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EVROPSKÁ UNIE
MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY
OP Vzdělávání pro konkurenceschopnost
INVESTICE DO ROZVOJE VZDĚLÁVÁNÍ

Název: Organické fluorescenční sloučeniny vhodné pro monitorovnání v živých organismech-úvod

Školitel: Pavel Kopel

Datum: 11.10.2013

Reg.č.projektu: CZ.1.07/2.3.00/20.0148

Název projektu: Mezinárodní spolupráce v oblasti "in vivo" zobrazovacích technik

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Název: Organic Fluorescent Compounds for in vivo Imaging - Introduction

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Datum: 11.10.2013

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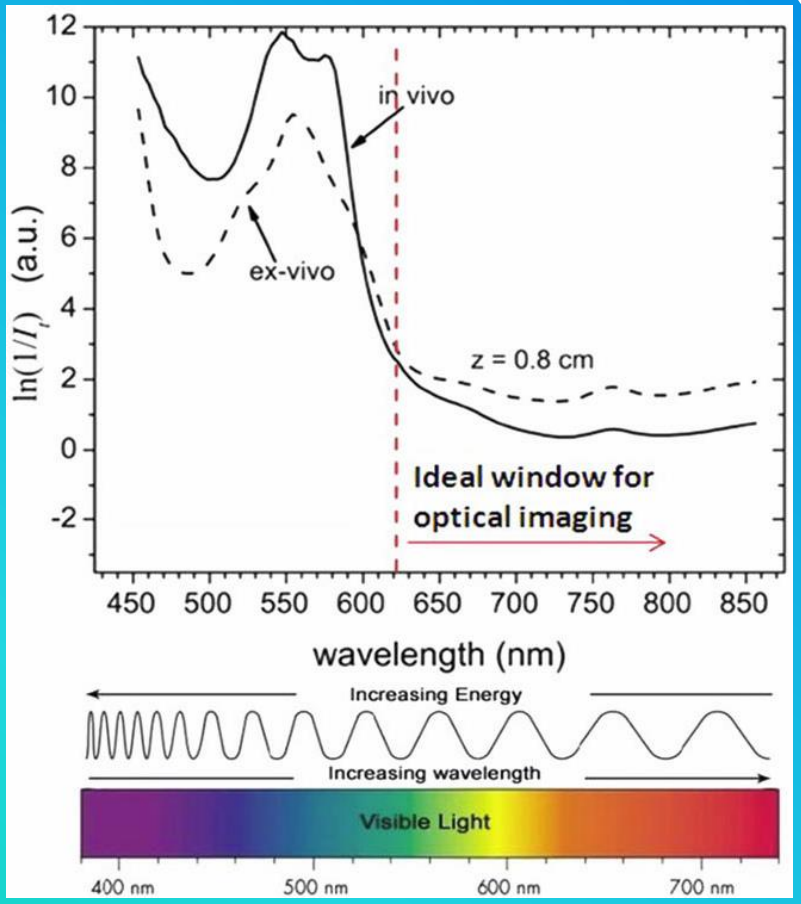
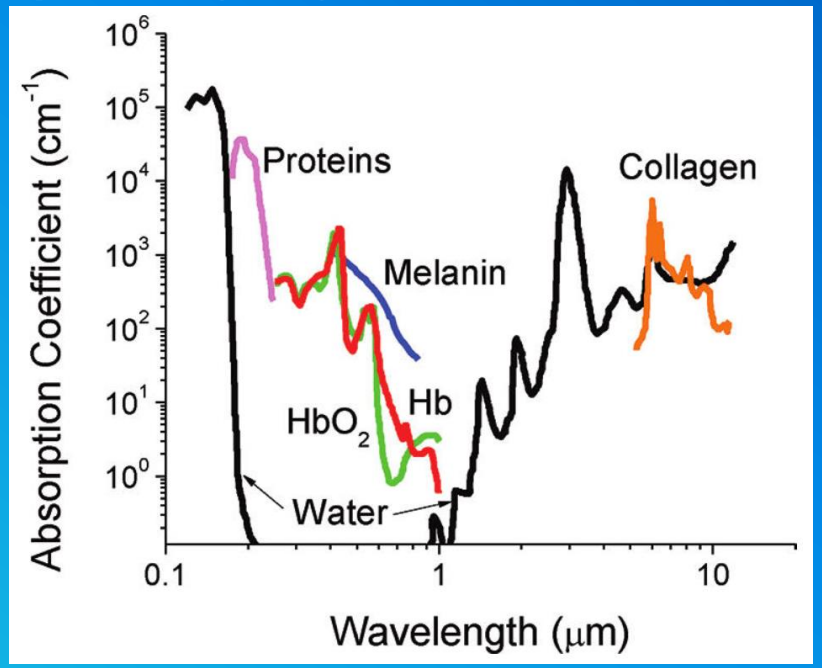
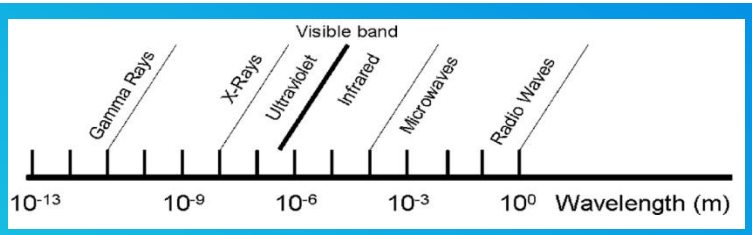
The importance of long wavelength and near-infrared (NIR) imaging



