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Neurodegeneration in Glaucoma: Progression and Calcium-Dependent Intracellular Mechanisms

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Abstract

Glaucoma is an age-related optic neuropathy involving sensitivity to ocular pressure. The disease is now seen increasingly as one of the central nervous system, as powerful new approaches highlight an increasing number of similarities with other age-related neurodegenerations such as Alzheimer's and Parkinson's. While the etiologies of these diseases are diverse, they involve many important common elements including compartmentalized programs of degeneration targeting axons, dendrites and finally cell bodies. Most age-related degenerations display early functional deficits that precede actual loss of neuronal substrate. These are linked to several specific neurochemical cascades that can be linked back to dysregulation of Ca^{2+}-dependent processes. We are now in the midst of identifying similar cascades in glaucoma. Here we review recent evidence on the pathological progression of neurodegeneration in glaucoma and some of the Ca^{2+}-dependent mechanisms that could underlie these changes. These mechanisms present clear implications for efforts to develop interventions targeting neuronal loss directly and make glaucoma an attractive model for both interrogating and informing other neurodegenerative diseases.

Glaucoma is a Neurodegenerative Disease

Disease Etiology: Age and Ocular Pressure

Optic neuropathies are the most common source of age-related loss of sensory activity in the central nervous system (CNS). Of these, glaucoma is by far the worst. This disease is the leading cause of irreversible (i.e., neurodegenerative) blindness worldwide. Age is the greatest risk factor: the likelihood of developing glaucoma increases nearly *7-fold* after 55 years. Thus, with the aging population, nearly 80 million people worldwide will be afflicted by 2020 (Quigley and Broman, 2006). The disease is also the number one cause of blindness (both irreversible and reversible) in African-Americans, occurring on average 10 years earlier than in the Caucasian population and at an incidence that is *4 to 5-fold* higher (Tielsch et al., 1991).

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Traditionally, glaucoma has been considered an “eye disease”. This is because elevated intraocular pressure (IOP) is the leading *modifiable* risk factor for the disease (Sommer, 1989; Gordon et al., 2002). Thus, animal models of the disease generally incorporate induced elevations or exploit naturally-occurring elevations (Pang et al., 2007; Sappington et al., 2010). However, glaucomatous pathology can also be induced by eliciting an auto-immune response (Wax et al., 2008). Lowering IOP is the most common treatment by far, generally through topical application of pharmaceuticals and/or surgery (Heijl et al., 2002).

The relationship between elevated IOP and vision loss is not straightforward as normal pressure glaucoma, or glaucomatous pathology without elevated IOP, represents about 50% of glaucoma diagnoses (Shields, 2008). Ocular hypertension, or chronically elevated IOP without nerve damage, is more common than glaucoma itself (reviewed in Heijl et al., 2002). Even more enigmatic from a neurobiological standpoint, results from the Early Manifest Glaucoma Trial indicate that lowering IOP for those with nominally normal pressure can be an effective preventive treatment for reducing the risk of the age-related neuropathy associated with glaucoma (Heijl et al., 2002), though it is not clear what fraction of large number of patients with normal IOP benefit from IOP-lowering (Anderson et al., 2003). Thus, IOP represents the primary modifiable risk factor whatever its magnitude. The Baltimore Eye Study concluded that the distinction between “low-tension” and “high-tension” glaucoma is artificial (Sommer et al., 1991). Even so, some estimates indicate that optic nerve degeneration may continue in as many as half of glaucoma patients treated with an IOP-lowering regimen (Leske et al., 2003). These observations suggest that we should consider glaucoma not as a disease involving *elevations* in pressure, but as a disease in which neurological sensitivity to *pressure itself*, independent of magnitude, is an additional insult against a back-drop of other age-related stressors in the system (Sappington et al., 2009).

The Neurobiological Roots of Vision Loss

Continued progression of visual loss in glaucoma even with lowered IOP has compelled the search for interventions that treat neurodegeneration directly (McKinnon et al., 2008). Through mechanisms that are not well understood, glaucoma selectively targets the 1.5 million retinal ganglion cell (RGC) neurons whose axons comprise the optic nerve. Because RGCs are the output neurons of the eye, their selective loss in glaucoma has reinforced the “eye disease” viewpoint. However, this perception has in fact created lost opportunities. Most of the retinal ganglion cell lies outside of the eye, and it is here that recent advances in understanding the progression of glaucomatous pathology have focused.

Since the retina and optic projection are part of the CNS, it is not surprising that glaucoma shares epidemiological and mechanistic similarities with other CNS neurodegenerations, including Alzheimer’s, Parkinson’s, amyotrophic lateral sclerosis (ALS), and Huntington’s disease. While the etiologies of these diseases are diverse, their progression involves many important common elements that are themselves potential targets for therapeutic interventions. Our recent work and that of other groups demonstrates that neurodegeneration in glaucoma shares these common components and that they hold promise as therapeutic targets (McKinnon, 2003; Steele et al., 2006; Buckingham et al., 2008; Kong et al., 2009; Crish et al., 2010). Thus, animal models of glaucoma are seen increasingly as emergent tools with which to inform studies of other age-related CNS neurodegenerations (Whitmore et al., 2005). In particular, models are useful for helping us understand (1) how aging biases the system towards irreversible loss of function, and (2) how deprivation of this sensory input affects a major portion of the aging brain.

Progression of Neurodegeneration

The Endgame First: Apoptosis in the Retina

Whatever the early events involved in RGC degeneration in glaucoma are, they ultimately result in down-stream caspase-dependent, mitochondrial-mediated apoptosis (Kerrigan et al., 1997; Quigley, 1999; Tatton et al., 2001; McKinnon et al., 2002; Tahzib et al., 2004; Waldmeier and Tatton, 2004; Huang et al., 2005a,b; Qu et al., 2010). Ample evidence supports the typical characterization of glaucoma as a disease inducing the eventual apoptotic loss of RGC somas in the retina. For example, histological examination of the RGC layer of the retina clearly demonstrates thinning of the cell body population; this is so for both human tissue and animal models (Kerrigan-Baumrind et al., 2000; John et al., 1998). Thus, as in other CNS conditions (see Rohn, 2010), investigations exploring the progression of glaucomatous neurodegeneration have focused on apoptotic elimination of RGCs (Quigley et al., 1995; Kerrigan et al., 1997; Huang et al., 2005a,b). Similarly, one of the most common techniques used historically to assay RGC somatic survival is straightforward tract tracing; applying a neuronal tracer such as fluoro-gold to the lateral geniculate body, superior colliculus or other central projection site and examining its localization in the retina some time later (Mittag et al., 2000; Vidal-Sanz et al., 2001; Danias et al., 2003; Filippopolous et al., 2006). However, these assays reflect better the state of retrograde axonal transport and axon survival. If axons are challenged early, and this seems to be case, then lack of retrograde-labeled RGCs does not necessarily indicate they are absent. For example, in the DBA2J mouse model of pigmentary glaucoma, RGC somas labeled by neuronal markers are seen to persist long after retrograde transport is depleted (Buckingham et al., 2008).

However, there is a growing movement away from viewing apoptosis as the cause of clinical presentation of disease (Gould et al., 2006; Jelinger, 2006; Brady and Morfini, 2010). Empirical support for compartmentalized degeneration of neuronal processes, including deficits in axonal transport and physiological dysfunction, is changing the way we view neurodegenerative disorders. This is so for glaucoma as well (Figure 1). For example, deletion of the pro-apoptotic gene BAX in the DBA/2J mouse model of glaucoma has a protective effect on the cell body but does not prevent RGC axon loss (Libby et al., 2005). Evidence supporting the idea that neuronal processes are affected separately from cell bodies and actually precede cell body loss has been steadily accumulating (Whitmore et al., 2005; Libby et al., 2005; Jakobs et al. 2005; Schlamp et al., 2006; Jakobs et al., 2006; Stevens et al., 2007; Howell et al., 2007; Buckingham et al., 2008; Soto et al., 2008, Fu et al., 2009; Crish et al., 2010; Baltan et al., 2010). Furthermore, structure in the optic projection persists after axonal transport is depleted (Buckingham et al., 2008; Crish et al., 2010). These and similar findings raise the question of if not cell death, what is blinding in glaucoma?

Early Deficits in Axonal transport

Functional deficits occur early in the progression of glaucoma, an intriguing finding that may change not only how glaucoma is viewed, but eventually how it is treated. The idea of deficits in axonal transport in glaucoma is not new, first appearing in the 1970's (Anderson and Hendrickson, 1974; Mincker et al., 1976; 1977). Since then, focus has been on the optic nerve head (ONH) as the site of impaired transport through obstruction of normal axoplasmic flow due to effectors such as mechanical blockade (Quigley and Addicks, 1980; Quigley et al., 1981; Hollander et al., 1995; Burgoyne et al., 2005). ONH blockade has figured prominently in a common view of glaucoma by which retrograde transport of pro-survival factors (most notably BDNF) from the RGC synaptic terminal in the brain to the cell body is challenged, thereby triggering apoptosis (Pease et al., 2000; Quigley et al.,

2000). This view is appealing, since RGC axons are naturally most vulnerable as they pass unmyelinated through the lamina cribrosa plates on their way to the optic nerve (reviewed in Whitmore et al., 2005). The nerve head is an early site for dramatic changes in glial reactivity (Son et al., 2010). Reactive glia in the nerve head are thought to induce compositional changes in the extracellular matrix of the lamina (Fuchshofer et al., 2005; Guo et al., 2005). This process may involve the release of inflammatory cytokines from astrocytes and microglia (Hernandez, 2000; Yuan and Neufeld, 2000; Tezel et al., 2001) and the death of oligodendrocytes (Nakazawa et al., 2006).

