Pegem Journal of Education and Instruction, Vol. 15, No. 1 , 2025 (pp. 371-382)

WWW.PEGEGOG.NET RESEARCH ARTICLE

Investigating the Effects of Eye Drops Containing CoQ10 on Tear Production, Intraocular Pressure, Eyeball Diameter, and the Degree of Cataracts as Assessed by Slit Lamp Examination in Experimental Cataracts Induced in Rabbits

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Abstract

Background: Cataracts are a leading cause of vision impairment. Coenzyme Q10 (CoQ10), known for its antioxidant properties, has shown potential for ocular health. However, its effects on cataract progression and ocular parameters require further investigation.

Objectives: This study aimed to evaluate the effects of CoQ10-containing eye drops on tear production, intraocular pressure, eyeball diameter, and cataract severity in rabbits with experimentally induced cataracts.

Methods: Twelve healthy adult New Zealand white rabbits were divided into experimental and control groups. The experimental group received CoQ10 eye drops (0.3% w/v, two drops every eight hours), while the control group received artificial tears. Sodium selenite was used to induce cataracts, and ocular parameters were monitored using ultrasonography and slit lamp examination every two days. Data were analyzed with SPSS software.

Results: The experimental group showed significantly smaller eyeball diameters initially but significantly larger diameters on days 3, 7, 10, 17, and 20 compared to the control group. Intraocular pressure was higher in the experimental group initially but significantly lower on days 7 and 10. Tear production was significantly greater on days 7 and 17 in the experimental group. Cataract severity decreased by day 17 in the experimental group.

Conclusions: CoQ10 eye drops demonstrated beneficial effects on ocular parameters and reduced cataract severity in rabbits. These findings suggest CoQ10 as a potential therapeutic agent for managing cataracts and related ocular conditions.

Key words: *experimental cataract, eye drops containing CoQ10, changes in tear production, intraocular pressure, eyeball diameter.*

Introduction

In animals that rely heavily on their daytime vision, a dark melanin pigment is found between the photoreceptors and the choroid in the epithelial layer. This pigment layer absorbs light that passes through the photoreceptors without

How to cite this article: Fereshteh Kashi, Vria Tohidi, Farrokhreza Kabir, Bita Vazir. Investigating the Effects of Eye Drops Containing CoQ10 on Tear Production, Intraocular Pressure, Eyeball Diameter, and the Degree of Cataracts as Assessed by Slit Lamp Examination in Experimental Cataracts Induced in Rabbits. Pegem Journal of Education and Instruction, Vol. 15, No. 1, 2025, 371-382

Source of support: Nil **Conflicts of Interest:** None. **DOI: 10.48047/pegegog.15.01.32**

Received: 12.11.2024 **Accepted:** 15.12.2024 **Published:** 01.01.2025

stimulating them. If the absorbed light spreads toward the exposed retina, it can weaken both visual acuity and overall eyesight. (1, 2, 3). In contrast, nocturnal animals and many domesticated mammals possess a reflective layer in the choroid called the tapetum. In these animals, the overlying epithelial layer does not contain light-absorbing pigment. This arrangement allows unabsorbed light to be reflected onto the retina, enhancing the utilization of available light and improving visual acuity. The reflection from the tapetum is what causes the eyes of nocturnal animals to shine in the dark (1, 2, 3). When the light passes through the transparent cornea, optical refraction occurs, which helps to focus the image on the retina. After passing through the cornea, the light enters the anterior chamber. Both anterior and posterior chambers are filled with aqueous humor, which is responsible for nourishing the cornea and lens. The iris, a pigmented structure made up of smooth muscle fibers, separates the anterior and posterior chambers. These muscle fibers are arranged in a way that allows them to contract and expand, thereby changing the pupil size (1, 2, 3).

Behind the iris lies the lens, which is held in place by suspensory ligaments attached to the ciliary body. The lens adjusts its shape to focus images of objects at varying distances onto the retina. Behind the lens is a chamber filled with vitreous fluid. The viscosity of this fluid, combined with the pressure from the aqueous humor and the relatively inelastic nature of the sclera and cornea, gives the eyeball its spherical shape. The vitreous fluid also contains phagocytic cells (1, 2, 3).