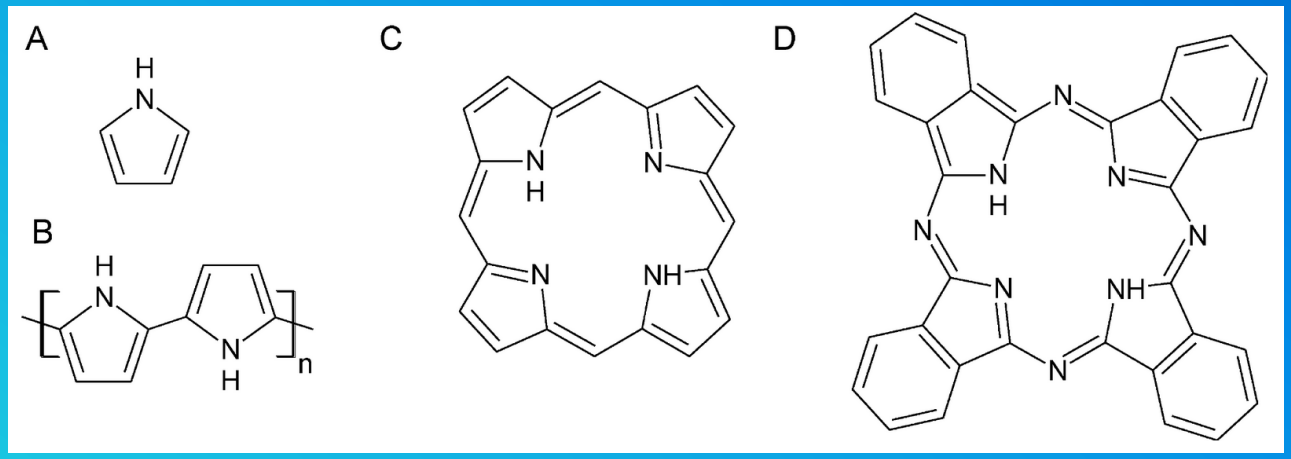


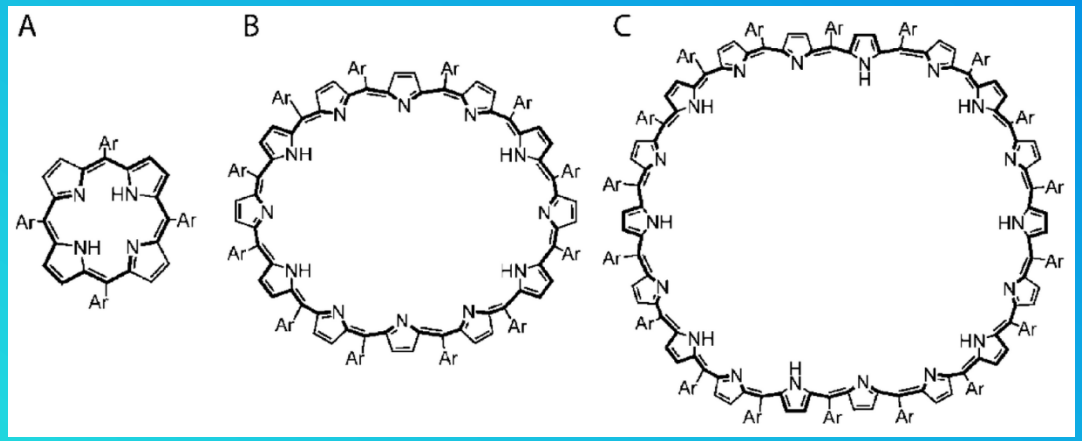


Absorbance of various tissue and blood components from 200 nm to 10 μm. The optical imaging window ranging from 650 to 1450 nm represents the range where tissue penetration is greatest.

