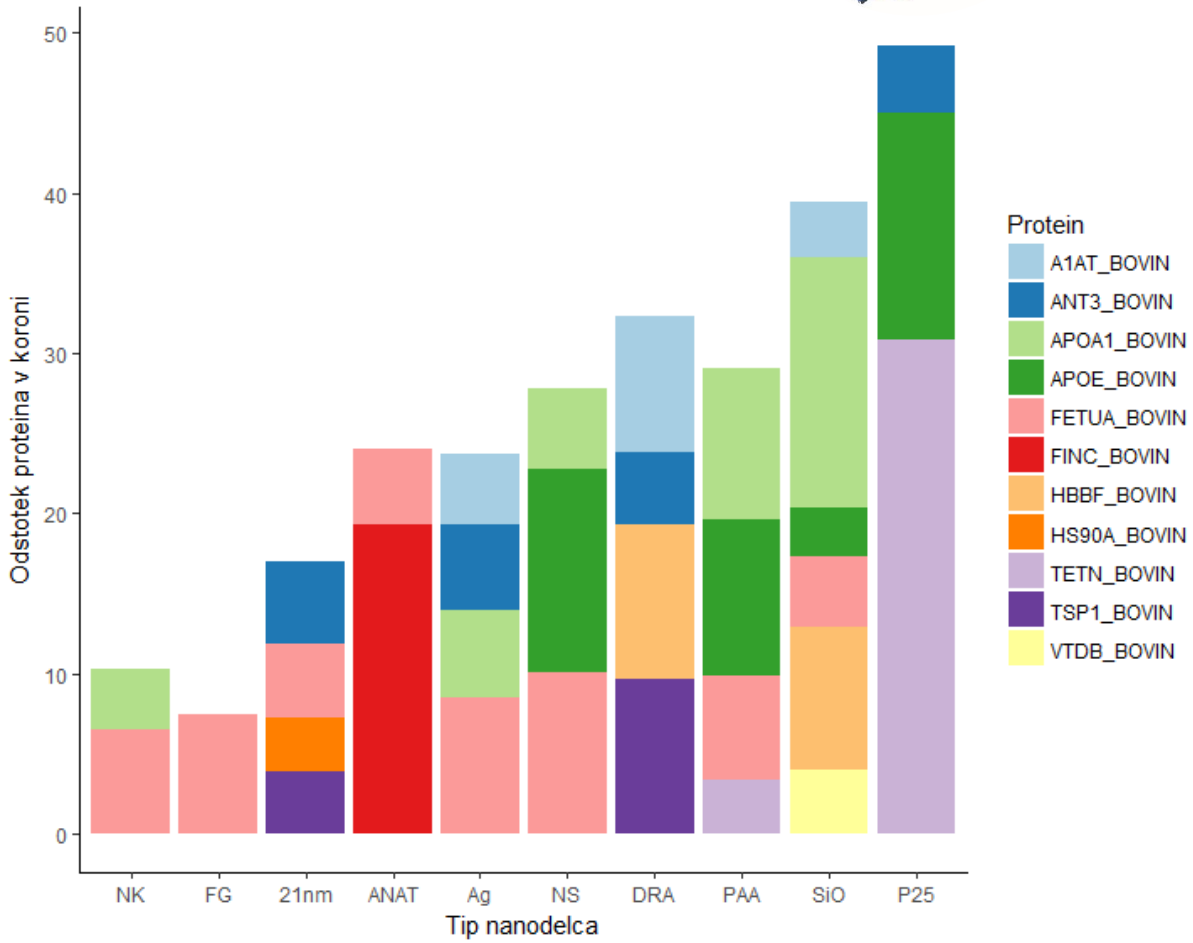
Laboratorij za sintezno biologijo in imunologijo