Vision loss in glaucoma develops in a well-defined manner, progressing from a paramacular lesion in the temporal field along the arcuate pattern formed by RGC axons and the retinal vasculature (Levin, 2001; Goldblum and Mittag, 2002). It has been argued that this general pattern correlates with anatomical features of the ONH, suggesting that glaucomatous field defects are primarily axogenic in origin, moving retrograde from the nerve head to the retina (Quigley, 1999). Several lines of evidence suggest that a component of glaucoma involves ischemic reperfusion injury at the ONH due to compromised vascular circulation (reviewed in Osborne et al., 2004). It is widely held that these processes represent the primary initiators of RGC death and are among the earliest events associated with RGC loss (Quigley, 1999; Schlamp et al., 2006; Mabuchi et al., 2004; see discussion in Pang et al., 2005).

In other neurodegenerative diseases, however, deficits in axonal transport also precede, and most likely cause, eventual axonal and somatic degeneration (Stokin et al., 2005; Coleman et al., 2005; Morfini et al., 2009). In these disorders active intra-axonal mechanisms downstream from the stressors are promoted as causative agents in defective transport. These include metabolic abnormalities or changes to the molecular motors and cytoskeletal structures that underlie axonal transport (Morfini et al., 2009) resulting in deficits occurring at locations far removed from the site of the stressor (Conforti et al., 2007; Cuchillo-Ibanez et al., 2008). Similar changes occur in the glaucomatous nerve head (Kashiwagi et al., 2003; Barron et al., 2004; Martin et al., 2006; Band et al., 2009) raising the possibility that these mechanisms can produce transport deficits outside of this region.

In animal models of glaucoma, impaired axonal transport first appears in the distal portion of the RGC projection in the brain, well removed from the ONH (Crish et al., 2010). In addition, anterograde axonal transport (cell body to synaptic terminal) appears to be affected earlier than retrograde transport (Danias et al., 2003; Filippopoulous et al., 2006; Buckingham et al., 2008; Crish et al., 2010). If transport blockade was due to mechanical factors at the nerve head, one would expect both anterograde and retrograde transport to be affected similarly. Instead, intra-axonal, functional and cytoskeletal defects affect anterograde transport disproportionally (Wang et al., 1995; Ross et al., 2006). Differences in the specifics of transport blockade, including location, are not trivial but result in very different patterns and mechanisms of degeneration, with deficits in anterograde transport producing axonopathy distal to the defective site (Coleman et al., 2005). This is not to say the ONH is not a critical site or even *the* critical site for pathogenesis, but rather that whatever ONH processes challenge axonal transport, the insult may be transduced within the axons themselves. We discuss various possibilities for this cascade below.

Neurophysiology

Though RGC cell bodies in the retina and axonal structures in the projection may persist until late in disease progression (Buckingham et al., 2008; Crish et al., 2010), physiological deficits can produce clinical symptoms that mirror degenerative processes. Since the goal of studying mechanisms of degeneration must be to identify targets for ameliorating loss of vision, this subtle aspect cannot be overlooked. The pattern electroretinogram (PERG) is a potential tool to detect early glaucomatous changes, though its use as a diagnostic measure is

actively debated (Johnson et al., 1989; Bach and Hoffman, 2008). PERG has produced intriguing results in animal models, showing dysfunctional RGC activity that precedes outright degeneration (Saleh et al., 2007); this has recently been supported in human glaucoma (North et al., 2010). Interestingly the association between elevated IOP and PERG outcome remains ambiguous. There is evidence of a positive relationship in that lowering IOP improves PERG amplitude (Nagaraju et al., 2007; Porciatti and Nagaraju, 2010), but other studies indicate no relationship between PERG amplitude and IOP elevation (Johnson et al., 1989; Sehi et al., 2010).

While these conflicting results may be due to species or experimental differences, it is quite possible that physiological changes in glaucoma are first reflected extra-retinally, e.g., in the RGC axon or axonal terminals. This view is supported by some direct measurements of nerve function (King et al., 2006; Baltan et al., 2010). Two salient points emerge in viewing these studies as a whole. First, the results support the idea that glaucoma is about sensitivity to IOP itself (as the clinical data suggest), rather than just elevated IOP, and second that physiological changes may not report actual degeneration or but rather earlier dysfunction. If these changes are functional in nature, elements of the neural substrate of the retinal projection still exist – and therefore do not need to be regenerated, at least at the scale of replacing cell bodies or long, convoluted axon segments. In this view, functional measures may serve as sensitive indicators of underlying pathology and as valid targets for intervention. It remains to be seen how attempts to reverse loss of function improve eventual outcome.

Distal Axonopathy

Compartmentalized degeneration in which neuronal processes and cell bodies are differentially affected is taking a major role as a mechanism of progression in CNS disorders (Fischer et al., 2004; Stokin et al., 2005; Coleman et al., 2005). Long or extensive processes present unique metabolic and logistical challenges to neurons, making compartments such as axons more susceptible to stressor-induced damage.

Axon degeneration traditionally has been described as occurring in one of two programs: Wallerian degeneration or dying back (see Coleman, 2005). Although current evidence blurs the distinction between these two programs, a convenient definition frames Wallerian degeneration as a synchronous event along the entire affected axon, while dying back occurs as a progressive distal-to-proximal cascade that begins at the synaptic terminals. Wallerian degeneration typically results from axon trauma. While transection is most often used to study Wallerian degeneration, it can arise from more subtle localized insults - thus its relevance for glaucoma. Insult at the nerve head could certainly result in axon degeneration that precedes somatic loss, causing Wallerian degeneration to occur along the entire post-ONH axon (Salinas-Navarro et al., 2010). However, typical morphological indications of Wallerian degeneration along the entire length such as axonal beading are not present prominently in glaucomatous human nerves or in animal models (Nickells, 2007; Crish et al., 2010). Progressive degenerative diseases exhibit distal axonopathy early followed by dying back (Fischer et al., 2004; Stokin et al., 2005; Coleman et al., 2005). RGC axons in the optic nerve and brain demonstrate distal axonopathy indicative of underlying dying back, even as more proximal axonal processes appear normal (Nickells, 2007; Crish et al., 2010).

Synaptic and Dendritic Remodeling

Of all the neuronal compartments, synapses are the most sensitive to modification – these mechanisms playing the major role in normal plastic changes in the nervous system. Therefore, one could expect synapses to be affected in disease, and this is certainly the case in other degenerative disorders (Spires and Hyman, 2004; Zaja-Milatovic et al., 2005; Zang

et al., 2005; Day et al., 2006; Knobloch and Mansuy, 2008). Glaucoma is no different, with RGC synapse elimination in the retina implicated early in disease progression (Stevens et al., 2007; Fu et al., 2009). Even so, MRI studies indicate that activity in the synaptic layer of the inner retina persists presumably well after synaptic pruning has started (Calkins et al., 2008). In the distal projection, little has been done examining RGC synaptic terminals in the brain. However, recent studies have indicated that RGC terminals are affected later in disease progression, with changes in NMDA physiology and synaptic loss occurring well after other functional pathologies emerge in glaucoma (Georgiou et al., 2010; Crish et al., 2010). It is not know how the dynamics of RGC terminal loss relate to those governing dendritic pruning for RGC-recipient neurons in central targets (Gupta et al., 2007). Another open question is how changes in synaptic terminals and their postsynaptic retinal targets relate to dendritic and synaptic changes in the retina.

While typically not as long as axons, dendritic processes are often very extensive, placing similar sorts of metabolic demands on a cell. Consequentially, early dendritic changes may be expected in glaucoma – and there is evidence to support this case in animal models. Jakobs et al. (2006) demonstrated dendritic field remodeling preceding cell body loss in the DBA/2J mouse model consisting of both loss of arborization and abnormal morphologies in existing processes. These changes appear to parallel the degeneration in the distal axon in the same model (Crish et al., 2010), but a direct comparison has not yet been made. Importantly, dendritic pruning (like axonal degeneration) can be mitigated at least in part by treatment with trophic factors (Weber et al., 2008; Weber et al., 2010).

