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EFFICACY OF ANTIOXIDANT TREATMENT IN THE MANAGEMENT OF ORAL LEUKOPLAKIA : SYSTEMATIC REVIEW

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1. RESUME

Introducción: La leucoplasia oral es el trastorno potencialmente maligno más frecuente de la cavidad oral. Su desarrollo y posible transformación maligna se han relacionado con el estrés oxidativo. En este contexto, los tratamientos antioxidantes han sido propuestos como una estrategia terapéutica para contrarrestar el daño oxidativo y prevenir la progresión de las lesiones.

Objetivos: Evaluar la eficacia clínica y bioquímica de los tratamientos antioxidantes en pacientes con leucoplasia oral.

Material y método: Se realizó una revisión sistemática siguiendo las directrices PRISMA. Se efectuó una búsqueda electrónica en las bases de datos PubMed, Scopus y Web of Science de estudios publicados entre 2000 y 2025. Se incluyeron ensayos clínicos aleatorizados, estudios clínicos controlados y estudios observacionales que evaluaran el uso de antioxidantes tópicos o sistémicos en pacientes con leucoplasia oral.

Resultados: Se incluyeron 11 estudios con un total de 695 pacientes. Los antioxidantes más investigados fueron el licopeno, la curcumina, la vitamina A y el fenretinida. La mayoría de los estudios reportaron mejoras clínicas significativas en cuanto a reducción del tamaño o regresión de la lesión. A nivel bioquímico, se observó una disminución del estrés oxidativo evidenciada por la reducción de los niveles de LPO, MDA y 8-OHdG, así como un aumento de las enzimas antioxidantes como la catalasa (CAT), vitaminas C y E. Algunos estudios mostraron una tasa elevada de apoptosis en el epitelio alterado. Los resultados sobre la superóxido dismutasa (SOD) fueron variables. La heterogeneidad en los diseños, dosis y duración del tratamiento limitó la comparación directa entre estudios.

Conclusión: Los antioxidantes representan una opción terapéutica prometedora para el manejo de la leucoplasia oral, no solo por su capacidad de mejorar los parámetros clínicos, sino también por su impacto positivo en los biomarcadores del estrés oxidativo. Su incorporación en protocolos terapéuticos podría contribuir a reducir el riesgo de progresión a carcinoma oral, aunque se necesitan más investigaciones.

2. ABSTRACT

Introduction: Oral leukoplakia is the most common potentially malignant disorder of the oral cavity. Its development and potential malignant transformation have been associated with oxidative stress. In this context, antioxidant treatments have been proposed as a therapeutic strategy to counteract oxidative damage and prevent lesion progression.

Objectives: To evaluate the clinical and biochemical efficacy of antioxidant treatments in patients with oral leukoplakia.

Material and Methods : A systematic review was conducted following the PRISMA guidelines. An electronic search was performed in the PubMed, Scopus, and Web of Science databases for studies published between 2000 and 2025. Randomized controlled trials, controlled clinical studies, and observational studies assessing the use of topical or systemic antioxidants in patients with oral leukoplakia were included.

Results: A total of 11 studies involving 695 patients were included. The most studied antioxidants were lycopene, curcumin, vitamin A, and fenretinide. Most studies reported significant clinical improvements in terms of lesion size reduction or regression. On a biochemical level, a decrease in oxidative stress was observed, evidenced by reductions in LPO, MDA, and 8-OHdG levels, as well as increases in antioxidant enzymes such as catalase (CAT), vitamins C and E. Some studies reported higher apoptosis rates in altered epithelium. Results concerning superoxide dismutase (SOD) were variable. The heterogeneity in study design, dosage, and treatment duration limited direct comparison between studies.

Conclusion: Antioxidants represent a promising therapeutic option for managing oral leukoplakia, not only for their ability to improve clinical outcomes but also due to their positive impact on oxidative stress biomarkers. Their inclusion in therapeutic protocols could help reduce the risk of progression to oral carcinoma, although further research is required.

3. KEYS WORDS

I. Oral leukoplakia

II. Leukoplakia

III. Antioxidants

IV. Antioxidant therapy

V. Antioxidant treatment

4. ABBREVIATIONS

ATP: Adenosine Triphosphate

CAT: Catalase

CI : Confidence interval

DNA: Deoxyribonucleic Acid

GPx: Glutathione Peroxidase

GSH: Glutathione

GSSH: Oxidized glutathione

HR : Hazard ratio

H_2O_2: Hydrogen peroxide

IL-6: Interleukin-6

LPO: Lipid peroxidation

MDA: Malondialdehyde

NO: Nitric Oxide

OH•: Hydroxyl Radical

OL: Oral Leukoplakia

OSCC: Oral Squamous Cell Carcinoma

O_2: Oxygen

O_2^-: Superoxide

PMD: Potentially malignant disorder

ROS: Reactive Oxygen Species

SOD: Superoxide Dismutase

TNF-α: Tumor Necrosis Factor Alpha

UV: Ultraviolet

8-OHdG: 8-hydroxy-2’-deoxyguanosine

5. INTRODUCTION

5.1. General Overview

Oral leukoplakia (OL) is one of the most common potentially malignant disorders (PMD) of the oral cavity, characterized by the presence of white patches on the mucosal surfaces that cannot be clinically or pathologically identified as any other disease. The lesions appear on the mucosa of the oral cavity and are usually asymptomatic, though they can cause discomfort in some cases. OL is considered a premalignant condition due to its potential for malignant transformation into oral squamous cell carcinoma (OSCC), a type of oral cancer that accounts for a significant proportion of cancers worldwide. Studies estimate that approximately 5-15% of individuals with OL may experience malignant transformation, which underscores the importance of early detection and appropriate management of this disorder (1,2).

The exact etiology of OL remains unclear, but it is thought to arise from an imbalance in the normal processes of keratinization in the oral mucosa. In many cases, OL is associated with significant lifestyle factors such as tobacco use (both smoked and smokeless forms) and excessive alcohol consumption, which are considered major risk factors for the development of OL (2,3). The prevalence of OL varies depending on the geographic location, the population studied, and the associated risk factors. In countries with high rates of tobacco and alcohol consumption, such as in Europe and Asia, the incidence of OL is notably high. Conversely, the condition is less common in regions with lower tobacco use and reduced alcohol intake (3,4). The lack of overt symptoms during the early stages of OL makes early diagnosis and intervention challenging, highlighting the need for continuous surveillance in high-risk populations.

Recent advances in molecular biology and epidemiology have increased the understanding of the condition. Research into the molecular mechanisms underlying OL has suggested that genetic mutations, oxidative stress, and alterations in signalling pathways all contribute to the development of the lesions (5). Additionally, a number of antioxidant-based therapies are now being investigated as potential treatment for OL, as they may help reverse oxidative damage and reduce the risk of malignant transformation (6).