- Vikram J. Pansare, Shahram Hejazi, William J. Faenza, Robert K. Prud'homme Chem. Mater. 2012, 24, 812−827

The importance of long wavelength and near-infrared (NIR) imaging





A, pyrrole; B, polypyrrole; C, porphyrin; D, phthalocyanine

Increasingly large porphyrin rings have absorption maxima that shift into the NIR (411−953 nm). A, porphyrin; B, dodecaphyrin; C, octadecaphyrin (Ar = C_6F_5).

Organic Fluorophores

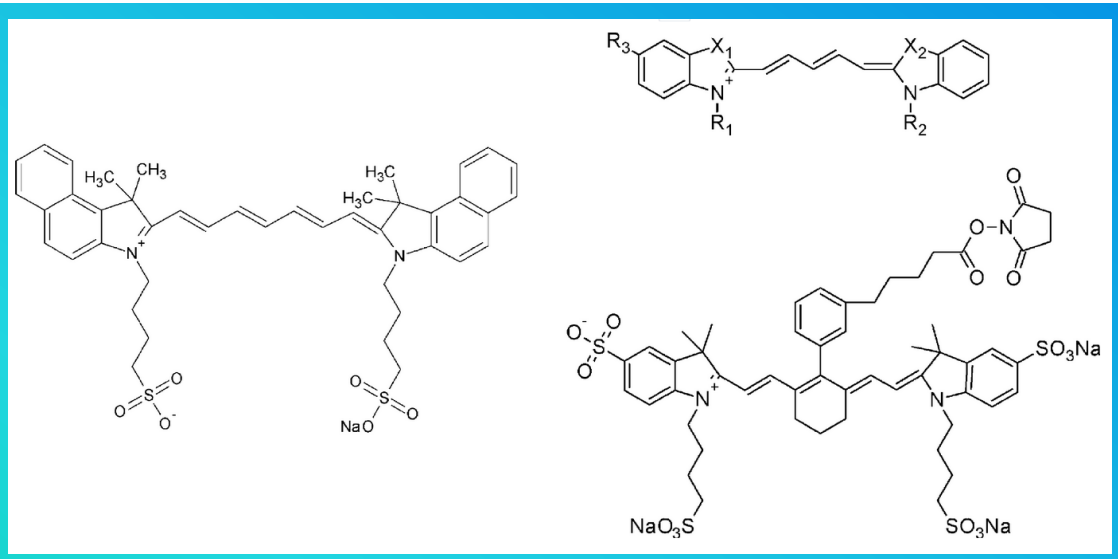
fluorophore	λ_{EX} (nm)	λ_{EM} (nm)	ε (M^{-1} cm^{-1})	QY[b] (%)
Cyanines				
indocyanine green (ICG)	775	831	113k[c]	1.3%
Cy5	648	666	250k	18%
Cy5.5	679	696	250k	24%
Cy7	745	775	250k	28%
DiD	648	669	>125k	33%
DiR	750	782	>125k	28%
IR Dye 800CW	778	794		
IR Dye 680RD/LT	680	694		
IR Dye 750	766	776		
IR Dye 800RS	770	786		
IR Dye 650	651	668		
heptamethine 3H-indolenine cyanine dyes	782–786	807–814	220k–250k	10–15%
large Stokes shift cyanine derivatives	602, 617, 783	757, 803	50k, 70k, 200k	47, 38, 17%
bis(heptamethine cyanine dyes)	780	800		
heptamethine dyes with robust C–C bond at center	770, 800	785, 811	220k–240k	8.8–10%
alkyl-thioether derivatized cyanine dyes	777–823	812–847	116k–174k	2%
Squaraines				
sulfonated squaraine derivatives	737–780	751–820	~200k	8–44%[d]
bis-squaraines with pyrene/ thiophene linker	thiophene: 695, 727 pyrene: 636, 647	thiophene: 750, 790 pyrene: 757, 763	thiophene: 74k–240k pyrene: 68k–100k	0.01–1%
tetralactam encapsulation of squaraines	631–661	650–704		8–74%[d]