In both humans and animals, ultrasonography can be used to examine the eyeball and pupil. To perform an ocular ultrasound, a transducer with frequencies higher than 7.5 MHz is required, along with a system that can record images in brightness and amplitude modes. Ultrasound equipment typically used for examining the heart and the ventricular area can also be utilized for ocular ultrasound in animals, provided it operates at frequencies above 7.5 MHz. Both sector transducers and linear transducers are suitable for ocular ultrasound. Standoff pads are generally unnecessary for ocular ultrasound; however, when using sector transducers to examine the anterior parts of the eye, it is advisable to utilize standoff pads (4, 5). The following parts of the eye can be evaluated using brightness-mode ultrasound: the cornea, anterior chamber, iris, ciliary bodies, lens, vitreous chamber, and the posterior part of the eyeball (4).

Various methods have been proposed to prevent cataracts. Mydriatic drugs, for example, increase the amount of light that passes through the lens, alleviating some symptoms of cataracts. Additionally, aldose reductase inhibitors are recommended for preventing cataracts related to diabetes. Studies have indicated that using this inhibitor—both locally and systemically—can slow

or halt the progression of cataracts in diabetic patients. Since oxidation plays a crucial role in the development of cataracts, antioxidants may also help prevent or stop their progression. Research has found that administering antioxidants such as selenium, vitamin E, orgotein, zinc ascorbate, and carnosine—both topically and systemically—can be effective in treating and preventing cataracts (6, 7).

Diagnosing cataracts is often straightforward. It is possible to diagnose cataracts before birth, although this can be challenging. Ultrasonography is an effective method for diagnosing cataracts in unborn children. Timely diagnosis of cataracts in infants immediately after birth is critical, as the absence of proper treatment could lead to blindness. Conventional ophthalmological tests are the most widely used method for diagnosing age-related cataracts. Since cataracts progress over time, they are often noticed only when vision begins to decline, making regular eye examinations especially important for the elderly (8, 9).

Cataracts refer to any opacity in the lens that leads to reduced vision. They occur due to the loss of the natural structure of the fibrous layer of the lens or its core, resulting in decreased transparency (10 and 11). There is a direct relationship between increasing age and the occurrence of cataracts; about half of the population over 80 years old in the United States suffers from this condition to varying degrees. The lens has a surface epithelium that grows inward at the equator. As the lens continuously produces new tissue, this tissue cannot penetrate elsewhere, causing significant compression of the lens with age. This compression is often associated with the deposition of yellow pigment. The presence of yellow pigment, along with compression, reduces light penetration and, consequently, visibility (12).

High osmotic pressure is caused by the excessive accumulation of sorbitol and fructose derived from glucose, particularly evident in cataracts induced by diabetes. In this condition, water infiltrates the lens. The increase in water within the lens fibers alters the transmission mechanism, ultimately leading to the rupture of the fibers (12, 13). Cataract-related changes involve a series of biochemical events in the lens, with oxidative damage being a key factor in the development of cataracts. These oxidative changes affect lens proteins, causing large insoluble proteins to aggregate and attach to the cell membrane of the lens fibers. In addition to common oxidants and superoxidants such as hydrogen peroxide, superoxides, and hydroxyl radicals, the lens also faces a challenge known as photooxidation. Epidemiological studies indicate that exposure to sunlight increases the incidence of cataracts by accelerating oxidative processes within the lens (12).

The formation of cataracts involves complex processes, including the Maillard reaction and autoxidation facilitated by carbonyl cofactors. The Maillard reaction is a series of chemical reactions

that begins with the combination of carbonyl and amino groups and culminates in the production of brown-colored melanoid polymeric compounds. These reactions occur between the carbonyl groups of carbohydrates—derived from aldehydic agents like glucose and fructose—and the free amino groups of proteins, particularly the amino group of the amino acid lysine in the protein polypeptide chain. This process alters the protein structure, modifies its biological properties, and forms new chemical connections between protein molecules (14). Since part of the carbonyl compounds are detoxified by glutathione, an enzymatic antioxidant, and NADPH, the amounts of glutathione and antioxidants will be significantly depleted as the carbonyl compounds accumulate. The Maillard reaction in cataracts, much like many other diseases, plays a central role in the emergence and progression of cataracts, and managing this process may greatly influence public health. Antioxidant enzymes such as catalase, superoxide dismutase peroxidase, glucose 6-phosphate, glutathione reductase, and dehydrogenase are particularly effective in protecting the lens membrane against oxidative stress. Catalase and superoxide dismutase inhibit the Na/K ATPase pump, which is in the cell membrane and is effective in the occurrence of protein and lipid oxidation (15, 16). Considering the abovementioned, the present study aims to explore the effects of eye drops containing CoQ10 on tear production, intraocular pressure, eyeball diameter, and the degree of cataracts as assessed by slit lamp examination in experimental cataracts induced in rabbits.

