

Closed Gateways – Can Neuroprotectants Shield the Retina in Glaucoma?

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Abstract

Neuroprotection for glaucoma is a therapeutic approach that aims to prevent optic nerve damage or cell death. An appropriate drug that reaches an adequate concentration across the blood retinal barrier is expected to shield the retina in glaucoma. Several *in vitro* and *in vivo* attempts in experimental models indicate the possibility of successful neuroprotection. However, clinical trials might not show the same level of neuroprotection as a result of subtherapeutic concentrations of the drug in the eye. The study by Zhong et al. in this issue of *Drugs in R&D* could not attribute the observed improvement in visual field indices to any one of the individual active constituents of *Erigeron breviscapus* (vant.) Hand. Mazz. (EBHM). One of the major constituents of EBHM is scutellarin, which is known to have poor oral bioavailability and an unclear ability to penetrate inside the eye. Therefore, before recognizing EBHM as a neuroprotectant in glaucoma for further clinical studies and practice, its active constituents and their pharmacokinetics (systemic as well as ocular) need to be explored.

Neuroprotection as a pharmacological strategy to shield retinal ganglion cell death has been a popular approach adopted by many researchers. Yet, its effectiveness in preventing retinal ganglion cell death and thereby preserving vision in patients with primary open-angle glaucoma has not been demonstrated.^[1]

Mitochondria are abundant in the unmyelinated part of the retinal ganglion cell axons to meet the higher energy demand for nerve conduction and for the maintenance of optimum neuronal function.^[2] Ischemia to the optic nerve head region can cause ganglion cell death at different rates. Therefore, effective intraocular pressure (IOP) control and blocking intracellular death triggers are both viable options. The right pharmacologic agent with a meaningful intraocular

penetration would rationalize the neuroprotection strategy in glaucoma.

For the last three decades, several attempts have been made to establish a successful neuroprotectant for glaucoma using various *in vitro* and *in vivo* models. A comprehensive PubMed search with the MeSH terms "glaucoma" and "neuroprotection" revealed the following compounds, factors and plant extracts: NMDA receptor antagonists (memantine, dexanabinol), an α 2-adrenergic receptor agonist (brimonidine), an α 1- and β -adrenergic receptor antagonist with a nitric oxide donor (niradilol), an angiotensin II type 1 receptor blocker (candesartan), a prostaglandin-related compound (tafluprost), calcium channel blockers (nilvadipine and cilnidipine), a sodium channel blocker (phenytoin), a dual

sodium and calcium channel blocker (NS-7), a choline precursor (citicoline), a tetracycline (minocycline), an anti-inflammatory agent (aspirin [acetylsalicylic acid]), an endothelin antagonist (sulfisoxazole), an inducible nitric oxide synthase inhibitor (aminoguanidine), α -tocopherol, and vinpocetine.

Endogenous compounds or their analogs, such as (endo)cannabinoids, granulocyte-colony stimulating factor (G-CSF), hematopoietic cytokine (erythropoietin), nerve growth factor, natural amino acid (taurine), glial cell line-derived neurotrophic factor and estrogen analogs (ZYC-1, ZYC-3), were also scrutinized.

Plant extracts and derived compounds, such as *Ginkgo biloba* extract, carotenoids (xanthophylls), epigallocatechin-gallate, cannabinoids, ferulic acid from *Ligusticum wallichii*, a muscarinic agonist (pilocarpine), a lignophenol derivative from bamboo lignin (Lig-8), aloe-emodin metabolites, *Lycium barbarum*, red wine-polyphenols (resveratrol), *Erigeron breviscapus*, flavonoids, and spider venom derived 2-amino-5-ureidopentanamide from *Parawixia bistrata*, were suggested as potential candidates and some of them were also evaluated using *in vitro* and *in vivo* experimental models for neuroprotection in glaucoma.

Although there is good laboratory evidence for neuroprotection in glaucoma by many agents, proof from randomized clinical trials is lacking.^[3] The reasons for the failure of any such clinical attempts are unclear as many of the results from these trials have not been published.

A potential neuroprotectant compound needs to effectively cross the blood retinal barrier (BRB) into the eye. Agents given systemically, such as antimicrobials, seldom reach effective concentrations in the uninflamed eye.^[4] For that reason, a neuroprotectant may require multiple doses at frequent intervals over a longer duration to achieve a significant concentration in the eye. In addition to the plasma kinetics of a compound, its vitreous disposition is also important in determining its therapeutic action. It is currently well recognized that the presence of drug transporters in the BRB are responsible for the efflux of the intravitreally injected drug and for resisting the influx of the drug into the eye

when it is given systemically. The presence of P-glycoprotein efflux transporters, cationic/anionic transporters, and other transporters in the BRB that resist the entry of systemically given substrates is well documented.^[5-7] Hence, for any predictable outcome in retinal neuroprotection, the ocular pharmacokinetic profile of the drug needs to be explored before initiating a clinical trial.