Organic Fluorophores

fluorophore	λ_{EX} (nm)	λ_{EM} (nm)	ε (M^{-1} cm^{-1})	QY[b] (%)
BODIPYs				
monomeric and polymeric BODIPY derivatives	monomer: 597, 665 polymer: 634–738	monomer: 631, 701 polymer: 669–760		monomer: 2.6, 4% polymer: 1.1–13%
heteroaryl fused BODIPYs	723, 509–690	738, 517–701	140k–316k	56%, 81–98%
BF_2-chelated tetraarylazadipyrromethenes	690–706	714–730	75k–95k	22–30%
BODIPY 650/665	647	664	102k	46%
Other				
Alexa Fluor 647	650	668	270k	33%
Alexa Fluor 660	664	691	130k	37%
Alexa Fluor 680	680	704	180k	36%
Alexa Fluor 700	694	720	190k	25%
Alexa Fluor 750	752	776	240k	12%
Alexa Fluor 790	784	814	260k	
Nile Blue	630	660	77k	27%[d]
CellMask Deep Red Plasma Membrane Stain	649	666		
FluoSpheres Dark Red	657	683		
FluoSpheres Infrared	715	755		
FocalCheck Double FarRed	669	693		
HCS CellMask DeepRed Stain	648	670		
HCS NuclearMask DeepRed	broad 638	broad 685		
LIVE/DEAD Fixable NIR Cell Stain	753	776		
MitoTracker DeepRed	640	662		
Qnuclear Deep Red Stain	642	656		
SYTO 60	649	681	>50k	16%
SYTOX	641	658		
TetraSpeck Dark Red Dye	656	684		
TO-PRO-3	642	657		
TOTO-3	643	660		
Vybrant DyeCycle Ruby	638	broad 686		
DRAQ5, DRAQ7	647	broad 665–800	21k	0.3–0.4%
X-Sight 650 Nanospheres[e]	650	673		
X-Sight 691 Nanospheres[e]	691	715		
X-Sight 761 Nanospheres[e]	761	789		
X-Sight 670 LSS Dye[e]	669	755		

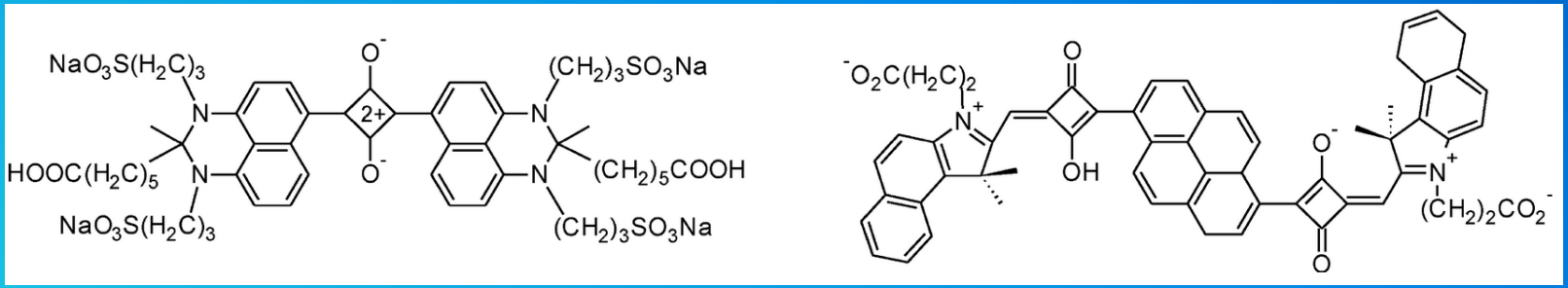
Organic Fluorophores

fluorophore	λ_{EX} (nm)	λ_{EM} (nm)	ε (M^{-1} cm^{-1})	QY[b] (%)
organic soluble perylenetetracarboxdiimide derivatives	604–712	636–768	41k–59k	NR[f]
water-soluble perylenetetracarboxdiimide derivatives	432–566	588–619	10k–33k	49–66%
fluorescent protein iRFP	690	713	85k–105k	5.9%
fluorescent protein IFP1.4	684	708	>90k	7%
upconverting phosphors	980	540, 655		N/A[78]
quantum dots	broad visible range	535–850	500k–2M	60%
Zn^{2+}-multiporphyrin	tunable: 410, 500, 690–800	tunable: 697–817	23k–126k	NR[f]
IRDye 700DX	680	687	170k	14%
single wall carbon nanotubes	broad 600–800	broad 950–1300	see refs 124, 125	0.01–0.1%

