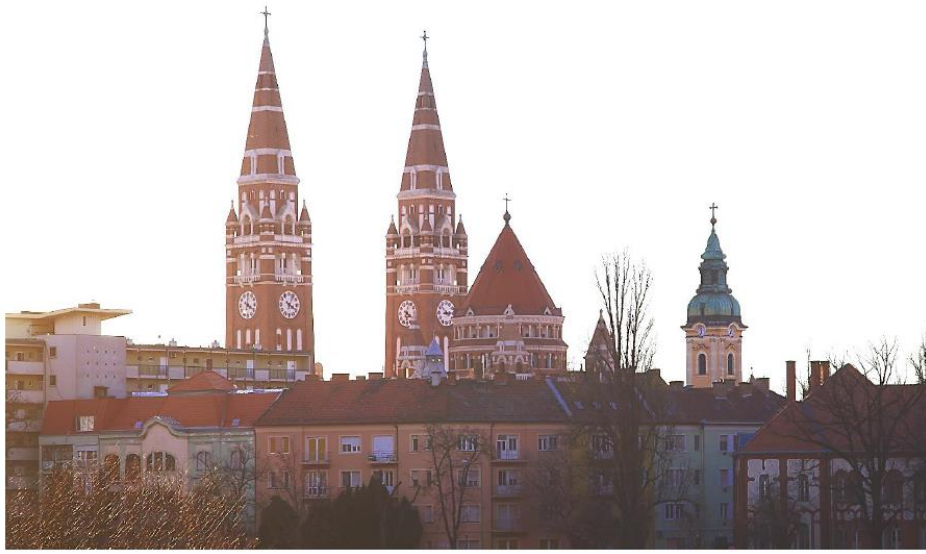




**4th Mini-Symposium on
The Blood-Brain Barrier
from Basic to Clinical Research**

24 March 2022

online meeting on [Zoom platform](#)

Organizers:

Maria A. Deli

Yoichi Morofuji

08:20 ~ 10:10 Plenary session II

Chair: Maria Deli

Keynote talk

08:20 (16:20 JST, 00:20 PDT)

Matthew Campbell (Trinity College, Dublin, Ireland)

Dysregulated CSF1R signalling drives cerebrovascular pathology

08:50 (16:50 JST, 00:50 PDT)

Shinya Dohgu (Fukuoka University, Fukuoka, Japan)

Involvement of P-glycoprotein in α -synuclein aggregate-induced inflammatory mediator release by brain pericytes

09:10 (17:10 JST, 01:10 PDT)

Sumio Ohtsuki (Kumamoto University, Kumamoto, Japan)

Regulation of the blood-brain barrier by membrane proteins

09:30 (17:30 JST, 01:30 PDT)

Andrej Kovac (Institute of Neuroimmunology, Slovak Academy of Sciences, Bratislava, Slovakia)

Analog of kynurenic acid decreases Tau pathology by modulating astrogliosis in rat model for tauopathy

09:50 (17:50 JST, 01:50 PDT)

Szilvia Veszeka (Biological Research Centre, Szeged, Hungary)

Triple ligand targeted nanoparticles cross the blood-brain barrier invitro and enter human midbrain organoids

10:10 ~ 10:20 Break

Plenary session II-5

Triple ligand targeted nanoparticles cross the blood-brain barrier *in vitro* and enter human midbrain organoids

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Nanoparticles hold great promise for drug delivery across the blood-brain barrier, but specific targeting is needed for the successful brain delivery of NPs, which is currently unsolved. There is a specific pattern of highly expressed nutrient transporters including solute carriers (SLC) on the surface of brain endothelial cells. Our hypothesis is that an optimal combination of transporter ligands may increase specifically the brain penetration of nanoparticles.

In this study, we demonstrated that labeling nanovesicles with the combination of the tripeptide glutathione, the vitamin ascorbic acid and the amino acid leucine increased the cellular uptake of a large protein cargo by more than 3-fold as compared to non-targeted nanoparticles. Moreover, we showed the ability of the triple-targeted nanovesicles to deliver their cargo into midbrain organoids after crossing the blood-brain barrier model. We revealed that the cellular uptake was temperature- and energy-dependent and partially mediated by endocytosis based on metabolic and endocytotic inhibition. The uptake of the cargo encapsulated in triple targeted nanoparticles increased after modification of the negative zeta potential of endothelial cells by treatment with a cationic lipid or after cleaving the glycocalyx with neuraminidase. We demonstrated that triple targeted nanoparticles elevated plasma membrane fluidity, indicating the fusion of nanovesicles with endothelial cell membranes. Our data indicate that labeling nanoparticles with three different ligands of multiple transporters of brain endothelial cells can enhance the transfer and delivery of molecules across the blood-brain barrier.

Keywords: blood-brain barrier; brain endothelial cell; drug targeting; nanoparticle; triple targeting ligand; solute carrier; glutathione; leucine; ascorbic acid; brain organoid