5.2. Oral Leukoplakia

OL is defined as a white patch or plaque on the oral mucosa that cannot be clinically or pathologically diagnosed as any other condition. The white appearance is due to an abnormal accumulation of keratin, a protein that helps protect the mucosal surfaces of the mouth. The condition most commonly affects the buccal mucosa, tongue, and floor of the mouth, and although many cases remain benign, a subset of patients with OL may develop malignant transformation, often resulting in OSCC. The pathogenesis of OL involves both environmental and genetic factors that contribute to abnormal mucosal keratinization and subsequent dysplasia (2).

5.2.1. Causes of Oral Leukoplakia

The causes of OL are multifactorial, with environmental, lifestyle, and genetic factors all playing a significant role. The most widely recognized risk factors for OL include tobacco use (both smoked and smokeless forms), alcohol consumption, and chronic mechanical irritation from poorly fitting dentures, rough teeth, or ill-fitting dental appliances. These factors contribute to the development of OL by increasing the local oxidative stress in the oral cavity, which promotes cellular damage and abnormal cell proliferation (3).

Tobacco use, in particular, is a major contributor to the development of OL, with studies showing that smokers have a significantly higher risk of developing OL compared to non-smokers. Tobacco use was found to be a significant risk factor for both the development of OL and its progression to oral cancer (1). Alcohol consumption, particularly when combined with tobacco use, further increases the risk of malignant transformation in individuals with OL. Additionally, exposure to ultraviolet (UV) radiation from the sun has been identified as a risk factor for OL, particularly in areas of the oral mucosa that are more exposed to sunlight, such as the lips (4).

Genetic predisposition also plays a role in the development of OL. Studies have identified mutations in genes related to cell cycle regulation, DNA repair, and apoptosis as contributing factors to the development of OL and its

progression to cancer. These genetic alterations may render individuals more susceptible to oxidative damage and carcinogenesis (5).

5.2.2. Symptoms of Oral Leukoplakia

OL is often asymptomatic in its early stages, making it difficult for patients to recognize the condition. The primary symptom is the presence of a white patch or plaque in the mouth, which can vary in size, shape, and texture. The lesions are typically painless, although some patients may experience mild discomfort or a burning sensation, especially when eating spicy or acidic foods. As the condition progresses and if malignant transformation occurs, patients may experience pain, difficulty eating, swallowing, or speaking, as well as the presence of ulcerations or bleeding in the affected area (7).

Although initial symptoms are often mild; chronic inflammation related to oxidative stress activates pro-inflammatory molecules such as interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α), which can contribute to pain and discomfort associated with OL. These cytokines play a crucial rôle in the inflammatory response and could also be involved in the progression of the lesions towards malignancy (3,8,9).

5.2.3. Diagnosis of Oral Leukoplakia

The diagnosis of OL is primarily clinical, based on visual inspection of the oral mucosa. A white lesion that cannot be attributed to other causes, such as candidiasis, lichen planus, or frictional keratosis, is suggestive of OL. However, a definitive diagnosis requires histopathological examination, as OL can resemble other conditions. A biopsy is essential to assess the presence of dysplasia, a precursor to cancer, and to determine the risk of malignant transformation. Dysplastic changes in the epithelium, including alterations in cell morphology and increased mitotic activity, are important indicators of the potential for malignancy (10).

In addition to biopsy, adjunctive diagnostic tools such as toluidine blue staining, autofluorescence, and brush cytology may be used to aid in the diagnosis and monitoring of OL. These tools can help identify areas of concern

that may require further investigation or intervention (3,10,11). Moreover, the evaluation of biomarkers associated with oxidative stress, such as glutathione (GSH), superoxide dismutase (SOD), and glutathione peroxidase (GPx), may offer additional diagnostic value. These biomarkers can provide insight into the oxidative damage and antioxidant capacity of the tissue, which may correlate with the severity and potential for malignant transformation of the OL lesion.

5.2.4. Treatment of Oral Leukoplakia

The management of OL focuses on reducing the risk of malignant transformation and alleviating any symptoms associated with the lesions. The most commonly used treatment modalities for OL include surgical excision, laser ablation, and cryotherapy. These procedures aim to remove the lesions and reduce the chances of recurrence. However, surgical treatment is often associated with risks such as scarring, pain, and the potential for recurrence of the lesions (2,3,12).

In light of these risks, medical therapies have been explored as alternative or adjunctive treatments for OL. Topical corticosteroids, retinoids, and antioxidant treatments have shown promise in reducing the size of OL lesions and promoting healing of the mucosa. These treatments may also help reduce the oxidative stress that contributes to the progression of the lesions (5,11,13).

5.2.5. Prevention and risk factors of Oral Leukoplakia

Prevention of OL primarily involves addressing modifiable risk factors such as smoking cessation and alcohol reduction. Public health initiatives aimed at reducing tobacco and alcohol consumption have been shown to decrease the incidence of OL in certain populations (3). Maintaining good oral hygiene and avoiding chronic irritation from poorly fitting dental appliances are also important preventive measures (2).

Regular dental check-ups and screenings are essential for the early detection of OL, as the condition often remains asymptomatic in its early stages. Early diagnosis allows for timely intervention, which can prevent malignant transformation. Furthermore, the potential role of diet and antioxidants in the

prevention of OL is an area of ongoing research. Diets rich in fruits and vegetables, which contain natural antioxidants, may help reduce the oxidative stress associated with the development of OL (3,6).

5.3. Oxidative stress

Oxidative stress is a biochemical phenomenon that results from an imbalance between the production of free radicals and reactive oxygen species (ROS) and antioxidant defense capacity to neutralize or repair the resulting damage. ROS include free radicals such as superoxide (O_2^-) and hydroxyl radical (HO•), as well as non-radical molecules like hydrogen peroxide (H_2O_2). Under normal conditions, these reactive species are naturally produced in the body, and play a crucial role in essential physiological processes. Primarily generated by normal cellular processes, such as mitochondrial respiration, where oxygen is used to produce energy (ATP) (9,14). Also cell signaling and immune defense use and produce ROS, for example, they are used by immune cells to destroy pathogens.

However, in situations of oxidative stress, an excess of ROS, or an inability to neutralize these reactive molecules, can lead to biomolecules, including lipids, proteins and DNA damage (9). These alterations can induce genetic mutations, disrupt cellular functions, and even promote chronic inflammatory processes. This accumulation of damage can lead to various pathologies, including cancer, cardiovascular diseases, and neurodegenerative disorders (13,15).

