ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN-POLONIA
VOL. LXI., N1, 42 SECTIO D 2006

2nd Department of Ophthalmology
Department of Pharmacognosy with the Laboratory of Medicinal Plants
Department of Market Gardening and Medicinal Plants, Medical University of Lublin

MAREK GERKOWICZ, OLAF KALISZ, TADEUSZ WOLSKI

Ginkgo biloba and glaucoma

Ginkgo biloba is one of the oldest tree species still present on earth. It is the only living representative of the order Ginkgoales, a group of gymnosperms composed of the family *Ginkgoaceae*. The plant originates from China, where it has been used in traditional medicine. The trees can reach 20–30 meters in height and are extremely long living – some individuals as many as 1500 years. *Ginkgo biloba* leaf extract has been used in traditional Chinese medicine since about 3000 B.C. The standard *Ginkgo biloba* extract contains many bioactive substances, the most important of which being flavonoids (about 24%) and the terpenes (about 6%). The terpenoids include bilobalids and gingkoloids.

The mechanism of *Ginkgo biloba* action results from its properties as a neuroprotective agent, antioxidant, membrane stabiliser and inhibitor of platelet-activating factor (1). Flavonoids are responsible for the antioxidant effect and for scavenging free radicals. The oxidant potential of *Ginkgo biloba* is comparable with ascorbic acid, glutathione, alpha-tocopherol and retinol acetate. *Ginkgo biloba* inhibits nitric oxide synthase (iNOS) and platelet activating factor. *Ginkgo biloba* extract exhibits neuroprotective action and inhibits apoptosis in cell culture systems (2). It also favourably affects neurotransmitter disturbances and neurone metabolism increasing anoxia tolerance. *Ginkgo biloba* improves both peripheral and cerebral blood flow decreasing blood viscosity, inhibiting platelet aggregation and thrombus formation. *Ginkgo biloba* preparation is also characterised by pharmacologic effect consisting in: endothelium relaxation mediated by inhibition of 3',5'-cyclic GMP (guanosine monophosphate phosphodiesterase). *Ginkgo biloba* is one of the most commonly used phytomedical preparations in Europe as well as the USA.

The plant is believed to be effective in the treatment of many pathologies, among others: decrease of cognitive function (9), disturbances of cerebral blood flow (15), peripheral vascular disorders (10), atopic dermatitis and to favourably affect non-pressure-dependent risk factors for glaucoma (11).

Ginkgo biloba is used in the therapy of vascular dementia, peripheral claudication and tinnitus of vascular origin. Clinical studies have shown that *Ginkgo biloba* extract is efficacious in the therapy of Alzheimer's diseaase (9). Some papers show positive effect of *Ginkgo biloba* on cognitive functions in Alzheimer patients. There are reports pointing to favourable effect of *Ginkgo biloba* extract on sexual function, macular degeneration, asthma and hypoxia.

NEUROPROTECTION AND NITRIC OXIDE

A number of papers confirm neuroprotective action of *Gingko biloba*. This effect has been demonstrated both *in vivo* and *in vitro* (1, 2, 8). The plant extract (Egb 761) reduces apoptosis (8), decreases ischemia-hypoperfusion in the brain, causes the increase of nitric oxide (NO) level by activation of endothelial nitric oxide synthase. EGb 761 also inhibits cyclic GMP (c-GMP) phosphodiestherase, and thus prolongs the vasodilatory effect of nitric oxide. Mice trials have shown that Egb 761 extract inhibited the degeneration of dopaminergic neurons in the striatum. This may

suggest that inhibition of reuptake of dopamine and of monoamine oxidase activity is connected with neuroprotective mechanism of Egb 761.

Oxidative stress is an important factor in many neurodegenerative diseases as well as in stroke, and the use of antioxidants is a significant element in the therapy of these diseases. There are studies confirming antioxidative effects of Egb 761 as well as its protective action against neuronal damage caused by oxidative stress (8).

