

## Viability of primary hippocampal neurons cultured from different knockouts of tumor necrosis factor receptor subtypes

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### Abstract

Tumor necrosis factor (TNF)- $\alpha$  is a major component of the inflammatory process responsible for neuronal cytotoxicity in Alzheimer's disease (AD); however, the specific mechanisms of TNF- $\alpha$ -induced neurotoxicity and neuroprotection are still unclear. TNF- $\alpha$  signaling through TNF receptor (TNFR) type 1 (TNFR1) and TNFR type 2 (TNFR2) has been reported to be involved in amyloid- $\beta$  peptide (A $\beta$ )- and glutamate-induced neurotoxicity. In the present study, we aimed to evaluate whether the TNFR subtypes differentially contribute to neuronal death induced by soluble A $\beta$ <sub>1-42</sub> oligomers and L-glutamate using primary cultures of hippocampal neurons from TNFR1 knockout (TNFR1  $-/-$ ), TNFR2 knockout (TNFR2  $-/-$ ), and wild-type mice. Morphological evaluation of neurons under a phase-contrast microscope and an assay for LDH release from neurons revealed that, with exposure to A $\beta$ <sub>1-42</sub> oligomers or L-glutamate, primary neurons from the TNFR1  $-/-$  mouse hippocampus grew more healthily than those from the wild-type mouse hippocampus, and primary neurons from the TNFR2  $-/-$  mouse hippocampus grew less healthily than those from the wild-type mouse hippocampus. Our present results suggest that TNFRs have some relationship with the processes of neuronal death induced by A $\beta$ <sub>1-42</sub> oligomers and L-glutamate, and that the receptor subtypes differentially contribute to the processes; that is, A $\beta$ - and glutamate-induced signaling pathways are thought to cooperate with the signaling pathways activated by binding of TNF- $\alpha$  to TNFR1 to promote neuron death, whereas the signaling pathways mediated by TNFR2 counteract the A $\beta$ - and glutamate-induced neurotoxicity. Two types of TNFRs are therefore potential targets for treating A $\beta$ - and glutamate-induced AD pathologies. Tottori J. Clin. Res. 6(1), 49-59, 2014

### Key Words

Tumor necrosis factor (TNF)- $\alpha$ , TNF receptors (TNFRs), neuron cultures, amyloid- $\beta$  peptide (A $\beta$ ), lactate dehydrogenase (LDH) release assay, glutamate neurotoxicity

### Introduction

TNF- $\alpha$  is a major component of the inflammatory process responsible for neuronal cytotoxicity, dysfunction and protection in a wide range of neurodegenerative disorders such as AD; however, the specific mechanisms underlying this

process are still unclear<sup>1, 2)</sup>. TNF- $\alpha$  is a pleiotropic pro-inflammatory cytokine that exerts multiple biological effects<sup>3)</sup>. The diverse regulatory functions of TNF- $\alpha$  are explained by the fact that it can bind to two structurally distinct membrane receptors expressed on many types of cells<sup>1, 4-6)</sup>. In