There is a defense system to protect cells from the harmful effects of ROS. This defense is organized into three lines. The first line of defense consists of antioxidant enzymes. These include superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx), which detoxify free radicals and peroxides, thereby reducing oxidative damage (11,16). SOD is an enzyme that catalyzes the conversion of superoxide (O_2^-) into hydrogen peroxide (H_2O_2) and molecular oxygen (O_2), thus reducing the harmful effects of superoxide. Glutathione peroxidase (GPx) is an enzyme that uses reduced glutathione (GSH) as a cofactor to reduce hydrogen peroxide (H_2O_2) and other lipid peroxide (17). Glutathione (GSH) is a tripeptide composed of glutamine, cysteine and glycine,

it is one of the most important antioxidants in the cell. It plays a major role in reducing ROS, particularly in reactions catalyzed by GPx.
It exists in two forms : reduced glutathione (GSH) and oxidized glutathione (GSSG), and the ratio between these two forms is a marker of the cell's redox state. This biomarker is particularly relevant in studies of oxidative stress and associated diseases (18,19). The second line of defense relies on dietary antioxidants, including vitamin C, is a water soluble antioxidant that protects cells by reducing free radicals. Also vitamin E, is a fat-soluble antioxidant that protects cell membranes from oxidative damage. The second line also includes carotenoids, and flavonoids, which help neutralize ROS and prevent their accumulations (9). Finally, the third line of defense involves enzymes that repair or eliminate oxidized biomolecules, contributing to maintaining cellular integrity (20). These defense systems work together to regulate oxidative stress and minimize cellular damage. However, while antioxidant therapies may offer benefits, they also have certain limitations, particularly in their effectiveness in treating diseases associated with oxidative stress.

In addition to these antioxidant defenses, inflammation plays a crucial role in the body's response to oxidative stress. Two major pro-inflammatory cytokines, interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α), are strongly involved in inflammatory responses (20). IL-6 is a cytokine that mediates immune responses and is critical in inflammation, while TNF-α is a potent pro-inflammatory molecule that plays a significant role in many inflammatory processes, contributing to the development of various diseases (15,17)

In the oral cavity, chronic oxidative stress can cause alterations in the cells of the oral mucosa, making them more vulnerable to precancerous conditions like oral leukoplakia. The impact of oxidative stress in this precancerous condition is well-documented and increasingly recognized as a major factor in the malignant transformation process (9).

5.3.1. Oxidative stress in OL

OL is considered a multifactorial disease, where oxidative stress plays a key role in the progression toward oral cancer. Studies have shown that the

activation of ROS and oxidative stress markers is particularly elevated in patients with OL. Which could accelerate the process of malignant transformation of oral cells (8).

Oxidative stress affects the cells of the oral mucosa through the excessive generation of free radicals, which damage DNA and alter proteins and lipids. This increases the likelihood of genetic mutations in the epithelial cells of the oral cavity, making these cells more prone to dysplasia and eventually carcinogenesis (6). Prolonged oxidative stress can also lead to chronic inflammation, which promotes the progression of precancerous lesions to invasive oral cancer (5).

Oxidative stress markers, such as lipid peroxidation products and oxidized protein levels, are used to assess the degree of oxidative stress in OL lesions. High levels of these biomarkers are associated with higher likelihood of progression to oral cancer, highlighting the importance of these indicators in monitoring the disease (8).

5.4. Antioxidants in Oral Leukoplakia

In the oral cavity, the balance between ROS production and antioxidant defense mechanisms is crucial to maintain the health of the oral mucosa and prevent diseases such as OL and potentially oral cancer (11).

In patients with OL, a deficiency in antioxidant defense systems is often observed. Studies have shown that the concentration of certain antioxidants, such as vitamin C, vitamin E and glutathione are significantly reduced in the saliva of these patients, which may increase their vulnerability to oxidative stress (6,9). Therefore, restoring an optimal antioxidant balance could offer a therapeutic strategy to prevent the progression of precancerous lesions.

Antioxidant supplementation, whether from dietary sources or pharmacological means, has shown promising results in counteracting the harmful effect of ROS. Polyphenols, flavonoids and carotenoids found in a variety of foods, particularly fruits, vegetables and spices are well-known antioxidants. These compounds play an important role in neutralizing free radicals and modulating oxidative stress, a key factor in precancerous lesions

progression(1,13). Polyphenols, such as the catechins in green tea and carotenoids, such as beta-carotene are known for their ability to protect oral epithelial cells from ROS-induced damage (13). By restoring a favorable redox balance in the oral cavity, these antioxidants may offer protection against OL progression. Targeted nutritional approaches that integrate these antioxidants into the diet could, therefore, play a key role in prevention and management of OL (1).

Furthemore, the topical application of antioxidant compounds such as vitamin C and E or other antioxidant extract, has been studied for its potential to reduce local oxidative stress. While specific research on the effect of topical treatments on OL lesions is limited, studies show that regulating oxidative stress in the oral cavity can help control local inflammation and limit cellular damage. These treatments, by improving the ability to neutralize ROS and reduce local inflammation, may reduce the progression of OL lesions to oral cancer (6,8,11).

In conclusion, managing oxidative stress through the use of antioxidants due to their potential to reduce oxidative stress, could represent a promising approach in the treatment and prevention of OL and other oral diseases associated with oxidative stress. However, research on the integration of antioxidant treatments into therapeutic strategies remains crucial to validate their effectiveness in managing these conditions.

6. JUSTIFICATION AND HYPOTHESIS OF THE STUDY

6.1. Justification and SDG

OL is a pre-cancerous condition characterized by white patches on the mucous membrane of the mouth and may progress into oral cancer. This pathology presents a major challenge in terms of prevention and treatment. While various treatments exist for OL, the use of antioxidant treatments remains a relatively under-explored area. This systematic review aims to assess the effectiveness of antioxidant treatments in managing this condition.

The importance of conducting this study lies in the fact that there is no clear consensus on the effectiveness of antioxidant treatments for OL. Current research on this topic is scattered, with sometimes conflicting results. A rigorous systematic review will provide a reliable synthesis of existing data and assess whether these treatments can truly play a role in reducing the progression of leukoplakia to cancer.

This systematic review will help clarify the role of antioxidants in the treatment of OL by gathering and analyzing the results of existing studies. It will determine whether the use of antioxidants is effective and safe for patients suffering from this condition. Additionally, this study may provide guidance for future clinical research and treatment recommendations in the management of OL.

This review will provide a comprehensive evaluation of the effectiveness of antioxidant treatments by examining the different types of antioxidants used (vitamins, plants, etc.) and their specific effects on OL. It will also help to better understand the underlying biological mechanisms and identify the most promising treatments in this field.

The primary beneficiaries will be healthcare professionals, including dentists, doctors, and researchers, who will have access to new information on the use of antioxidants in the treatment of OL. Patients suffering from this condition will indirectly benefit from the study through improved therapeutic strategies and reduced risks of severe complications, such as oral cancer.

This work will help resolve the uncertainty surrounding the effectiveness of antioxidant treatments for OL, an area still lacking robust evidence. It will standardize knowledge and guide clinical practices by providing reliable data on the efficacy and safety of these treatments.

This study addresses an important gap in the existing literature. While systematic reviews on the use of antioxidants in OL have been conducted, the most recent one dates back to 2013. This review aims to update the previous findings by providing a comprehensive and critical analysis of the most recent data available.