Nitric oxide (NO) is responsible for damage of many cells including cerebral and ocular tissue. In the central nervous system NO is responsible for the development of neurodegenerative diseases such as Alzheimer's disease or multiple sclerosis. Nitric oxide produced in excess can cause death of nervous cells and cytotoxicity is associated with activation of poly (ADP-ribose) polymerase and the formation of poeroxynitrite by reaction with superoxide. Low NO concentration can have protective effect on the nervous tissue, but increased NO concentration may be toxic for nervous tissue.

Nitric oxide is a compound synthetised by nitric oxide synthase (NOS) occurring in three isoforms – neuronal (nNOS, NOS-1), inducible (iNOS, NOS-2) and endothelial (eNOS, NOS-3). Nitric oxide derived at low levels from eNOS synthase acts as a vasodilatator. iNOS activity causes inductive NO production in excessive quantities which results in endothelial cell damage. It is primarily expressed in macrophages – a high level of nitric oxide can be sustained for a prolonged period of time. The effect of *Ginkgo biloba* extract (EGb 761) consists in inhibiting excessive NO production liberated by macrophages (iNOS activation), whereas they did not affect eNOS-mediated nitric oxide production. It seems probable that the effect of EGb 761 extract consisting in increasing cerebral blood flow results from the increase of eNOS synthase activity.

There are reports confirming *Ginkgo biloba* protective effect on nervous cells counteracting toxic damage of NO (2). EGb 761 extract has properties of removing NO and decreasing NO liberation after periods of cerebral and cardiac ischemia as well as from macrophages by iNOS mRNA reduction and protein expression.

GINKGO BILOBA AND GLAUCOMA

Glaucoma is one of the main causes of blindness in the world. Normal tension glaucoma is such a form of primary open-angle glaucoma in which optic nerve damage and visual field loss occurs despite the maintenance of intraocular pressure values within normal limits. The exact mechanism of glaucoma damage in this form of pathology has not been fully explained yet. Both the vascular factor – decrease of blood flow or mechanical factor – relatively high intraocular pressure can be considered here. In glaucoma treatment we have often to do with progression of the disease in spite of efficacious reduction of intraocular pressure. A similar situation occurs in normal tension glaucoma. Development of the disease leads to the progression of optic nerve damage and to selective retinal ganglion cell death (4). This injury is often irreversible and, therefore, neuroprotective action seems to play the crucial role here. A hypothesis has appeared claiming that glaucoma may result from neurotoxic effect of NO on the optic nerve head and retina, which may cause degenerative changes of the optic nerve and visual field loss. Nitric oxide synthetised by NOS-2 synthase (enzyme form not encountered in cells under normal conditions) and responsible for neurodegenerative processes in many diseases may also be held responsible for glaucoma development (7). Recent work on acute retinal ischemia in rats has demonstrated that NOS-2 participates in retinal destruction. Clusters of NOS-2 positive cells have been found in human glaucomatous optic nerve heads. It has been found out that EGb 761 extract has dose-dependent effect inhibiting NO synthesis by inhibiting inducible NO (iNOS) synthase (2).

Another explanation of favourable *Ginkgo biloba* effect in patients with normal tension glaucoma can be its effect on cognitive function improvement. It may be especially important in patients with normal tension glaucoma because such patients have been shown to suffer from cerebral ischemia of small vessels more frequently than healthy people (14). The findings of other papers also confirm a deficit of ocular blood flow (5) and pre-capillary ischemia in patients with normal tension glaucoma or with primary open-angle glaucoma. *Ginkgo biloba* extract seems to

have a potential role in the improvement of visual function as biologic activity of *ginkgo* include increasing neuronal tolerance to anoxia, having a favourable effect on neurotransmitter disturbances and preventing damage of cell membranes caused by free radicals.

In the clinical trial with 27 patients (54 eyes) suffering from normal tension glaucoma with bilateral visual field damage, the effect of *Ginkgo biloba* on preexisting visual field damage was evaluated (11). Patients received 40 mg *Ginkgo biloba* extract three times daily for four weeks. It was shown that the administration of *Ginkgo biloba* statistically significantly improved vision (visual field assessment) – mean deviation value was at the beginning of the investigations 11.4+−3.27 dB while after *Ginkgo biloba* therapy it was 8.78+−2.56 dB. (t=8.86, P=0.0001, chi-square test); corrected pattern standard deviation (CPSD) at baseline was 10.93+−2.12 and after therapy 8.13+−2.12 dB (t=9.98, P=0.0001, chi-square test). No significant changes were observed in the values of intraocular pressure, general pressure or pulse compared with placebo group.

