Finding evidence of efficacy and safety of antioxidant vitamin supplementation in preventing and slowing the progression of age-related cataract

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Diploma thesis

Academic year:
2021/2022

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LIST OF ABBREVATIONS

ARNC – age-related nuclear cataract

CAT – catalase

IOL – intraocular lens

IOP – intraocular pressure

LDL – low density lipoprotein

NADPH – reduced nicotinamide adenine dinucleotide phosphate

RCT – randomized controlled trial

RDA – recommended dietary allowance

RNS – reactive nitrogen species

ROS – reactive oxygen species

SOD – superoxide dismutase

1. INTRODUCTION

1.1 Cataract

1.1.1 Epidemiology

The most prevalent etiology of visual impairment and blindness in humans worldwide are lens cataracts (1). Even though lens cataracts are commonly a curable disease (2), in 2014 they affected an estimated 95 million people worldwide (3).

According to studies there are 36 million blind people worldwide and 12 million of these people are blind due to cataracts (4,5). By estimation 68% of people over 79 years of age have some type of cataract or reduced lens opacity (6). Women are at a higher risk than men to have an impaired eyesight, around the globe the prevalence ration of this between female and male individuals is 1.5 to 2.2 (7). Over the last 20 years, an improvement of active surgical initiatives and techniques has led to a decline in cataract prevalence (8).

1.1.2 Etiology

The major risk factor for cataracts in humans is age, but in general it is considered to be "multifactorial" (9). Environmental and genetic factors increasing the risk of developing cataract include: ultraviolet light exposure, smoking cigarettes, an array of disease e.g., diabetes, uveitis, but also IOP-lowering medications/surgery, steroid usage, trauma and lastly certain occupations (10). Medications known to start cataractogenesis besides corticosteroids, include busulfan, miotics, phenothiazines and rarely amiodarone (7). Factors about genetic susceptibility in age-related cataract are not well researched yet (11).

1.1.3 Structure and function of the lens

The lens helps to focus and refract light onto the retina, for this purpose it is biconvex and transparent in the healthy eye. It consists of a thin capsule surrounding fibers supported by zonules on the sides. The lens fibers migrate to the center from the periphery and are created from the lens epithelium (3). This means that newest fibers are on the outermost layer, the cortex, and the oldest fibers are in the nucleus, the center (3).

1.1.4 Classification

In an eye diseased with cataract the lens is opacified and significantly interferes with vision (9). If we classify cataracts by the cause, we have three different types: 1. age-related cataract, 2. pediatric cataract and 3. cataracts secondary to other causes.

The most common type of cataract in adults is age-related cataract with an usual age of onset between 45 and 50 years (3). As the direct consequence of oxidative stress, we get the opacification of the lens (12).
Age-related cataracts can be subdivided on account of the location of the opacification in the lens into nuclear cataract (ARNC), cortical cataract, mixed cataract (nuclear and cortical) and posterior subcapsular cataract, making the main four different types of cataracts (1).

The cells of the human eye lens, which have the highest metabolic activity are the epithelial cells. They undergo insolubilization, crosslinking and oxidation. Migrating to the equator of the lens, these cells get gradually compressed centrally and form an opacity and result in nuclear sclerosis and the so-called nuclear cataract (12), which is usually colored (9).
For cortical cataract it is characteristic that it starts of at the cortex, often wedge-shaped and then extends to the center of the lens (3). When in the axial posterior cortical layer, a plaque-like opacity develops, we diagnose the posterior subcapsular cataract (3). More than one type of cataract is found in the majority of patients (3).

1.1.5 Signs and symptoms of age-related cataract

The most common symptoms of patients with cataract are blurred vision as well as haloes and glares from lights (3). Distance vision is usually more affected than near vision in nuclear cataracts, while near vision is typically more reduced with posterior subcapsular cataracts (3). Furthermore, the eye gets more myopic, because the cataractous lens with progressive nuclear sclerotic changes can refract more light than the healthy lens (3). This change in the refractive index can be corrected with glasses, if it is not corrected the affected person will experience a decline in the ability of distance vision and a paradoxical improvement in the ability of near vision (3). Glare is frequent in people with posterior subcapsular cataracts. When the refractive index of the opacification of the human eye lens varies between eyes, the affected subject can complain of monocular diplopia. Other patients may only experience visual disability when attempting to perform daily activities for example driving or reading (3).

1.1.6 Treatment of age-related cataracts

The present-day standard of treatment for cataracts significantly affecting vision is the surgical removal of the human eye lens diseased with cataract and its following replacement with an IOL. A human lens cataract is considered to significantly affect vision at a visual acuity of 20/40 or worse (13). Worldwide it is one of the most frequently performed procedures and

between the oldest procedures as well as counting as one of the most successful treatments in medicine (14). This surgery is indicated when the impact and severity of the loss of vision override the risks of the surgery (3). The outcome of these surgeries is unrelated to preoperative visual acuity (15). Positive surgical outcomes depend, even with improving surgical techniques and technologies, still on preoperative assessment, and proper intraoperative and postoperative care and support as well as accurate intraocular lens power measurements.

Cataract surgery is often performed as a day-case procedure (16) meaning that the patient gets admitted to the hospital for the planned surgical procedure and returns home on the same day. No routine medical investigation is required to take place before cataract surgery as it does not affect the outcome (17), nor does anticoagulant and antiplatelet medication have to be discontinued, as there was no increase in vision-threatening hemorrhages (18).

1.1.6.1 Preoperative assessment

The patient is examined for ocular comorbidities that could influence postoperative prognosis with a detailed ophthalmic examination including a slit lamp examination, fundus assessment, intraocular pressure, refraction and visual acuity testing. If no direct visualization of the retina is possible a B-scan ultrasonography can be performed (3).

After removal of the cataract an intraocular lens is implanted which can also correct refractive errors, for this biometry has to be performed, which is the accurate calculation of intraocular lens power (3).

There are three different anesthesia options in cataract surgery: general, local and topical (3). The most frequently used anesthesia in developing countries is local (3).

1.1.6.2 Cataract extraction

The evolution of cataract surgery reaches from intracapsular cataract extraction over extracapsular cataract extraction to phacoemulsification (3). Intracapsular cataract extraction is at this point in time mostly used in less-developed countries and has been replaced mostly by modern cataract surgery (19). During extracapsular cataract extraction the posterior capsule remains intact and it enables the implantation of the intraocular lens to in the capsular bag, which is made possible by a limbal incision followed by an anterior capsulotomy, from which the lens cortex and nucleus are expressed manually. This procedure has the advantage of

enhanced anatomic stability (3). This procedure does not require expensive equipment, which makes it very popular (20).

The procedure of choice today is phacoemulsification. In phacoemulsification the lens capsule is opened anteriorly, which is also called capsulorhexis. After this an ultrasonic handpiece is used to emulsify the lens and to aspirate it through the incision with the dimensions 2.2-3.2 mm.
Before the implantation of the intraocular lens can be made possible, the anterior chamber of the eye gets injected with an ophthalmic viscoelastic device, which replaces the aqueous humour. The use of this is to maintain the intraocular spaces to have room for all the instruments and their safe passage into the eye. Through the use of the ophthalmic viscoelastic device and its properties the corneal endothelium is protected from damage. In modern cataract surgery the use of these devices is obligatory, they are considered non-toxic and are optically clear.
Following this part of the procedure the next step is the implantation of an IOL into the bag of the capsule (3). For this to happen trough the smaller incision, foldable IOLs have been developed and can be inserted either by a special forceps into the capsular bag or they can be loaded into the cartridge of an intraocular lens injector and then be injected into their place. After this last step the ophthalmic viscoelastic device can be aspirated out of the anterior chamber (3).
Phacoemulsification's advantage over extracapsular cataract extraction is the smaller incision, which reduces surgical complications and accelerates visual rehabilitation (3).

The newest procedure is femtosecond laser-assisted cataract surgery, which first made an occurrence in 2010. It offers the possibility of automating some steps of cataract surgery through the use of a laser and before continuing with phacoemulsification. Since the clinical benefits of this procedure do not outweigh the negative cost-effectiveness, it is not widely used yet (3).

1.1.6.3 Postoperative management and follow-up

The representative follow-up schedule for age-related cataract consists of patient examinations on day one, after one week and after one month and three months following the surgery, but this schedule is not internationally standardized. The accompanying pharmacological treatment consists of topical antibiotics, corticosteroids or non-steroidal anti-

inflammatory dungs for one to four weeks, with this duration varying as well between countries, doctors and patients and being longer used in patients with complications (3).

Usually, patients have a delayed sequential bilateral cataract surgery, which means one eye is operated on and the second eye is scheduled some time after the first eye, to avoid bilateral complications such as endophthalmitis and to give the possibility of improvement and for tailoring the second procedure or choice of intraocular lens for the second eye with the experience made during the procedure on the first eye. When both eyes undergo cataract surgery at the same time, we call it immediate sequential bilateral cataract surgery, which provides quicker visual rehabilitation, saves costs and time, but is still a highly debated issue and not the current standard of care.

1.1.6.4 Complications

Complications and their prevalence are listed in the table below.

	Prevalence
Intraoperative complications	
Posterior capsule rupture with or without vitreous loss	0·5–5·2%
Intraoperative iris floppy syndrome or iris prolapse	0·5–2·0%
Iris or ciliary body injury	0·6–1·2%
Lens materials dropped into vitreous	0·002–0·2%
Suprachoroidal effusion with or without haemorrhage	0–0·4%
Early postoperative complications	
Transient elevated intraocular pressure	0·3–18·1%
Corneal oedema	0·1–5·4%
Toxic anterior segment syndrome	0·1–2·1%
Intraocular lens decentration or dislocation	0·1–1·7%
Retained lens materials	0·5–1·7%
Wound leak or rupture	0·02–1·1%
Hyphaema	0·02–0·1%
Endophthalmitis	0·006–0·04%
Late postoperative complications	
Posterior capsule opacification	0·3–28·4%
Clinical cystoid macular oedema	1·2–11·0%
Pseudophakic bullous keratopathy	0·3–5·4%
Anterior capsule fibrosis and phimosis	0·47–3·3%
Chronic uveitis	1·1–1·8%
Retinal tear or detachment	0·1–1·3%
Endophthalmitis	0·017–0·05%

Table: Prevalence of complications of cataract surgery

Table 1. The prevalence of various complications of cataract surgery (3).

1.1.6.5 Outcomes

With cataract surgery, the best-corrected visual acuity, that can be achieved as an outcome in 84-94% of eyes is 20/30 or better at six months after surgery, which makes it a safe and effective procedure. Good long-term visual rehabilitation is proven by studies reporting the 10-year and 15-year outcomes (21,22). A beneficial effect on the patient's quality of life, especially the social and emotional life as well as a decrease in all-cause mortality and concurrent extended long-term survival for old people are additional outcomes of cataract surgery (23–25).

1.1.7 Costs of cataract surgery and quality of life

With regard to the aging population around the globe, not only the economic but also the social costs of cataract surgeries are quiet staggering and the demands for surgery exceed by far the limited healthcare resources (26). Furthermore, cataract surgery improves not only vision, but quality of life, through improving the performance of critical daily tasks, for example: recognizing people, reading the newspaper or books, watching TV, driving, cooking, sewing, knitting, doing handcrafts, noticing traffic and negotiating steps (23).

1.2 Oxidative stress

1.2.1 Introduction

In a state of disbalance between oxidants and antioxidants, with antioxidants making up the majority of the share, we get a condition named oxidative stress. The two routes leading to oxidative stress are either a surplus of oxidants or a lack of antioxidants (27). Oxidative stress stands in relation to various aging processes and disease, among them also age-related cataract (27).

1.2.2 Oxidants

In the processes of cell signaling and cell metabolism reactive species are generated including free radicals and non-radicals, the most popular being reactive nitrogen species (RNS) and reactive oxygen species (ROS) (27). Any chemical species with an unpaired electron is defined as a free radical (28). These unpaired electrons change the chemical reactivity of atoms and molecules to an increased state (28). Immoderate amounts of reactive species can lead to oxidative stress and be potentially harmful, while smaller amounts of the same are needed for proper cell functioning and cell signaling (27). Targets for oxidation are not only long-chain polyunsaturated fatty acids, but also proteins and nucleic acids (27).

ROS production is influenced by endogenous factors such as genetics, mitochondria, peroxisomes, inflammation (cytokines) and antioxidant activity, as well as by environmental (exogenous) factors like radiation, tobacco, pollutants, xenobiotics and drugs and dietary antioxidants (27).