The Calcium Hypothesis

Ca^{2+} and Neurodegeneration

In healthy neurons, Ca^{2+}-dependent cascades influence a variety of cellular functions, including exocytosis, gene transcription, membrane trafficking and intracellular respiration (Berridge et al., 2000). Normally, the concentration of cytosolic Ca^{2+} is roughly 10,000 times lower than that in the extracellular space (Hernandez-Fonseca and Massieu, 2005). An important, common component of axonopathy across neurodegenerative disorders is increased influx of extracellular Ca^{2+}, which triggers cytoskeletal degradation through enzymatic activity (Coleman, 2005). Excessive levels of neuronal Ca^{2+} lead to breakdown of Ca^{2+} homeostasis and to a series of cytoplasmic processes that promote caspase-dependent neuronal cell death (Bredesen et al., 2006). Importantly, while intra-axonal Ca^{2+} increases prior to degeneration, the source of the influx can be either directly through the axon or through the neuronal cell body and dendrites (Coleman and Perry, 2002). There are many mechanisms that could contribute to increased influx, which are likely to depend on neuronal cell type and the nature of the initiating insult. For example, in Alzheimer's disease, neurons bearing neurofibrillary tangles demonstrate higher levels of free and bound Ca^{2+}, activated Ca^{2+}-dependent proteases, and the Ca^{2+}-activated enzyme transglutaminase (reviewed in Mattson and Chan, 2003). Elevated intracellular Ca^{2+} could arise through direct conductance through amyloid β protein channels (Kawahara and Kuroda, 2000).

Axonal degeneration in glaucoma bears several striking similarities to that in other neurological diseases (Tatton et al., 2003; Whitmore et al., 2005). These include abnormal processing of APP (McKinnon, 2003), dependence on target-derived trophic support (Quigley et al., 2000), and involvement of various Ca^{2+}-mediated cascades (Whitmore et al., 2005). For example, cleavage of calcineurin, a Ca^{2+}-dependent protein phosphatase, occurs in response to elevated IOP, and inhibiting calcineurin systemically inhibits pressure-induced RGC axon loss in the optic nerve (Huang et al., 2005). Relevant to transport deficits and axonopathy is Ca^{2+} activation of calpains. Calpains are a class of Ca^{2+}-dependent proteases that have been implicated in a number of neurodegenerative conditions (Vosler et

al., 2008), including glaucoma (Huang et al., 2008). While Ca^{2+} dysregulation-induced calpain activation can elicit a number of pathological changes in the cell, most relevant here are its activities on the cytoskeleton. Calpains act directly on synaptic components (Lu et al., 2000; Jourdi et al., 2005) and breaks down the structural proteins alpha-II-spectrin and heavy chain neurofilaments (Siman et al., 1984; Chan and Mattson, 1999). Calpains also activate kinases such as cyclin dependent kinase 5 (Lee et al., 2000), extracellular signal regulated kinases (Veeraana et al., 2004), and stress activated protein kinases (Goni-Oliver et al., 2007). The latter serve to phosphorylate neurofilaments and the microtubule associated protein Tau, slowing or blocking axonal transport (Figure 2; Shea et al., 2004, Shea and Chan, 2008). Recent evidence demonstrates that in experimental glaucoma, calpains are activated with cleavage of associated substrates in RGCs (Huang et al., 2010).

Ca^{2+} and Oxidative Stress

With aging, the CNS becomes increasingly susceptible to oxidative damage to DNA and protein (Cakatay et al., 2001). Oxidative stress markers have been implicated in several animal studies involving acutely induced elevations in IOP. These includes higher levels of lipid peroxidation and protein carbonyl content (Moreno et al., 2004; Ko et al., 2005; Tezel et al. 2005; Ferreira et al., 2010). In the DBA2J mouse, ceruloplasmin, an important antioxidant implicated in other neurodegenerative conditions, is strongly upregulated (Steele et al., 2006; Stasi et al., 2007). Ceruloplasmin exerts much of its effect on oxidative stress by converting ferrous (Fe^{2+}) iron into the ferric (Fe^{3+}) form. This reduces the iron-mediated formation of reactive oxygen species (ROS) and allows iron to interact with transferring or storage proteins such as transferrin and ferritin. All three iron-associated proteins act as antioxidants and are elevated in monkeys with chronically elevated IOP as well as in human glaucoma (Farkas et al., 2004; Stasi et al., 2007). Several anti-oxidative markers are also elevated in the aqueous humor of glaucoma patients, including catalase, glutathione peroxidase, superoxide dismutase, and malondialdehyde (Ferreira et al., 2004; Ghanem et al., 2010). In human glaucomatous retinas and optic nerve heads, glial-related oxidative stress pathways are upregulated (Tezel et. al., 2007).

Oxidative stress can arise from Ca^{2+} dysregulation through several mechanisms, including increasing metabolic rate (Zundorf et al., 2009) and activation of ROS-producing enzymes such as nitric oxide synthase and nicotinamide adenine dinucleotide phosphate oxidase (Feissner et al., 2009; Abramov et al., 2007). ROS formation can directly damage proteins, lipids, and nucleic acids. Even worse, oxidative stress creates a positive feedback loop with Ca^{2+} dysregulation. ROS impair mitochondrial respiration and depolarize the mitochondrial membrane, thereby decreasing the organelle's ability to buffer Ca^{2+} (Ward et al., 2000; Murchison and Griffith, 2007; Esin, 2007). In addition, ROS promote Ca^{2+} release from internal stores due to effects on the ryanodine and inositol triphosphate receptors (Missiaen et al., 1992; Abramson et al., 1995; Feng et al., 2000; Bultynk et al., 2004). Finally, ROS damage plasma membrane proteins, such as Ca^{2+} ATPase and the Na-Ca exchanger, responsible for maintaining and restoring the large differential in Ca^{2+} concentrations across the membrane (Zaidi and Michaelis, 1999; Huschenbett et al., 1998). Together these mechanisms maintain, and even exacerbate, abnormal Ca^{2+} levels in the cell (Figure 2). The neuroprotective drug deprenyl apparently acts by promoting transcription of Bcl2, which over time increases the Ca^{2+}-buffering capacity of mitochondria (Rodnitsky, 1999). Bcl2 expression increases 3-fold in RGCs exposed to elevated pressure (Sappington et al., 2006), suggesting that an increase in intracellular Ca^{2+} could initiate a similarly protective pathway.

Implications for Neuroprotective Therapies

For years it has been recognized that many drugs used to lower IOP or reduce vasoconstriction also have a secondary, direct action on RGCs themselves by modulating cation influx and accumulation of intracellular Ca^{2+} over time. For example, non-selective β-adrenoceptor antagonists in addition to lowering IOP also blunt both the influx of Na^{+} and Ca^{2+} into RGCs (Wood et al., 2003; Osborne et al., 2004). In the DBA2J mouse, the β-blocker timolol promotes RGC axonal survival almost as well as the NMDA antagonist memantine (Schuettauf et al., 2002). Iganidipine is a broad Ca^{2+} antagonist that reduces vascular constriction in the retina and nerve head induced by endothelin-1 and so is thought to relieve pressure-induced ischemic insult (Ishii et al., 2003). Studies of pressure-related ischemic/reperfusion injury at the nerve head indicate that influx of extracellular Ca^{2+} into the RGC axoplasm is critical to the pathology associated with ischemic insult (Stys et al., 1992). In addition to reducing ischemic injury through their action as vasodilators, β-blockers also counteract ischemia by reducing Ca^{2+} influx into the RGC (Tomita, 2000). This has been demonstrated directly in spectrometry studies comparing the efficacy of various β-adrenoceptor antagonists in attenuating RGC intracellular Ca^{2+} (Wood et al., 2003). Optical imaging studies have demonstrated that betaxolol, a β1-antagonist, is highly effective at reducing Ca^{2+} influx to RGCs induced by glutamate receptor agonism (Zhang et al., 2003). In axonal injury, an action of Na^{+} is thought to involve leakage through non-NMDA channels that induces a reversal of the intra-axonal Na^{+}/Ca^{2+} exchanger, leading to a rise in intracellular Ca^{2+} (Stys et al., 1992).

In so-called normal pressure glaucoma, the absence of elevated IOP has compelled several hypotheses to explain the progression of visual field loss and nerve damage, which are nearly identical to those with elevated IOP (Heijl et al., 2002). A significant risk factor is nonuse of Ca^{2+} channel antagonists (Tomita, 2000). A focus has been on ocular blood flow through the optic nerve in the absence of elevated pressure, since most β-adrenoceptor antagonists modulate hemodynamics (Tomita et al., 1999). Other evidence suggests a more likely mechanism may be direct modulation of RGC intracellular Ca^{2+}. An earlier investigation compared the effects of nimodipine, a Ca^{2+} channel antagonist, on ocular blood flow using Doppler imaging in normal tension glaucoma patients (Boehm et al., 2003). These investigators found that while the drug did not affect blood flow or IOP, contrast sensitivity was dramatically improved for patients in the treatment group. Their conclusion was similar to that derived from other studies using Ca^{2+} channel antagonists: that the efficacy is a direct result of decreasing Ca^{2+} influx to the RGC or its axon. An open question is whether other types of neuroprotective therapies also modulate Ca^{2+}-dependent processes more directly, such as those involving modulation of glial signals (Bosco et al., 2008), trophic factors (Morquette and di Polo, 2008; Saragovi et al., 2009), or mitochondrial dysfunction (Ju et al., 2010).

