p50-deficient mice (Figure 2 and Figure 3). Even though RGC-5 cells were stimulated by glutamate, the pre-treatment with tacrolimus reduced activated caspase 3 and Bax expression in RGC-5 cells. The amount of Bax in RGCs and TUNEL-positive RGCs of p50-deficient mice was markedly decreased by the chronic administration of tacrolimus (Figure 3C and Figure 4D). These research findings suggest that a CaN signal cascade involving impaired NF-Bactivation induces spontaneous RGC death (Figure 5).

Several studies have shown the potential role of the immune system, i.e., autoantibodies against ocular antigens, in the pathogenesis of glaucoma [2]. The p50-deficient mice exhibited multiple immune response defects [7]. Our previous data demonstrated the possible production of autoantibodies in the retinas of p50-deficient mice [25]. Tacrolimus is an immunosuppressive reagent whose main use is after allogenic organ transplant to reduce the activity of the patient's immune system, and so lower the risk of organ rejection. Therefore, another possibility is that tacrolimus inhibits the immune system, especially autoimmunity. To clarify the mechanism by which tacrolimus promotes RGC survival, further studies must be performed.

One important issue that remains to be resolved in neuronal physiology is how a signal that is released by neurotransmitters binding to their receptors at the synapse, and is transduced into the nucleus to activate gene transcription. Despite the importance of NF-κB to neuronal function and survival, little is known about the signaling pathways leading to its activation. In this study, we demonstrated that p50 expression is required for optic nerve cell survival, suggesting a connection between the loss of p50 expression and optic neuropathy. The mechanism whereby CaN-dependent pathways activate NF-κB is still unknown; we have chosen to utilize p50-deficient mice as well as RGC-5 cells to investigate the mechanisms by which the CaN signal cascade stimulates NF-κB-mediated neuronal cell death. The factors that act on the CaN-dependent pathway might become targets in the clinical treatment of NTG. Clinical studies have shown that there is a correlation between NTG and migraine with aura (MA): significant evidence of alinkage was found between MA phenotype and the marker D4S1647 on 4q24, where the NF-κBp50-coding gene is located [44]. Unfortunately, scientific evidence which directly shows a correlation between defective p50 expression and NTG has yet to be obtained.

In conclusion, tacrolimus protects RGCs from chronic spontaneous optic neuropathy, and then a CaN signal cascade including NF-κB activation induced chronic spontaneous optic neuropathy. Our results provide a new framework for understanding how cross-talk integrates a variety of signals in a physiologic context for the initiation of chronic optic neuropathy. These findings provide strong additional information regarding therapeutic targets for NTG.

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Appendix 1. Supplemental Information.

To access the data, click or select the words "Appendix 1." This will initiate the download of a compressed (pdf) archive that contains the file.

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