A common ROS and free radical produced among other things during cell metabolism by many cells is the superoxide anion ($O_2^{\bullet-}$). When superoxide reacts to form hydrogen peroxide (H_2O_2), which is a non-radical, it can in turn generate hydroxyl (OH•), which is a very reactive radical that can then cause oxidative damage. An example for a RNS would be superoxide reacting with nitric oxide (NO•) to form peroxynitrite (ONOO-). Nitric oxide is known in the body as a very important molecule in cell signaling and blood pressure regulation in the central nervous system (27).

1.2.3 Antioxidants

Substances that have the ability to prevent, delay or remove oxidative damage from a target molecule are antioxidants (27). Antioxidants can donate one or more electrons to reactive species', reducing the formation of the same, they can also take part in redox-reactions (27). Oxidized forms of antioxidants can, by other antioxidant enzymes or systems, be reduced back into their reduced/active state (27).

Antioxidants can be either produced in the body, so called endogenous antioxidants or they can be exogenous, meaning they are dietary. These two types of antioxidants can combine into a complex network and interdependently act against oxidative stress (29). Antioxidants can be further divided into enzymatic and non-enzymatic antioxidants (27).

Antioxidant activity is influenced by different factors, *inter alia*, the redox-state of a cell, genetic factors, health conditions and diet (27).

Transcription factors for inflammation and oxidative stress play an important role in the cellular defense against oxidative stress as they regulate the expression of various antioxidants. Examples of these include a metabolite of vitamin A (retinoic acid), nuclear factor κβ, and nuclear factor E2-related factor 2 (Nrf2) (30,31). When transcription factors bind to responsive elements on the genes, this eventuates in the activation of signaling pathways, an example for this would be Nrf2 binding to an antioxidant response element, enabling a sequence found on a gene encoding antioxidant enzymes (32).

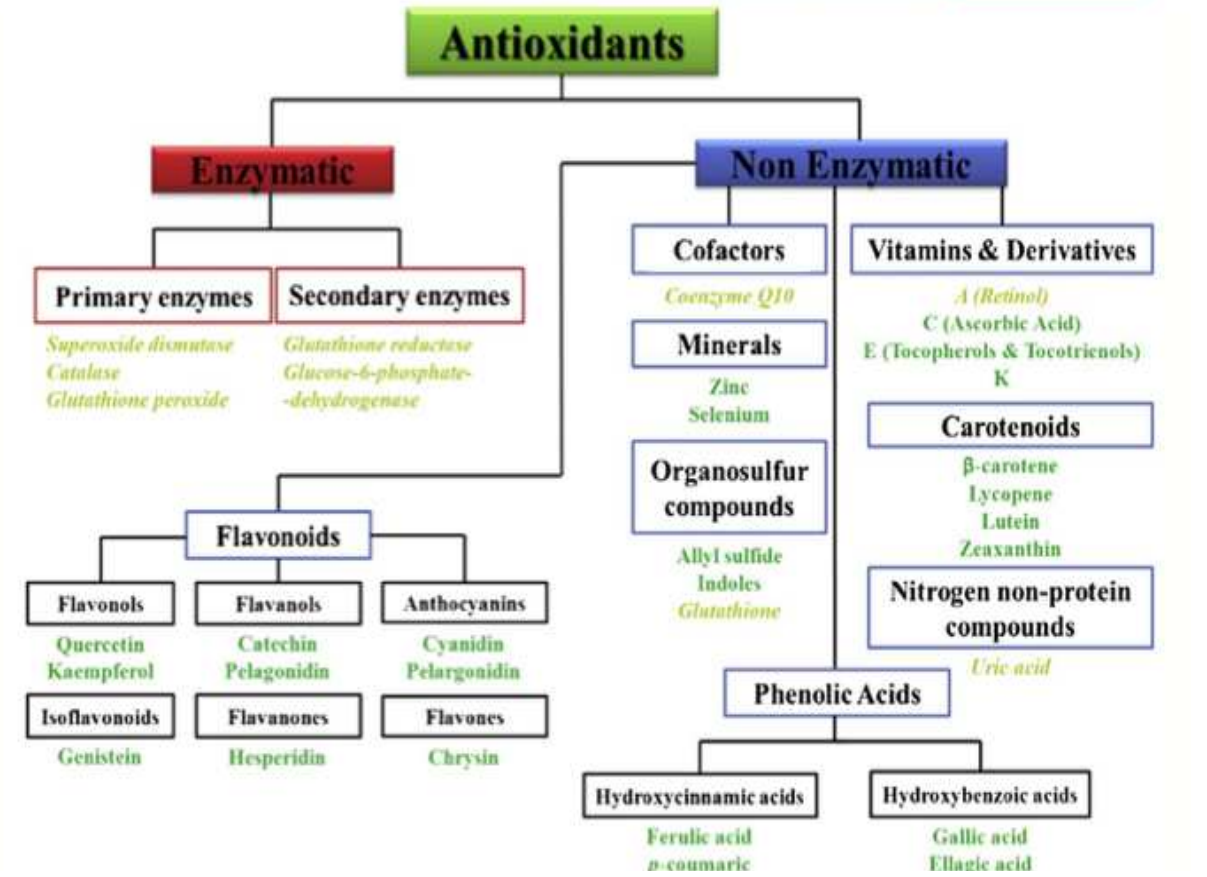

Figure 1: Different classes of antioxidants (33)

1.2.3.1 Endogenous antioxidants

Endogenous antioxidants can be either enzymatic or non-enzymatic (27). Belonging to the enzymatic antioxidants that neutralize the reactive species are catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase and glutathione reductase (33).

1.2.3.1.1 Superoxide dismutase

Superoxide dismutase catalyzes the transition of superoxide anion, a radical, into hydrogen peroxide, a non-radical (33). It belongs to the family of multimeric metalloenzymes and is located in cytosol and mitochondria and extracellular (27,33). Superoxide dismutases are further divided into different groups: Cu-SOD, Cu-Zn-SOD, Fe-SOD, Mn-SOD and Ni-SOD. The various superoxide dismutases are found in several locations (33).

Cu-Zn-SOD is essential for the existence of aerobic life, it is mostly present in the cytosol and chloroplast of eukaryotic cells. Furthermore, it has been established that it is among the most important and strongest providers of defense against oxygen toxicity inside the cell (34). Mn-SOD is predominantly present in cytosol of bacteria and the matrix of mitochondria. The SOD

found in prokaryotes and some plants is Fe-SOD (35). In general SODs have a key-role in shielding proteins and enzymes against oxygen toxicity in eukaryotes as well as prokaryotes (33,36–38).

1.2.3.1.2 Catalase

Catalase catalyzes the formation of H_2O and O_2 from H_2O_2 (hydrogen peroxide), but it also reacts with many other substrates (39). It mostly exists in the peroxisomes of mammalian cells, and it was the first antioxidant enzyme that was ever discovered and is thus the oldest known one (39).

1.2.3.1.3 Glutathione peroxidase and glutathione reductase

Glutathione peroxidase requires reduced glutathione as a substrate. Its action against oxidative stress is considered a second line defense (40). Glutathione peroxidase catalyzes the reduction of H_2O_2 to H_2O as well as corresponding alcohols, while using glutathione as a cofactor (41).

It performs as a multicomponent antioxidant defense system protecting the cell membrane against polyunsaturated fatty acids (42). Moreover, it can be subdivided into selenium dependent and selenium independent glutathione peroxidase, which are both found in the mitochondria and the cytosol (33).

Glutathione reductase reduces oxidized glutathione by using NADPH. It is known as a cytosolic protein with a distribution pattern similar to that of glutathione peroxidase. Its main role is the maintenance of the ration of glutathione to oxidized glutathione, when this ratio shifts and we have a surplus of oxidized glutathione inside of the cell it can lead to protein denaturation, lipid peroxidation and even DNA damage (43).

1.2.3.1.4 Glutathione

An important non-enzymatic antioxidant is glutathione. It is produced in the body from various amino acids. Its role is the reduction of disulfide bonds to prevent oxidative damage (27). It occurs in two states: in the reduced one as glutathione and in the oxidized one as glutathione disulfide (27). An additional role of glutathione is its importance for the proper functioning of other antioxidants like glutaredoxins/thioltransferases (44).

1.2.3.2 Exogenous antioxidants

Antioxidants derived from dietary sources, mostly vegetables and fruits, are exogenous antioxidants (27). Valuable antioxidants for the human body include, but are not limited to, Vitamin A (provitamin A as beta-carotene and retinol), vitamin C (ascorbic acid) and Vitamin E (tocotrienol and tocopherol) (33). The exogenous antioxidant system completes the endogenous antioxidant system, which means there is an ongoing demand in the body for exogenous antioxidants to prevent oxidative stress (45). Exogenous antioxidants can have a defined recommended dietary allowance (RDA) which informs about the average daily intake level of various nutrients to meet the requirement of almost all (97-98%) healthy people (46).

1.2.3.2.1 Beta-carotene

Beta-carotene is a carotenoid. Carotenoids are found in fungi, plants and algae as natural pigments (27). Carotenoids are provitamins of Vitamin A and in the liver, they are converted to this vitamin (47). Their bioavailability is variable and easily influenced by the preparation of the fresh fruit (48). Carotenoids can be found in *e.g.*, carrots, mangoes, papayas and yams or the vegetables which include spinach, kale and many more green leaves (49). In the role of an antioxidant, carotenoids scavenge radicals created from lipid peroxidation (50). Besides beta-carotene the carotenoids, also called pro-vitamin A, in the human body are alpha-carotene and beta-cryptoxanthin, which can be converted into vitamin A as well (24). In general, physiological functions of carotenoids in the body are related to vision, growth, development and the immune system (27). A vitamin A deficiency causes “night blindness” and abnormal epithelial cell growth (47), as well as xerophthalmia and an impairment of immune response and dermatological problems in general (49). Beta-carotene has no RDA defined by the responsible authorities in the United States, the “U.S Institute of Medicine of the National Academy of Sciences”, only Vitamin A has an RDA, which is set at 700 micrograms retinol for females in the adult period of life and 900 micrograms of retinol for males in the adult period of life. 900 micrograms of retinol correspond to 3000 IU (International Units) and 700 micrograms of retinol correspond to 2300 IU. One microgram of retinol is equivalent to twelve micrograms of all-trans-beta carotene in food or two micrograms as a supplement (49).

1.2.3.2.2 Vitamin C

Vitamin C is also well-known under then name ascorbic acid. Found in most fruits and vegetables, its existence in these decreases with prolonged storage times or cooking (27). It is water-soluble and a very powerful free radical scavenger (33), preventing oxidative damage of

happening to DNA, proteins and lipids (51). The substances scavenged by vitamin C are mainly: peroxynitrite, nitric oxide, hypochlorous acid, as well as superoxid, hydroxyl radicals and oxygen, it additionally participates in the reduction of hydrogen peroxide to water via ascorbate peroxidase reaction (51).

Other roles of vitamin C are the preservation of LDL (low density lipoprotein) and the maintenance of vitamin E levels in the cell membrane (52,53), and the restoration of alpha-tocopherol, glutathione, urate and beta-carotene, which also act as antioxidants (33).

Nevertheless, in the companionship of metal ions or in high concentrations vitamin C's antioxidant capacity changes to a pro-oxidant state (inducing radicals) (54,55).

The RDA for a male individual in the adult period of life is 90 mg of Vitamin C and for a female individual in the adult period of life the RDA is 75 mg. Vitamin C deficiency causes scurvy (49).

1.2.3.2.3 Vitamin E

Vitamin E consists of a collection of eight lipophilic compounds with similar structures, including tocopherols and tocotrienols (27). Vitamin E can be found in nuts, cereals and vegetable oils (27). Their amount of vitamin E decreases with cooking and long-term storage just as for vitamin C (27).

This antioxidant is lipid soluble and found in the plasma membrane. There it performs as a chain breaker in lipid peroxidation of cell membranes (56). To protect the cell membranes from lipid peroxidation it donates a hydrogen atom to the radical, directly scavenging lipoperoxyl radicals (33). The then oxidized tocopheryl radical can be converted back to tocopherol by a pathway facilitated by ascorbic acid (57).

Just as Vitamin C, also Vitamin E can have prooxidative effects at high concentrations (58,59).

For Vitamin E the RDA for adolescents and adults is 15 mg (22.4 IU). Impaired immune responses, neuromuscular disorders and red blood cell lysis can be caused by vitamin E deficiency (49).

1.2.4 Oxidative stress in the lens

Oxidative damage of the lens has been shown to play a role in the age-related cataract etiology**,** while glycation of proteins and lipid oxidation have been connected to cataract development (44,54,60). In lenses diseased with cataract several biomarkers of oxidative stress have been found in higher levels than in the lenses not diseased with cataract in humans as well as in animals (54,60–62).