Odsek za nanostrukturne materiale IJS

Imunski odziv

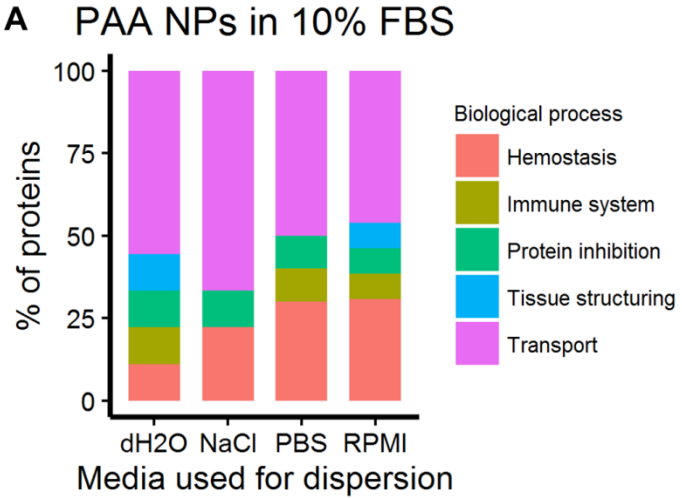
Proteinska korona

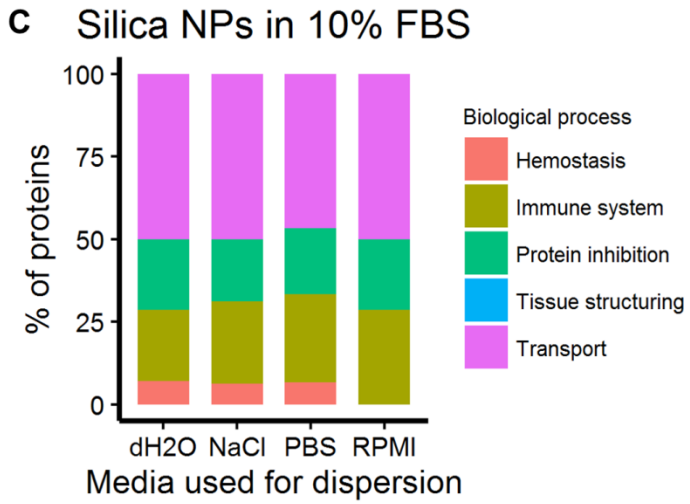


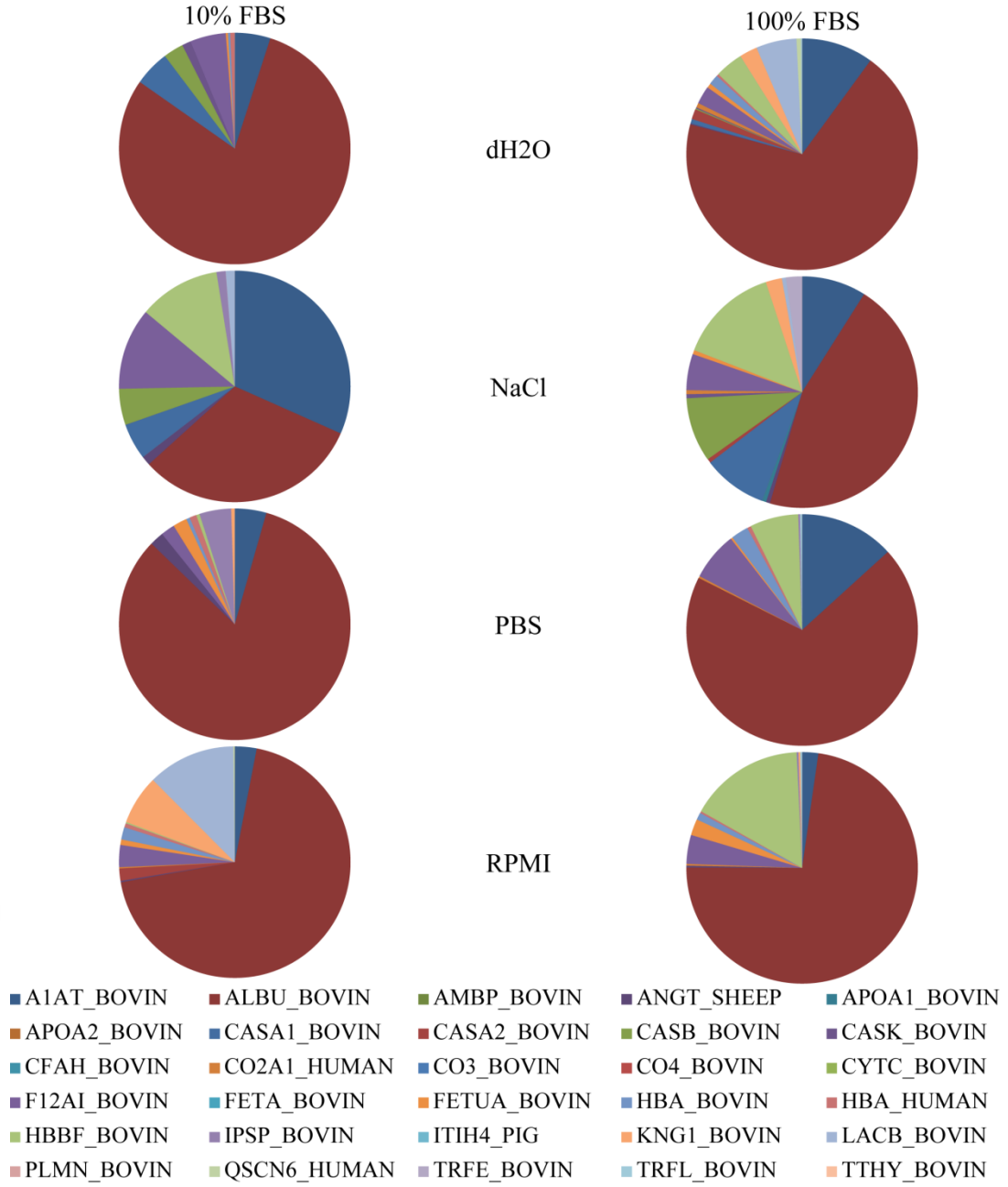


Laboratorij za sintezno biologijo in imunologijo

Odsek za nanostrukturne materiale IJS

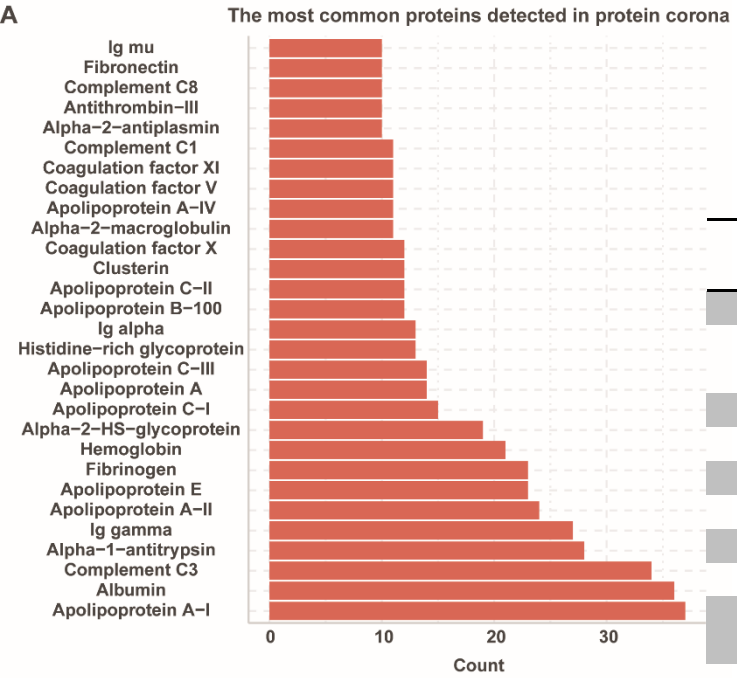


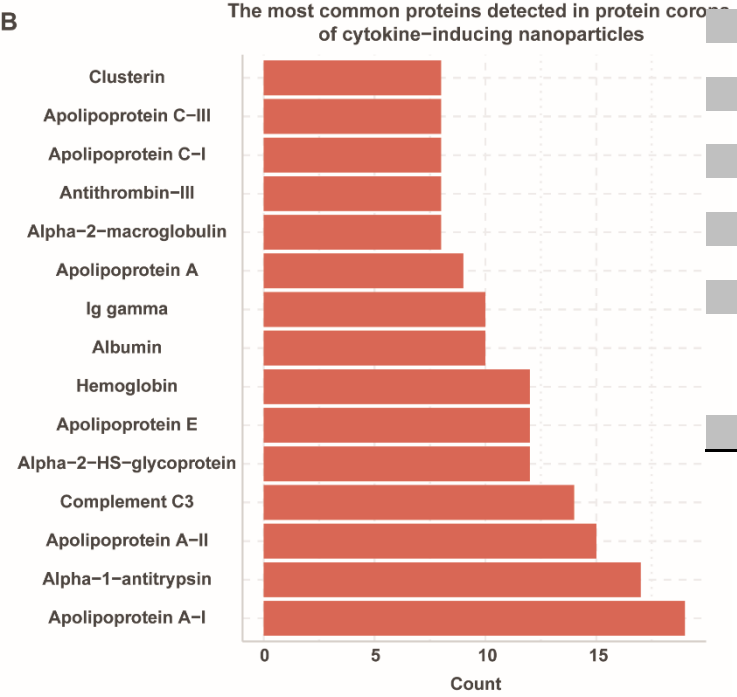




Odsek za molekularne in biomedicinske znanosti, IJS

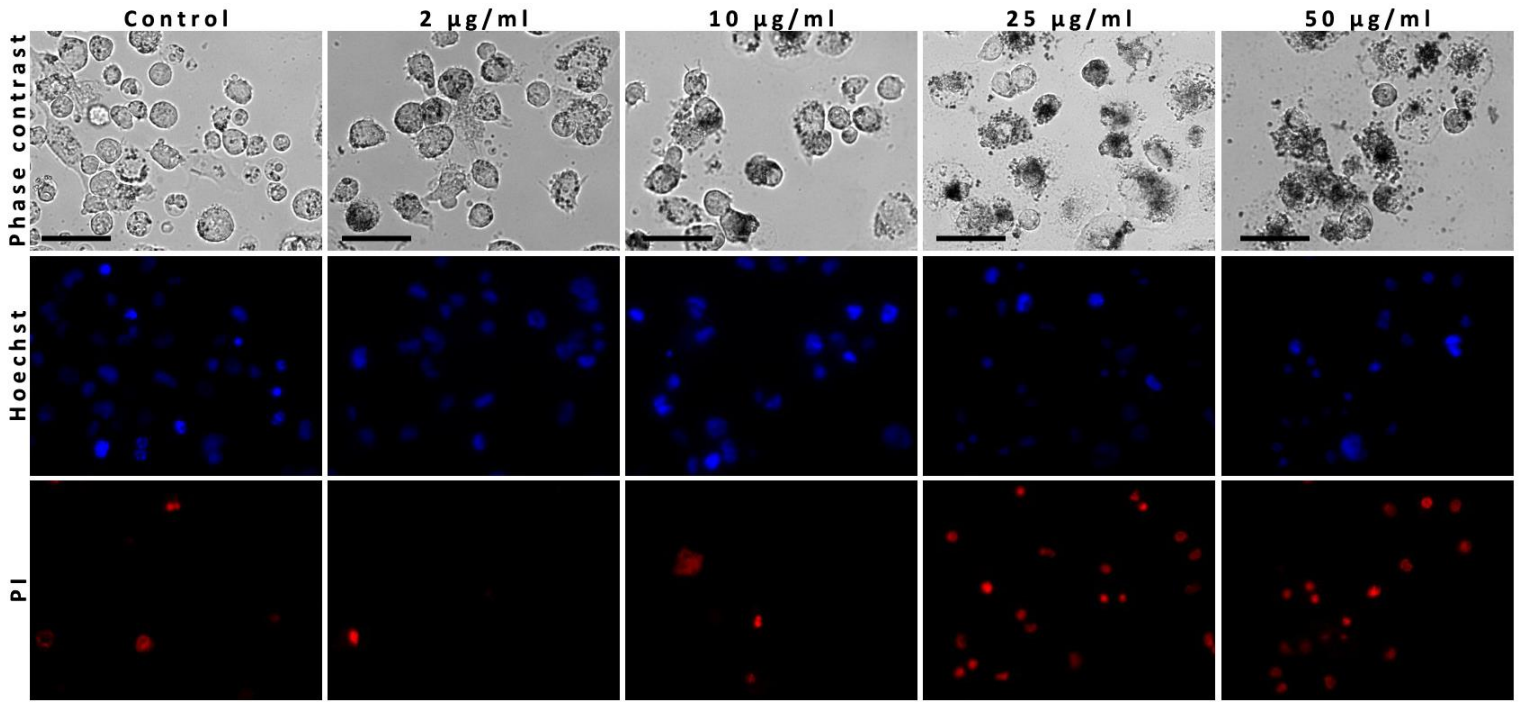
Proteinska korona



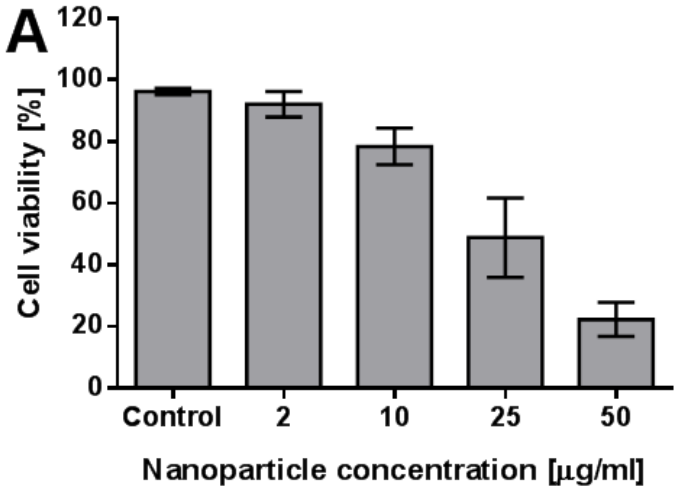


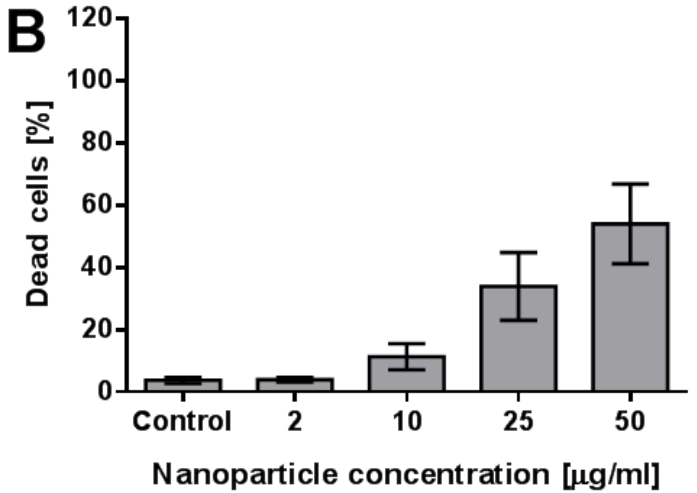