Materials and Methods

To perform the present research, first, 12 healthy adult male New Zealand white rabbits, each weighing approximately 2 to 3 kg, were acquired from the Pasteur Institute of Tehran. The rabbits were then transported to a designated area where they underwent a clinical examination to confirm their health status. Each rabbit was weighed and received an anti-parasitic pill, and its information was documented on a special form. The rabbits were carefully numbered and housed individually in separate cages for two weeks. During this period, they were fed with a high-quality ration for rodents. After two weeks and renewal of clinical examinations, the following were performed to ensure their eye health: ophthalmological examinations using an ophthalmoscope and a slit-lamp biomicroscope equipped with a digital camera; examination of the external surface of the eye to confirm the absence of corneal injury and lens opacity; and measurement of intraocular pressure using tonometry.

After ensuring that their eyes were healthy, the normal shape of the cornea, the normal size of the lens, and other eye structures were examined using ultrasonography. The ultrasound examination was conducted without the need for anesthesia or local anesthetics; only physical restraint was utilized. To perform corneal ultrasound, sufficient sterile

ultrasound gel was used and the probe was covered with a strile cover. A high-frequency transducer was positioned horizontally on the cornea.

All examinations were performed and recorded by a single person on images where the cornea, anterior capsule of the lens, posterior capsule of the lens, and the optic disc were in the same direction. The ultrasound settings remained consistent throughout all tests, with depths set at 1.5 and 115 Gain, and a frequency ranging from 11.3 to 16 using a linear probe. After each ultrasound, the rabbits' eyes were rinsed with a normal saline solution for 45 seconds.

Then, the rabbits were divided into two groups: a control group that received artificial tear drops and a experimental group that received eye drops containing CoQ10. After accurately weighing the rabbits, a 0.1% solution of sodium selenite, manufactured by Merck, Germany, was prepared by dissolving 99.3% sodium selenite in injectable normal saline. This solution was injected subcutaneously into all the rabbits at the back of the neck, with a dosage of 1 mg/kg of body weight. On the third and sixth days after re-weighing the rabbits, sodium selenite was re-injected subcutaneously with the same dosage of 1 mg/kg of live weight. In the experimental group, eye drops containing CoQ10 were used daily, with two drops administered every eight hours. A pharmacologist prepared eye drops containing CoQ10. For this purpose, 300 mg of soluble CoQ10 was dissolved in sufficient distilled water to obtain a solution with a concentration of 0.3% w/v. To preserve the solution, 0.001% Benzalkonium chloride was added. The solution was then sterilized by passing it through a 0.2 µm pore size filter and placed into a sterile dropper for use.

Eye drops containing CoQ10 were administered to the experimental group starting on the first day of sodium selenite injection. Intraocular lens turbidity, aqueous turbidity, retinal conditions, corneal thickness, lens thickness, eyeball diameter, and changes in the anterior and posterior lens capsules were examined using a SonoScape ultrasound machine every two days. Slit-lamp biomicroscopy was performed every three days, and cataracts were graded according to Table 1.

Table 1: The results of the Shapiro-Wilk test for examining the normality of research variables in the treatment and control groups

Variable	Experimental group		Control group	
	Shapiro-Wilk	sig	Shapiro-Wilk	sig
Eyeball diameter	0.96	0.78	0.91	0.34
Intraocular pressure	0.90	0.27	0.98	0.91
Tear production	0.86	0.25	0.85	0.10

Intraocular pressure was measured every other day using a fixed iCare tonometer. Tear production was assessed every other day with Schirmer strips. All experiments continued for 28 days, and on the 28th

day, final grading was conducted for each sample, with results recorded separately for each group.

The data were analyzed using SPSS software. Central indicators such as mean and bar charts were utilized to assess the data. The Shapiro-Wilk test was performed to examine the normality of the variables and independent-sample t-tests were carried out to compare the variables between the two groups based on measurement frequency.