The lens is protected from oxidative stress by antioxidants in the eye (44). The endogenous antioxidants we can find in the human lens are superoxide dismutase (SOD) and glutathione (44). The majority of SOD in the human lens is cytosolic CuZn-SOD and SOD's activity in the lens is 15-fold lower than in the average human tissue (63). Human lenses affected by cataract have been shown to have decreased levels of CuZn-SOD activity in comparison to lenses not affected by cataract (64), so this enzyme seems to have a role in protecting the eye against oxidative stress.

Another endogenous antioxidant, glutathione, may in collaboration with vitamin C, function to scavenge ROS and can be found in high levels in the lens (44).
Additionally exogenous antioxidants have been found in various parts of the human lens, among them vitamin C (65), vitamin E, retinol and the fat-soluble lutein/zeaxanthin (carotenoids) (66). In younger and metabolically active epithelial/cortical layers of the lens the concentration of fat-soluble antioxidants has been shown to be 2-3-fold higher than in older nuclear layers of the lens (27).
As mentioned before ascorbic acid can have prooxidant properties besides being able to scavenge ROS such as superoxide (65), so it can contribute to the aging of the lens crystallin notably even more so in the presence of high metal concentrations (54,55). For the protection of ultraviolet radiation induced cataract vitamin E supplementation has shown to be protective in a dose-response manner (67). In the function of decreasing oxidative stress high concentrations of lutein/zeaxanthin have proved to be effective (68).

2. OBJECTIVES

2.1 AIMS

The aim of this study was to evaluate the available randomized controlled trials and systematic reviews on the efficacy and safety of antioxidant vitamin supplementation in preventing and slowing the progression of age-related cataract.

2.2 HYPOTHESIS

There will be enough evidence found for the efficacy and safety of supplementation with antioxidant vitamins as a preventative treatment of age-related cataract.

3. MATERIALS AND METHODS

3.1 Data collection

This study was planned and executed as a "systematic review", a secondary study design which aims to summarize published data in a qualitative approach. The focus was set on exploring the extent on RCTs and systematic reviews that discuss the efficacy and safety of antioxidant vitamin supplementation in preventing and slowing the progression of age-related cataract.

3.1.1 Literature search

The start of the literature search, revolved around the topic of antioxidant vitamin supplementation in connection with age-related cataract and was made online in the Cochrane library in the section "Eyes & Vision", where we found the article "Antioxidant vitamin supplementation for preventing and slowing the progression of age-related cataract (Review)". With this article as a base, we commenced the further search for articles published after this review (2012) yielding new results. The goal was to find systematic reviews and randomized controlled trials published after the systematic review found on Cochrane (49), with the same inclusion and exclusion criteria as the systematic review (49).

A systematic search was performed using three databases: PubMed (http://www.ncbi.nlm.nih.gov/pubmed/), CENTRAL (https://www.cochranelibrary.com) and DARE (https://www.crd.york.ac.uk/CRDWeb/HomePage.asp). The search was performed on 13 June 2022.

The search on PubMed was built with these terms: multivitamin supplements AND age-related cataract OR antioxidant vitamin supplementation AND age-related cataract OR vitamin c AND age-related cataract OR ascorbic acid AND age-related cataract OR vitamin e AND age-related cataract OR tocopherol AND age-related cataract OR vitamin a AND age-related cataract OR provitamin a AND age-related cataract OR carotenoids AND age-related cataract NOT mice NOT rats. The next step in the process of searching was to set the year from after the Cochrane article, 2012 until now and the results were filtered to only show randomized controlled trials and systematic reviews.

The obtained results were then analyzed so that publications that did not correspond to the topic of the use of antioxidant vitamin supplements in the slowing or preventing of age-related cataract were rejected (e.g., prevention of age-related cataract with antioxidant supplements,

which are not vitamins). Then the remaining results were analyzed with respect to the summary. After another round of exclusions, the then remaining publications were read in full text and were analyzed with respect to the input and output criteria of the Cochrane systematic review (49).

CENTRAL was searched with the words “antioxidant vitamin supplementation” and “age-related cataract” for the publication years 2012-2022, antioxidants that were not vitamins were excluded again.

DARE was searched with the word “age-related cataract (any field)” for the years 2012-2022.

Following a literature search, a quality assessment of the Cochrane systematic review was performed using the R-AMSTAR quality assessment form (69).

3.2 Criteria used for considering studies for the Cochrane systematic review (49)**.**

Methods	Type of studies: RCTs with a one-year minimum follow-up Types of participants: independent of co-morbidities and demographics Types of interventions: supplementation with • beta- carotene or • vitamin C or • vitamin E alone or in any combination, in any dosage or form for at least one year or longer, compared to no supplementation or placebo or a different antioxidant vitamin Categories of outcome measures: a) Primary: 1. Incidence of cataract (defined by the respective study); 2. Incidence of cataract extraction (by definition surgery for the removal of a vision impairing lens opacity, vision impairment defined by the respective study) b) Secondary:

	1. Progression of cataract: any clearly defined measurement of progression (dependent on the mode of presentation by the authors) 2. Loss of vision: any clearly defined measurement of visual acuity (dependent on the mode of presentation by the authors) c) Adverse effects: reported for vitamin E and beta-carotene supplementation

3.3 Search methods for identifications of studies for the Cochrane systematic review (49)

Electronic searches	Eight different electronic sources were used: ● CENTRAL 2012, Issue 2 ● MEDLINE (1950-2012) ● EMBASE (1980-2012) ● LILACS ("Latin American and Caribbean Literature on Health Sciences") (1982 - 2012) ● Open Grey ● mRCT ("the metaRegister of Controlled Trials") ● ClinicalTrials.gov ● ICTRP (the WHO "International Clinical Trials Registry Platform") No search limitations by language or date. Last date of database-search for trials: 2 March 2012
Searching other resources	The list of ongoing trials and the reference lists of included studies were searched to identify additional trials. "Science Citation Index" was used for the identification of trials referencing these trials. For information on unreported and additional trials the investigators of the included trials were contacted.

3.4 Data collection and analysis of data from the Cochrane systematic review (49)

Selection of studies	The titles and abstracts obtained by the searches were independently screened. From definitely relevant and probable trials the full-text versions were gathered and evaluated. The methodological

	quality was evaluated for each article that satisfied the requirements for inclusion.
Data extraction and management	Data was gathered separately by two review authors using a "Cochrane Eyes and Vision Group"-form. RevMan 5 was used by one author for data entry and all values were verified by another author.
Assessment of study characteristics	The studies were assessed by extraction of the following characteristics: • Methods: study design; method of randomization; unit of randomization (individuals/eyes); method of allocation concealment; number randomized; exclusions after randomization; number analyzed; masking (blinding); losses to follow-up; unit of analysis (individuals/eyes). • Participants: country; age; gender; inclusion/exclusion criteria. • Interventions: treatment (including dose and schedule); control; duration of treatment; length of follow-up (planned/ actual); compliance. • Outcomes: relevant outcomes (definition, method of assessment, statistical methods used); eye examined for the outcome (worse/better/average); intervals at which each outcome was assessed; quality control for outcome assessment; adverse effects. • Notes: study period; general health status of study population; types of subgroup analyses; control group event rate for dichotomous outcomes; power calculation (Yes/No, if yes whether appropriate); quality of life indicators; funding sources.
Assessment of risk of bias in included studies	Included trials were evaluated for potential systematic bias by two authors independently in accordance with the guidelines of the "Cochrane Handbook for Systematic Reviews of Interventions", Chapter 8. Criteria, for which the trials were assessed:

	• Selection bias: allocation concealment and sequence generation • Performance bias: masking of recipients of care and care providers • Detection bias: masking of outcome assessors • Attrition bias: incomplete outcome data • Reporting bias: selective outcome reporting • Other sources of bias: adherence to treatment, intention-to-treat analysis, equivalence of baseline characteristics Every criterion was judged as unclear (insufficient information for assessment), high risk of bias or low risk of bias. Discussions were used to overcome disagreements. To resolve issues that were unclear, based on the data in the original articles, the authors of the respective articles were contacted. The methodological quality was assessed based on the available original information, if no contact could be established with the primary investigators within six weeks.
Measurement of treatment effect	For primary outcomes percentages and numbers, unadjusted risk ratios and Cox proportional hazard ratios were gathered. For secondary outcomes unadjusted risk ratios, unadjusted odds ratios, difference in slope, mean change from baseline and mean of last value were gathered. The Mantel-Haenszel risk ratio was calculated for PHS II 2010 (70), with 95% confidence limits for cataract incidence. The relative risk and 95% confidence limits (from 99% confidence intervals) were calculated for the cataract extraction incidence for AREDS 2001 (71). From values in the published study, the cataract extraction incidence, was calculated for VECAT 2004 (72). From values gathered through personal communication, the cataract extraction incidence for PPP 2001 (73), was calculated. To perform said calculations RevMan 5 was used. These data transformations led to minor differences at the second decimal place.

Unit of analysis issues	In all studies included in the systematic review, the individual equated the unit of analysis.
Dealing with missing data	For not reported or missing outcomes and study methods the primary authors of the respective trials were contacted. 95% confidence intervals (CI) and relative risks (RR) were calculated. Data was not imputed.
Assessment of heterogeneity	Clinical heterogeneity was examined by country of origin, participant gender and age and by antioxidant type for the included trials. Due to the variation in the clinical properties of the various antioxidant vitamin groups, the results were not pooled. Using the I2 statistics and Chi2 test statistical heterogeneity was examined.
Assessment of reporting biases	In order to investigate reporting biases, there was the intention to examine a funnel plot in conjunction with study characteristics or other factors that may contribute to funnel plot asymmetry. Due to the limited amount of included studies the choice to not include a funnel plot was made.
Data synthesis	For the incidence of cataract extraction and cataract the summary relative risk was calculated *via* the fixed-effect model (the generic inverse variance method). For secondary outcomes results were not pooled, due to discrepancies in the mode of definition of the outcomes, along with noticeable variation in the mode of presentation and analysis of the data. “Other data tables” were used for data presentation in aforementioned cases and in instances where only adjusted estimates and summary data were disclosed in the trial report.
Investigation of heterogeneity and subgroup analysis	No subgroup analysis was planned, but it was performed after all according to type of cataract (posterior subcapsular, nuclear and cortical) in cases, in which this information was available from the published article.
Sensitivity analysis	Sensitivity analyses was planned to be conducted by exclusion of the trials with high risk of bias. No sensitivity analysis was conducted, due to most pooled trials being of high methodological quality.

3.5 Quality assessment of systematic reviews

Systematic reviews are of great value in summarizing evidence and providing information on its quality. Systematic reviews can be used to put the best possible evidence into practice. As the number of systematic reviews increases, the question of their quality arises (74).

A systematic review is not the same as a literature review because it is based on an objective and transparent approach, which is based on the science of research synthesis with the specific intention and goal of minimizing bias. Therefore, most systematic reviews are based on explicit quantitative analysis of measurable data (e.g., acceptable sample analysis, meta-analysis). Despite these investments, certain threats to bias (publication bias) remain. Also, a significant number of systematic reviews are of a qualitative nature and, while respecting accepted standards for the collection, evaluation and publication of evidence, do not allow for quantitative evaluation (69).

To address this issue, a number of quality evaluation instruments have been developed and validated and have eventually led to an AMSTAR form that assesses the 11 most important features of a systematic review (75). Although AMSTAR is convenient and easy to use, it does not provide a quantitative assessment of the quality of a systematic review and therefore AMSTAR has been revised. The result was R-AMSTAR used to quantify the quality of systematic reviews (69).

Quantitative measure of R-AMSTAR are points for each of the 11 individual domains of the original instrument. The number of points for each domain can be from 1-4 (maximum), and the total number of points can be in the range of 11-44 points (maximum). A total score of 11 points indicates that none of the AMSTAR criteria are met. In contrast, the total score 44 indicates that all criteria of excellence of the systematic review are met (69).