Perspective

The evidence summarized here points towards a progression of pathology in glaucoma that mirrors recent findings in other major neurodegenerative disorders. Degeneration and functional loss appear to be compartmentalized, affecting the neuronal processes well before the cell body in the retina (Figure 1). This has important ramifications for the search for new neuronal-directed treatments. Addressing loss of function or axonal dystrophy are far more tractable problems than replacing lost cell bodies in the retina or long stretches of their axons in the optic projection. Indeed an exciting avenue of therapeutics seeks to supplant loss of function by supplying exogenous ionic activity (Corredor and Goldberg, 2009). Compartmentalized pathogenesis allows for a considerably more generous therapeutic window between the onset of the disease and irreversible degeneration (Adalbert et al., 2009). The primary message gleaned from completed work is that (1) axonal injury is early

with deficits in active transport among the first events, (2) axon injury can occur independently of cell body loss, and (3) synapse loss and dendritic pruning precede drop-out in the retina. However, much work remains. The temporal relationship between axonal and retinal events remains an important issue to resolve, as does that between synaptic and dendritic pruning.

Glaucoma is a multi-faceted syndrome that is etiologically complex. We have focused on describing known relationships between degenerative events in the RGC and possible dysregulation of Ca^{2+}-dependent processes, including oxidative stress mechanisms. Our review of pre-clinical and clinical literature indicates that targeting these processes may have relevance as therapeutic opportunities, especially for those who do not respond to IOP-lowering regimens (Baltmr et al., 2010). They are, however, downstream of key initiating events we do not as yet understand. If the disease involves sensitivity to IOP, what are the transducing elements? Are they extrinsic to the RGC, perhaps mediated exclusively by glial cells of the nerve head and retina? If not, RGCs themselves may sense changes in their own microenvironment independent of glial signaling. RGCs utilize a rich variety of cation channels, including many with a robust Ca^{2+} conductance, for processing excitatory signals from bipolar neurons. Recent results indicate that RGCs also express a number of additional membrane channels that in other neuronal systems respond directly to pressure-related insults. For example, the vanilloid-1 transient receptor potential (TRPV1) channel increases in RGCs with elevated IOP and contributes to increased intracellular Ca^{2+} with exposure to pressure (Sappington et al., 2009). The channel may also mediate secretion of protective cytokines (Sappington and Calkins, 2008). Similarly, pressure-induce release of ATP in the retina has been linked to activation of pannexin hemichannels (Reigada et al., 2008). Whether these or similar channels form a substrate through which RGCs can transduce pressure stimuli and respond directly to glaucomatous insults in vivo remains to be seen. Even so, it seems likely that the best bet for novel neuroprotective opportunities lies in improving our understanding of how RGCs respond to the various stressors associated with glaucoma.

Highlights

Glaucoma is an age-related neurodegenerative disorder

The disease involves degeneration of the retinal ganglion cell projection

Ganglion cell degeneration is compartmentalized, with axonal and retinal programs

Calcium dysregulation is an emerging theme in glaucomatous neurodegeneration

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References

Abramov AY, Scorziello A, Duchen MR. Three distinct mechanisms generate oxygen free radicals in neurons and contribute to cell death during anoxia and reoxygenation. J Neuro 2007;27:1129–1138.

Abramson JJ, Zable AC, Favero TG, Salama G. Thimerosal interacts with the Ca2+ release channel ryanodine receptor from skeletal muscle sarcoplasmic reticulum. J Biol Chem 1995;270(50):29644–29467. [PubMed: 8530347]

Adalbert R, Nogradi A, Babetto E, Janeckova L, Walker SA, Kerschensteiner M, Misgeld T, Coleman MP. Severely dystrophic axons at amyloid plaques remain continuous and connected to viable cell bodies. Brain 2010;132(Pt 2):402–416. [PubMed: 19059977]

Anderson DR, Hendrickson A. Effect of intraocular pressure on rapid axoplasmic transport in monkey optic nerve. Invest Ophthalmol Vis Sci 1974;13(10):771–783.

Anderson DR, Graham S, Pillunat L. Normal-tension glaucoma. J Glaucoma 2003;12(2):164–166. [PubMed: 12671472]

Bach M, Hoffmann MB. Update on the pattern electroretinogram in glaucoma. Optom Vis Sci 2008;85(6):386–395. [PubMed: 18521020]

Baltan S, Inman DM, Danilov CA, Morrison RS, Calkins DJ, Horner PJ. Metabolic vulnerability disposes retinal ganglion cell axons to dysfunction in a model of glaucomatous degeneration. J Neurosci 2010;30(16):5644–5652. [PubMed: 20410117]

Band LR, Hall CL, Richardson G, Jensen OE, Siggers JH, Foss AJ. Intracellular flow in optic nerve axons: a mechanism for cell death in glaucoma. Invest Ophthalmol Vis Sci 2009;50(8):3750–3758. [PubMed: 19407018]

Barron MJ, Griffiths P, Turnbull DM, Bates D, Nichols P. The distributions of mitochondria and sodium channels reflect the specific energy requirements and conduction properties of the human optic nerve head. Br J Ophthalmol 2004;88(2):286–290. [PubMed: 14736793]

Baltmr A, Duggan J, Nizari S, Salt TE, Cordeiro MF. Neuroprotection in glaucoma - Is there a future role? Exp Eye Res 2010;91(5):554–566. [PubMed: 20800593]

Berridge MJ, Lipp P, Bootman MD. Signal transduction. The calcium entry pas de deux. Science 2000;287(5458):1604–1605. [PubMed: 10733429]

Boehm AG, Breidenbach KA, Pillunat LE, Bernd AS, Mueller MF, Koeller AU. Visual function and perfusion of the optic nerve head after application of centrally acting calcium-channel blockers. Graefes Arch Clin Exp Ophthalmol 2003;241(1):34–38. [PubMed: 12545290]

Bosco A, Inman DM, Steele MR, Wu G, Soto I, Marsh-Armstrong N, Hubbard WC, Calkins DJ, Horner PJ, Vetter ML. Reduced retina microglial activation and improved optic nerve integrity with minocycline treatment in the DBA/2J mouse model of glaucoma. Invest Ophthalmol Vis Sci 2008;49(4):1437–1446. [PubMed: 18385061]

Bredesen DE, Rao RV, Mehlen P. Cell death in the nervous system. Nature 2006;443(7113):796–802. [PubMed: 17051206]

Brady S, Morfini G. A perspective on neuronal cell death signaling and neurodegeneration. Mol Neurobiol 2010;42(1):25–31. [PubMed: 20480262]

Buckingham BP, Inman DM, Lambert W, Oglesby E, Calkins DJ, Steele MR, Vetter ML, Marsh-Armstrong N, Horner PJ. Progressive ganglion cell degeneration precedes neuronal loss in a mouse model of glaucoma. J Neurosci 2008;28(11):2735–2744. [PubMed: 18337403]

Bultynck G, Szlufcik K, Kasri NN, Assefa Z, Callewaert G, Missiaen L, Parys JB, De Smedt H. Thimerosal stimulates Ca2+ flux through inositol 1,4,5-trisphosphate receptor type 1, but not type 3, via modulation of an isoform-specific Ca2+-dependent intramolecular interaction. Biochem J 2004;381:87–96. [PubMed: 15015936]

Burgoyne CF, Downs JC, Bellezza AJ, Suh JK, Hart RT. The optic nerve head as a biomechanical structure: a new paradigm for understanding the role of IOP-related stress and strain in the pathophysiology of glaucomatous optic nerve head damage. Prog Retin Eye Res 2005;24(1):39–73. [PubMed: 15555526]

Cakatay U, Telci A, Kayali R, Tekeli F, Akcay T, Sivas A. Relation of oxidative protein damage and nitrotyrosine levels in the aging rat brain. Exp Gerontol 2001;36:221–229. [PubMed: 11226738]

Calkins DJ, Horner PJ, Roberts R, Gradianu M, Berkowitz BA. Manganese-enhanced MRI of the DBA/2J mouse model of hereditary glaucoma. Invest Ophthalmol Vis Sci 2008;49(11):5083–5088. [PubMed: 18552381]

Chan SL, Mattson MP. Caspase and calpain substrates: roles in synaptic plasticity and cell death. J Neurosci Res 1999;58(1):167–190. [PubMed: 10491581]

Coleman M. Axon degeneration mechanisms: commonality amid diversity. Nat Rev Neurosci 2005;6(11):889–898. [PubMed: 16224497]

Coleman MP, Perry VH. Axon pathology in neurological disease: a neglected therapeutic target. Trends Neurosci 2007;25(10):532–537. [PubMed: 12220882]

Conforti L, Adalbert R, Coleman MP. Neuronal death: where does the end begin? Trends Neurosci 2007;30(4):159–166. [PubMed: 17339056]

Corredor RG, Goldberg JL. Electrical activity enhances neuronal survival and regeneration. J Neural Eng 2009;6(5):055001. [PubMed: 19721179]

Crish SD, Sappington RM, Inman DM, Horner PJ, Calkins DJ. Distal axonopathy with structural persistence in glaucomatous neurodegeneration. Proc Natl Acad Sci U S A 2010;107(11):5196–5201. [PubMed: 20194762]

Cuchillo-Ibanez I, Seereeram A, Byers HL, Leung KY, Ward MA, Anderton BH, Hanger DP. Phosphorylation of tau regulates its axonal transport by controlling its binding to kinesin. FASEB J 2008;22(9):3186–3195. [PubMed: 18511549]

Danias J, Lee KC, Zamora MF, Chen B, Shen F, Filippopoulos T, Su Y, Goldblum D, Podos SM, Mittag T. Quantitative analysis of retinal ganglion cell (RGC) loss in aging DBA/2NNia glaucomatous mice: comparison with RGC loss in aging C57/BL6 mice. Invest Ophthalmol Vis Sci 2003;44(12):5151–5162. [PubMed: 14638711]