6.1.1. SDG 3 Objective : Health and Well-being.

Oral health plays a fundamental role in overall well-being, directly aligning with **Sustainable Development Goal (SDG) 3: Good Health and Well-Being**. OL, a potentially malignant lesion, is a significant concern due to its risk of transforming into OSCC, one of the most common and deadly cancers worldwide. Early diagnosis and effective management of OL are essential in preventing malignant transformation and reducing the incidence of OSCC. In this context, research into alternative treatments, such as antioxidant supplementation and nutraceutical therapies, is crucial. These therapies provide promising non-invasive options that can be more accessible than traditional treatments. By investigating such approaches, this study directly contributes to the goals of SDG 3, aiming to improve oral health outcomes, reduce cancer risk, and enhance the quality of life for individuals affected by OL.

6.2. Hypotheses of the Study

The hypothesis of the study is to consider that antioxidant treatments may be effective in reducing the progression and malignant transformation of oral leukoplakia, also, improving clinical outcomes for patients like pain, inflammation, size reduction and oral function.

7. OBJECTIVES

General Objective

The main objective of this systematic review is to analyze the effectiveness of antioxidant treatments in preventing the progression of oral leukoplakia and improving related clinical outcomes.

Specific Objectives:

1. To evaluate the effectiveness of antioxidant treatments in reducing the size of oral leukoplakia lesions.
2. To determine whether antioxidant treatments can prevent the progression of oral leukoplakia, malignant transformation to more severe forms, such as dysplasia or cancerous lesions.
3. To measure the improvement of clinical symptoms associated with oral leukoplakia (pain, inflammation, difficulty eating or speaking) and improvement of oral functions after antioxidant treatment..
4. To examine changes in biological markers of oxidative stress, such as salivary or blood biomarkers, after antioxidant treatment in patients with oral leukoplakia.

8. MATERIALS AND METHODS

A systematic review was conducted following the guidelines outlined in the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) Statement, which provides a set of recommendations aimed at optimizing the quality and transparency of reports for systematic reviews and meta-analyses. The PRISMA Statement includes a 27-item checklist that covers key aspects such as the title, abstract, introduction, methods, results, and discussion. It also features a flow diagram that illustrates the process of selecting studies for inclusion in the review. The goal is to facilitate critical appraisal and reproducibility of systematic reviews and meta-analyses by readers, editors, and reviewers. Additionally, this work was registered in the international prospective register of systematic reviews, PROSPERO, under the registration number CRD42025649017.

8.1. Identification of the Research Question (PICO Format)

This study aims to answer a question formulated according to the PICO method (Patient, Intervention, Comparison, Outcome), based on the selected articles. The question following the PICO method is as follows (Table 1):

“How effective are antioxidant treatments in preventing the progression of oral leukoplakia and improving clinical outcomes?”

Table 1. Description of PICO question

Population	**Intervention**	**Comparison**	**Outcomes**
Adult patients diagnosed with oral leukoplakia.	Antioxidant treatments (e.g., vitamin A, curcumin, among others) administered topically or systemically.	Placebo, control group (no treatment), or conventional treatment (active surveillance, surgery, corticosteroids). Comparison between different antioxidant treatments.	O1: Reduction in the size of oral leukoplakia lesions. O2: Prevention of progression to more severe forms, such as dysplasia or cancer. O3: Improvement in clinical symptoms (pain, inflammation, difficulty eating or speaking). O4: Changes in biomarkers of oxidative stress (levels in saliva or blood).

8.2. Eligibility Criteria

Inclusion Criteria

The following criteria were used to determine the studies eligible for inclusion in this systematic review.

- Study Design:
  - Randomized controlled trials (RCTs), controlled studies, and observational studies.
  - Studies published in English, Spanish, and French.
- Population :
  - Patients with oral leukoplakia.
  - Studies conducted on human subjects.
- Intervention :
  - Antioxidant treatments (vitamin A, curcumin, green tea, vitamin C, vitamin E) for oral leukoplakia, administered either topically or systemically.
- Comparison :
  - Placebo, control group (no treatment), or conventional treatments (active surveillance, surgery, corticosteroids).
  - Studies comparing different antioxidant treatments.
- Outcome :
  - Reduction in lesion size.
  - Prevention of malignant transformation, including progression to dysplasia or cancer.
  - Improvement in clinical symptoms such as pain relief, reduction of inflammation, and functional benefits (easier eating or speaking).
  - Biological markers of oxidative stress reduction, such as changes in salivary or blood biomarkers.
- Publication date :
  - Studies published between 2000 and 2025.

Exclusion Criteria

Studies were excluded based on the following criteria:

- Studies involving patients with other oral conditions (e.g., oral cancer).
- Studies without antioxidant treatment.
- Studies lacking a comparison group or measurable outcomes related to lesion regression.
- Studies with low methodological quality, including those with significant risk of bias or insufficient data reporting.

- Studies with less than 4 patients. (n<4)
- Studies published in languages that are inaccessible for analysis (languages other than English, Spanish, or French).

8.3. Information sources and search strategy

For this systematic review, an automated search was conducted across three academic databases, PubMed, Web Of Science (WOS), and SCOPUS, with the following keys terms: “oral leukoplakia”, “Leukoplakia”, “antioxidants”, “antioxidant therapy”, “antioxidant treatment”.

The keywords were combined using Boolean operators AND and OR, and NOT, to construct the search equation. In this case the operator « NOT » wasn’t used. Along with controlled vocabulary terms MeSH (*Medical Subject Headings* – A controlled vocabulary thesaurus used by the U.S. National Library of Medicine for indexing and cataloging biomedical literature in the PubMed database and other resources) for PubMed to ensure the best comprehensive search results. The final search equation for all databases would be:

- ("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment")

The search equation was executed in each of the selected sources, adapting the terms and operators according to the characteristics of each database.

The search in PubMed was as follows: (("oral leukoplakia"[All Fields] OR ("leucoplakias"[All Fields] OR "leukoplakia"[MeSH Terms] OR "leukoplakia"[All Fields] OR "leucoplakia"[All Fields] OR "leukoplakias"[All Fields])) AND ("antioxidant s"[All Fields] OR "antioxidants"[Pharmacological Action] OR "antioxidants"[MeSH Terms] OR "antioxidants"[All Fields] OR "antioxidant"[All Fields] OR "antioxidating"[All Fields] OR "antioxidation"[All Fields] OR "antioxidative"[All Fields] OR "antioxidatively"[All Fields] OR "antioxidatives"[All Fields] OR "antioxidizing"[All Fields] OR "antioxidant therapy"[All Fields] OR "antioxidant treatment"[All Fields])) AND (2000:2025[pdat])

The search in Web of Science was as follows: ("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment") (Topic) and Review Article (Exclude – Document Types) and 1988 or 1989 or 1991 or 1992 or 1993 or 1994 or 1995 or 1997 or 1998 or 1999 (Exclude – Publication Years)

The search in Scopus was as follows: TITLE-ABS-KEY (("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment")) AND PUBYEAR > 1999 AND PUBYEAR < 2025 AND (LIMIT-TO (DOCTYPE , "ar")) AND (LIMIT-TO (LANGUAGE , "English"))

Refining the search by applying filters based on the pre-defined eligibility criteria. To limit the search to randomized clinical trials published in the last 25 years (2000-2025), the following filters were used:

- PubMed: (Randomized Controlled Trial[ptyp]) AND ("2000/01/01"[PDAT] : "2025/12/31"[PDAT]
- SCOPUS: (randomized controlled trial) AND (LIMIT-TO (PUBYEAR, 2000-2025))

In Table 1, included in the appendix section, a summary of the searches from each of the consulted databases is shown.