Type[1]	Protein source[2]	Cell line	Complement activation	Detected cytokines
metal	FBS	Human macrophages	NA	IL-1β, IL-6, IL-10
metal, combined	FCS	THP-1	NA	TNF-α, IL-1β, IL-6
polymeric	HS	NA	No	NA
combined	HP	THP-1	NA	TNF-α, IL-8
silica	FBS	RAW 264.7, MH-S	NA	TNF-α, IL-1β, IL-6
metal	HP	NA	No	NA
metal	HP	Human blood cells	Yes	none detected
magnetic	HP	THP-1	NA	none detected
silica	FCS	Primary human monocytes	NA	IL-1β
metal	HP	NA	No	NA
polymeric	FBS	J774	No	none detected
silica	HS	RAW 264.7	NA	TNF-α
metal	FBS	J774	NA	TNF-α, MIP-2
metal	FBS, BSA	RAW 264.7	NA	none detected
other	HP, BF	THP-1, PBMC	NA	TNF-α, IL-1β, IL-7
polymeric	HS	Human macrophages	NA	none detected
magnetic	HP	Human macrophages	NA	none detected
silica	FBS	THP-1	NA	TNF-α, IL-1β, IL-6
liposomal	HS	NA	Yes/No	NA
polymeric	FBS	THP-1	NA	TNF-α, IL-1β, IL-6, IL-2, IL-8, IL-9, IL-17A, Eotaxin, IFN-γ, MIP-1α, MIP-1β
polymeric	HS	NA	Yes	NA

Nano srebro, koloidno srebro



Silver powder has been classified as hazardous (Aquatic Chronic 1 and Aquatic Acute 1) with the following Hazard Statements (GHS): H410: Very toxic to aquatic life with long lasting effects and H400: Very toxic to aquatic life. [https://echa.europa.eu]





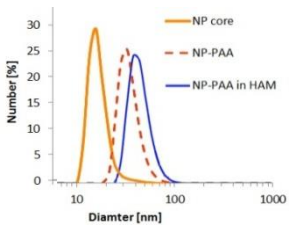
Nanoportal

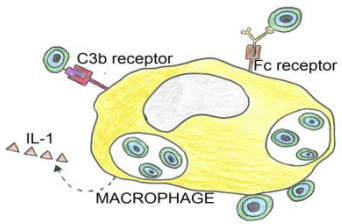
http://www.uk.gov.si/si/delovna_podrocja/nanoportal/

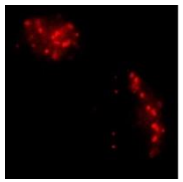
NANOPORTAL
Definicija nanomaterialov
Lastnosti in klasifikacija nanomaterialov
Uporaba nanomaterialov
Izvor nanodelcev
Karakterizacija nanodelcev in nanomaterialov
Monitoring nanodelcev v zraku in vodi
Zakonodaja na področju nanomaterialov
Nanodelci in nanovarnost
Ognjemeti in druga zabavna pirotehnika
Koristne povezave
Seznam raziskovalcev za nanomateriale
8. POSVET KEMIJSKA VARNOST ZA VSE: NANOVARNOST – Ali smo dovolj previdni z nano?