R-AMSTAR checklist (76)

1. **Was an "a priori" design provided?**

   If it satisfies 3 criteria -> 4 points

   If it satisfies 2 criteria -> 3 points

   If it satisfies 1 criterion -> 2 points

   If it satisfies 0 criteria -> 1 point

Criteria:

(A) A clearly focused (PICO-based) question
(B) Description of inclusion criteria
(C) Study protocol is published and/or registered in advance

2. Was there duplicate study selection and data extraction?

If it satisfies 3 criteria -> 4 points

If it satisfies 2 criteria -> 3 points

If it satisfies 1 criterion -> 2 points

If it satisfies 0 criteria -> 1 point

Criteria:

(A) At least two persons independently extracted the data, explicitly stated
(B) Statement of consensus procedure for disagreements
(C) Disagreements among extractors resolved properly as stated or implied

3. Was a comprehensive literature search performed?

If it satisfies 4 or 5 criteria -> 4 points

If it satisfies 3 criteria -> 3 points

If it satisfies 2 criteria -> 2 points

If it satisfies 1 or 0 criteria -> 1 point

Criteria:

(A) At least two electronic sources are searched
(B) Years and databases used are mentioned
(C) Key words and/or MESH terms are stated and where feasible the search strategy outline is provided
(D) Searches should are supplemented by consulting current contents, reviews, textbooks, registers and by reviewing the references in the studies found
(E) Journals are hand-searched or manual searched

4. **Was the status of publication (i.e., grey literature) used as an inclusion criterion?**

If it satisfies 3 or 4 criteria -> 4 points

If it satisfies 2 criteria -> 3 points

If it satisfies 1 criterion -> 2 points

If it satisfies 0 criteria -> 1 point

Criteria:

(A) The authors state that they searched for reports regardless of their publication type.
(B) The authors state whether or not they excluded any reports based on their publication status, language etc.
(C) "Non-English papers were translated" or readers sufficiently trained in foreign language
(D) No language restriction or recognition of non-English articles

5. **Was a list of studies (included and excluded) provided?**

If it satisfies 4 criteria -> 4 points

If it satisfies 3 criteria -> 3 points

If it satisfies 2 criteria -> 2 points

If it satisfies 1 criterion -> 1 point

Criteria:

(A) Table/list/figure of included studies, a reference list does not suffice
(B) Table/list/figure of excluded studies either in the article or in a supplemental source
(C) Satisfactory/ sufficient statement of the reason for exclusion of the seriously considered studies
(D) Reader is able to retrace the included and excluded studies anywhere in the article bibliography, reference or supplemental source

6. **Where the characteristics of the included studies provided?**

   If it satisfies 3 criteria -> 4 points

   If it satisfies 2 criteria -> 3 points

   If it satisfies 1 criterion -> 2 points

   If it satisfies 0 criteria -> 1 point

   Criteria:

(A)	In an aggregated form such as a table, data from the original studies are provided on the participants, interventions/exposure and outcomes
(B)	Ranges are provided of the relevant characteristics in the studies analyzed
(C)	The information provided appears to be complete and accurate

7. **Was the scientific quality of the included studies assessed and documented?**

   If it satisfies 4 criteria -> 4 points

   If it satisfies 3 criteria -> 3 points

   If it satisfies 2 criteria -> 2 points

   If it satisfies 1 or 0 criteria -> 1 point

   Criteria:

(A)	"A priori" methods are provided
(B)	The scientific quality of included studies appears to be meaningful
(C)	Discussion/recognition/awareness of level of evidence is present
(D)	Quality of evidence is rated/ranked based on characterized instruments

8. **Was the scientific quality of the included studies used appropriately in formulating conclusions?**

   If it satisfies 4 criteria -> 4 points

   If it satisfies 3 criteria -> 3 points

   If it satisfies 2 criterion -> 2 points

   If it satisfies 1 or 0 criteria -> 1 point

Criteria:

(A) The scientific quality is considered in the analysis and the conclusions of the review
(B) The scientific quality is explicitly stated in formulating recommendations
(C) Conclusions integrated/drives towards practice guidelines
(D) Clinical consensus statement drives towards revision or confirmation of practice guidelines

9. Were the methods used to combine the findings of studies appropriate?

If it satisfies 4 or 5 criteria -> 4 points

If it satisfies 3 criteria -> 3 points

If it satisfies 2 criteria -> 2 points

If it satisfies 1 or 0 criteria -> 1 point

Criteria:

(A) Statement of criteria that were used to decide that the studies analyzed were similar enough to be pooled
(B) For the pooled results, a test is done to ensure the studies were combinable, to assess their homogeneity
(C) A recognition of heterogeneity or lack thereof is present
(D) If heterogeneity exists a “random effects model” is used and/or the rationale of combining is taken into consideration
(E) If homogeneity exists, author state a rationale or a statistical test

10. Was the likelihood of publication bias assessed?

If it satisfies 3 criteria -> 4 points

If it satisfies 2 criteria -> 3 points

If it satisfies 1 criterion -> 2 points

If it satisfies 0 criteria -> 1 point

Criteria:

(A) Recognition of publication bias or file-drawer effect
(B) Graphical aids (e.g. funnel plot)
(C) Statistical tests (e.g. Egger regression test)

11. Was the conflict of interest included?

If it satisfies 3 criteria -> 4 points

If it satisfies 2 criteria -> 3 points

If it satisfies 1 criterion -> 2 points

If it satisfies 0 criteria -> 1 point

Criteria:

(A) Statement of sources of support
(B) No conflict of interest. This is subjective and may require some deduction or searching.
(C) An awareness/statement of support or conflict of interest in the primary inclusion studies

4. RESULTS

4.1 Results of the Cochrane systematic review (49)

Selection of trials	The electronic searches resulted in a total of 1861 found trial reports. Manual searches were conducted concerning ongoing and included trials and study authors were approached for data on other ongoing trials or completed trials. Abstracts and titles were screened according to the inclusion criteria. Of 31 trial reports full text evaluations were performed, as well as for the description of one trial. 21 trials were excluded. Eleven trials qualified for inclusion, among these, one complete trial with unfinished data analysis and one trial, that was not open for participant recruitment at that point of time. In the qualitative synthesis nine trials were included. In the quantitative analysis six trials were included.
Included studies	Nine trials, with a total of 117,272 individuals: ● APC 2006: *"The Antioxidants in Prevention of Cataracts Study: effects of antioxidant supplements on cataract progression in South India"* (77), ● AREDS 2001: *"A randomized, placebo-controlled, clinical trial of high-dose supplementation with vitamins C and E and beta carotene for age-related cataract and vision loss: AREDS report no. 9"*(71), ● ATBC 1998: "*Incidence of cataract operations in Finnish male smokers unaffected by alpha tocopherol or beta carotene supplements"* (78), ● PHS I 2003: "*A randomized trial of beta carotene and age-related cataract in US physicians"* (79) ● PHS II 2010: "*Age-related cataract in a randomized trial of vitamins E and C in men"* (70) ● PPP 2001: "*Epidemiological feasibility of cardiovascular primary prevention in general practice: a trial of vitamin E and aspirin. Collaborative group of the Primary Prevention Project"* (73)

	● REACT 2002: "*The Roche European American Cataract Trial (REACT): a randomized clinical trial to investigate the efficacy of an oral antioxidant micronutrient mixture* to slow progression of age-related cataract" (80) ● VECAT 2004: "*Vitamin E supplementation and cataract: randomized controlled trial"* (72) ● WHS 2004/8: "*Vitamin E and age-related cataract in a randomized trial of women"* (81) were part of the review. The trials were conducted in the United States of America (70,71,79–81), Australia (72), Italy (73), India (77), Finland (78) and the United Kingdom (80), in the years from 1982 to 2010. Across these trials the duration of treatment and follow-up ranged from 2.1 to 12 years. Tables with the characteristics of the included studies can be found in "supplement 1".
Types of Participants	The age range among the participants in the included trials was ≥35 years. Three trials had exclusively male participants (70,78,79) and one trial had exclusively female participants (81). In the other five studies the participant population was made up of more females than males (71–73,77,80). One trial (78) only included subjects, that consumed over five cigarettes per day and in two trials (72,80) a requirement for participation was the presence of any stage of age-related cataract. One study (77) excluded patients with previous intraocular surgery. In VECAT 2004 (72), patients with previous cataract surgery were excluded. From REACT 2002 (80), patients expected to undergo cataract extraction in the first two years after enrollment were excluded. Four trials (70,77–80) excluded patients already consuming vitamin supplements.
Types of interventions	No trial had a "no treatment" control. Every included trial was either controlled with an alternate treatment or with placebo. Beta-carotene alone was evaluated in 3 trials (78,79,81). Vitamin C alone was evaluated in one trial (70).

Vitamin E alone was evaluated in five trials (70,72,73,78,81).

Beta-carotene and vitamin E in combination were evaluated in one trial (78).

Vitamin C and vitamin E in combination were also evaluated in one trial (70).

Beta-carotene, vitamin C and vitamin E in combination were evaluated in three trials (71,77,80).

In every trial, the antioxidant vitamin dosage surpassed the RDA.

Study	*Beta-carotene*	*Vitamin C*	*Vitamin E*
PHS I 2003 (79)	50mg every other day	-	-
WHS 2004/8 (81)	50 mg every other day	-	600 IU every other day
ATBC 1998 (78)	20 mg once daily	-	50 mg once per day
PHS II 2010 (70)	-	500mg once daily	400 IU every other day
PPP 2001 (73)	-	-	300 mg/day
VECAT 2004 (72)	-	-	500 IU daily
APC 2006 (77)	15 mg 3 times per week	500mg 3 times per week	400 IU 3 times per week
AREDS 2001(71)	15 mg daily	500mg daily	400 IU daily
REACT 2002 (80)	6 mg	250mg	200 mg (alpha-tocopherol) three times daily, as a capsule

Types of outcome measures	**Primary outcomes** 1. **Incidence of cataract:** evaluated in four trials (70,72,79,81). 2. **Incidence of cataract extraction:** evaluated in eight trials (70–73,77–79,81). **Secondary outcomes** 1. **Progression of cataract:** evaluated in four trials (71,72,77,80). 2. **Loss of visual acuity:** evaluated in three trials (71,77,80).
Excluded studies	21 trials were excluded after full-text review. Following a precise methodological review, two studies were excluded (82,83). One more trial (84) was excluded after finding no reported outcomes concerning eyes/vision.

4.1.1 Risk of bias of the studies included in the Cochrane systematic review (49)

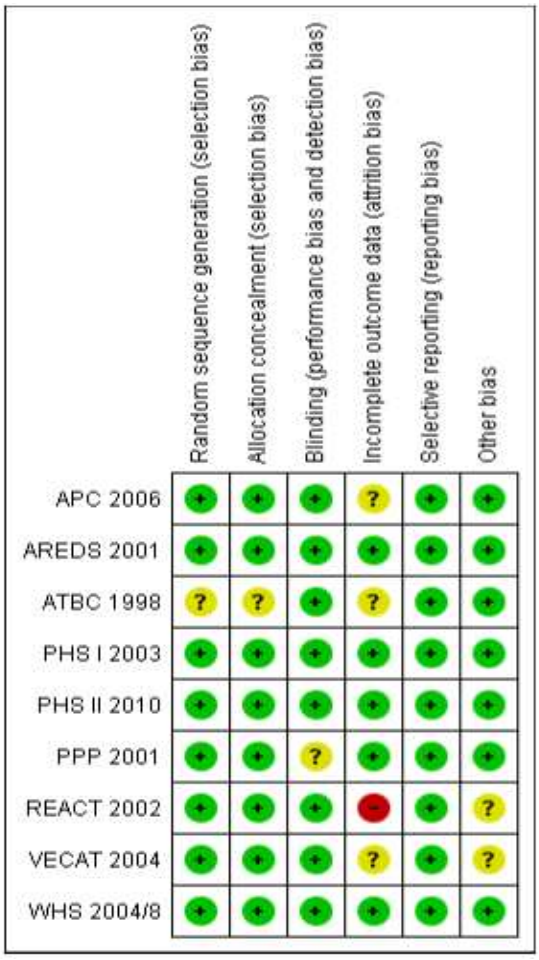

Figure 2: "Risk of bias" summary: review authors' judgements about each risk of bias item for each individual study (49).