Import bibliographic references and eliminate possible duplicates that may exist between different information sources. References were imported into reference management software (Zotero), and duplicates were removed using the Find duplicates option.

8.4. Study Selection Process

The study selection process followed a structured and transparent approach to ensure that only studies directly relevant to the research question were included. This process was conducted in two stages:

- Stage 1: Title and Abstract Screening

  During the first stage, studies were initially evaluated based on their titles and abstracts. The aim was to identify studies that potentially met the inclusion criteria, which included the study design (randomized controlled trials, observational studies, etc.), the target population (patients diagnosed with OL), the intervention (antioxidant treatments), and relevant outcome measures (such as lesion size reduction and prevention of malignant transformation). Studies that did not meet these criteria or lacked sufficient information for further evaluation were excluded. The number of selected studies and the reasons for exclusion were documented, ensuring transparency in the process.
- Stage 2: Full-Text Assessment

  In the second stage, the full texts of the studies selected in Stage 1 were reviewed to confirm their eligibility. The same eligibility criteria from Stage 1 were applied, along with additional factors such as publication language, date of publication, and methodological quality. Studies that did not meet these criteria or could not be accessed in full were excluded. The number of studies deemed eligible and those excluded at this stage, along with the reasons for their exclusion, were also recorded

The process of selection will be documented by means of flow chart, following the PRISMA Guidelines model, showing the number of records identified, selected, full-text retrieved, eligible and finally included in the systematic review.

8.5. Data extraction

The following information was extracted from the articles and organized in tables according to the type of antioxidant (lycopene, curcumin, moringa, vitamin A, etc.). The data include: authors and year of publication, study type (mainly randomized clinical trials), number of patients, doses and frequencies of treatments, conventional treatment (if applicable), primary outcomes (regression of OL lesions and reduction in their size), secondary outcomes (inflammatory and oxidative markers, such as selenium, ceruloplasmin, and other biomarkers), follow-up time (in months), adverse effects (number, type and management), and comparisons with placebo or conventional treatments. Statistically significant discrepancies between groups were also noted to assess the efficacy of the treatments.

8.5.1. Main Variable

Clinical Improvement: The amount of clinical improvement in leukoplakia lesions, such as reduction in lesion size, reduction in plaque thickness, and lesion regression, by comparing the initial measurement before the application of antioxidant treatments (lycopene, curcumin, vitamins A, etc.) with the final measurement at the end of the treatment. This evaluation can be performed through visual assessment or photographic evaluation of the lesions, or by using clinical methods such as direct lesion examination and measurement of their diameter and thickness. Furthermore, improvement in the texture and color of the lesions, which typically become less white and more homogeneous, can also be a key indicator of treatment efficacy. When these measurements are reported, they should be performed in a standardized manner, using appropriate clinical measuring tools such as probes or validated visual assessment scales.

8.5.2. Secondary variable

Reduction of Oxidative stress and/or inflammation: The extent of reduction in oxidative stress and inflammation is measured by associated biomarkers, such as GSH, SOD, GPx. The reduction of inflammatory and oxidative markers is compared between the baseline values before the application of antioxidant treatment (such as Lycopene, curcumin, vitamin A,C,E)

and the final values after treatment. Measurements should be reported in concentration units or percentage changes, using biological fluids such as blood, serum, or gingival cervical fluid.

8.6. Valoration of quality

To assess the quality of randomized clinical trials on the efficacy of antioxidants for the treatment of oral leukoplakia, the Cochrane Risk of Bias (RoB 2.0) tool was used. This tool, developed by the Cochrane Collaboration, is specifically designed to evaluate the risk of bias in randomized trials. It covers five key domains: bias arising from the randomization process, deviations from intended interventions, missing outcome data, measurement of the outcome, and selection of the reported result. Each domain is judged as “low risk,” “some concerns,” or “high risk” of bias, based on signaling questions with predefined criteria. The overall risk of bias for each study is then determined based on the combined judgments across all domains. This approach provides a systematic and transparent framework for assessing the internal validity of included studies

For observational studies, the Newcastle-Ottawa Scale is used, as it is specifically designed for assessing the quality of nonrandomized studies. The scale was critically evaluated by Stang (2010) in the article “Critical evaluation of the Newcastle-Ottawa scale for the assessment of the quality of nonrandomized studies in meta-analyses” (European Journal of Epidemiology, 2010; 25:603–05). This scale assesses three key domains : selection of study groups, comparability of the groups, and the ascertainment of outcomes. Each domain is given a set number of « stars », and the overall quality of the study is determined by the total number of stars.

Quality Classification:

- Studies will be considered to have low risk of bias and good quality if they score more than 6 stars on the Newcastle-Ottawa Scale for observational studies or were assessed as having a low risk of bias according to the Cochrane Risk of Bias Tool for randomized controlled trials.
- Studies will be considered to have high risk of bias and poor quality if they score 6 stars or fewer on the Newcastle-Ottawa Scale or if they were

assessed as having a high risk of bias according to the Cochrane Risk of Bias Tool for randomized controlled trials.

Selection Criteria: Only studies that meet the low and medium risk of bias criteria will be included in the final analysis. Studies that are rated as having a high risk of bias will be excluded from the review to ensure the reliability and validity of the conclusions drawn from the systematic review.

8.7. Data synthesis

To analyse the collected data, a mixed methodology was used, combining both qualitative and quantitative approaches. A PRISMA flow chart will also be created to illustrate the study selection process based on the inclusion and exclusion criteria for each database search. This flowchart will provide a clear overview of the study selection process. Additionally, a table will be compiled to list the selected studies, and synthesize the results obtained. This table will allow for a comparison of the different antioxidants used in the studies and an analysis of their effects on primary and secondary outcomes, such as lesion regression and oxidative stress markers.

9. RESULTS

9.1. Study selection

The initial search process identified a total of 236 articles: from PubMed (n=87), Scopus (n=89), and Web of Science (n=60). Furthermore, 8 other studies were added following a hand-search of references. After removing 83 duplicates, 161 articles remained.

Of these, 132 were excluded: 114 for tittle and 13 for abstract, and 4 for no full text accessibility. As a result, 25 articles were retained for full-text review.

After full-text assessment, 14 articles were excluded, mainly due to their observational or in vitro design with no active antioxidant treatment.

In total, 11 studies were included in the qualitative synthesis. Figure 1 presents the PRISMA flow diagram, illustrating this selection process. Additionally, the information on the excluded articles is provided in Table 2.