Day M, Wang Z, Ding J, An X, Ingham CA, Shering AF, Wokosin D, Ilijic E, Sun Z, Sampson AR, Mugnaini E, Deutch AY, Sesack SR, Arbuthnott GW, Surmeier DJ. Selective elimination of glutamatergic synapses on striatopallidal neurons in Parkinson disease models. Nat Neurosci 2006;9(2):251–259. [PubMed: 16415865]

Esin MM. Ageing and the brain. J. Pathol 2007;211:181–187. [PubMed: 17200950]

Feissner, et al. Crosstalk signaling between mitochondrial Ca2+ and ROS. Front Biosci 2009;14:1197–1218. [PubMed: 19273125]

Feng W, Liu G, Allen PD, Pessah IN. Transmembrane redox sensor of ryanodine receptor complex. J Biol Chem 2000;275(46):35902–35907. [PubMed: 10998414]

Ferreira SM, Lerner SF, Brunzini R, Evelson PA, Llesuy SF. Oxidative stress markers in aqueous humor of glaucoma patients. Am J Ophthalmol 2004;137(1):62–69. [PubMed: 14700645]

Ferreira SM, Lerner SF, Brunzini R, Reides CG, Evelson PA, Llesuy SF. Time course changes of oxidative stress markers in a rat experimental glaucoma model. Invest Ophthalmol Vis Sci 2010;51(9):4635–4640. [PubMed: 20357192]

Filippopoulos T, Danias J, Chen B, Podos SM, Mittag TW. Topographic and morphologic analyses of retinal ganglion cell loss in old DBA/2NNia mice. Invest Ophthalmol Vis Sci 2006;47(5):1968–1974. [PubMed: 16639004]

Fischer LR, Culver DG, Tennant P, Davis AA, Wang M, Castellano-Sanchez A, Khan J, Polak MA, Glass JD. Amyotrophic lateral sclerosis is a distal axonopathy: evidence in mice and man. Exp Neurol 2004;185:232–240. [PubMed: 14736504]

Flammer J, Orgül S, Costa VP, Orzalesi N, Krieglstein GK, Serra LM, Renard JP, Stefánsson E. The impact of ocular blood flow in glaucoma. Prog Retin Eye Res 2002;21(4):359–393. [PubMed: 12150988]

Fu QL, Li X, Shi J, Xu G, Wen W, Lee DH, So KF. Synaptic degeneration of retinal ganglion cells in a rat ocular hypertension glaucoma model. Cell Mol Neurobiol 2009;29(4):575–581. [PubMed: 19172389]

Fuchshofer R, Birke M, Welge-Lussen U, Kook D, Lütjen-Drecoll E. Transforming growth factor-beta 2 modulated extracellular matrix component expression in cultured human optic nerve head astrocytes. Invest Ophthalmol Vis Sci 2005;46(2):568–578. [PubMed: 15671284]

Georgiou AL, Guo L, Cordeiro MF, Salt TE. Changes in NMDA receptor contribution to synaptic transmission in the brain in a rat model of glaucoma. Neurobiol Dis 2010;39(3):344–351. [PubMed: 20451613]

Ghanem AA, Arafa LF, El-Baz A. Oxidative stress markers in patients with primary open-angle glaucoma. Curr Eye Res 2010;35(4):295–301. [PubMed: 20373896]

Goldblum D, Mittag T. Prospects for relevant glaucoma models with retinal ganglion cell damage in the rodent eye. Vision Res 2002;42(4):471–478. [PubMed: 11853763]

Goñi-Oliver P, Lucas JJ, Avila J, Hernández F. N-terminal cleavage of GSK-3 by calpain: a new form of GSK-3 regulation. J Biol Chem 2007;282(31):22406–22413. [PubMed: 17569662]

Gordon MO, Beiser JA, Brandt JD, Heuer DK, Higginbotham EJ, Johnson CA, Keltner JL, Miller JP, Parrish RK 2nd, Wilson MR, Kass MA. The Ocular Hypertension Treatment Study: baseline factors that predict the onset of primary open-angle glaucoma. Arch Ophthalmol 2002;120(6):714–720. [PubMed: 12049575]

Gould TW, Buss RR, Vinsant S, Prevette D, Sun W, Knudson CM, Milligan CE, Oppenheim RW. Complete dissociation of motor neuron death from motor dysfunction by Bax deletion in a mouse model of ALS. J Neurosci 2006;26:8774–8786. [PubMed: 16928866]

Guo L, Tsatourian V, Luong V, Podoleanu AG, Jackson DA, Fitzke FW, Cordeiro MF. En face optical coherence tomography: a new method to analyse structural changes of the optic nerve head in rat glaucoma. Br J Ophthalmol 2005;89(9):1210–1206. [PubMed: 16113384]

Heijl A, Leske MC, Bengtsson B, Hyman L, Bengtsson B, Hussein M. Early Manifest Glaucoma Trial Group. Reduction of intraocular pressure and glaucoma progression: results from the Early Manifest Glaucoma Trial. Arch Ophthalmol 2002;120(10):1268–1279. [PubMed: 12365904]

Hernandez MR. The optic nerve head in glaucoma: role of astrocytes in tissue remodeling. Prog Retin Eye Res 2000;19(3):297–321. 2000 May. [PubMed: 10749379]

Hernández-Fonseca K, Massieu L. Disruption of endoplasmic reticulum calcium stores is involved in neuronal death induced by glycolysis inhibition in cultured hippocampal neurons. J Neurosci Res 2005;82(2):196–205. [PubMed: 16175570]

Holländer H, Makarov F, Stefani FH, Stone J. Evidence of constriction of optic nerve axons at the lamina cribrosa in the normotensive eye in humans and other mammals. Ophthalmic Res 1995;27(5):296–309. [PubMed: 8552370]

Howell GR, Libby RT, Jakobs TC, Smith RS, Phalan FC, Barter JW, Barbay JM, Marchant JK, Mahesh N, Porciatti V, Whitmore AV, Masland RH, John SW. Axons of retinal ganglion cells are insulted in the optic nerve early in DBA/2J glaucoma. J Cell Biol 2007;179(7):1523–1537. [PubMed: 18158332]

Huang W, Fileta JB, Dobberfuhl A, Filippopolous T, Guo Y, Kwon G, Grosskreutz CL. Calcineurin cleavage is triggered by elevated intraocular pressure, and calcineurin inhibition blocks retinal ganglion cell death in experimental glaucoma. Proc Natl Acad Sci U S A 2005a;102(34):12242–12247. [PubMed: 16103353]

Huang W, Dobberfuhl A, Filippopoulos T, Ingelsson M, Fileta JB, Poulin NR, Grosskreutz CL. Transcriptional up-regulation and activation of initiating caspases in experimental glaucoma. Am J Pathol 2005b;167(3):673–681. [PubMed: 16127148]

Huang W, Fileta J, Rawe I, Qu J, Grosskreutz CL. Calpain activation in experimental glaucoma. Invest Ophthalmol Vis Sci 2008;51(6):3049–3054. [PubMed: 20107181]

Huschenbett J, Zaidi A, Michaelis ML. Sensitivity of the synaptic membrane Na+/Ca2+ exchanger and the expressed NCX1 isoform to reactive oxygen species. Biochim Biophys Acta 1998;1374(1–2):34–46. [PubMed: 9814850]

Ishii K, Matsuo H, Fukaya Y, Tanaka S, Sakaki H, Waki M, Araie M. Iganidipine, a new water-soluble Ca2+ antagonist: ocular and periocular penetration after instillation. Invest Ophthalmol Vis Sci 2003;44(3):1169–1177. [PubMed: 12601046]

Jakobs TC, Libby RT, Ben Y, John SW, Masland RH. Retinal ganglion cell degeneration is topological but not cell type specific in DBA/2J mice. J Cell Biol 2005;171(2):313–325. [PubMed: 16247030]

Jellinger KA. Challenges in neuronal apoptosis. Curr Alzheimer Res 2006;3:377–391. [PubMed: 17017868]

John SW, Smith RS, Savinova OV, Hawes NL, Chang B, Turnbull D, Davisson M, Roderick TH, Heckenlively JR. Essential iris atrophy, pigment dispersion, and glaucoma in DBA/2J mice. Invest Ophthalmol Vis Sci 1998;39(6):951–962. [PubMed: 9579474]

Johnson MA, Drum BA, Quigley HA, Sanchez RM, Dunkelberger GR. Pattern-evoked potentials and optic nerve fiber loss in monocular laser-induced glaucoma. Invest Ophthalmol Vis Sci 1989;30(5):897–907. [PubMed: 2722446]

Jourdi H, Lu X, Yanagihara T, Lauterborn JC, Bi X, Gall CM, Baudry M. Prolonged positive modulation of alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors induces calpain-mediated PSD-95/Dlg/ZO-1 protein degradation and AMPA receptor down-

regulation in cultured hippocampal slices. J Pharmacol Exp Ther 2005;314(1):16–26. [PubMed: 15784649]

