One of the therapeutics for acute cerebral ischemia is tissue plasminogen activator (t-PA). Using t-PA after 3h time window increases the chances of hemorrhage, involving multiple mechanisms. For most ischemic stroke patients edaravone is used as a neuroprotectant in Japan.

For in vivo study, normal Wistar rats intracerebrally received vehicle, plasmin or t-PA, followed by intravenous edaravone or vehicle. At 6h after the administration rat brains were perfused hematoxylin and eosin staining, and immunostainings for oxidative stress markers (4-HNE, HEL, 8-OHdG, AOE) and neurovascular unit markers (NAGO, occludin, collagen IV, MMP-9, GFAP) were performed. In in vitro study, after an administration of vehicle, plasmin, t-PA or edaravone from upper or lower insert, Na-F (natrium-fluorescein) assay, NAGO, occludin, claudin 5 and GFAP immunostainings were performed.

Plasmin and t-PA damaged rat brain with most prominent injury in the t-PA group on 4-HNE, HEL, and 8-OHdG immunostainings. Such brain damages were strongly decreased in t-PA plus edaravone group. For the neurovascular unit immunostainings, a total area of occludin and collagen IV stainings were decreased in single plasmin or t-PA group, which greatly recovered in t-PA plus edaravone group. In contrast, MMP staining was the strongest in t-PA group, less in plasmin, and was the least prominent in t-PA plus edaravone group. In vitro data showed a strong damage to tight junctions for occludin and claudin 5 in both administration groups, while there were no changes for endothelial (NAGO) and perivascular (GFAP) stainings. Such damage to tight junctions was also recovered in t-PA plus edaravone group with similar recovery in Na-F permeability assay.

Administration of t-PA caused oxidative stress damage to lipids, proteins and DNA, and led to disruption of outer parts of neurovascular unit, greater than the effect in plasmin administration. Additive edaravone ameliorated such an oxidative damage by t-PA with protecting outer layers of BBB (in vivo) and tight junctions (in vitro).

Summary

t-PA is a potentially effective agent for acute ischemic stroke. Administration of t-PA causes oxidative stress damage to lipids, proteins and DNA, and leads to disruption of outer parts of the neurovascular unit, greater than the effect of plasmin administration. Additive edaravone ameliorates such oxidative damage by t-PA with protecting outer layers of BBB (in vivo) and tight junctions (in vitro).