Results

The Shapiro-Wilk test was used to examine the distribution of research variables in the control and experimental groups. The results are listed in Table (1).

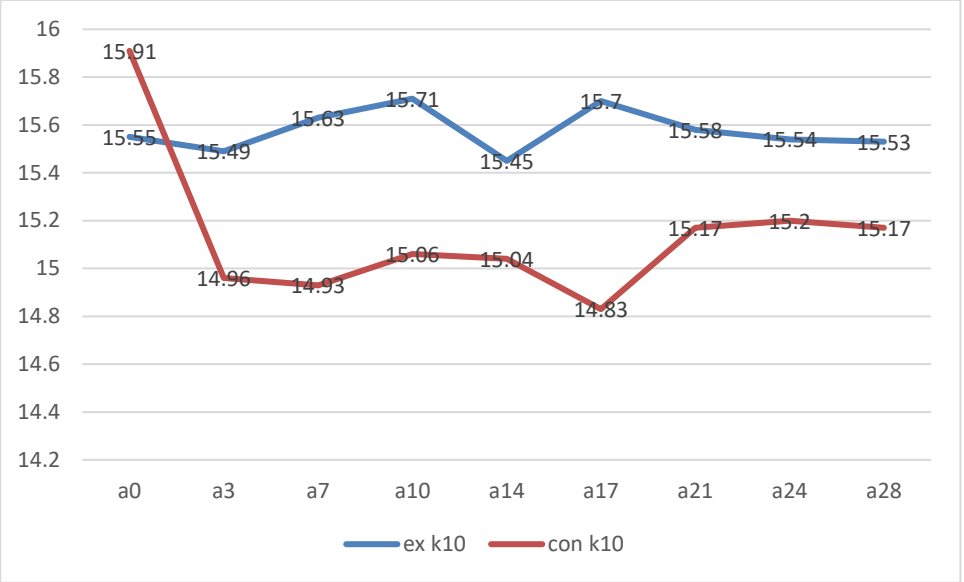
Table (1) shows Sig.>0.05 for the research variables in the treatment and control groups, implying that the distribution of research variables in the treatment and control groups was not significantly different from the normal distribution at the significance level of 0.05 (sig. >0.05).

According to Table (2), Diagram (1), and the results of the t-test, in the first measurement, the average eyeball diameter in the experimental group was significantly less than in the control group. On the third, seventh, tenth, seventeenth, and twentieth days, the eye diameter in the experimental group was significantly greater than in the control group. No significant difference was observed between the treatment and control groups in the eyeball diameter on the 14th, 24th, and 28th days.

Table 2: Examination of eyeball diameter in the treatment and control groups

group	a0	a3	a7	a10	a14	a17	a21	a24	a28
ex	15.55	15.49	15.63	15.71	15.45	15.70	15.58	15.54	15.53
con	15.91	14.96	14.93	15.06	15.04	14.83	15.17	15.20	15.17
t	-2.742	3.872	6.925	4.620	1.756	7.161	2.134	1.815	1.877
p	0.021	0.000	0.000	0.000	0.057	0.000	0.048	0.065	0.061