Ju WK, Kim KY, Duong-Polk KX, Lindsey JD, Ellisman MH, Weinreb RN. Increased optic atrophy type 1 expression protects retinal ganglion cells in a mouse model of glaucoma. Mol Vis 2010;16:1331–1342. [PubMed: 20664796]

Kashiwagi K, Ou B, Nakamura S, Tanaka Y, Suzuki M, Tsukahara S. Increase in dephosphorylation of the heavy neurofilament subunit in the monkey chronic glaucoma model. Invest Ophthalmol Vis Sci 2003;44(1):154–159. [PubMed: 12506068]

Kawahara M, Kuroda Y. Molecular mechanism of neurodegeneration induced by Alzheimer's beta-amyloid protein: channel formation and disruption of calcium homeostasis. Brain Res Bull 2000;53(4):389–397. [PubMed: 11136994]

Kerrigan LA, Zack DJ, Quigley HA, Smith SD, Pease ME. TUNEL-positive ganglion cells in human primary open-angle glaucoma. Arch Ophthalmol 1997;115(8):1031–1035. 1997 Aug; [PubMed: 9258226]

Kerrigan-Baumrind LA, Quigley HA, Pease ME, Kerrigan DF, Mitchell RS. Number of ganglion cells in glaucoma eyes compared with threshold visual field tests in the same persons. Invest Ophthalmol Vis Sci 2000;41(3):741–748. [PubMed: 10711689]

King WM, Sarup V, Sauvé Y, Moreland CM, Carpenter DO, Sharma SC. Expansion of visual receptive fields in experimental glaucoma. Vis Neurosci 2006;23(1):137–142. [PubMed: 16597357]

Knobloch M, Mansuy IM. Dendritic spine loss and synaptic alterations in Alzheimer's disease. Mol Neurobiol 2008;37(1):73–82. [PubMed: 18438727]

Ko ML, Peng PH, Ma MC, Ritch R, Chen CF. Dynamic changes in reactive oxygen species and antioxidant levels in retinas in experimental glaucoma. Free Radic Biol Med 2005;39(3):365–373. [PubMed: 15993335]

Kong GY, Van Bergen NJ, Trounce IA, Crowston JG. Mitochondrial dysfunction and glaucoma. J Glaucoma 2009;18(2):93–100. [PubMed: 19225343]

Lee MS, Kwon YT, Li M, Peng J, Friedlander RM, Tsai LH. Neurotoxicity induces cleavage of p35 to p25 by calpain. Nature 2000;405(6784):360–364. [PubMed: 10830966]

Leske MC, Heijl A, Hussein M, Bengtsson B, Hyman L, Komaroff E. Early Manifest Glaucoma Trial Group. Factors for glaucoma progression and the effect of treatment: the early manifest glaucoma trial. Arch Ophthalmol 121(1):48–56. [PubMed: 12523884]

Levin LA. Relevance of the site of injury of glaucoma to neuroprotective strategies. Surv Ophthalmol 2001;45 Suppl 3:S243–S249. [PubMed: 11377443]

Libby RT, Li Y, Savinova OV, Barter J, Smith RS, Nickells RW, John SW. Susceptibility to neurodegeneration in a glaucoma is modified by Bax gene dosage. PLoS Genet 2005;1(1):17–26. [PubMed: 16103918]

Lu X, Rong Y, Baudry M. Calpain-mediated degradation of PSD-95 in developing and adult rat brain. Neurosci Lett 2000;286(2):149–153. [PubMed: 10825658]

Mabuchi F, Aihara M, Mackey MR, Lindsey JD, Weinreb RN. Regional optic nerve damage in experimental mouse glaucoma. Invest Ophthalmol Vis Sci 2004;45(12):4352–4358. [PubMed: 15557443]

Gupta N, Ly T, Zhang Q, Kaufman PL, Weinreb RN, Yücel YH. Chronic ocular hypertension induces dendrite pathology in the lateral geniculate nucleus of the brain. Exp Eye Res 2007;84(1):176–184. 2007 Jan. [PubMed: 17094963]

Martin KR, Quigley HA, Valenta D, Kielczewski J, Pease ME. Optic nerve dynein motor protein distribution changes with intraocular pressure elevation in a rat model of glaucoma. Exp Eye Res 2006;83(2):255–262. [PubMed: 16546168]

Mattson MP, Chan SL. Neuronal and glial calcium signaling in Alzheimer's disease. Cell Calcium 2003;34(4–5):385–397. [PubMed: 12909083]

Mattson MP, Chan SL. Calcium orchestrates apoptosis. Nat Cell Biol 2003;5(12):1041–1043. [PubMed: 14647298]

McKinnon SJ, Lehman DM, Kerrigan-Baumrind LA, Merges CA, Pease ME, Kerrigan DF, Ransom NL, Tahzib NG, Reitsamer HA, Levkovitch-Verbin H, Quigley HA, Zack DJ. Caspase activation

and amyloid precursor protein cleavage in rat ocular hypertension. Invest Ophthalmol Vis Sci 2002;43(4):1077–1087. [PubMed: 11923249]

McKinnon SJ. Glaucoma: ocular Alzheimer's disease? Front Biosci 2003;8:s1140–s1156. [PubMed: 12957857]

McKinnon SJ, Goldberg LD, Peeples P, Walt JG, Bramley TJ. Current management of glaucoma and the need for complete therapy. Am J Manag Care 2008;14(1 Suppl):S20–S27. [PubMed: 18284312]

Missiaen L, Taylor CW, Berridge MJ. Luminal Ca2+ promoting spontaneous Ca2+ release from inositol trisphosphate-sensitive stores in rat hepatocytes. J Physiol 1992;455:623–640. [PubMed: 1484365]

Minckler DS, Tso MO, Zimmerman LE. A light microscopic, autoradiographic study of axoplasmic transport in the optic nerve head during ocular hypotony, increased intraocular pressure, and papilledema. Am J Ophthalmol 1976;82(5):741–757. [PubMed: 63246]

Minckler DS, Bunt AH, Johanson GW. Orthograde and retrograde axoplasmic transport during acute ocular hypertension in the monkey. Invest Ophthalmol Vis Sci 1977;16(5):426–441. [PubMed: 67096]

Mittag TW, Danias J, Pohorenec G, Yuan HM, Burakgazi E, Chalmers-Redman R, Podos SM, Tatton WG. Retinal damage after 3 to 4 months of elevated intraocular pressure in a rat glaucoma model. Invest Ophthalmol Vis Sci 2000;41(11):3451–3459. [PubMed: 11006238]

Moreno MC, Campanelli J, Sande P, Sánez DA, Keller Sarmiento MI, Rosenstein RE. Retinal oxidative stress induced by high intraocular pressure. Free Radic Biol Med 2004;37(6):803–812. [PubMed: 15384194]

Morfini GA, Burns M, Binder LI, Kanaan NM, LaPointe N, Bosco DA, Brown RH Jr, Brown H, Tiwari A, Hayward L, Edgar J, Nave KA, Garberrn J, Atagi Y, Song Y, Pigino G, Brady ST. Axonal transport defects in neurodegenerative diseases. J Neurosci 29(41):12776–12786. [PubMed: 19828789]

Morquette JB, Di Polo A. Dendritic and synaptic protection: is it enough to save the retinal ganglion cell body and axon? J Neuroophthalmol 2008;28(2):144–154. [PubMed: 18562848]

Murchison D, Griffith WH. Calcium buffering systems and calcium signaling in aged rat basal forebrain neurons. Aging Cell 2007;6(3):297–305. [PubMed: 17517040]

Nagaraju M, Saleh M, Porciatti V. IOP-dependent retinal ganglion cell dysfunction in glaucomatous DBA/2J mice. Invest Ophthalmol Vis Sci 2007;48(10):4573–4579. [PubMed: 17898280]

Nakazawa T, Nakazawa C, Matsubara A, Noda K, Hisatomi T, She H, Michaud N, Hafezi-Moghadam A, Miller JW, Benowitz LI. Tumor necrosis factor-alpha mediates oligodendrocyte death and delayed retinal ganglion cell loss in a mouse model of glaucoma. J Neurosci 2006;26(49):12633–12641. [PubMed: 17151265]

Neufeld AH, Liu B. Glaucomatous optic neuropathy: when glia misbehave. Neuroscientist 2003;9(6):485–495. [PubMed: 14678581]

North RV, Jones AL, Drasdo N, Wild JM, Morgan JE. Electrophysiological evidence of early functional damage in glaucoma and ocular hypertension. Invest Ophthalmol Vis Sci 2010;51(2):1216–1222. [PubMed: 19850843]

Osborne NN, Wood JP, Chidlow G, Casson R, DeSantis L, Schmidt KG. Effectiveness of levobetaxolol and timolol at blunting retinal ischaemia is related to their calcium and sodium blocking activities: relevance to glaucoma. Brain Res Bull 2004;62(6):525–528. [PubMed: 15036567]

Pang IH, Clark AF. Rodent models for glaucoma retinopathy and optic neuropathy. J Glaucoma 2007;16(5):483–505. [PubMed: 17700292]

Pease ME, McKinnon SJ, Quigley HA, Kerrigan-Baumrind LA, Zack DJ. Obstructed axonal transport of BDNF and its receptor TrkB in experimental glaucoma. Invest Ophthalmol Vis Sci 2000;41(3):764–774. [PubMed: 10711692]