The mechanism explaining improvement in former visual field disorders after EGb 761 therapy is not quite clear, but it has been shown that the extract affects the improvement of blood flow in the ophthalmic artery of healthy people (3). This may probably at least partly account for obtained results. *Ginkgo biloba* extract has probably a limited duration of action when discontinued and may have to be administered chronically to maintain its effect. The improvement in visual field indices was not maintained after 8 weeks.

Advantages connected with therapeutic effect of *Ginkgo bilooba* in ophthalmology are not limited to glaucoma. In the eye EGb 761 decreases ischemia reperfusion injury in the retina and inhibits preretinal proliferation in experimental tractional retinal detachment. There are reports about protective action of the extract (EGb 761) against lipoperoxidation, against damages caused by argon laser photocoagulation, progression of diabetic retinopathy and against light induced damages. In elderly people there has been noted improvement in visual field examination after *Ginkgo biloba* administration (12). A study on healthy volunteers has shown statistically significant improvement of end diastolic blood velocity in the ophthalmic artery (3). The difference in comparison with the initial value was about 24%.

A study on rats has shown that therapy with the ginkgo extract has neuroprotective action in chronic glaucoma (6). Increased intraocular pressure was obtained by closing 3 episcleral veins. Assessments of degenerative changes were performed after 5 months in two groups – one treated with EGb 761 in the dose 100 mg/kg/day and the other that was not treated. Retinal ganglion cell death in eyes with increased intraocular pressure in animals in the group not treated compared with the other eye was 29.8+−1.5%, while in the group given the extract (EGb 761) 4.6+−4.5%. Dose analysis was based on the results of studies on mice where it was shown that multiple dose of *Ginkgo biloba* extract in the range from 50 to 100 mg/kg/day efficiently inhibits monamine oxidase in mice producing protective action for ischemia reperfusion injury in the retina.

In another study carried out on 11 healthy volunteers the researchers have shown that oral administration of the extract in the dose 40 mg three times a day for 2 days causes statistically significant increase of diastolic blood flow velocity in the ophthalmic artery compared with placebo group (3). No adverse effects have been observed connected with *Ginkgo biloba* therapy. EGb 761 extract administration did not affect arterial pressure values, puls or intraocular pressure.

Ginkgo biloba has a number of properties that should theoretically favourably influence glaucoma mechanisms unconnected with intraocular pressure: increase of ocular flow, antioxidative action, antagonistic action to platelet activating factor, inhibition of nitric oxide effect, neuroprotective action (13).

CONCLUSIONS

Ginkgo biloba seems to be an interesting raw material with great therapeutic potential and carrying out of investigations on a numerous group of patients would let dispel therapeutic doubts concerning its efficacy in glaucoma treatment.