Diagram 1: The average eyeball diameter in the treatment and control groups

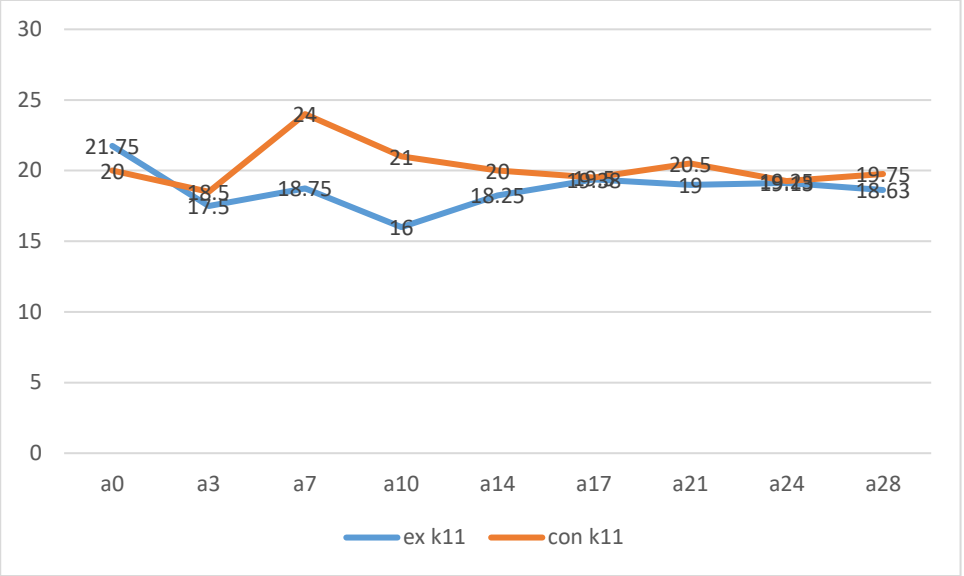


According to Table (3), Diagram (2), and the results of the t-test, in the first measurement, the average intraocular pressure in the experimental group was significantly higher than in the control group. On the seventh and tenth days, it was significantly lower in the experimental group than in the control group. No significant difference was observed between the treatment and control groups in the intraocular pressure on the remaining days.

Table 3: Examination of intraocular pressure in the treatment and control groups

	Mean								
Group	a0	a3	a7	a10	a14	a17	a21	a24	a28
Ex	21.75	17.50	18.75	16.00	18.25	19.38	19.00	19.13	18.63
Con	20.00	18.50	24.00	21.00	20.00	19.50	20.50	19.25	19.75
T	2.932	-1.557	-11.560	-7.454	-1.790	-0.196	-1.879	-0.189	-1.978
P	0.015	0.151	0.000	0.000	0.104	0.849	0.090	0.854	0.076

Diagram 2: The average intraocular pressure in the treatment and control groups

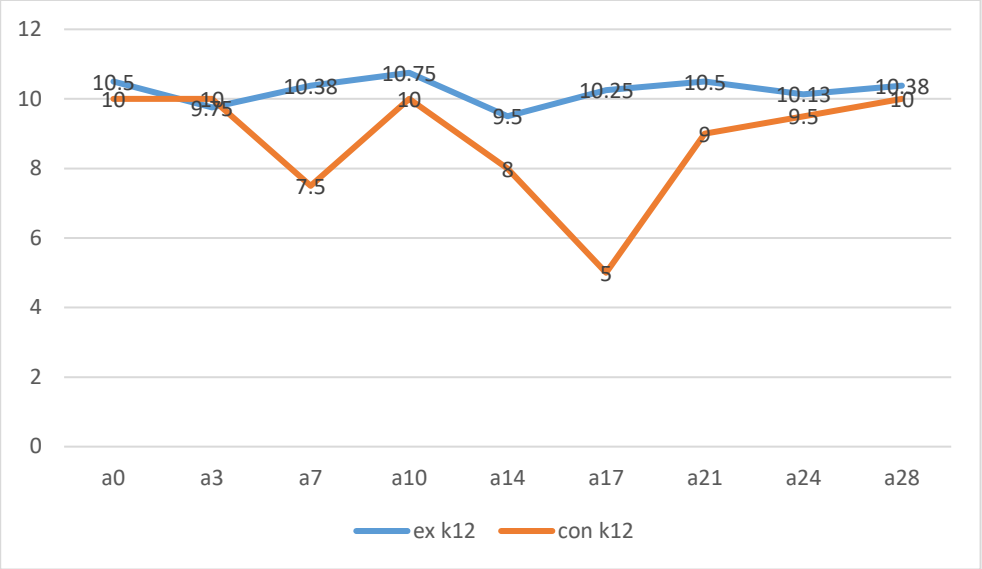


According to Table (4), Diagram (3), and the results of the t-test, on the seventh and seventeenth days, the average tear production in the experimental group was significantly greater than in the control group. There was no significant difference between the treatment and control groups in the average tear production on the other days.