Porciatti V, Nagaraju M. Head-up tilt lowers IOP and improves RGC dysfunction glaucomatous DBA/2J mice. Exp Eye Res 2010;90(3):452–460. [PubMed: 20036238]

Qu J, Wang D, Grosskreutz CL. Mechanisms of retinal ganglion cell injury and defense in glaucoma. Exp Eye Res 2010;91(1):48–53. [PubMed: 20394744]

Quigley HA, Addicks EM. Chronic experimental glaucoma in primates. II. Effect of extended intraocular pressure elevation on optic nerve head and axonal transport. Invest Ophthalmol Vis Sci 1980;19(2):137–152. [PubMed: 6153173]

Quigley HA, Addicks EM, Green WR, Maumenee AE. Optic nerve damage in human glaucoma. II. The site of injury and susceptibility to damage. Arch Ophthalmol 1981;99(4):635–649. [PubMed: 6164357]

Quigley HA, Nickells RW, Kerrigan LA, Pease ME, Thibault DJ, Zack DJ. Retinal ganglion cell death in experimental glaucoma and after axotomy occurs by apoptosis. Invest Ophthalmol Vis Sci 1995;36(5):774–786. [PubMed: 7706025]

Quigley HA. Neuronal death in glaucoma. Prog Retin Eye Res 1999;18(1):39–57. [PubMed: 9920498]

Quigley HA, McKinnon SJ, Zack DJ, Pease ME, Kerrigan-Baumrind LA, Kerrigan DF, Mitchell RS. Retrograde axonal transport of BDNF in retinal ganglion cells is blocked by acute IOP elevation in rats. Invest Ophthalmol Vis Sci 2000;41(11):3460–3466. [PubMed: 11006239]

Quigley HA, Broman AT. The number of people with glaucoma worldwide in 2010 and 2020. Br J Ophthalmol 2006;90(3):262–267. [PubMed: 16488940]

Reigada D, Lu W, Zhang M, Mitchell CH. Elevated pressure triggers a physiological release of ATP from the retina: Possible role for pannexin hemichannels. Neuroscience 2008;157(2):396–404. [PubMed: 18822352]

Rodnitzky RL. Can calcium antagonists provide a neuroprotective effect in Parkinson's disease? Drugs 1999;57(6):845–849. [PubMed: 10400399]

Rohn TT. The role of caspases in Alzheimer's disease; potential novel therapeutic opportunities. Apoptosis. 2010 2010 Feb 3. [Epub ahead of print].

Ross JL, Wallace K, Shuman H, Goldman YE, Holzbaur EL. Processive bidirectional motion of dynein-dynactin complexes in vitro. Nat Cell Biol 2006;8:562–570. [PubMed: 16715075]

Saleh M, Nagaraju M, Porciatti V. Longitudinal evaluation of retinal ganglion cell function and IOP in the DBA/2J mouse model of glaucoma. Invest Ophthalmol Vis Sci 2007;48(10):4564–4572. [PubMed: 17898279]

Salinas-Navarro M, Jiménez-López M, Valiente-Soriano FJ, Alarcón-Martínez L, Avilés-Trigueros M, Mayor S, Holmes T, Lund RD, Villegas-Pérez MP, Vidal-Sanz M. Retinal ganglion cell population in adult albino and pigmented mice: a computerized analysis of the entire population and its spatial distribution. Vision Res 2009;49(6):637–647. [PubMed: 19948111]

Sappington RM, Chan M, Calkins DJ. Interleukin-6 protects retinal ganglion cells from pressure-induced death. Invest Ophthalmol Vis Sci 2006;47(7):2932–2942. [PubMed: 16799036]

Sappington RM, Calkins DJ. Contribution of TRPV1 to microglia-derived IL-6 and NFkappaB translocation with elevated hydrostatic pressure. Invest Ophthalmol Vis Sci 2008;49(7):3004–3017. [PubMed: 18362111]

Sappington RM, Sidorova T, Long DJ, Calkins DJ. TRPV1: contribution to retinal ganglion cell apoptosis and increased intracellular Ca2+ with exposure to hydrostatic pressure. Invest Ophthalmol Vis Sci 2009;50(2):717–728. [PubMed: 18952924]

Sappington RM, Carlson BJ, Crish SD, Calkins DJ. The microbead occlusion model: a paradigm for induced ocular hypertension in rats and mice. Invest Ophthalmol Vis Sci 2010;51(1):207–216. [PubMed: 19850836]

Schuettauf F, Quinto K, Naskar R, Zurakowski D. Effects of anti-glaucoma medications on ganglion cell survival: the DBA/2J mouse model. Vision Res 2002;42(20):2333–2337. [PubMed: 12350421]

Schlamp CL, Li Y, Dietz JA, Janssen KT, Nickells RW. Progressive ganglion cell loss and optic nerve degeneration in DBA/2J mice is variable and asymmetric. BMC Neurosci 2006;7:66. [PubMed: 17018142]

Sehi M, Grewal DS, Feuer WJ, Greenfield DS. The impact of intraocular pressure reduction on retinal ganglion cell function measured using pattern electroretinogram in eyes receiving latanoprost 0.005% versus placebo. Vision Res. 2010 2010 Sep 8. [Epub ahead of print].

Shea TB, Yabe JT, Ortiz D, Pimenta A, Loomis P, Goldman RD, Amin N, Pant HC. Cdk5 regulates axonal transport and phosphorylation of neurofilaments in cultured neurons. J Cell Sci 2004;117(Pt 6):933–941. [PubMed: 14762105]

Shea TB, Chan WK. Regulation of neurofilament dynamics by phosphorylation. Eur J Neurosci 2008;27(8):1893–1901. [PubMed: 18412610]

Shields MB. Normal-tension glaucoma: is it different from primary open-angle glaucoma? Curr Opin Ophthalmol 2008;9(2):85–88. [PubMed: 18301279]

Siman R, Baudry M, Lynch G. Brain fodrin: substrate for calpain I, an endogenous calcium-activated protease. Proc Natl Acad Sci U S A 1984;81(11):3572–3576. [PubMed: 6328521]

Sommer A. Intraocular pressure and glaucoma. Am J Ophthalmol 1989;107(2):186–188. [PubMed: 2913813]

Sommer A, Tielsch JM, Katz J, Quigley HA, Gottsch JD, Javitt J, Singh K. Relationship between intraocular pressure and primary open angle glaucoma among white and black Americans. The Baltimore Eye Survey. Arch Ophthalmol 1991;109(8):1090–1095. [PubMed: 1867550]

Son JL, Soto I, Oglesby E, Lopez-Roca T, Pease ME, Quigley HA, Marsh-Armstrong N. Glaucomatous optic nerve injury involves early astrocyte reactivity and late oligodendrocyte loss. Glia 2010;58(7):780–789. [PubMed: 20091782]

Soto I, Oglesby E, Buckingham BP, Son JL, Roberson ED, Steele MR, Inman DM, Vetter ML, Horner PJ, Marsh-Armstrong N. Retinal ganglion cells downregulate gene expression and lose their axons within the optic nerve head in a mouse glaucoma model. J Neurosci 2008;28(2):548–561. [PubMed: 18184797]

Spires TL, Hyman BT. Neuronal structure is altered by amyloid plaques. Rev Neurosci 2004;15(4): 267–278. [PubMed: 15526551]

Stasi K, Nagel D, Yang X, Ren L, Mittag T, Danias J. Ceruloplasmin upregulation in retina of murine and human glaucomatous eyes. Invest Ophthalmol Vis Sci 2007;48(2):727–732. [PubMed: 17251471]

Steele MR, Inman DM, Calkins DJ, Horner PJ, Vetter ML. Microarray analysis of retinal gene expression in the DBA/2J model of glaucoma. Invest Ophthalmol Vis Sci 2006;47(3):977–985. [PubMed: 16505032]

Stevens B, Allen NJ, Vazquez LE, Howell GR, Christopherson KS, Nouri N, Micheva KD, Mehalow AK, Huberman AD, Stafford B, Sher A, Litke AM, Lambris JD, Smith SJ, John SW, Barres BA. The classical complement cascade mediates CNS synapse elimination. Cell 2007;131(6):1164–1178. [PubMed: 18083105]

Stokin GB, Lillo C, Falzone TL, Brusch RG, Rockenstein E, Mount SL, Raman R, Davies P, Masliah E, Williams DS, Goldstein LS. Axonopathy and transport deficits early in the pathogenesis of Alzheimer's disease. Science 2005;307(5713):1282–1288. [PubMed: 15731448]

Stys PK, Waxman SG, Ransom BR. Ionic mechanisms of anoxic injury in mammalian CNS white matter: role of Na+ channels and Na(+)-Ca2+ exchanger. J Neurosci 1992;12(2):430–439. [PubMed: 1311030]

Tahzib NG, Ransom NL, Reitsamer HA, McKinnon SJ. Alpha-fodrin is cleaved by caspase-3 in a chronic ocular hypertensive (COH) rat model of glaucoma. Brain Res Bull 2004;62(6):491–495. [PubMed: 15036563]

Tatton WG, Chalmers-Redman RM, Tatton NA. Apoptosis and anti-apoptosis signalling in glaucomatous retinopathy. Eur J Ophthalmol 2001;11 Suppl 2:S12–S22. [PubMed: 11592526]