REFERENCES

1. Ahlemeyer B., Kriegelstein J.: Neuroprotective effects of *Ginkgo biloba* extract. In: L.D. Lawson, R. Bauer: Phytomedicines of Europe: Chemistry and Biological Activity. Washington, D.C. American Chemical Society, 210, 1998.
2. Bastianetto S. et al.: The *Ginkgo biloba* extract (EGb 761) protects and rescues hippocampal cells against nitric oxide-induced toxicity: involvement of its flavonoid constituents and protein konase C. J. Neurochem., 74, 2268, 2000.
3. Chung H.S. et al.: Ginkgo biloba extract increases ocular blood flow velocity. J. Ocul. Pharmacol. Ther., 15, 233, 1999.
4. Glovinsky Y. et al.: Retinal ganglion cell loss is size dependent in experimental glaucoma. Invest. Ophthalmol. Vis. Sci., 32, 198, 1991.
5. Kaiser T.J. et al.: Blood-flow velocities of the extraocular vessels in patients with high-tension and normal-tension open-angle glaucoma. Am. J. Ophthalmol., 123, 320, 1997.
6. Kazuyuki H. et al.: The *Ginkgo biloba* extract (EGb 761) provides a neuroprotective effect on retinal ganglion cells in a rat model of chronic glaucoma. Current Eye Research, 28, 153, 2004.
7. Neufeld A.H.: New conceptual approaches for pharmacological neuroprotection in glaucomatous neuronal degeneration. J. Glaucoma, 7, 434, 1998.
8. Ni Y. et al.: Preventive effect of *Ginkgo biloba* extract on apoptosis in rat cerebellar neuronal cells induced by hydroxyl radicals. Neurosci. Lett., 214,115, 1996.
9. Oken B.S. et al.: The efficacy of *Ginkgo biloba* on cognitive function in Alzheimer disease. Arch Neurol., 55, 1409, 1998.
10. Pittler M.H., Ernst E.: *Ginkgo biloba* extract for the treatment of intermittent claudication: a meta-analysis of randomized trials. Am. J. Med., 108, 276, 2000.
11. Quaranta L. et al.: Effect of *Ginkgo biloba* extract on preexisting visual field damage in normal tension glaucoma. Ophthalmology, 110, 359, 2003.
12. Raabe A. et al.: Therapeutic follow-up using automatic perimetry in chronic cerebroretinal ischemia in elderly patients. Protective double blind study with graduated dose *Ginkgo biloba* treatment. Klin. Monatsbl. Augenheilkd., 199, 432, 1991.
13. Ritch R.: Potential role for *Ginkgo biloba* extract in the treatment of glaucoma. Med. Hypotheses, 54, 221, 2000.
14. Stroman G.A. et al.: Magnetic resonance imaging in patients with low-tension glaucoma. Arch. Ophthalmol., 113, 168, 1995.
15. Vesper J., Hansgen K.D.: Efficacy of *Ginkgo biloba* in 90 outpatients with cerebral insufficiency caused by old age. Result of a placebo controlled double blind trial. Phytomedicine, 1, 9, 1994.

SUMMARY

Normal tension glaucoma is still a big therapeutic problem. In this form of glaucoma nervous cells damage is also caused by risk factors not associated with intraocular pressure. A significant role seems to be played here by the vascular factor as well as neuroprotective and antioxidant effect on the degree of glaucoma damage of the optic nerve. *Ginkgo biloba* is a raw material with potentially essential therapeutic action in the treatment of glaucoma due to such properties as: increase of peripheral and cerebral blood flow, neuroprotective and antioxidant action, inhibition of platelet-activating factor, apoptosis inhibition and decrease of blood viscosity. Promising results of former studies seem to justify considerable interest in *Ginkgo biloba* extract and its possible application in many diseases, including normal tension glaucoma.

Ginkgo biloba a jaskra

Jaskra z normalnym ciśnieniem wewnątrzgałkowym ciągle stanowi istotny problem terapeutyczny. W tej postaci jaskry za uszkodzenie komórek nerwowych odpowiedzialne są także niezwiązane z ciśnieniem wewnątrzgałkowym czynniki ryzyka. Istotne znaczenie wydaje się mieć czynnik naczyniowy oraz wpływ działania neuroprotekcyjnego i antyoksydacyjnego na stopień uszkodzenia jaskrowego nerwu wzrokowego. *Ginkgo biloba* jest surowcem o potencjalnie istotnym znaczeniu terapeutycznym w leczeniu jaskry ze względu na swoje właściwości, takie jak: zwiększenie obwodowego i mózgowego przepływu krwi, działanie neuroprotekcyjne i antyoksydacyjne, aktywność inhibitora czynnika płytkowego oraz hamowanie apoptozy i zmniejszanie lepkości krwi. Obiecujące wyniki dotychczasowych badań uzasadniają duże zainteresowanie wyciągiem z miłorzębu oraz jego potencjalnym zastosowaniem w wielu chorobach, w tym także w jaskrze z normalnym ciśnieniem wewnątrzgałkowym.