4.1.2 Effects of interventions in the studies included in the Cochrane systematic review (49)

4.1.2.1 Primary outcomes

1) Incidence of cataract	Evaluated by four trials (70,72,79,81). Results were pooled for vitamin E and beta-carotene.
Beta-carotene versus placebo	In the United States, 22,071 male physicians in the age range from 40 to 84 years were evaluated (79) and no difference was found between placebo and beta-carotene (50 mg every other day) regarding the risk of cataract incidence. The study had a twelve-year follow-up. For aspirin assignment the Cox proportional hazard ratio was 1.0 (95% confidence interval 0.91-1.09). 39,876 female health professionals aged 45-years and older were assessed in the United States (81) and no difference was found between beta-carotene (50 mg every second day) and placebo in a median period of 2.1 years in the risk for cataract incidence. For vitamin E and aspirin assignment the Cox proportional hazard ratio was 0.95 (95% CI 0.75-1.21). No evidence could be found of the effect of beta-carotene supplementation in reducing the risk of cataract incidence in the pooled analysis of two trials (79,81) with a total of 57,703 patients. 0.99 was the summary relative risk (RR) (95% CI 0.91-1.08). The test for heterogeneity was not statistically significant (Chi2 = 0.15, P = 0.69; I2 = 0%).
Vitamin C versus placebo	No difference was found between daily supplementation with 500mg Vitamin C and placebo for the risk of cataract incidence over a mean period of eight years among 14,641 male physicians in the age-range of 50 years and older. 1.02 (95% CI 0.91-1.14) was the adjusted Cox proportional hazard ratio and there was no difference in risk according to various types of cataracts.
Vitamin E versus placebo	One study (70) showed no difference between daily 400 IU of vitamin E and placebo for cataract incidence in a mean

	follow-up period of eight years with an value of 0.99 for the adjusted Cox proportional hazard ratio (95% CI 0.88-1.11). In Australia, 1204 volunteers in the age range of 55 to 80 years were evaluated and no difference was found between placebo and 500 IU of vitamin E daily for the incidence of cataract. The follow-up period encompassed four years 1.0 (95% CI 0.8 to 1.4) was the risk ratio. One study (81) showed no difference between the supplementation with 600 IU vitamin E every second day and placebo over an average follow-up of 9.7 years. For aspirin and beta-carotene, the adjusted Cox proportional hazard ratio was 0.96 (95% CI 0.88-1.04). Regarding risks by types of cataract no difference could be found. Three trials were pooled for analysis, made up of a total of 50,059 participants and no evidence could be found for the effect of supplementation with vitamin E for the reduction of the risk of cataract incidence. 0.97 was the summary RR with a 95% CI of 0.91 to 1.04. The heterogeneity-test showed no statistical significance and the subgroup analysis also deemed no difference in effect in regard to type of cataract.
Vitamin C and vitamin E versus placebo	One study (70) combined 500 mg of vitamin C daily and 400mg IU on alternate days of vitamin E and found no difference compared to placebo for the cataract incidence over a mean follow-up period of eight years. The Mantel-Haenszel risk ratio was 0.98 (95% CI 0.84-1.15) and by type of cataract there was no difference in risk.
2) Cataract extraction incidence	Evaluated by eight trials: (70–73,77–79,81). Pooled results were created for vitamin E and beta-carotene.
Beta-carotene versus placebo	29,133 male smokers were evaluated in Finland in the age range of 50 to 69 years (78). Over a median follow-up period of 5.7 years, no difference was found between 20 mg beta-carotene daily and placebo for the cataract extraction

	incidence. The Cox proportional hazard ratio adjusted for risk factors for cataract was 0.97 (95% CI 0.79-1.19) One study (79) had the same results over a 12-year follow-up and the Cox proportional hazard ratio adjusted for aspirin assignment was 1.00 (95% CI 0.89 to 1.12). One more study (81) had the same results as well, but over a median period range of 2.1 years and the Cox proportional hazard ratio adjusted for aspirin and vitamin E assignment was 1.04 (95% CI 0.78-1.39). No evidence could be found for a risk reduction of cataract extraction by supplementation with beta-carotene in the pooled analysis of these three trials involving 86,836 patients. The summarized RR was 1.0 (95% CI 0.91-1.10) and the test for heterogeneity did not show statistical significance.
Vitamin C versus placebo	No difference was found between supplementation with vitamin C and supplementation with placebo for the cataract extraction incidence in the mean follow-up period of eight years (70). 0.97 (95% CI 0.85-1.12) was the calculated adjusted Cox proportional hazard ratio. When distinguishing various types of cataract no differences in risk could be found.
Vitamin E versus placebo	Two studies (70,78) did not detect a difference between the supplementation of 50 mg vitamin E once daily and placebo for the cataract extraction incidence over a median follow-up period of 5.7 and eight years. Adjusted for risk factors of lens cataract, the Cox proportional hazard ratio was 0.91 (95% CI 0.74-1.11) (78) and 0.96 (95% CI 0.83-1.10) (70) and by types of cataract there was no difference in risk. 4495 volunteers in the age range of 50 years and above were evaluated in Italy (73) and found no disparities between supplementation with vitamin E and placebo with a mean

	period of follow-up of 3.6 years. 1.03 (95% CI 0.73-1.46) was the unadjusted risk ratio of this study. One more study (72) also did not find any difference in between vitamin E supplementation and placebo with a follow-up of 4 years and a RR of 1.09 (95% CI 0.69-1.72). Another study (81) did not find any difference either with a follow-up of 9.7 years on average and a Cox proportional hazard ratio of 1.00 (95% CI 0.91-1.11), which was adjusted for beta-carotene and aspirin assignments. The various types of cataract had no different risk. An analysis was pooled from the five trials, involving a total of 83,956 patients with no support for effect of supplementation of vitamin E in risk reduction of cataract extraction. 0.98 (95% CI 0.9-1.05) was the pooled RR. No statistical significance was obtained by the test for heterogeneity. Likewise, in the subgroup analysis in two trials (70,81) no disparity of effect could be established between the various types of lens cataract. The RRs (95% CI) were 1.02 (0.89-1.16) for posterior subcapsular cataract, 0.99 (0.91-1.07) for nuclear cataract and 0.92 (0.81-1.05) for cortical cataract.
Beta-carotene + vitamin E versus placebo	One study (78) did a comparison of this combination and found no difference in the risk of cataract extraction incidence over a median follow-up period of 5.7 years. For the cataract extraction incidence, the rate ratio was 0.92 (95% CI 0.7 to 1.21).
Beta-carotene + vitamin C + vitamin E versus placebo	This comparison was done by one study (77) but the data was unavailable. AREDS 2001 (71) evaluated 4757 volunteers between 55 and 80 years old in the United States and found no difference over a mean period of 6.3 years with a Cox proportional hazard ratio adjusted for age, race, sex and smoking status was 0.97 (95% CI 0.83-1.13).

4.1.2.2 Secondary outcomes

1) Progression of cataract	Evaluated by four trials (71,72,77,80). Results were not pooled due to variations in the analysis and the outcome definitions of presentation of data. Neither Vitamin C, nor beta-carotene were compared with placebo in any trials.
Vitamin E versus placebo	In one study (72) no difference was found over a follow-up period of four years. The risk ratio was 1.0 (95% CI 0.7-1.3). There was no difference in risk by types of cataract.
Beta-carotene + vitamin C + vitamin E versus placebo	798 volunteers with ages ranging from 35 to 50 years were evaluated in India by one trial (77) and no difference between treatment and placebo was found in the risk of progression of cataract in a timeframe of five years. Results were similar by age group and type of cataracts. Another trial (71) also did not find any difference in between treatment and placebo for any lens event over a timeframe of 6.3 years. Adjusted for baseline smoking status, gender, race, age and age-related macular degeneration category the odds ratio (OR) was 1.0 (95% CI 0.90-1.11). For a severe lens event the results were similar with an OR of 0.95 (95% CI 0.82 to 1.10). No difference could be found in the risk by cataract type. Over 40 years, 297 patients from the United Kingdom and the United States were evaluated by one trial (80), and found to favor antioxidants in comparison to placebo, but without statistical significance. Further, independent of follow-up length, a beneficial effect was reported among subgroups of participants with no or early lens cataract, and moderate to more advanced lens cataracts, in both countries. This study lost 22% of participants after two follow-up-years and 47% after three follow-up-years. No statistical significance is shown in the results by type of cataract.

2) Loss of the visual acuity	In total, 3 studies (71,77,80) evaluated this and all examined a combination of all three antioxidants supplements or placebo.
Beta-carotene + vitamin C/E vs. placebo	Treatment and placebo showed no difference in the visual acuity (71,77,80).

4.1.2.3 Adverse effects

- Hypercarotenodermia while on beta-carotene: 8.6% in AREDS 2001 (71), 8.8% in ATBC 1998 (78), 15.8% in PHS I 2003 (79), 7.4% in REACT 2002 (80) and 10.7% in WHS 2004/8 (81)
- Increased risk of epistaxis while on vitamin E supplementation in WHS 2004/8 (81)

4.2 Literature search after the Cochrane systematic review

Figure 3 shows the literature search procedure in the PubMed, CENTRAL and DARE databases. Details of the search method are described in the materials and methods section.

The search of the CENTRAL database found the systematic review already used (49) and four trials of which three had to be eliminated by title. From this search one RCT was included in this work.

The search of the DARE database found nine systematic reviews, one of these was again the Cochrane systematic review already used (49) and the remaining eight were found to not match the topic after reading the title.

The search of the PubMed database yielded 14 results. After excluding one duplicate from CENTRAL and the Cochrane systematic review already used (49), and one article not available in full text, eleven studies remained. Out of the eleven studies, one systematic review (85) and one RCT (86) were seriously considered for inclusion in this work but eliminated after full text review.

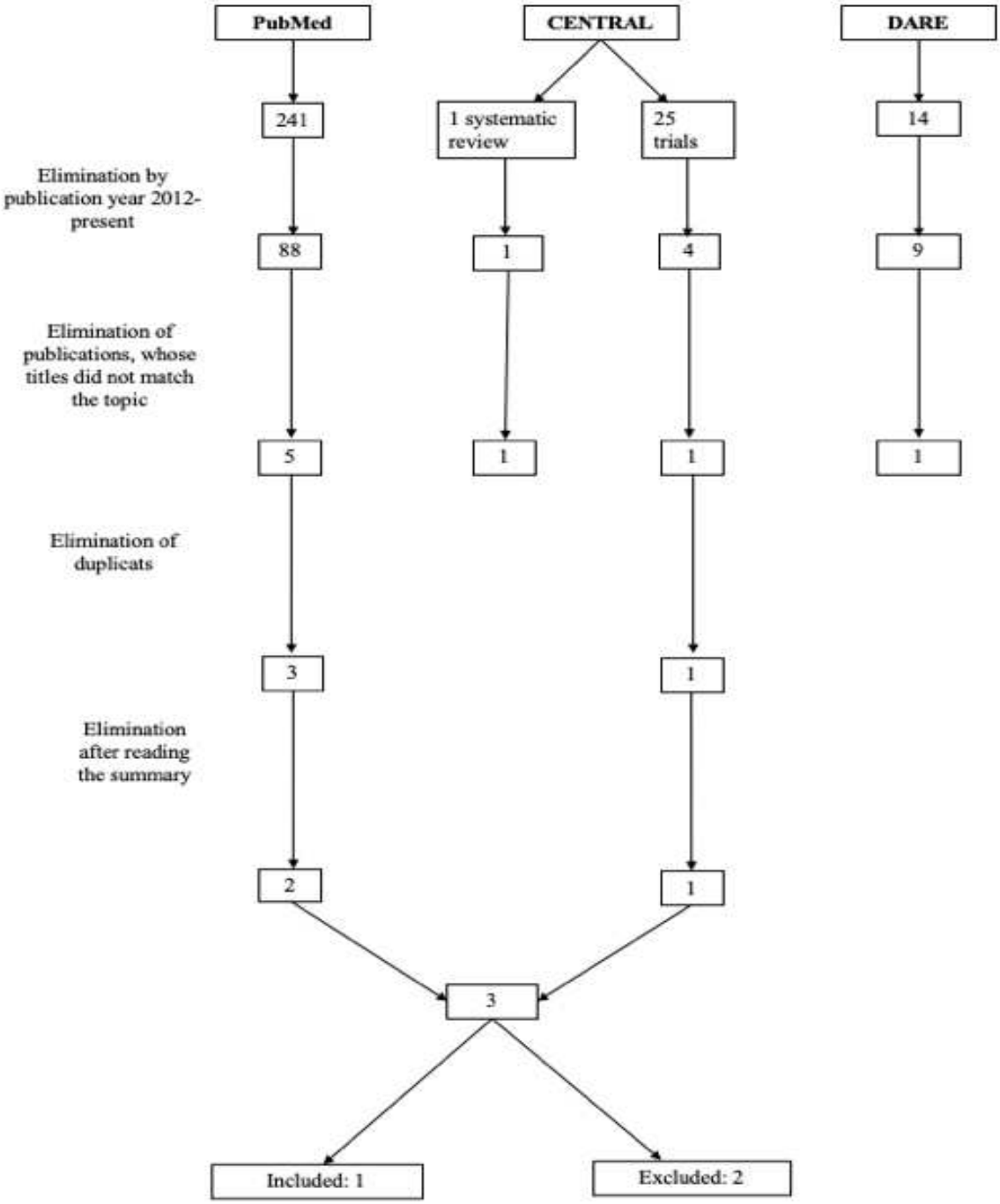

Figure 3: Flowchart of the search of the databases.

