Acute and sustained excitoxicity differentially influence riluzole's neuroprotective effect

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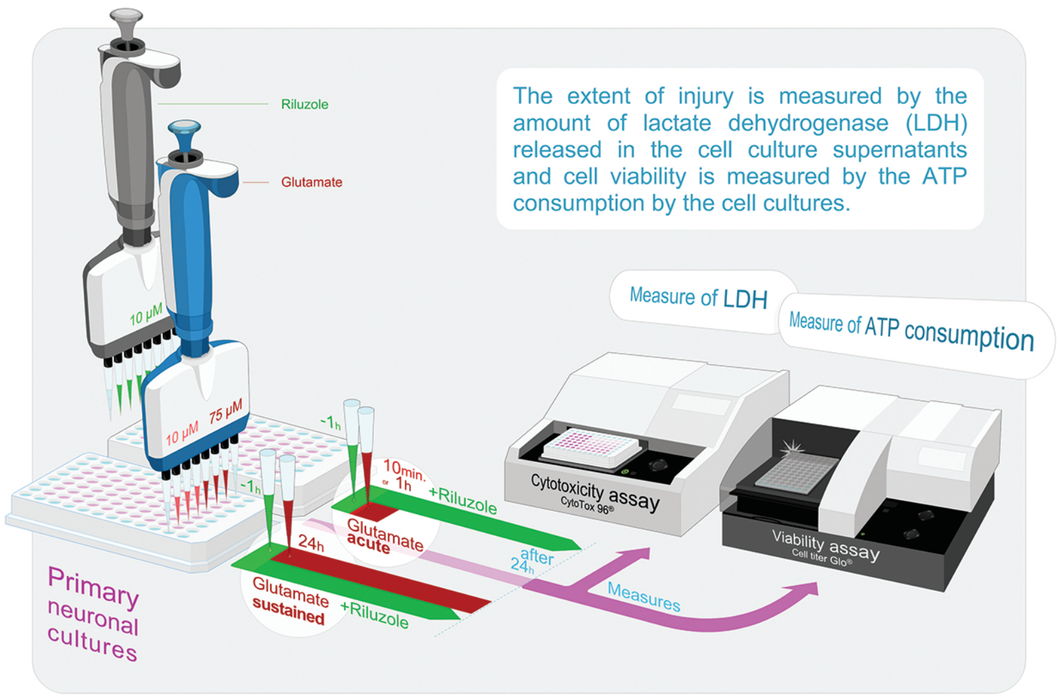
Introduction

Excitotoxicity is initially defined as an acute insult to neurons that leads to their death by excessive activation of glutamate receptors. Acute and massive glutamate release is thought to occur and play a role in various severe insults including cerebral ischemia, traumatic brain injury, hypoglycemia, and epilepsy. Amyotrophic lateral sclerosis (ALS) is a chronic neurodegenerative disease where implication of excitotoxicity has been reported. In contrast to the acute excitotoxicity, more chronic and sustained exposure to milder elevations of glutamate is believed to mediate excitotoxicity in neurodegenerative disease such as ALS.

Objective

The present study is to investigate the impact of acute (10 - 60 min) and sustained (24 h) exposure to low (10 µM) and high (75 µM) concentration of glutamate on the injury and viability of primary neuronal cultures. The neuroprotective effect of riluzole (the unique FDA-approved ALS drug) is comparatively assessed under the two glutamate exposure conditions.

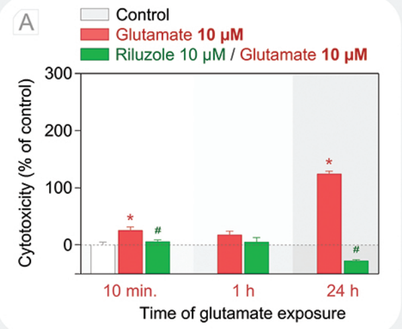
Material and Methods

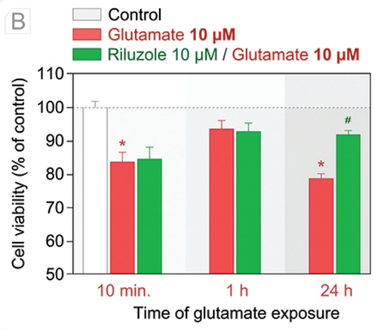


The extent of injury is measured by the amount of lactate dehydrogenase (LDH) released in the cell culture supernatants and cell viability is measured by the ATP consumption by the cell cultures.

Results

Figure 1 : Impact of **acute** and **sustained exposure** to **low concentration of glutamate** on cell injury and cell viability
Assessment of the neuroprotective effect of **riluzole**





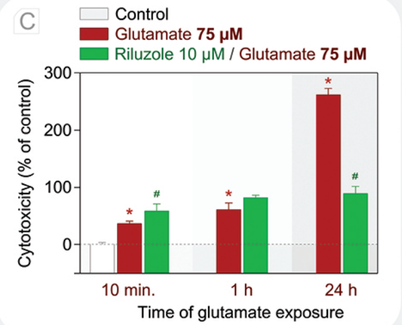
A Cytotoxicity

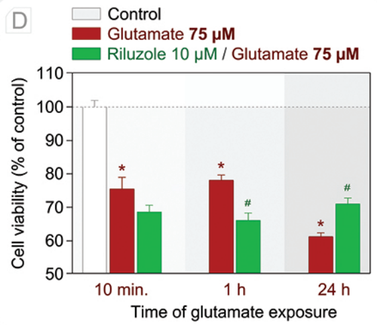
Sustained exposure with low concentration of glutamate produces 4 fold increase of cytotoxicity as compared to acute exposure.
Riluzole significantly reduces the cytotoxicity of acute exposure but it fully prevents the insult under sustained glutamate exposure.

B Cell viability

Acute and sustained exposure with low concentration of glutamate produces comparable reduction of neuronal viability. Whilst riluzole is ineffective against the insult induced by acute exposure, it markedly protects cell viability from sustained glutamate insult.

Figure 2 : Impact of **acute** and **sustained exposure** to **high concentration of glutamate** on cell injury and cell viability
Assessment of the neuroprotective effect of **riluzole**





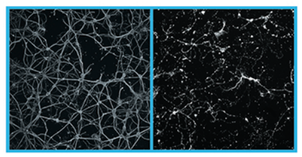
C Cytotoxicity

Sustained exposure with high concentration of glutamate produces 4-7 fold increase of cytotoxicity compared to acute exposure.
Whilst Riluzole is ineffective against the insult induced by the acute exposure, it markedly reduces the cytotoxicity of sustained glutamate exposure to a level comparable to that induced by the acute condition.

D Cell viability

Sustained exposure with high concentration of glutamate is associated with an enhanced reduction (about 50%) of cell viability as compared to the acute exposure condition.
Whilst riluzole is ineffective against the insult during the acute exposure, it markedly protects cell viability from sustained glutamate insult. Riluzole under sustained exposure significantly improves cell viability.

Key points



The mechanism of neuronal damage induced by acute exposure to glutamate is largely insensitive to riluzole.

In contrast, the neuronal damage induced by sustained exposure to low concentration of glutamate is largely riluzole-sensitive mechanism. This observation is in accordance with the argument in favor of the chronic slow excitotoxicity hypothesis in ALS.

Neuronal damage induced by exposure to high concentration of glutamate is mediated by mechanisms sensitive and insensitive to riluzole treatment.