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Hypoglycemia causes widespread white matter injury – an imaging study in transgenic mice.

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Insulin-induced hypoglycemia presents the most important limitation to effective treatment for diabetes. Acute severe hypoglycemia may cause transient or permanent brain dysfunction such as confusion, cognitive impairment, seizures and coma. Deprivation of cellular glucose also contributes to the pathophysiology of ischemic brain injury. Recent neuroimaging and pathological studies of patients with severe hypoglycemic episodes suggest that white matter is also vulnerable to hypoglycemia. Although hypoglycemic brain injury is well documented in gray matter, little is known of mechanisms of injury in white matter deprived of glucose. In this study, we investigated the time course in vivo of axon and oligodendrocyte injury in a model of cerebral white matter injury in acute brain slices from adult transgenic mice.

Acute coronal brain slices (400μm) including corpus callosum were prepared from transgenic mice with neuron specific expression of YFP controlled by the Thy1 promoter or from oligodendrocyte-specific expression of GFP (PLP-EGFP) controlled by a proteolipid protein promoter. Perfused brain slices were visualized under confocal microscopy and which permitted high resolution time-lapse fluorescence imaging of intact axons and oligodendrocytes in white matter slices with minimal photodamage.

Transient glucose deprivation (45 min) caused delayed structural disruption of YFP-labeled axons, which appeared as beading, fragmentation, and loss of fluorescence intensity 30-60 min after restoration of glucose levels. Application of the AMPA/kainate antagonist, 30μM NBQX, reduced axonal injury even if started immediately following glucose deprivation. Confocal microscopy also allowed visualization of structural changes of oligodendrocytes in slices from PLP-transgenic mice.

Brain slices permit direct access to white matter cellular components within an intact three-dimensional relationship and are therefore ideal to examine white matter injury without regard to alterations in cerebral vasculature or blood flow, which are known to be effected by diabetes. Thy1-F labeling of axons in white matter proved as a very sensitive marker for visualization of axonal injury *in vivo*. These results indicate that glucose deprivation causes delayed structural disruption in axons, mediated in part by activation of AMPA/kainate glutamate receptors. Transgenic expression of fluorescent proteins allows direct observation of cell-specific structural changes in living tissue.