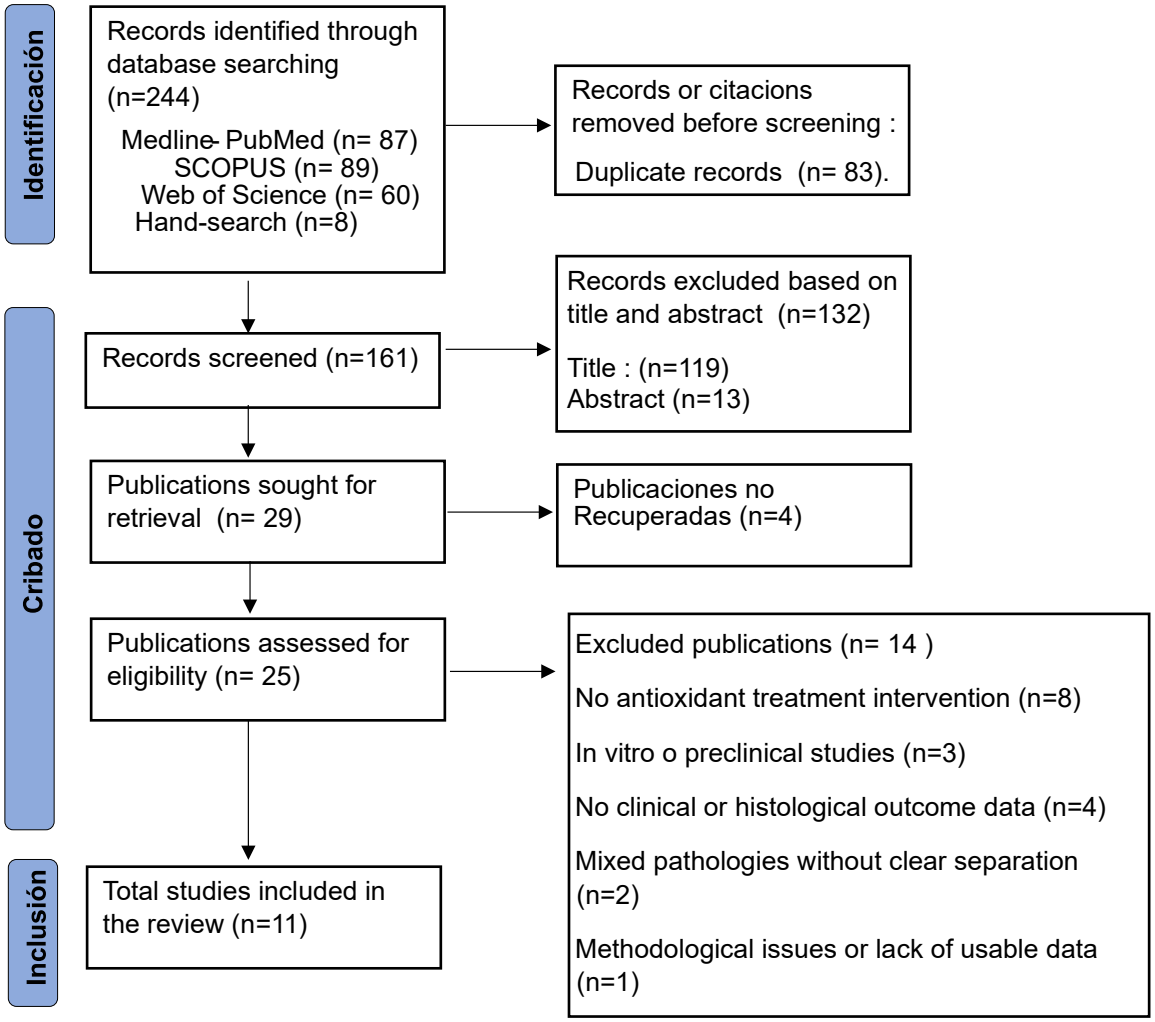

Fig. 1. Flow diagram of the search and selection process of titles during the systematic review.

Table 2 : Excluded articles in this systematic review

Author and year	Publication	Reason for exclusion
Babiuch et al. (2019) (6)	*Evaluation of enzymatic and non-enzymatic antioxidant status and biomarkers of oxidative stress in saliva of patients with oral squamous cell carcinoma and oral leukoplakia: a pilot study*	Observational/pilot study; no antioxidant intervention
Srivastava et al. (2014) (21)	*Oxidant-antioxidant status in tissue samples of oral leukoplakia*	Observational; no therapeutic intervention
Bose et al. (2012) (22)	*Plasma zinc antioxidant vitamins, glutathione levels and total antioxidant activity in oral leukoplakia*	Cross-sectional biomarker study; no treatment
Nagao et al. (2000) (23)	*Serum antioxidant micronutrients and the risk of oral leukoplakia among Japanese*	Case–control observational study
Srivastava KC (2019) (24)	*Comparative Evaluation of Saliva's Oxidant-Antioxidant Status in Patients with Different Clinicopathological Types of Oral Leukoplakia*	Observational pilot study; no treatment intervention
Agrawal et al. (2010) (25)	*Role of GSTM1 and GSTT1 polymorphism: susceptibility to oral submucous fibrosis in the North Indian population*	Genetic polymorphism/observational study
Rai S et al. (2015) (26)	*Estimation of serum antioxidant enzymes in histopathological grades of oral leukoplakia, oral submucous fibrosis, and oral cancer: A clinicopathologic study*	Observational clinicopathologic study; no randomizedd clinical trial
Mampilly et al. (2021) (27)	*Assessment of Serum Selenium and Ceruloplasmin in Potentially Malignant Disorders and Oral Cancer.*	Observational biomarker study only (no antioxidant intervention tested). No comparison group or

		lesion-regression outcomes reported.
Shah et al. (2017) (28)	*Determination of role of ceruloplasmin in oral potentially malignant disorders and oral malignancy-A cross-sectional study.*	Cross-sectional design (no in vivo intervention). Measures serum ceruloplasmin only, without evaluating an antioxidant therapy. Includes both OPMDs and oral cancers in cohort. No clinical or histologic outcomes on lesion size or regression.
Balasundaram et al. (2024) (29)	*Effectiveness of Fenugreek as an Adjuvant in the Management of Oral Potentially Malignant Disorders: A Randomized Controlled Trial.*	No differentitate between oral leukoplakia ad other oral potentially malignant disorders
Mallery et al. (2008) (30)	*Topical application of a bioadhesive black raspberry gel modulates gene expression and reduces cyclooxygenase 2 protein in human premalignant oral lesions*	Biomolecular focus without relevant clinical outcome data.
Yang et al. (2024) (31)	*Cucurbitacin B induces ferroptosis in oral leukoplakia via the SLC7A11/mitochondrial oxidative stress pathway.*	Preclinical in vitro study
Ansari et al. (2023) (32)	*Comparison of Effectiveness of Moringa Oleifera Leaves Extract Gel (2%) with Retino A (0.1%) Cream for Treatment of Oral Leukoplakia: Double Blinded Randomized Control Trial.*	Methodological issues and lack of complete data.
Khaera et al. (2022) (33)	*The effectiveness of watermelon rind extracts waste (citrullus lanatus l.) in removing free radical in oral leukoplakia patients*	Only measured lesion size as the outcome, without assessing histological changes, oxidative stress markers, or long-term follow-up.