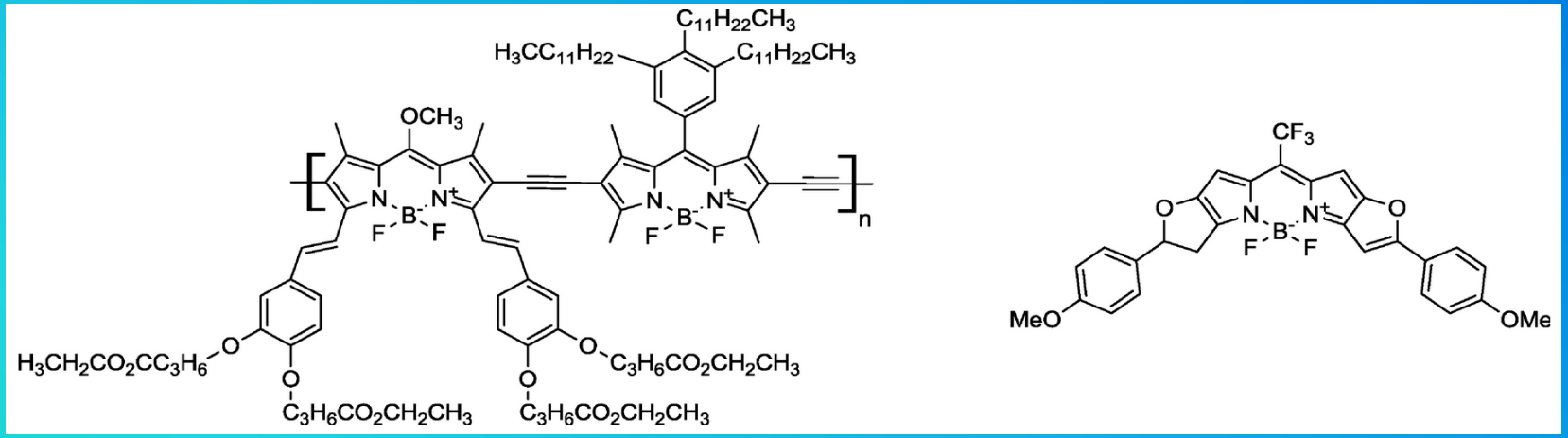


- Cyanine dyes ICG, Cy5, IRDye 750 NHS Ester

Organic Fluorophores



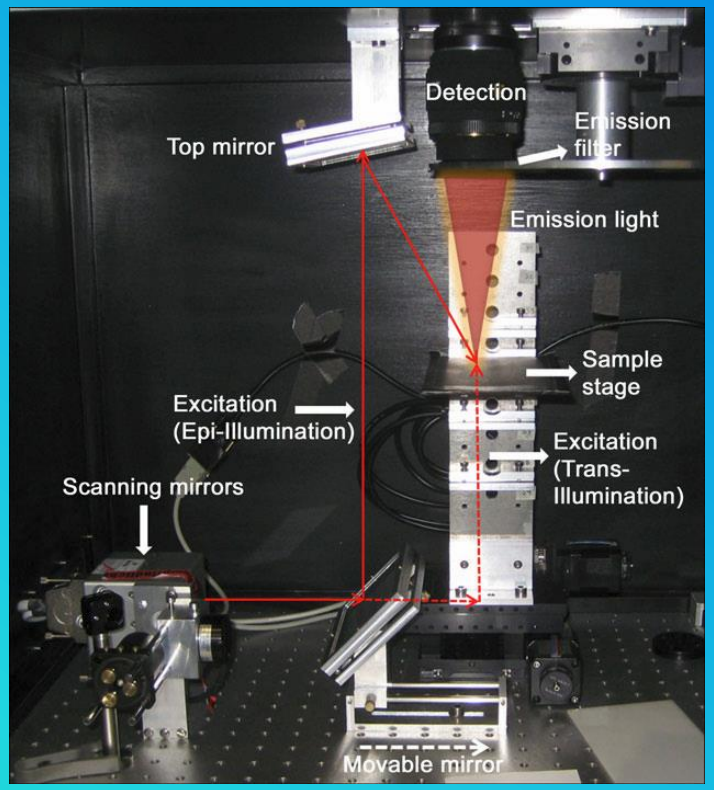
- Examples of squaraines



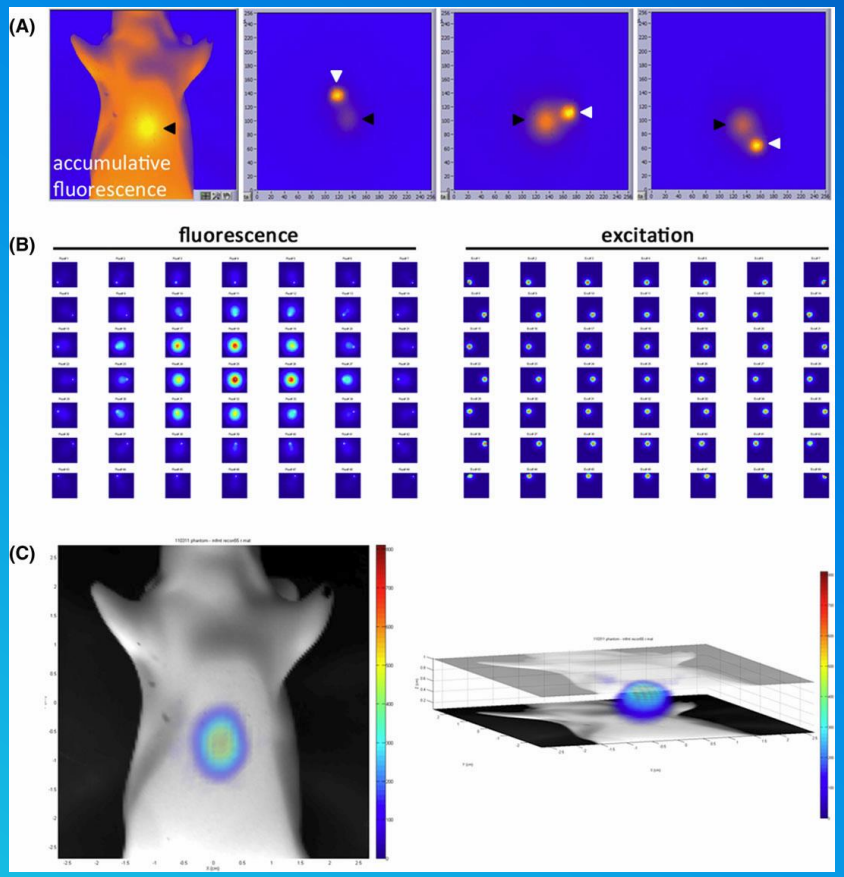
- BODIPY (borondipyrromethene) class dyes

They generally have sharp absorbance and emission profiles and high quantum yields approaching 100%. However, few are water-soluble. Most of them emit below 600 nm, making them unsuitable for deep tissue imaging

Fluorescence Molecular Tomography (FMT)