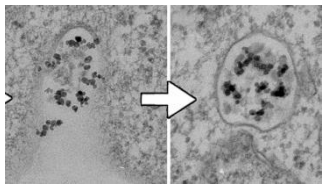
Nanoportal pripravljamo v sodelovanju z:
- prof. dr. Majo Remškar, Institut Jožef Stefan
- prof. dr. Urško Lavrenčič Štangar, Univerza v Novi Gorici
- prof. dr. Damjano Drobne, Biotehniška fakulteta, Univerza v Ljubljani
- doc. dr. Mojco Pavlin, Fakulteta za elektrotehniko, Univerza v Ljubljani

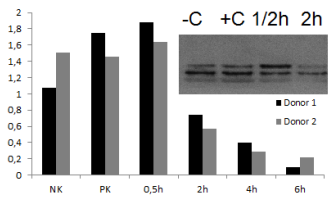
- Ključno zadostno poznavanje potencialne nanotoksičnosti
- Kaj je nano? Velik pomen ustrezne karakterizacije, standardizacije (smernice EU, standardi, itd...): http://www.euncl.eu/
- Kako poročati nevtralno?
- Vsaka formulacija nanomateriala svoj problem
- Velika razlika med industrijskimi nanomateriali ter nanodelci, ki so v osnovi razviti za stabilnost pri pH 7 - večina industrijskih materialov agregira v fizioloških razmerah
- Relevantne doze in vivo in vitro – prenosljivost rezultatov
- Način izpostavitve/administracije, prah/suspenzija, kronična izpostavitev, akumulacija
- Vendar, problem dolgotrajnega kopičenja ND (npr. v možganih, pljučih) – nujnost modelov in protokolov z dolgotrajno izpostavitvijo (*Magnetite pollution nanoparticles in the human brain PNAS 2016 113 (39) 2016*)
- Nanomateriali „prehiteli" zadostno karakterizacijo ter razumevanje potencialne nanotoksičnosti (hrana – 1 mg TiO2/dan)
- Določeni nanodelci okoljsko problematični (izpuhi, voda, zemlja)
- Imunogenost - zelo kompleksno











Skupina za nano in biotehnološke aplikacije

Doc. Dr. Mojca Pavlin
znan. sod. Dr. Vladimir B Bregar
Dr. Jasna Lojk
Maruša Bizjak
Klemen Strojan
Klemen Dolinar

Dr. Matej Skočaj
Petra Malavašič
Dr. Vid Šuštar
Nives Škorja

Klavdija Glavač
Urša Kešar
Dominik Dekleva
Jerneja Kladnik
Suzana Semič
Blaž Oblak

Špela Granda
Ajda Demšar
Maja Okretič
Jan Gregorec
Nina Roštan

Javna agencija
za raziskovalno dejavnost
Republike Slovenije

Hvala!

Kemijski Inštitut
Laboratorij za biotehnologijo
Prof. Dr. Roman Jerala
Dr. Iva Hafner Bratkovič

Inštitut za patološko fiziologijo,
Laboratorij za molekularno nevrobiologijo MF-UL
Doc. Dr. Sergej Pirkmajer
Prof. Dr. Tomaž Marš
Prof. Dr. Zoran Grubič
Dr. Katarina Miš

Katedra za organsko kemijo FKKT UL
prof. dr. Jurij Svete

Inštitut za biologijo celice, MF-UL
Prof. Dr. Peter Veranič
Izr. Prof. Dr. Mateja Erdani Kreft

Odsek za molekularne in biomedicinske znanosti, IJS
Prof. Dr. Igor Križaj
Dr. Adriana Leonardi

Odsek za biotehnologijo IJS
Prof. Dr. Boris Rogelj

Odsek za nanostrukturne materiale IJS
Znan. svet. Dr. Saša Novak
Dr. Nataša Drnovšek

Fakulteta za informatiko UL
Doc. dr. Luka Šajn
Dr. Uroš Čibej

Inštitut za biofiziko
P1-0055

Javna agencija za raziskovalno dejavnost Republike Slovenije

10. cikel

13. September 2017 ob 19h

Izbira

Dr. Renata Salecl, Inštitut za kriminologijo

20. September 2017 ob 19h

Znanstveni slam