9.2 Study caracteristics

A total of 11 studies were included, focusing on the use of antioxidants in the treatment of oral leukoplakia. These studies examined a variety of antioxidant interventions, including lycopene (n=3), curcumin (n=5), high-dose vitamin A (n=1), fenretinide (n=2), and other approaches such as enzymes-base assays and nutraceutical combinations (n=2). The studies included randomized controlled trials (RCTs), non-randomized prospective studies and phase II pilot trial. The primary outcomes measured were clinical response, lesion size reduction, histological improvements, oxidative stress markers, such as MDA and 8-OHdG. Some studies also evaluated long-term outcomes, including recurrence rates, malignant transformation, and the prevention of cancer development.

The sample sizes varied across studies, with most involving between 6 and 300 patients. The duration of treatment ranged from 1 month to 1 year, with follow-up periods from a few months to up to 5 years. Most studies employed control or placebo groups to compare the effectiveness of the antioxidant treatments. In terms of methodology, clinical and histopathological evaluations were the primary tools used to assess the efficacy of the treatments.The general characteristics of the included studies are summarized in Table 3.

Table 3. Characteristics of the Selected Articles:

Variable / Characteristic	Lycopene (n=3)	Curcumin (n=5)	Vitamin A (n=1)	Fenretinide (n=2)	Others (n=2)	Total (n=11)
Type of study: RCT	3	2	0	1	0	6
Type of study: Non-randomized prospective	0	1	1	0	2	4
Type of study: Phase II / Pilot	0	0	0	1	0	1
Sample size (min–max)	41-300	25–300	6	35–170	10-50	6-300
Lesion size reduction	3	3	1	2	2	11
Histological improvement	2	2	1	1	2	4
Oxidative stress markers	1	2	0	0	1	3
Adverse events / Safety	0	0	0	1	0	1
Recurrence / New lesions	0	0	0	1	0	1

9.3. Risk of bias in studies

In order to ensure the reliability of the conclusions of this systematic review, a rigorous assessment of the risk of bias was conducted for each of the included studies, according to their methodological type.

For randomized controlled trials (RCTs), the risk of bias was assessed using the Cochrane Risk of Bias tool, which examines criteria such as random sequence generation, allocation concealment, blinding of participants and personnel, participant follow-up, and selective outcome reporting. In our sample, most randomized studies presented an overall moderate risk of bias, mainly due to a lack of detail regarding blinding methods (of participants, personnel, and

investigators), and in some cases, insufficient description of randomization procedures (Table 4).

For observational studies with a non-randomized control group, the risk of bias was evaluated using the Newcastle-Ottawa Scale (NOS), which considers group selection, comparability, and the quality of outcome measurement. These studies obtained variable scores, generally classified as moderate risk, due to limited cohort representativeness or insufficient control of confounding variables (Table 5).

Finally, cohort studies without a control group were also assessed using an adapted version of the NOS scale, focusing on cohort representativeness, quality of exposure measurement, and follow-up duration. Overall, these studies showed a higher risk of bias due to the absence of direct comparison and the potential influence of uncontrolled external factors (Table 6).

These results reflect the variability in the methodological quality of the included studies and highlight the effectiveness of antioxidant treatments in the management of oral leukoplakia.

Table 4: Risk of Bias Assessment for Randomized Studies According to the Cochrane Handbook.

	Generation of randomized sequence (election bias)	Allocation concealment (selection bias)	Binding of participants and personnel (detection bias)	Blinding of outcome assessment (detection bias)	Follow-up and exclusions (attrition bias)	Selective reporting (reporting bias)	Other bias
Fathima et al. (2022) (34)	?	−	?	?	?	?	+
Singh et al. (2017) (35)	+	+	+	+	?	+	+
Patel et al. (2014) (36)	+	+	+	+	?	+	?
Chiesa et al. (2005) (37)	+	+	?	?	?	?	?
Kuriakose et al. (2016) (38)	+	+	+	+	+	+	+
Ahmad et al. (2023) (39)	+	?	−	?	?	+	?

Table 5 : Risk of bias assessment of non-randomized observational studies using the Newcastle-Ottawa scale – observational studies with a non-randomized control group.

	Case definition adequate	Representtiveness of cases	Selection of controls	Definition of controls	Comparability (main factor)	Comparability (other factors)	Ascertainment of exposure	Same method of ascertainment cases and	Drop-out rate	Total
Jain et al. (2011) (40)	☆	☆	-	☆	-	-	☆	☆	☆	6

Table 6 : Risk of bias assessment of non-randomized observational studies using the Newcastle-Ottawa scale – observational cohort studies without a control group.

	Represetativeness of exposed cohort	Selection of the non-exposed cohort	Ascertainment of exposure	Demonstrationthat outcome of interest not present at start	Comparability of cohort based on design or analysis	Comparability of cohort based on any additional factors	Assessment of outcome	Sufficient follow-up	Drop-out rate	Total
Leunig et al. (2000) (41)	☆	-	☆	☆	-	-	☆	☆	☆	6
Kapoor, S et al. (2019) (42)	☆	-	☆	☆	-	-	☆	☆	☆	6
Rai, Balwant et al (2010) (43)	☆	-	☆	☆	-	-	☆	☆	☆	6
Lippman et al. (2006) (44)	☆		☆	☆			☆	☆	☆	6

9.4. Result synthesis

To synthesize the results, a summary of the outcomes of the variables from the 11 selected articles for review on the treatment of oral leukoplakia with antioxidants is presented (table 7) :

The primary objective of this review was to assess clinical improvement of oral leukoplakia lesions via reduction in lesion size and rates of partial or complete regression. Across the 11 included studies, al lot them produced clinical responses and 8 of them showed quantifiable responses.

Several studies evaluated the clinical efficacy of lycopene administered either orally or as a topical gel, either alone or with co-antioxidants in the treatment of oral leukoplakia (35,36,39). In a study (35), two topical antioxidant treatment were compared in patients with tabacco-induced homogeneous oral leukoplakia. Group I received Calendula officinalis gel, and Group II received

lycopene gel. Both formulations contained 2 mg of active ingredient per gram of gel and were applied once daily for one month. The mean reduction in lesion size was 2.0 ± 1.0 cm in the Calendula group and 1.57 ± 0.87 cm in the lycopene group. Although both groups showed a significant clinical improvement, the difference between them was not statistically significant ($p > 0.05$). No adverse effects were reported, and no placebo control group was included in this study. Similarly in a RCT (36), comparing a group treated with a combined treatment consisting of lycopene (3mg), vitamin E (200 I.U.), and selenium (100 mg) twice daily to a placebo group. A significant reduction in mean lesion size was observed in the treatment group (from 9.85 cm^2 to 1.49 cm^2), while the placebo group showed an increase in lesion size (from 7.58 cm^2 to 8.55 cm^2). The clinical improvement rate was 86% in the lycopene group versus only 20% in the placebo group. Lastly, in a RCT (39), three groups were compared: one treated with curcumin, one with lycopene, and one with a combination of both. After 90 days, cure rates were 63% in the lycopene group, 51% in the curcumin group, and 72% in the combination group. Although precise numerical data on lesion size reduction were not provided, the authors reported evident clinical improvement across all antioxidant-treated groups.