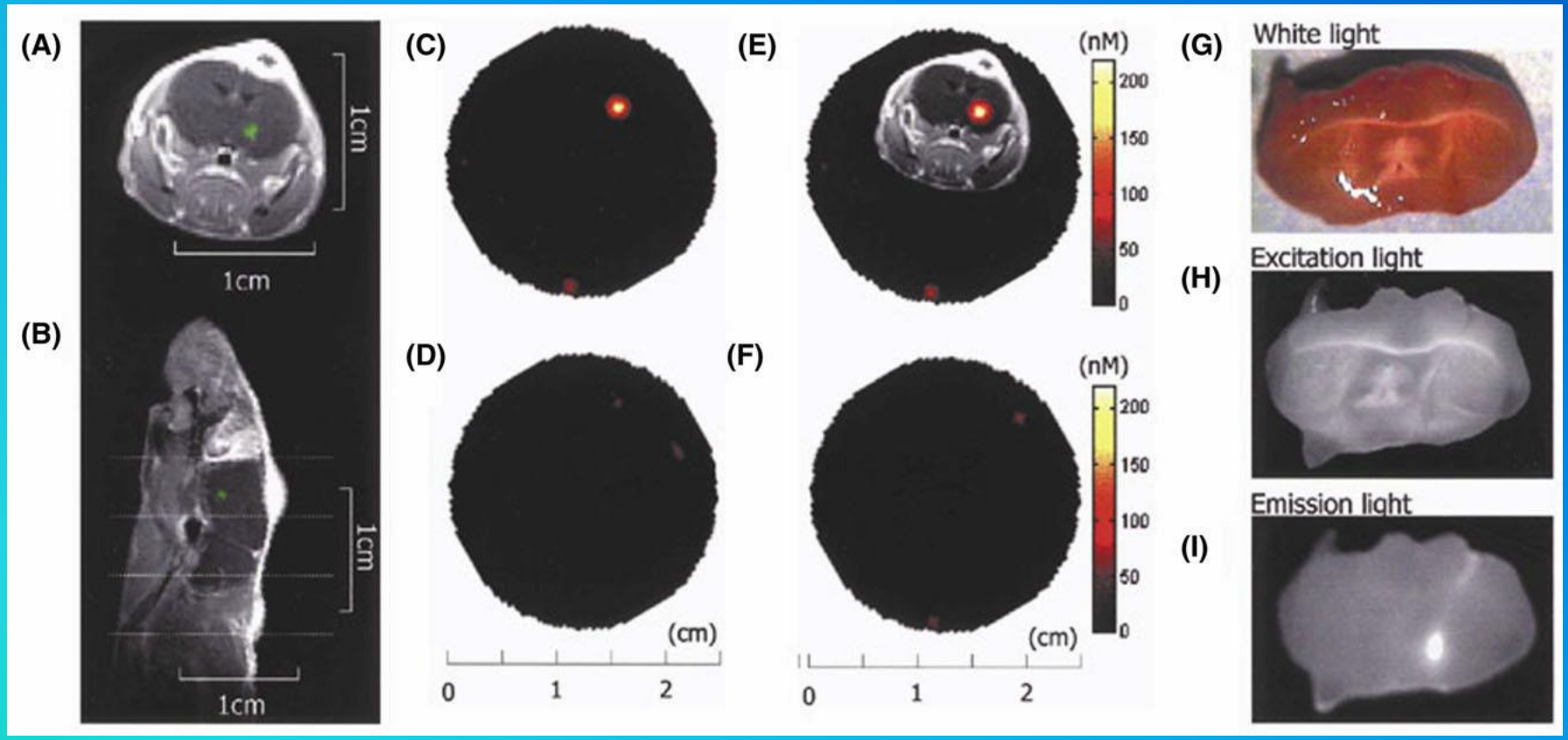


FMT apparatus



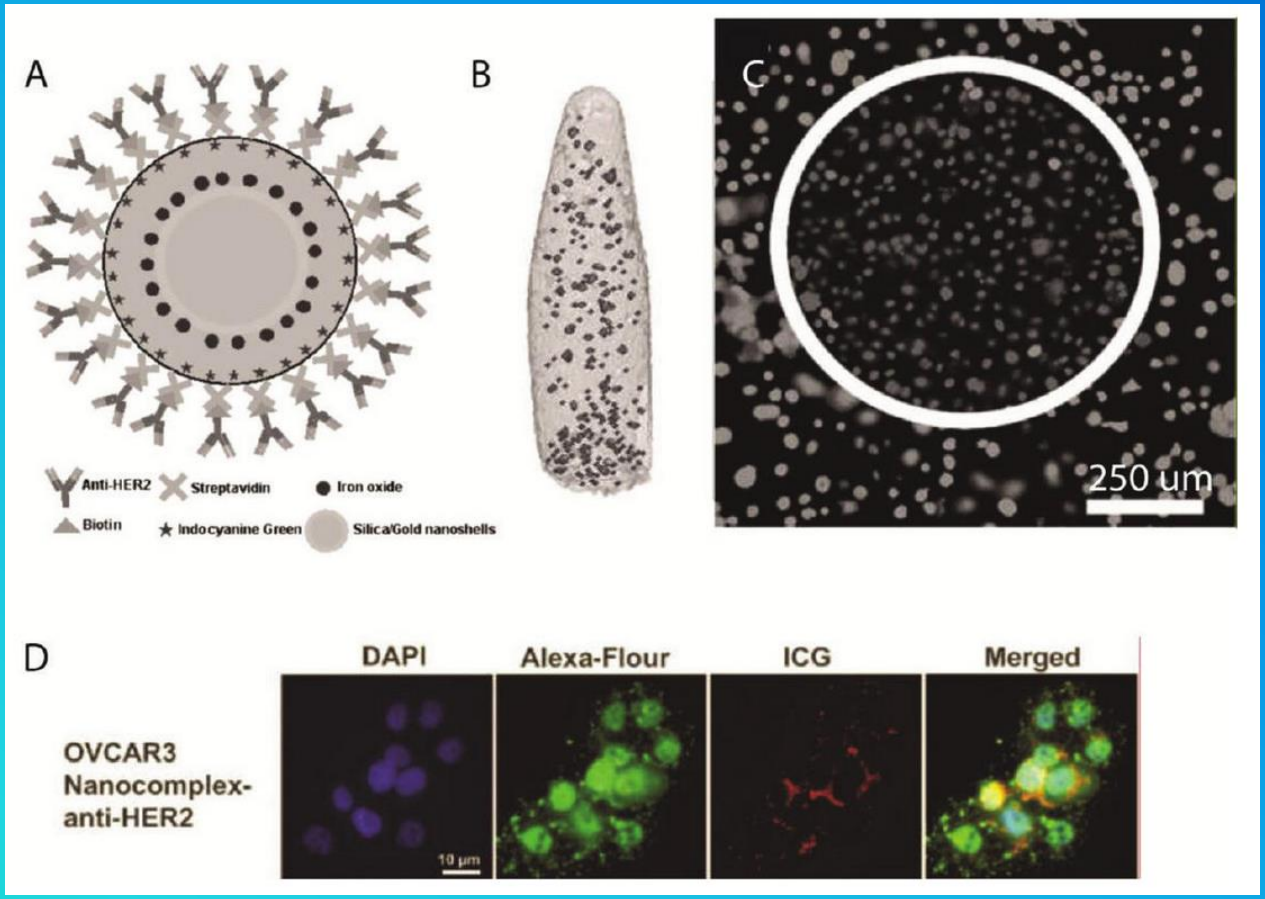
Projections and reconstruction in FMT imaging, by a phantom. (A) Snapshots of raw data during image acquisition. (B) Representation of all acquired projections. Both fluorescence and excitation measurements are shown. Each image in the fluorescence panel corresponds to the equivalent image in the excitation panel. (C) Volume reconstruction of the data.