Systemic treatment with memantine of experimental glaucoma in monkeys showed that it was safe and effective in reducing the functional loss of the retina, although it had no effect on IOP. This study concluded that peak plasma concentration (C_{\max}) of memantine reached 1 $\mu\text{mol/L}$ at 4–15 months after the initiation of an oral dose of 4 mg/kg/day and similar levels were also observed in the vitreous humour after 16 months.^[8] Surprisingly, memantine, which is approved for the treatment of Alzheimer's disease, has failed to show any significant improvement in glaucoma in subsequent clinical trials as a neuroprotectant.^[9]

A single 20 mg dose of memantine resulted in a C_{\max} of 0.14 $\mu\text{mol/L}$ in healthy human volunteers, a time to C_{\max} of 6.9 hours and a half-life of 62 hours.^[10] A human study conducted by Kornhuber and Quack^[11] reported that a 20 mg/day oral dose of memantine over 14 days resulted in concentrations of 0.37 $\mu\text{mol/L}$ in the plasma and 0.207 $\mu\text{mol/L}$ in the cerebrospinal fluid (CSF) at 2–3 hours after the last maintenance dose, and a CSF to serum ratio of 0.55 was proposed. A target concentration of memantine 1 $\mu\text{mol/L}$ is expected to prevent excitotoxicity in neurons.^[12] The failure of memantine in the clinical trial for neuroprotection in glaucoma might have been due to an inadequate concentration of memantine in the vitreous after the oral dose of 20 mg/day.

The randomized, double-blind, clinical trial reported by Zhong et al.^[13] in this issue of *Drugs in R&D* concluded that the Chinese traditional herb *Erigeron breviscapus* (vant.) Hand. Mazz. (EBHM) showed a partial protective effect in the visual field of glaucoma patients with controlled IOP. Breviscapine is a commercially produced plant extract from the Chinese herb

EBHM. Scutellarin is a flavone glucuronide (5,6,4'-trihydroxyflavone-7-O-glucuronide) and is reported to be the main active component of EBHM.^[14] The percentage of scutellarin varies according to the source of production. In experimental studies, it has been reported to have estrogenic and neuroprotective properties.^[15] Breviscapine has been reported to attenuate neuroapoptosis and regulate the protein expression related to apoptosis after transient focal cerebral ischemia at a dose of 50–100 mg/kg in rats.^[16] Neuroprotective effects of scutellarin on brain ischemic injury-induced apoptosis have been reported to be associated with inhibition of PARP-dependent mitochondrial dysfunction and subsequent translocation of the apoptosis-inducing factor pathway.^[17]

Rat brain drug distribution studies showed that 22.3% of orally and 29.18% of tail vein administered radiolabeled breviscapine reached the brain tissues when it was administered at a dose of 0.4 mg/kg.^[18] Interestingly, an experimental study in rats also reported that protective effects of scutellarin on cardiovascular and cerebrovascular ischemia were better than those of the extract mixture (breviscapine).^[19] Another study reported that the relative bioavailabilities of orally administered scutellarin and scutellarein conjugates in rats were very low (<11%).^[20]

In this issue of *Drugs in R&D*, Zhong et al.^[13] could not attribute the observed activity to any one of the individual active constituents of EBHM. They have correlated the activity to 4'-hydroscutellarein, 4'-hydrobaicalein-7- β -D-plamyagin, 4'-hydroscutellarein-7- β -D-glycuron methyl ester, pyromeconic acid, and several kinds of flavones and flavonoids. Chen et al.^[21] reported that after a single oral administration of scutellarin 60 mg to 20 healthy subjects, plasma concentrations of scutellarin and its major metabolite (isoscutellarin) were found to reach a mean C_{\max} of <5.0 and 87 ng/mL, respectively. They have also indicated that after hydrolysis in the gastrointestinal system by bacterial enzymes, the aglycone of scutellarin (scutellarein) was detected in serum as glucuronic acid conjugates, which showed regioselectivity and species difference. Considering the limited oral absorption of its major

constituent (scutellarin), EBHM needs rigorous analysis regarding the active constituent(s) that get absorbed into the systemic circulation and, in turn, reach the vitreous humor for the projected activity.

Before promoting EBHM as a neuroprotectant for glaucoma and commencing with long-term studies, it is essential to identify the active constituent(s) and to determine whether the compounds have the ability to cross the closed gateways of the BRB to reach an effective concentration to shield the retina.

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