Tatton W, Chen D, Chalmers-Redman R, Wheeler L, Nixon R, Tatton N. Hypothesis for a common basis for neuroprotection in glaucoma and Alzheimer's disease: anti-apoptosis by alpha-2-adrenergic receptor activation. Surv Ophthalmol 2003;48 Suppl 1:S25–S37. [PubMed: 12852432]

Tezel G, Hernandez MR, Wax MB. In vitro evaluation of reactive astrocyte migration, a component of tissue remodeling in glaucomatous optic nerve head. Glia 2001;34(3):178–189. [PubMed: 11329180]

Tezel G, Yang X, Cai J. Proteomic identification of oxidatively modified retinal proteins in a chronic pressure-induced rat model of glaucoma. Invest Ophthalmol Vis Sci 2005;46(9):3177–3187. [PubMed: 16123417]

Tezel G. TNF-alpha signaling in glaucomatous neurodegeneration. Prog Brain Res 2008;173:409–421. [PubMed: 18929124]

Tielsch JM, Sommer A, Katz J, Royall RM, Quigley HA, Javitt J. Racial variations in the prevalence of primary open-angle glaucoma. The Baltimore Eye Survey. JAMA 1991;266(3):369–374. [PubMed: 2056646]

Tomita G. The optic nerve head in normal-tension glaucoma. Curr Opin Ophthalmol 2000;11(2):116–120. [PubMed: 10848217]

Veeranna, Kaji T, Boland B, Odrljin T, Mohan P, Basavarajappa BS, Peterhoff C, Cataldo A, Rudnicki A, Amin N, Li BS, Pant HC, Hungund BL, Arancio O, Nixon RA. Calpain mediates calcium-induced activation of the erk1,2 MAPK pathway and cytoskeletal phosphorylation in neurons: relevance to Alzheimer's disease. Am J Pathol 2004;165(3):795–805. [PubMed: 15331404]

Vidal-Sanz M, Lafuente MP, Mayor-Torroglosa S, Aguilera ME, Miralles de Imperial J, Villegas-Pérez MP. Brimonidine's neuroprotective effects against transient ischaemiainduced retinal ganglion cell death. Eur J Ophthalmol 2001;11 Suppl 2:S36–S40. [PubMed: 11592529]

Vosler PS, Brennan CS, Chen J. Calpain-mediated signaling mechanisms in neuronal injury and neurodegeneration. Mol Neurobiol 2008;38(1):78–100. [PubMed: 18686046]

Waldmeier PC, Tatton WG. Interrupting apoptosis in neurodegenerative disease: potential for effective therapy? Drug Discov Today 2004;9(5):210–218. [PubMed: 14980539]

Wang Z, Khan S, Sheetz MP. Single cytoplasmic dynein molecule movements: characterization and comparison with kinesin. Biophys J 1995;69(5):2011–2023. [PubMed: 8580344]

Wang X, Ng Y, Tay S. Factors contributing to neuronal degeneration in retinas of experimental glaucomatous rats. J Neurosci Res 2005;82:674–689. [PubMed: 16273539]

Ward MW, Rego AC, Frenguelli BG, Nicholls DG. Mitochondrial membrane potential and glutamate excitotoxicity in cultured cerebellar granule cells. J Neurosci 2000;20(19):7208–7219. [PubMed: 11007877]

Wax MB, Tezel G, Yang J, Peng G, Patil RV, Agarwal N, Sappington RM, Calkins DJ. Induced autoimmunity to heat shock proteins elicits glaucomatous loss of retinal ganglion cell neurons via activated T cell-derived Fas-ligand. J. Neurosci 2008;28:12085–12096. [PubMed: 19005073]

Weber AJ, Harman CD, Viswanathan S. Effects of optic nerve injury, glaucoma, and neuroprotection on the survival, structure, and function of ganglion cells in the mammalian retina. J Physiol 586(Pt 18):4393–4400. [PubMed: 18565994]

Weber AJ, Viswanáthan S, Ramanathan C, Harman CD. Combined application of BDNF to the eye and brain enhances ganglion cell survival and function in the cat after optic nerve injury. Invest Ophthalmol Vis Sci 2010;51(1):327–334. 2010 Jan; [PubMed: 19710411]

Whitmore AV, Libby RT, John SW. Glaucoma: thinking in new ways-a role for autonomous axonal self-destruction and other compartmentalised processes? Prog Retin Eye Res 24(6):639–662. [PubMed: 15953750]

Wood JP, Schmidt KG, Melena J, Chidlow G, Allmeier H, Osborne NN. The beta-adrenoceptor antagonists metipranolol and timolol are retinal neuroprotectants: comparison with betaxolol. Exp Eye Res 2003;76(4):505–516. [PubMed: 12634114]

Yuan L, Neufeld AH. Tumor necrosis factor-alpha: a potentially neurodestructive cytokine produced by glia in the human glaucomatous optic nerve head. Glia 2000;32(1):42–50. [PubMed: 10975909]

Zaidi A, Michaelis ML. Effects of reactive oxygen species on brain synaptic plasma membrane Ca(2+)-ATPase. Free Radic Biol Med 1999;27(7–8):810–821. [PubMed: 10515585]

Zaja-Milatovic S, Milatovic D, Schantz AM, Zhang J, Montine KS, Samii A, Deutch AY, Montine TJ. Dendritic degeneration in neostriatal medium spiny neurons in Parkinson disease. Neurology 2005;64(3):545–547. [PubMed: 15699393]

Zang DW, Lopes EC, Cheema SS. Loss of synaptophysin-positive boutons on lumbar motor neurons innervating the medial gastrocnemius muscle of the SOD1G93A G1H transgenic mouse model of ALS. J Neurosci Res 2005;79(5):694–699. [PubMed: 15668955]

Zhang J, Wu SM, Gross RL. Effects of beta-adrenergic blockers on glutamate-induced calcium signals in adult mouse retinal ganglion cells. Brain Res 2003;959(1):111–119. [PubMed: 12480164]

Zündorf G, Kahlert S, Bunik VI, Reiser G. alpha-Ketoglutarate dehydrogenase contributes to production of reactive oxygen species in glutamate-stimulated hippocampal neurons in situ. Neuroscience 2009;158:610–616. [PubMed: 18996448]

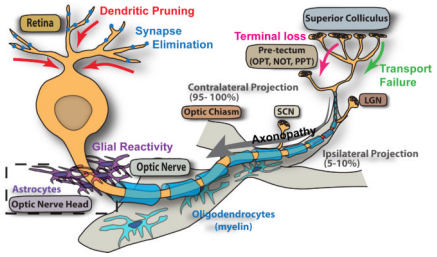

Figure 1. Degenerative Events Affecting the RGC Projection in Glaucoma

The RGC axon passes unmyelinated through a plexus of astrocytes in the nerve head that becomes neurochemically reactive in glaucoma. This is an established zone of vulnerability to IOP- and age-related stressors. All or nearly all RGCs project contralaterally to the superior colliculus, with small collaterals terminating in more anterior sites. These include the suprachiasmatic nucleus (SCN), lateral geniculate nucleus (LGN), and the pretectal nuclei: olivary pretectal (OPT), nucleus of optic tract (NOT), and posterior pretectal (PPT). A small fraction of RGCs also form ipsilateral projections. Early degenerative events in glaucoma include failure of anterograde transport and axonopathy, both of which progress from distal projection sites towards the optic nerve and retina. RGC axon terminals and their synapses eventually degrade. In the retina, degeneration includes loss of excitatory synapses and dendritic pruning.

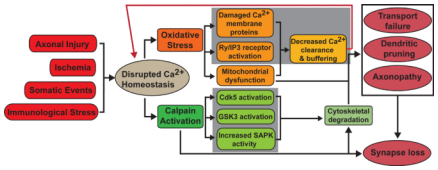

Figure 2. Possible Ca^{2+}-Dependent Mechanisms that Could Contribute to Glaucomatous Neurodegeneration

Multiple stressors relevant in glaucoma could disrupt homeostasis of RGC intracellular Ca^{2+}. Two immediate consequences include oxidative stress and activation of calpains. Oxidative stress exacerbates Ca^{2+} dysregulation by reducing the capacity for mitochondria to buffer Ca^{2+}, increasing release of Ca^{2+} from internal stores through interactions with the ryanodine (Ry) and inositol-triphosphate (IP3) receptors, and reducing clearance of intracellular Ca^{2+} by damaging membrane-bound maintenance proteins such as Ca^{2+} ATPase and the Na^{+}/Ca^{2+} exchanger. Activated calpains increase the activity of cyclin-dependent kinase 5 (Cdk5), glycogen synthase kinase 2 (GSK3), and stress-activated protein kinases (SAPK) which phosphorylate cytoskeletal elements, slowing or stopping their axonal transport and promoting their aggregation. Calpains also directly degrade both cytoskeletal scaffolding and synapses. Cytoskeletal degradation and reduced mitochondrial function further compromise axonal transport and maintenance of dendrites, thus likely promoting synapse elimination. These degenerative components trigger the eventual apoptotic elimination of the cell body. Components in grey boxes represent likely targets not directly established for glaucoma.