

Neurotrophin Signalling Protects CLL Cells from Death

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Abstract

Neurotrophin signaling can protect B lymphocytes from death. We show that use of neurotrophic pathway inhibitors induces non-apoptotic RIP1-dependent death of the Mec1 chronic lymphocytic leukemic (CLL) cell line.

1. Introduction

Chronic lymphocytic leukemia (CLL) is the most common leukemia in the Western world, It is a fatal malignancy resulting from dysregulated B cell death [1].

Nerve growth factor (NGF), a neurotrophin, can rescue B lymphocytes from apoptosis [2]. Neurotrophins have has two receptors - p75^{NTR} and Trk receptors, which can direct cells towards survival or apoptosis, in a cell-type specific manner [3].

Here we investigate the effect of neurotrophin inhibitors on Mec1 CLL cell survival.

2. Results

2.1. NGF signalling inhibition leads to decreased cell viability

On treatment with Ro 08-2750 (which binds NGF, inhibiting NGF-receptor binding), Tat-pep5 (a chemical inhibitor of p75) (Fig. 1) or K252a (a pan-Trk phosphorylation inhibitor; data not shown) for 48 h, Mec1 cells showed a decrease in viability.

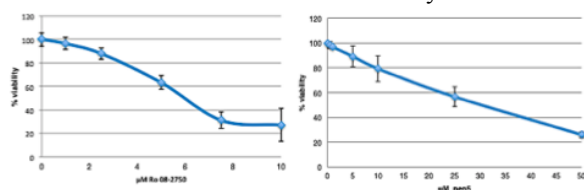


Figure 1. NGF signaling inhibition leads to a decrease in cell viability of Mec1 cells. Inhibition of NGF or p75^{NTR} for 48 h. Viability was analyzed by MTT assay.

2.2. NGF signalling inhibition induces cell death, but not apoptosis

On treatment with Ro 08-2750, Mec1 cells had a variety of cellular morphologies (data not shown), indicating induction of numerous modes of cell death. Apoptosis induction was investigated by Western blotting and caspase activity assays (Fig. 2). On use of the RIP1

inhibitor nec1-S, cell death was reduced (data not shown).

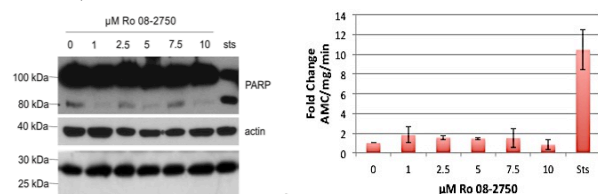


Figure 2. NGF inhibition of Mec1 cells does not induce apoptosis. Western blotting and DEVDase activity assay for caspase induction on 48 h NGF inhibition.

3. Discussion

Inhibition of NGF signalling induces cell death in Mec1 cells, on inhibition of NGF, p75^{NTR} and Trk. All three inhibitors resulting in cell death suggest that both receptors are involved in Mec1 cell survival. Lack of caspase activation and various cellular morphologies suggest the inhibitors are inducing non-apoptotic cell death, supporting the evidence that CLL cells have dysregulated apoptosis. Reduced cell death with RIP1 inhibition suggests that necroptosis may be the mode of death induced.

Future work aims to confirm the induction of necroptosis on treatment with these inhibitors, and confirm these findings in primary CLL cells.

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5. References

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