Table 4: Examination of tear production in the treatment and control groups

Group	a0	a3	a7	a10	a14	a17	a21	a24	a28
Ex	10.50	9.75	10.38	10.75	9.50	10.25	10.50	10.13	10.38
Con	10.00	10.00	7.50	10.00	8.00	5.00	9.00	9.50	10.00
T	0.443	-0.309	2.977	0.835	1.142	22.136	1.826	0.937	0.562
P	0.667	0.764	0.014	0.423	0.280	0.000	0.098	0.371	0.587

Diagram 3: The average tear production in the treatment and control groups



Based on studies conducted using a slit-lamp biomicroscope, all rabbits in the control group developed incipient cataracts in both eyes by the 10th day. The condition worsened by the 14th day, with immature cataracts being observed in both eyes. In contrast, the experimental group experienced a delayed onset of cataracts. By the 14th day, cataracts were noted in only 50% of the rabbits in the experimental group, with half of the eyes exhibiting incipient cataracts and the other half showing immature cataracts. By the 17th day, the severity of cataracts in the affected rabbits had decreased; incipient cataracts were seen in only one eye of the involved rabbits. From the 21st day, no symptoms of cataracts were observed in any of the groups during the slit-lamp biomicroscopy examination.

Discussion

The results indicated that in the first measurement, the average eyeball diameter in the experimental group was significantly less than in the control group. On the third, seventh, tenth, seventeenth, and twentieth days, the eye diameter in the experimental group was significantly greater than in the control group. No significant difference was observed between the treatment and control groups in the eyeball diameter on the 14th, 24th, and 28th days. Furthermore, in the first measurement, the average intraocular pressure in the experimental group was significantly higher than in the control group. On the seventh and tenth days, it was significantly lower in the experimental group than in the control group. No significant difference was observed between the treatment and control groups in the intraocular pressure on the remaining days. On the seventh and seventeenth days, the average tear production in the experimental group was significantly greater than in the control group. There was no significant difference between the treatment and control groups in the average tear production on the other days.

The results also revealed that, By the 17th day, the severity of cataracts in the affected rabbits had decreased; incipient cataracts were seen in only one eye of the affected rabbits. From the 21st day, no symptoms of cataracts were observed in any of the groups during the slit-lamp biomicroscopy examination.

There are reports regarding the control of cataract progression through non-surgical methods, utilizing various substances experimentally in rats and rabbits. In these studies, researchers induced cataracts using methods such as feeding large amounts of glucose to create diabetic cataracts, exposing subjects to ionizing radiation, administering subcutaneous injections of sodium selenite in immature rats, and feeding naphthalene to rabbits. Following the induction of cataracts, they attempted to slow or halt the progression using various inhibitors, such as carbonic anhydrase enzyme, and antioxidants like erdosteine, N-acetyl carnosine, and vitamin E. The results indicated success in slowing down cataract progression. (17,18).

Rita Mencucci et al. (2014) demonstrated that CoQ10 functions as an antioxidant, protecting cells from damage caused by free radicals. Their study found that eye drops containing CoQ10 can help reduce damage from ultraviolet radiation both in vitro and in vivo. Additionally, the use of CoQ10 eye drops was shown to maintain mitochondrial efficiency during ultraviolet exposure and to enhance wound healing following the removal of the epithelium (19).

BAHA'A A. ABDUL-HUSSEIN et al. (2017) conducted research demonstrating that CoQ10 exhibits antioxidant properties when used as eye drops and can help prevent lens opacity, which is a primary cause of cataracts resulting from oxidative stress. In their study, they found that the average lens opacity score in the control group, which used distilled water, was 4. In contrast, the average score for the test group that used CoQ10 was only 1.5, highlighting a significant difference between the two groups. Lens opacity was assessed and graded through ophthalmoscopy. The cataract induced in this study was identified as posterior subcapsular (PCS) and was observed 48 to 72 hours after the disease was induced (20).

Kabir et al. (2009) found that injecting sodium selenite at a dosage greater than 1.5 mg/kg of a rabbit's live weight to induce experimental cataracts led to the death of the rabbit within a maximum of 18 hours (21). As a result, they used lower doses, discovering that a subcutaneous injection of 0.01% sodium selenite at a dosage of 1 mg/kg of body weight on the zero, third, and sixth days could lead to the formation of Posterior Subcapsular Cataract (PSCC). It was observed that this dosage induced cataracts in both immature and adult rabbits, while in the tested mice, nuclear cataracts occurred only in newborns. In this study, all of the rabbits (100%) developed cataracts by the fourth day, which peaked on the eleventh day. Furthermore, none of the rabbits died, and no

abnormal complications were noted in other parts of their bodies.

Conclusion

The results of ultrasound and slit-lamp biomicroscopy examinations indicate that using eye drops containing CoQ10 not only delays the onset of cataracts but also significantly reduces both the progression and severity of cataract development.

Conflict of interest

The authors declare that they have no conflict of interest.

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