4.2.1 Characteristics of the articles published after the Cochrane systematic review (49) fulfilling the inclusion criteria

SEE 2016 (87)

"Age-related Cataract in a Randomized Trial of Selenium and Vitamin E in Men: The SELECT Eye Endpoints (SEE) Study"

Methods	Design: phase III randomized, placebo-controlled, four arm trial Method of randomization: randomized block scheme, the block was the study side Method of allocation concealment: unavailable Number randomized to SELECT: 35,533 Exclusions: 1) Site not participating in SEE: 21,034 2) Participant refused all substudies: 953 3) Participant reported prior cataract or extraction: 2,279 Men Eligible for SEE (analyzed): 11,267 Placebo (no antioxidant): 2829 Vitamin E alone (antioxidant): 2844 Selenium + Vitamin E: 2789 Selenium alone: 2805 Masking: no data available Losses to follow-up: no data available Unit of analysis: no data available
Participants	Country: USA, Canada, Puerto Rico Age: median: 61; range: 50 years and older for African American men and 55 and older for all other men Gender: all men Inclusion criteria: no prior diagnosis of prostate cancer, 4 ng/mL or less of PSA in serum, digital rectal examination not suspicious for cancer, reported no current use of anticoagulant therapy other than 175 mg/d or less of acetylsalicylic acid or 81 mg/d or less of acetylsalicylic acid

	with clopidogrel bisulfate, no history of hemorrhagic stroke, normal blood pressure Exclusion criteria: previously reported cataract or extraction
Interventions	Treatment: (a) selenium (200 μg/d from L-selenomethionine) (b) vitamin E (400 IU/d of all rac-α-tocopheryl acetate) (c) combination Control: placebo Duration: 5.6 years on average
Outcomes	1. Incident cataract, defined as a lens opacity, age-related in origin, responsible for a reduction in best-corrected visual acuity to 20/30 or worse based on self-report confirmed by medical record review 2. cataract extraction, defined as the surgical removal of an incident cataract. Outcomes were assessed every 6 months
Notes	SEE 2016 was an ancillary study of SELECT (SWOG-coordinated Selenium and Vitamin E Cancer Prevention Trial) Study period: September 2003 to October 2008 Study population: middle-aged to older men, apparently well-nourished Subgroup analysis: by categories of baseline variables consisting of possible risk factors for cataract Funding: National Eye Institute

4.2.2 Characteristics of excluded studies

Study name	Exclusion criteria
„The effect of multivitamin/mineral supplements on age-related cataracts: a systematic review and meta-analysis"(85)	This systematic review and meta-analysis did not state clearly which supplements were used in the included studies, which made it impossible to extricate the effects of vitamin antioxidants and they reviewed cohort studies, this were both reasons for exclusion.
"Effects of multivitamin supplement on cataract and age-related macular degeneration in a randomized trial of male physicians"(86)	This randomized trial used multivitamin supplements and it was not possible to extricate the effects of vitamin antioxidants, which was the reason it was excluded.

4.3 R-AMSTAR quality assessment of the systematic review

"Antioxidant vitamin supplementation for preventing and slowing the progression of age-related cataract (Review)"

1. Criteria fulfilled: A, B → 3 points
2. Criteria fulfilled: A, B, C → 4 points
3. Criteria fulfilled: A, B, C, D → 4 points
4. Criteria fulfilled: B, D → 3 points
5. Criteria fulfilled: A, B, C, D → 4 points
6. Criteria fulfilled: A, B, C → 4 points
7. Criteria fulfilled: A, B → 2 points
8. Criteria fulfilled: A, C → 2 points
9. Criteria fulfilled: A, C → 2 points
10. Criteria fulfilled: A, B → 3 points
11. Criteria fulfilled: A, B, C → 4 points

OVERALL QUALITY ASSESSMENT OF THE SYSTEMATIC REVIEW: **35 points**

5. DISCUSSION

In the search for answers to specific questions, some research methods provide evidence of higher quality than other research methods and thus the validity of research results also varies according to method. For evaluation of the effectiveness of an intervention randomized controlled trials are considered to provide the most reliable evidence, because the processes used in conducting RCTs are constructed to minimize the risk of confounding factors affecting the results. Consequently, the results of RCTs are more likely to be closer to true effects than findings from other research methods (88). A comprehensive summary of the best available evidence on a specific issue is a systematic review (89). For conduction of a systematic review a search strategy and a comprehensive and detailed plan are usually defined a priori, with a focus on reduction of bias by identifying, evaluating and synthesizing all relevant studies on a particular topic. Meta-analysis' use statistical techniques to synthesize data from several studies into a single quantitative summary of effect size and are often included in systematic reviews (90). These are the reasons, why for this work RCTs and systematic reviews were used.

The search for evidence of the safety and efficacy of antioxidant vitamin supplementation for the prevention and slowing of age-related cataract started with the analysis of Cochranes systematic review on this topic (49), which states that the antioxidant vitamin supplementation failed to have the desired effect (49).

Regarding the incidence of cataracts, no evidence of effect could be found by any of the four trials examining this, not for vitamin C (70), nor for beta-carotene (79,81), vitamin E (70,72,81) and not even for vitamin E and C in combination (70). The type of cataract was shown to be irrelevant in the supplementation with vitamin E and C in combination or as single agents.

Moreover, no evidence of effect could be observed by a fixed-effect meta-analysis for vitamin E or beta-carotene on the incidence of cataract.

The systematic review contained eight trials examining the cataract extraction incidence and could not detect any evidence for the effect of any antioxidant vitamin supplementation alone or in combination of all three or of vitamin E and beta-carotene combined. In addition, no evidence could be found for various cataract forms with supplementation of vitamin E or C either. Lastly the fixed-effect meta-analysis on incidence of cataract extraction for vitamin E and beta-carotene supplementation did not show any evidence.

The Cochrane article contained four trials documenting the progression of cataract, independent of type, for vitamin E alone and all three antioxidants in combination and could not find any proof of positive effect for that either, but every trial also defined the progression of cataract individually.

One trial (80) from the systematic review reported in favor of antioxidant supplementation's role to slow down the development of age-related cataract, but the test for statistical significance of these results yielded only a "borderline" probability. No statistically significant effect estimates for lens cataract progression (as a secondary outcome) could be found. Statistical significance was not found for results by type of cataract either.
Lastly, no proof of beneficial effect on the loss of visual acuity by combination of all three antioxidants could be found.
It needs to be mentioned that all studies included in the systematic review dosed the antioxidant supplementation at values above the RDA.
Special characteristics of the studies included in the systematic review that could have influenced the results are:

1) Participants were 35 years and older, possibly the supplementation would need to start earlier in life to have a beneficial effect.

2) The time needed for manifestation of protective effects of antioxidant supplementation could be longer than the time tested in the trials.

3) All studies included, but one took place in the developed world with seemingly healthy individuals and probably different nutritional status than in individuals in the developing world.

4) Three vitamin antioxidant supplementations were tested, vitamin C, vitamin E and beta-carotene, but a lot of other antioxidants exists as well as substances with antioxidant properties and it is possible that one of them might have a beneficial effect in regard to age-related cataracts in humans.

The results of this study should not be transferred onto recommendations for dietary intake of vegetables and fruits. Fruits and vegetables are naturally wealthy in antioxidants and have doses not exceeding the RDA.

The R-AMSTAR quality assessment of this systematic review (49) yielded 35 of 44 possible points. This high score indicates a high quality of the systematic review, which gives more value to its results.

The literature search found only one additional RCT (87) which examined the role of Vitamin E in the prevention and progression of age-related cataract, but also the role of Selenium, and Vitamin E and Selenium in combination.

Men assigned to the vitamin E group were near the null value of 1.0 with a 95% CI excluding with reasonable certainty the beneficial effects greater than 17% for both cataract and cataract extraction. These findings were consistent with the negative findings of the previous randomized trials. The dose of vitamin E was more than 26 times the dose of the RDA of 15mg. Observational studies reported beneficial effects with a median intake of 12 mg of vitamin E (91). These findings cannot be applied to women as the studied population consisted exclusively of men. Once more it should be noted that the population of this study, just as the studies from the Cochrane systematic review, was conducted among a well-nourished population.

Unfortunately, no other and new RCTs and systematics reviews matching the criteria of the original Cochrane systematic review could be found.

One newer article (1) found on MEDLINE regarding nutritional strategies for prevention of lens cataracts acknowledges the inefficiency of supplementation of the lens with antioxidants as well and tried to explain it: reactive oxygen species seem to act as important modulators of redox signaling, principal in maintaining normal cellular processes and metabolism (1). This would mean that antioxidant supplementation would be in fact counterproductive and removing this important natural occurring stimulus for normal redox signaling and cellular function (1). Hence, a therapy for age-related cataract would not only need to consist of antioxidant against the oxidative damage but it would also need to be delivered at levels effectively restoring antioxidant balance in different lens regions where this is needed (1)

Another article from 2020 (26) supports the value of a well-balanced diet rich in vegetables and fruits in the prevention and slowing of age-related cataract.

A meta-analysis of 13 observational studies from 2013 (92) observed the blood levels of antioxidants and the risk of age-related cataract. It explained that the assessment of antioxidant intake was imprecise and that the blood levels were better for evaluation of nutritional status. Furthermore, this meta-analysis could not single out which antioxidant had the protective effect on the lens, as many were used in combination and different studies correlated the blood levels of different antioxidants with a beneficial effect. This article concluded that on general the blood levels of certain antioxidants were inversely associated with the risk of age-related cataract but acknowledged the that the supplementation with antioxidants/vitamins in this regard needed further research.

The last three mentioned articles (1,26,92) could not be included in the work, because they did not match the criteria of systematic reviews or RCTs, which was chosen to have the best possible quality of evidence as mentioned above.

6. CONCLUSION

Not enough evidence could be found to prove the safety and efficacy of antioxidant vitamin supplementation for preventing and slowing the progression of age-related cataract.

The supplementation of the antioxidants vitamin C, vitamin E and beta-carotene did not show a beneficial effect for the prevention and slowing of age-related cataract.

Other research however suggested that the intake of these supplements does not always correlate with their blood levels and that vitamins from food, that is from fruit and vegetables have a superior quality than the vitamins from supplements. Hence, more high-quality research should be done in the form of randomized controlled trials in this field.

Furthermore, most studies were conducted among well-nourished populations, consequently another field of interest would be randomized controlled trials with antioxidant supplementation among not well-nourished populations.

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8. SUMMARY

Objectives: The aim of this study was to evaluate the available RCTs and systematic reviews regarding the efficacy and safety of antioxidant vitamin supplementation in preventing and slowing the progression of age-related cataract and to examine if we could find enough evidence for this.

Materials and methods: The Cochrane systematic review "Antioxidant vitamin supplementation for preventing and slowing the progression of age-related cataract" was used as a base and according to its inclusion criteria we tried to find RCTs and systematic reviews published after it, yielding additional proof of the safety and efficacy of antioxidant vitamin supplementation regarding age-related cataracts. We searched PubMed, CENTRAL and DARE. After the search, we made a quality assessment of the systematic review using R-AMSTAR.

Results: The Cochrane systematic review involved nine trials, with 117,272 participants overall and an age range of ≥ 35 years. The trial follow-up ranged from 2.1 to 12 years and the trials were conducted in Italy, Finland, the United Kingdom, Australia, India and the United States. The antioxidant vitamins were dosed above the recommended daily allowance. No evidence was found supporting that antioxidant vitamin supplementation could reduce the risk of progression of cataract, cataract development, cataract extraction or that it could slow the loss of visual acuity. No evidence of effect of supplementation of beta-carotene in reducing the risk of cataract could be found in the pooled analysis (RR 0.99, 95%, CI 0.91-1.08; number of participants: 57,703) nor could be evidence found for a reduction of the risk of cataract extraction (RR 1.00, 95% CI 0.91-1.10; number of participants: 86,836). For the supplementation with vitamin E no risk reduction in cataract incidence could be found in the pooled analysis either (RR 0.97, 95% CI 0.91-1.04; number of participants: 50,059) or of cataract extraction (RR 0.98, 95% CI 0.91 - 1.05; number of participants: 83,956). 7.4 - 15.8% of patients on beta-carotene developed hypercarotenodermia (yellowing of skin). Only one additional RCT was found involving 11,267 exclusively male participants from the United States, Canada and Puerto Rico, with an age range of 50 years and older. Only part of this RCT was of interest for us, the part concerning supplementation with vitamin E. It lasted 5.6 years.

Conclusion: Not enough evidence could be found to prove the safety and efficacy of supplementation with antioxidant vitamins for the slowing and the prevention of age-related cataract. Further research among non-well-nourished populations or well-nourished populations but regarding antioxidant blood levels rather than supplementation could be conducted in the future.

9. CROATIAN SUMMARY

Naslov: Pronalaženje dokaza o učinkovitosti i sigurnosti suplementacije vitamina antioksidansa u prevenciji i usporavanju napredovanja katarakte povezane sa starenjem

Ciljevi: Cilj ove studije bio je procijeniti dostupne RCT-ove i sustavne preglede koji se odnose na učinkovitost i sigurnost suplementacije vitamina antioksidansa u prevenciji i usporavanju napredovanja katarakte povezane sa starenjem te ispitati možemo li pronaći dovoljno dokaza za to.