In vivo combined FMT and MRI



Cathepsin B expression levels are detected by an activatable probe in 9L gliosarcomas stereotactically implanted into unilateral brain hemispheres of nude mice. (A) and (B) Axial and sagittal MR slices of an animal implanted with a tumour, which is shown in green after gadolinium enhancement. (C), (D) and (F) Consecutive FMT slices

Future Perspectives



Novel theranostic agent. A) nanoparticles containing 70 nm Au nanoshell with a silica shell doped with superparamagnetic iron oxide and ICG and surface-decorated with anti-HER2 antibodies for targeting.

B) magnetic resonance imaging of the nanoparticles in vitro

C) photothermal ablation capabilities of the theranostic system in vitro.

D) fluorescence visualization after targeted uptake into OVCAR3 cells.

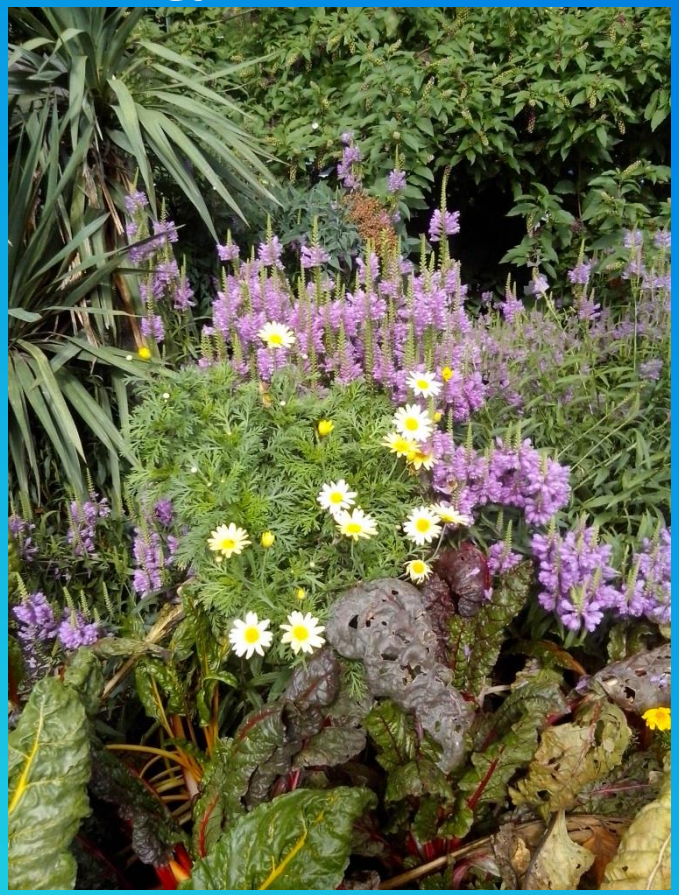
Conclusions

- combining increasingly bright and stable NIR fluorophores
- effective delivery systems enhancing the fluorescence
- prolongation of circulation time and photochemical stability

- powerful tools for fundamental biological studies as well as diagnostic applications
- addition of drug - theranostic agents
- complete solution for diagnostics and disease therapy

Acknowledgements

All the members of Laboratory of Metalomics and Nanotechnology



CYTORES GA CR P301/10/0356, NANIMEL GA CR 102/08/1546, NANOLABSYS CZ.1.07/2.3/.00/20.148, CEITEC CZ.1.05/1.1.00/02.0068

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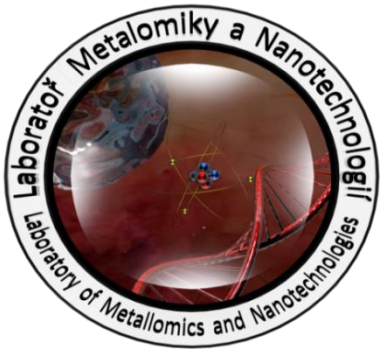

Thank you for your attention

Mendel University in Brno

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