氏名（本籍）	Arnela Mujagic
学位の種類	博士（ 医学 ）
学位記番号	博甲第　9962　号
学位授与年月	令和　3　年　3　月　25　日
学位授与の要件	学位規則第４条第１項該当
審査研究科	人間総合科学研究科
学位論文題目	Antioxidant Nanomedicine with Cytoplasmic Distribution in Neuronal Cells Shows Superior Neurovascular Protection Properties for Ischemic Stroke in Mice (神経細胞の細胞質に分布する抗酸化剤ナノメディシンはマウスの虚血性脳卒中において優れた神経血管保護特性を示す)

主　査	筑波大学教授	医学博士	井上　貴昭
副　査	筑波大学准教授	博士（医学）	石津　智子
副　査	筑波大学講師	博士（医学）	冨所　康志
副　査	筑波大学助教	博士（理学）	塩見　健輔

論文の内容の要旨
Abstract of thesis

In this doctoral dissertation, Arnela Mujagic describes the evaluation of an anti-oxidant nanomedicine (RNPs; nitroxide radical-containing nanoparticles) and examines the ability of RNPs to prevent neurovascular unit impairment caused by reactive oxygen species (ROS) after cerebral ischemia-reperfusion.

The summary is as follows:

Purpose: Development of the new brain protective drug RNPs and elucidation of its neuroprotective mechanism.

Methods: The author underwent transient middle cerebral artery occlusion (tMCAO) with C57BL/6J mice. After 60 min of occlusion and 20 min of reperfusion, the author randomly divided mice and injected intra-arterial of RNPs 9 mg/kg; edaravone 3mg/kg. Also, the author injected phosphate-buffered saline PBS as control group. Survival rate and neurological deficit were evaluated 24 h after the injection of the medicines. The author evaluated distribution of RNPs using immunofluorescence staining, and evaluated BBB disruption using the assay of Evans blue extravasation and FITC dextran 4kD permeability assay. Then the author set up the immunofluorescence staining and calculated total number of M1 and M2 cells to evaluate the effect of RNPs and edaravone on the microglia polarization into neurotoxic microglia M1 and neuroprotective microglia M2. The author used MULTIS method, and evaluated ROS scavenging capacity in brain homogenates of RNP and edaravone treated animals. Also, the author measured scavenging effects of RNPs and edaravone on mitochondria generated superoxide radical.

Results: The author showed RNPs significantly improved survival rate and neurological deficit 24h after the injection, significantly protected BBB from disruption and supported polarization of microglia into neuroprotective M2 microglia compared to edaravone. Also, the author showed that RNPs was detected by immunofluorescence analysis and localized on the endothelial cells, perivascular space, cytoplasm of neuronal cells, astrocytes and microglia. Finally, the author showed ROS scavenging capacities were significantly higher in RNPs treated group compared to edaravone treated group.

Conclusion: The author concluded that improving survival rate and neurological deficit, protecting BBB from disruption and improving scavenging capacities of ROS, RNPs showed that it could be a future anti-oxidant and neuroprotective nanomedicine for cerebral ischemia-reperfusion injury after mechanical thrombectomy in acute ischemic stroke.

審査の結果の要旨
Abstract of assessment result (Note: about 150 words)

（批評　General Comments）

This study shows the effect of nitroxide radical-conjugating nanoparticles, RNPs as an antioxidant nanomedicine with cytoplasmic distribution in neuronal cells for the ischemia-reperfusion brain injury model of mouse with survivability, neurological function, and immune staining histology. The authors clearly demonstrate the effect of RNP comparing Edaravone commercially available in the clinical setting of stroke. In addition, the results of this study have the potential to be clinically applied as a new drug for cerebral ischemia, and the author has a large project to develop for this big goal, and great development is expected in the future.

（最終試験の結果　Assessment）

The final examination committee conducted a meeting as a final examination on Dec, 22, 2020. The applicant provided an overview of dissertation, addressed questions and comments raised during Q&A session. All of the committee members reached a final decision that the applicant has passed the final examination.

（結論　Conclusion）

The final examination committee approved that the applicant is qualified to be awarded Doctor of Philosophy in Medical Sciences.