Materijali i metode: Cochraneov sustavni pregled „Dopuna antioksidativnim vitaminima za prevenciju i usporavanje napredovanja katarakte povezane sa starenjem" korišten je kao osnova i prema njegovim kriterijima za uključivanje pokušali smo pronaći RCT-ove i sustavne preglede objavljene nakon Cochranovom sustavnom pregledu, koji su dali dodatne dokaze o sigurnost i učinkovitosti antioksidativnih vitaminskih suplementacija u pogledu katarakte povezane sa starenjem. Pretražili smo PubMed, CENTRAL i DARE. Nakon pretrage napravili smo procjenu kvalitete sistematskog pregleda pomoću R-AMSTAR-a.

Rezultati: Cochrane sustavni pregled uključivao je devet ispitivanja s ukupno 117 272 osobe u dobi od 35 godina i više. Praćenje ispitivanja kretalo se od 2,1 do 12 godina, a ispitivanja su provedena u Australiji, Finskoj, Indiji, Italiji, Ujedinjenom Kraljevstvu i Sjedinjenim Državama. Doze antioksidativnih vitamina premašile su preporučenu dnevnu dozu. Nije bilo dokaza o učinku dodataka vitamina antioksidansa u smanjenju rizika od katarakte, ekstrakcije katarakte, progresije katarakte ili usporavanja gubitka vidne oštrine. U zbirnim analizama nije bilo dokaza o učinku suplementacije beta-karotena u smanjenju rizika od katarakte (dva ispitivanja) (relativni rizik 0,99, 95% interval pouzdanosti 0,91-1,08; broj ispitanika 57 703) ili u smanjenju rizika od ekstrakcije katarakte (tri ispitivanja). Udio sudionika koji su razvili hiperkarotenodermiju (žutilo kože) dok su uzimali beta-karoten kretao se od 7,4% do 15,8%.
Pronađen je samo jedan dodatni RCT koji je uključivao 11.267 isključivo muških sudionika iz Sjedinjenih Država, Kanade i Portorika, s dobnim rasponom od 50 godina i više. Zanimao nas je samo dio ovog RCT-a, dio koji se odnosi na suplementaciju vitaminom E. Trajao je 5,6 godina.

Zaključak: Ne može se pronaći dovoljno dokaza za sigurnost i učinkovitost antioksidativnih vitaminskih suplemenata za prevenciju i usporavanje katarakte povezane sa starenjem. U budućnosti bi se mogla provesti daljnja istraživanja među neuhranjenim ili dobro uhranjenim stanovništvom, ali u vezi s razinama antioksidansa u krvi, a ne suplementaciji.

11. SUPPLEMENT

Supplement 1: Characteristics of the studies included in the Cochrane systematic review (49)

APC 2006

"The Antioxidants in Prevention of Cataracts Study: effects of antioxidant supplements on cataract progression in South India"(77)

Methods	Design: multicenter, parallel-arm RCT Method of randomization: in blocks of 40 Method of allocation concealment: the placebo tablets were identical to active tablets in appearance and taste Number randomized: 798 Exclusions after randomization: none Number analyzed: 798; Group 1 = 398; Group 2 = 400 Masking: participants, study workers, investigators, biostatistician masked Losses to follow-up: equal across treatment groups (personal communication) Unit of analysis: analysis conducted for each eye separately
Participants	Country: India Age: range: 35 to 50 years Gender (% female): 63.8% in Group 1, 58.5% in Group 2 Inclusion criteria: age 35 to 50 years, best corrected visual acuity of 20/40 or better in both eyes Exclusion criteria: diabetes mellitus, intraocular surgery, radiation therapy, steroid therapy, active use of vitamin supplements, presence of congenital or traumatic cataract, active infectious keratitis, narrow anterior chamber angle
Interventions	Treatment: vitamin C: 500 mg; vitamin E: 400 IU; beta-carotene: 15 mg 3 times a week Control: placebo Duration of treatment/length of follow-up: 5 years
Outcomes	<u>Primary:</u> 1. Change in nuclear opalescence from baseline using Lens Opacities Classification System III

	Secondary: 1. Change from baseline of nuclear color 2. Change from baseline of cortical cataract 3. Change from baseline of posterior subscapular cataract 4. Change from baseline of best corrected spectacle visual acuity 5. Change from baseline of refraction 6. Failure of treatment defined as cataract progression to a point necessitating surgery or best corrected visual acuity of 20/400 or worse 7. Cataract surgery was offered if best corrected visual acuity decreased to 20/60 or worse or if decreased visual acuity caused problems with everyday functioning	
Notes	Study period: 5 years, 1999 to 2004 Study population: majority were middle class or lower in rural South India Subgroup analysis: results stratified by age Control group event rate: continuous outcomes were used Equivalence of baseline characteristics: important baseline characteristics appear equally distributed Quality of life indicators: none reported Funding: Francis I. Proctor Foundation, Aravind Eye Hospital, Peierls Foundation, Jack and DeLoris Lange Foundation, Harper Inglis Trust	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Randomization was completed in blocks of 40 and within this group half were assigned to each treatment
Allocation concealment (selection bias)	Low risk	The placebo tablets were identical to active tablets in appearance and taste

Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, study workers, investigators, biostatistician were masked
Incomplete outcome data (attrition bias) All outcomes	Unclear risk	Data on losses to follow-up were unavailable, but were balanced across treatment groups
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes described in the methods section
Other bias	Low risk	Met other parameters of quality that were assessed

AREDS 2001

"A randomized, placebo-controlled, clinical trial of high-dose supplementation with vitamins C and E and beta carotene for age-related cataract and vision loss: AREDS report no. 9" (71)

Methods	Design: multicenter, parallel-arm RCT Method of randomization: computer-generated list (AREDS2 Advantage Electronic Data Capture system (AdvantageEDCSM)). Method of allocation concealment: the study tablets were identical in external and internal appearance and taste Number randomized: 4757 Exclusions after randomization: 128 Number analyzed: 4629; antioxidants = 2304; no antioxidants = 2325 Masking: participants, care providers and outcomes assessors masked Losses to follow-up: 15% (includes those lost to follow-up and current smokers who withdrew from the study) Unit of analysis: individuals
Participants	Country: USA Age: median: 68 years; range: 55 to 80 years

	Gender (% female): 56% Inclusion criteria: best corrected visual acuity of 20/32 or better in at least one eye, at least one eye was free from eye disease that could complicate assessment of age-related macular degeneration, lens opacity progression or visual acuity Exclusion criteria: illness or disorders such as history of cancer with a poor 7-year prognosis, major cardiovascular or cerebrovascular event within the last year or hemochromatosis
Interventions	Treatment: vitamin C: 500 mg; vitamin E: 400 IU; beta-carotene: 15 mg daily 58% (n = 853) of those in the antioxidant group also received 80 mg of Zinc Control: placebo Duration of treatment/length of follow-up: average 6.3 years
Outcomes	1. Incidence of a cortical, nuclear or posterior subcapsular event 2. Incidence of any lens event 3. Best-corrected visual acuity 4. Incidence of cataract surgery 5. Incidence of any severe lens event Outcomes were assessed at 6 months and annually Some participants had 1 eye enrolled in the study and others had 2 study eyes
Notes	Study period: 1992 to 2001 Study population: apparently well-nourished older cohort Subgroup analysis: (a) on eyes with no or minimal opacity in one eye (n = 823), (b) no opacity in both eyes (n = 338) at baseline, (c) by type of cataract Control group event rate: 34% for any lens event over 5 years Equivalence of baseline characteristics: important baseline characteristics appear equally distributed. Approximately, 66% of AREDS participants chose to take CENTRUM, a

	commercially available multivitamin-mineral supplement, the use was balanced across treatment groups Quality of life indicators: reported Funding: National Eye Institute, National Institutes of Health, USA and Bausch and Lomb Inc	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Computer-generated list
Allocation concealment (selection bias)	Low risk	The study tablets were identical in external and internal appearance and taste
Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, care providers and outcomes assessors were masked
Incomplete outcome data (attrition bias) All outcomes	Low risk	90% had at least five years of follow- up. The losses to follow-up were balanced across treatment groups
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes described in the methods section
Other bias	Low risk	Met other parameters of quality that were assessed

ATBC 1998

"Incidence of cataract operations in Finnish male smokers unaffected by alpha tocopherol or beta carotene supplements" (78)

Methods	Design: 2 X 2 factorial RCT Method of randomization: unavailable Method of allocation concealment: unavailable Number randomized: 29,133 Exclusions after randomization: 199 Number analyzed: 28,934

	Masking: participants, care providers and outcomes assessors masked Losses to follow-up: 28.4% in the alpha-tocopherol alone group to 29.4% in the beta- carotene alone group Unit of analysis: individuals
Participants	Country: Finland Age: median: 57 years; range: 50 to 69 years Gender: all men Inclusion criteria: smokers of at least 5 cigarettes per day, absence of lung cancer as determined by an X-ray Exclusion criteria: those with a history of cancer or serious disease limiting the ability to participate, those taking supplements of vitamin E, vitamin A or beta-carotene in excess of predefined doses, those on treatment with anticoagulants
Interventions	Treatment: (a) Alpha-tocopherol: 50 mg once daily (b) Beta-carotene: 20 mg once daily (c) Combination: once daily Control: placebo Duration of treatment/length of follow-up: 5 to 8 years; median: 5.7 years; 1,59,199 person-years
Outcomes	1. Incidence of cataract extraction Outcome was identified from the National Hospital Discharge Registry using International Classification of Diseases codes
Notes	Study period: 1986 to 1992 Study population: apparently healthy male smokers over 50 years Subgroup analysis: age Control group event rate: 1.44% over a median period of 5.7 years

	Equivalence of baseline characteristics: important baseline characteristics appear equal. There were fewer participants with diabetes in the placebo group Quality of life indicators: none reported Funding: National Public Health Institute of Finland and National Cancer Institute, National Institutes of Health, USA Cataract extraction rates may differ by type of cataract, e.g., Posterior Subcapsular Cataract is treated early because of early decrease in vision	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Unclear risk	Information unavailable
Allocation concealment (selection bias)	Unclear risk	Information unavailable
Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, care providers and outcomes assessors were masked
Incomplete outcome data (attrition bias) All outcomes	Unclear risk	Losses to follow-up were greater than 28%, but were roughly equal across treatment groups
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes described in the methods section
Other bias	Low risk	Met other parameters of quality that were assessed

PHS I 2003

"A randomized trial of beta carotene and age-related cataract in US physicians"(79)

Methods	Design: 2 X 2 factorial RCT Method of randomization: computer-generated list of random numbers Method of allocation concealment: study pills in the treatment arms were identical except for the active agent in the beta-carotene group Number randomized: 22,071 Exclusions after randomization: 1103; aspirin and beta-carotene: 286, aspirin and placebo: 278, beta-carotene and placebo: 275, placebo only: 264 Number analyzed: 20,968; aspirin and beta-carotene = 5231, aspirin and placebo = 5242, beta-carotene and placebo = 5244, placebo only = 5251 Masking: participants, care providers and outcomes assessors masked Losses to follow-up: 99.2% provided information on morbidity after 11 years Unit of analysis: individuals
Participants	Country: USA Age: mean: 52.6 years; range: 40 to 84 years Gender: all male Inclusion criteria: no history of cancer (except non-melanoma skin cancer), myocardial infarction, stroke or transient cerebral ischemia Exclusion criteria: current use of vitamin A supplement
Interventions	Treatment: (a) Beta-carotene: 50 mg on alternate days (b) Aspirin: 325 mg on alternate days Control: placebo, aspirin Duration of treatment/length of follow-up: 12 years; range: 11.6 to 14.2 years
Outcomes	1. Incidence of age-related cataract over 12 years

	2. Extraction of age-related cataract over 12 years Outcome assessment was based on self-reports confirmed by medical record review Outcome assessed in the worse eye is used in the analysis	
Notes	Study period: 1982 to 1995 Study population: apparently healthy male medical professionals over 40 years Subgroup analysis: (a) age (b) baseline smoking status Control group event rate: 9.7% over 12 years for incident cataract Equivalence of baseline characteristics: important baseline characteristics appear equally distributed Quality of life indicators: none reported Funding: National Institutes of Health, USA	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Computer-generated list of random numbers
Allocation concealment (selection bias)	Low risk	Study pills in the treatment arms were identical except for the active agent in the beta- carotene group
Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, care providers and outcomes assessors masked
Incomplete outcome data (attrition bias) All outcomes	Low risk	Morbidity follow-up rate was 99.2% and balanced across treatment groups
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes described in the methods section
Other bias	Low risk	Met other parameters of quality that were assessed

PHS II 2010

"Age-related cataract in a randomized trial of vitamins E and C in men"(70)

Methods	Design: 2 X 2 X 2 X 2 factorial RCT Method of randomization: computer-generated list of random numbers Method of allocation concealment: study pills in the treatment arms were identical Number randomized: 14,641 Exclusions after randomization: 3096; vitamin C and vitamin E: 771, vitamin C: 759, vitamin E: 773, placebo: 793 Number analyzed: 11,545; vitamin C and vitamin E: 2885, vitamin C: 2914, vitamin E: 2886, placebo: 2860 Masking: participants, care providers and outcomes assessors masked Losses to follow-up: 95.3% for morbidity and 97.7% for mortality Unit of analysis: individuals
Participants	Country: USA Age: mean: 62 years, standard deviation: 7.9 Gender: all male Inclusion criteria: no history of cancer (except non-melanoma skin cancer), cardiovascular disease, current liver disease, renal disease, peptic ulcer or gout Exclusion criteria: unwillingness to avoid use of non-study supplements
Interventions	Treatment: (a) Vitamin C: 500 mg daily (b) Vitamin E: 400 IU on alternate days Control: placebo, vitamin C, vitamin E Duration of treatment/length of follow-up: mean: 8 years
Outcomes	1. Incidence of age-related cataract over 12 years 2. Extraction of age-related cataract over 12 years Outcome assessment was based on self-reports confirmed

	by medical record review Outcome assessed in the worse eye is used in the analysis	
Notes	Study period: 1997 to 2007 Study population: apparently healthy male medical professionals over 50 years Subgroup analysis: (a) age (b) type of cataract and possible risk factors Control group event rate: vitamin C: 10.1%, vitamin E: 10.3% over 8 years for incident cataract Equivalence of baseline characteristics: important baseline characteristics appear equally distributed Quality of life indicators: none reported Funding: National Institutes of Health, USA, BASF Corporation	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Computer-generated list of random numbers
Allocation concealment (selection bias)	Low risk	Study pills in the treatment arms were identical
Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, care providers and outcome assessors masked
Incomplete outcome data (attrition bias) All outcomes	Low risk	Morbidity follow-up rate was 95.3% and balanced across treatment groups
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes described in the methods section
Other bias	Low risk	Met other parameters of quality that were assessed

PPP 2001

"Epidemiological feasibility of cardiovascular primary prevention in general practice: a trial of vitamin E and aspirin. Collaborative group of the Primary Prevention Project"(73)

Methods	Design: 2 X 2 factorial RCT Method of randomization: computer-generated randomization table Method of allocation concealment: treatments were centrally assigned on telephone verification of the correctness of inclusion criteria with a separate computer-generated randomization table produced for each physician in random permuted blocks of 12 Number randomized: 4495 Exclusions post randomization: none Number analyzed: 4495 Masking: open label trial. Clinical events were validated by an expert committee masked to treatment assignment, unclear if it extended to the outcome of interest in this review Losses to follow-up: vitamin E group: 14, placebo group: 17 Unit of analysis: individuals
Participants	Country: Italy Age: mean: 64.5, standard deviation: 7.6 - 7.7 Gender: both male and female Inclusion criteria: age over 50 years with at least one of the major cardiovascular risk factors Exclusion criteria: treatment with antiplatelet drugs, anti-inflammatory agents or anti- coagulants, those with diseases with poor short-term prognosis
Interventions	Treatment: vitamin E: 300 mg daily Control: placebo Duration of treatment/length of follow-up: mean of 3.6 years
Outcomes	1. Incidence of cataract surgery Outcome assessment was validated by chart review

Notes	Study period: 1993 to 1998 Study population: individuals with at least one risk factor for cardiovascular disease Subgroup analysis: none Control group event rate: 2.7% Equivalence of baseline characteristics: important baseline characteristics appear equally distributed Quality of life indicators: none reported Funding: public source	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Computer-generated list of random numbers
Allocation concealment (selection bias)	Low risk	Treatments were centrally assigned
Blinding (performance bias and detection bias) All outcomes	Unclear risk	Open-labeled trial
Incomplete outcome data (attrition bias) All outcomes	Low risk	Losses to follow-up were 0.6% and 0.75% in the treatment and placebo groups
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes described in the methods section
Other bias	Low risk	More than 13% of those randomized to vitamin E discontinued the medication, which is likely to bias the results towards no effect

REACT 2002

"The Roche European American Cataract Trial (REACT): a randomized clinical trial to investigate the efficacy of an oral antioxidant micronutrient mixture to slow progression of age-related cataract" (80)

Methods	Design: multicenter, parallel-arm RCT Method of randomization: Efron's biased coin method Method of allocation concealment: those involved in preparing the randomization scheme were not associated with determining eligibility, administering the intervention or assessing the outcomes Number randomized: 297 Exclusions after randomization: none (see number analyzed and losses to follow-up) Number analyzed: completers of the study at 3 years: antioxidants = 81; placebo = 77 Masking: participants, care providers and outcomes assessors masked Losses to follow-up: 66 (22%) after two years, 139 (47%) after 3 years, 261 (88%) after 4 years Unit of analysis: individuals
Participants	Country: USA and UK Age: USA: mean: 64.7 years, standard deviation: 9.1 years, UK: mean: 67.9 years, standard deviation: 8.5 years Gender (% female): USA: 62.4%, UK: 55.7% Inclusion criteria: age, 40 years or older, at least at eye satisfying the following criteria: cataract extraction unlikely within the next 2 years, immature idiopathic senile cataract present in at least one or both eyes, logMAR acuity < = 0.5, no clinical signs of glaucoma Exclusion criteria: use of vitamin supplements
Interventions	Treatment: Beta-carotene: 6 mg; vitamin C: 250 mg; all-rac alpha-tocopherol acetate: 200 mg capsules 3 times per day Control: placebo

	Duration of treatment/length of follow-up: mean = 34 months; standard deviation: 12 months Planned: 2 years, decided on 3 years used for the primary analysis (after the results of the interim analysis suggested a difference in effect)	
Outcomes	1. Progression of cataract 2. Logarithm of Minimum Angle of Resolution visual acuity Outcomes were assessed approximately every 4 months It is not clear whether outcome data were from the worse eye, or the average of both eyes	
Notes	Study period: 1990 to 1995 Study population: apparently healthy people over 40 years with some degree of age- related cataract Subgroup analysis: (a) type of cataract (b) severity of cataract (based on % Pixels Opaque - Anterior) Control group event rate: not applicable Equivalence of baseline characteristics: those in the United Kingdom were slightly older, had lower serum proteins, poorer liver function, lower vitamin levels, less brunescent lenses and more nuclear and cortical opacification Quality of life indicators: none reported Funding: Industry: Hoffmann-La Roche	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Efron's biased coin method
Allocation concealment (selection bias)	Low risk	Those involved in preparing the randomization scheme were not associated with determining eligibility, administering the intervention or assessing the outcomes

Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, care providers and outcomes assessors were masked
Incomplete outcome data (attrition bias) All outcomes	High risk	Losses to follow-up were 22% after 2 years and 47% after 3 years
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes de- scribed in the methods section
Other bias	Unclear risk	Those in the United States were apparently healthier with less mature cataracts at baseline

VECAT 2004

"Vitamin E supplementation and cataract: randomized controlled trial" (72)

Methods	Design: parallel-arm RCT Method of randomization: using permuted blocks Method of allocation concealment: the allocation list was stored at a remote site and medication was dispensed in identical containers Number randomized: 1204 Exclusions after randomization: 11 Number analyzed: vitamin E = 595, placebo = 598; completers: vitamin E = 443, placebo = 456 Masking: participants, care providers and outcomes assessors masked Losses to follow-up: withdrawn: vitamin E: 78, placebo: 72; discontinued: vitamin E: 74, placebo: 70 Unit of analysis: individuals
Participants	Country: Australia Age: mean: 65.67 years; range: 55 to 80 years Gender (% female): 56% Inclusion criteria: age between 55 and 80 years

	Exclusion criteria: prior cataract surgery, advance cataract in both eyes, glaucoma, known sensitivity to vitamin E, long-term treatment with steroids and anti-coagulants
Interventions	Treatment: (a) Vitamin E, 500 IU natural vitamin E in soybean oil daily Control: placebo Duration of treatment/length of follow-up: 4 years
Outcomes	1. Incidence of cataract (nuclear, cortical and posterior subcapsular, clinical grading and digital assessment) over 4 years 2. Cataract extraction over 4 years 3. Progression of cataract (nuclear, cortical and posterior subcapsular, clinical grading and digital assessment) over 4 years Outcomes were assessed annually Incidence and prevalence were assessed using data from the worse eye, but progression rates were not derived from the eye with the most advanced cataract change at baseline
Notes	Study period: 1995 to 2000 Study population: apparently healthy people over 55 years with some degree of age- related cataract Subgroup analysis: (a) type of cataract Control group event rate: 16.7% over 4 years for incident cataract Equivalence of baseline characteristics: the vitamin E group had a statistically significant greater number of cases of cortical and any cataract at baseline. Other baseline characteristics appear equally distributed Quality of life indicators: assessed using health-related quality of life SF-36 and visual function 14 questionnaires. Data not reported Funding: National Health and Medical Research Council of Australia and other foundations
Risk of bias	

Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	Randomization schedule was prepared by a biostatistician using permuted blocks allocation scheme
Allocation concealment (selection bias)	Low risk	The allocation list was stored at a remote site and medication was dispensed in identical containers
Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, care providers and outcomes assessors were masked
Incomplete outcome data (attrition bias) All outcomes	Unclear risk	Losses to follow-up were greater than 23%, but were roughly balanced across treatment groups
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes de- scribed in the methods section
Other bias	Unclear risk	The vitamin E group had a statistically significant greater number of cases of cortical and any cataract at baseline and these were excluded from the analysis for incidence of cataract

WHS 2004/8

"Vitamin E and age-related cataract in a randomized trial of women" (81)

Methods	Design: 2 X 2 X 2 factorial RCT Method of randomization: computer-generated list of random numbers Method of allocation concealment: study pills in the treatment arms were identical except for the active agent in

	the beta-carotene group Number randomized: 39,876 Exclusions after randomization: Beta-carotene component: 3141; beta-carotene: 1534; placebo: 1607 Vitamin E component: 2201; vitamin E: 1137; placebo: 1064 Number analyzed: Beta-carotene component: 36,735; beta-carotene: 18,405; placebo: 18,330 Vitamin E component: 37,675; vitamin E: 18,800; placebo: 18,875 Masking: participants, care providers and outcome assessors masked Losses to follow-up: the beta-carotene component was terminated early and for the vita- min E component, mortality and morbidity follow-up were 97.2% and 99.4% respectively Unit of analysis: individuals
Participants	Country: USA Age: mean: beta-carotene component: 53.2 years; vitamin E component: 54.1 years; range: 45 years and older Gender: all female Inclusion criteria: no history of cancer (except non melanoma skin cancer), coronary heart disease or cerebrovascular disease Exclusion criteria: see above
Interventions	Treatment: (a) Beta-carotene, 50 mg on alternate days (b) Vitamin E: 600 IU on alternate days (c) Aspirin 100 mg on alternate days Control: placebo, aspirin, beta-carotene, vitamin E Duration of treatment/length of follow-up: Beta-carotene component: median: 2.1 years; range: 0.00 to

	2.72 years Vitamin E component: average: 9.7 years Compliance: Beta-carotene component: 87% reported taking at least 2/3rd of study capsules; vitamin E component: 78.9% reported taking at least 2/3rd of study capsules at 5 years and 71. 6% at 10 years	
Outcomes	1. Incidence of age-related cataract 2. Extraction of age-related cataract Outcomes were assessed every 6 months for first year and annually thereafter Outcome assessment was based on self-reports confirmed by medical record review Outcome assessed in the worse eye is used in the analysis	
Notes	Study period: 1993 to 2004 Study population: apparently health women health professionals Subgroup analysis: Beta-carotene component: (a) age, (b) baseline smoking status Vitamin E component: (a) age, (b) type of cataract Control group event rate: Beta-carotene component: 0.007% over 2.1 years for incident cataract Vitamin E component: 6.5% over 9.7 years for incident cataract Equivalence of baseline characteristics: important baseline characteristics appear equally distributed Quality of life indicators: none reported Funding: National Institutes of Health, USA	
Risk of bias		
Bias	**Authors' judgement**	**Support for judgement**
Random sequence generation (selection bias)	Low risk	Computer-generated list of random numbers
Allocation concealment (selection bias)	Low risk	Study pills in the treatment arms were identical except

		for the active agent in the beta- carotene group
Blinding (performance bias and detection bias) All outcomes	Low risk	Participants, care providers and outcome assessors were masked
Incomplete outcome data (attrition bias) All outcomes	Low risk	Morbidity follow-up rates for the beta- carotene and vitamin E arms were 99% and 97.2% respectively
Selective reporting (reporting bias)	Low risk	Results were reported for outcomes de- scribed in the methods section
Other bias	Low risk	Met other parameters of quality that were assessed