Several studies have evaluated the clinical efficacy of curcumin as an antioxidant treatment for oral leukoplakia (34,38,39,42,43). One of the articles investigated the use of topical curcumin gel combined with antioxidants in a group of patients vs a group of patient treated with bleomycin with antioxidant s. The results showed significant histopathological improvement in dysplasia in the curcumin group ($p = 0.01$), although clinical resolution was not as substantial as with the bleomycin treatment. These findings suggest that while curcumin may not be as effective in clinical resolution, it can contribute positively to the histological improvement of lesions (34).

Another study conducted a RCT with 60 patients, divided into three groups. Group A included patients with oral leukoplakia, Group B with oral lichen planus and Group C with oral submucous fibrosis (OSMF). All group received oral curcumin (400 mg daily) for three months. The study reported a significant

reduction in lesion size in Group A, from 4.05 ± 1.69 cm to 1.29 ± 1.10 cm (mean difference = 2.76 cm), demonstrating significant clinical response in the leukoplakia group (42).

A further study evaluated the effects of oral curcumin in patients diagnosed with oral leukoplakia, submucous fibrosis and lichen planus compared to normal healthy individuals. The participants received curcumin 1gr caplets (900mg curcumin, 80mg desmethoxycurcumin and 20mg bisdesmethoxycurcumin). Clinical and histopathological examination during the treament demonstrated significant clinical improvement in lesion size reduction ($p > 0.05$) with clinical curation after 131 days. This study also found significant increases in serum vitamins C from 8.78 µmol/L to 9.09 µmol/L and from 8.01 µmol/L to 8.99 µmol/L in serum vitamin E. While salivary vitamin C increase from 1.08 µmol/L to 1.54 µmol/L, and from 0.65 µmol/L to 0.96 µmol/L in case of salivary vitamin E. Concurrently, Salivary MDA levels decrease from 0.36 µmol/L to 0.13 µmol/L, and serum level decrese from 1.23 µmol/L to 0.97 µmol/L. Similar reductions were observed in 8-OHdG levels, with salivary concentrations dropping from 0.34 ng/ml to 0.12 ng/ml and serum level from 2.13 ng/ml to 1.88 ng/ml. Reductions in oxidative stress markers such as MDA and 8-OHdG ($p < 0.05$), confirming the antioxidant effect of curcumin in precancerous oral lesions (43).

In a larger study, 3.6 g/day of oral curcumin were administered for six months to 111 patients with oral leukoplakia. The clinical response defined as complete or partial response was significantly higher in the curcumin group (67.5%) compared to the placebo group (55.3%) ($p = 0.03$). A combined clinical and histological response was also noted, suggesting that curcumin had a durable clinical effect. However, the histological response between the curcumin and placebo groups was not statistically significant (HR = 0.88, 95% CI: 0.45–1.71, $p = 0.71$), and no additional benefits were observed with prolonged treatment (38).

Vitamin A (Retinyl Palmitate) was investigated for its efficacy in treating oral leukoplakia. In this study, (41), a small sample size of 6 patients was treated with increasing doses of retinyl palmitate, starting with 300×10^3 IU/day for the first week and increasing to 1.5×10^6 IU/day by the fifth week, followed by maintenance therapy with 150×10^3 IU/day. The results indicated a complete

histological response, with no dysplasia observed in follow-up biopsies. However, the clinical reduction in lesion size was not quantified, and there were no reported adverse events or other clinical outcomes. Despite the small sample size, the study demonstrated a positive histological response monitored using fluorescence, suggesting that retinyl palmitate may be effective in managing oral leukoplakia.

Fenretinide (4-HPR) has been evaluated in two key studies for its ability to prevent recurrence and promote lesion regression in oral leukoplakia (37,44). In a long-term multicenter randomized trial (37), a control group was compared to fenretinide 200 mg/day over one year in 170 post-surgical patients. Although lesion size reduction was not explicitly quantified, the fenretinide group experienced significantly fewer recurrences, new lesion formations, and carcinomas compared to controls, demonstrating a substantial prophylactic effect against malignant progression over extended follow-up.

In a separate Phase II trial, (44), retinoid-resistant leukoplakia patients was treated with fenretinide 200 mg/day for three months. The overall three-month clinical response rate (complete + partial) was 34.3% (95% CI: 19.2–52.4%), a notable result considering the refractory nature of these lesions. Interestingly, patients with adquired resistance showed better responses than those with primary resistance (p=0.015). Although the increase in apoptosis (from 0.35% to 1.18%, p=0.001) was statistically significant, it did not correlate directly with clinical outcomes. Despite the modest response rate, the treatment was well tolerated, and the findings suggest potential utility for fenretinide in this difficult-to treat population.

Other antioxidants such as beta-carotene, vitamin E, vitamin C, copper, manganese, zinc, and selenium have also been evaluated for their potential in the treatment of oral leukoplakia (40). In a clinical study patients diagnosed with oral Leukoplakia received a daily oral capsule containing beta-carotene (10mg (5000IU)), vitamin E acetate I.P. (25IU), Vitamin C acetate I.P. (100mg), Copper (from copper sulphate) (1mg), Manganese (1.5mg), Zinc 7.5mg and Selenium (150mcg) for one year. Patents were categorized into subgroups based on the histological grade of dysplasia: no dysplasia (Group BI), mild dysplasia (Group

BII), and moderate dysplasia (Group BIII). Following treatment, partial or complete clinical regression was observed in 74.19% of patients. Although histopathological improvement was not reported, significant changes were observed in oxidative stress biomarkers across all subgroups. In patients with no dysplasia (BI), LPO levels decreased from 0.532 ± 0.110 to 0.436 ± 0.102 nmol/mg protein ($p < 0.001$), SOD levels decreased from 65.3 ± 2.78 to 50.6 ± 2.10 U/g tissue ($p < 0.001$), and CAT activity increased from 120.1 ± 13.6 to 144.3 ± 17.7 U/g tissue ($p < 0.001$). Similar trends were observed in the mild dysplasia (BII) and moderate dysplasia (BIII) subgroups, both in oral mucosa and serum samples. In the mucosal tissue, significant reductions in LPO levels were observed (from 0.316 to 0.268 nmol/mg protein in BII and from 0.289 to 0.239 nmol/mg protein in BIII), along with decreased SOD activity and increased CAT levels (e.g., CAT increased from 48.8 to 66.1 U/g in BII and from 43.4 to 79.9 U/g in BIII; $P < 0.001$). In the serum, these antioxidant effects were also evident: LPO decreased significantly (from 0.628 to 0.560 in BII and from 0.468 to 0.396 in BIII), SOD levels declined (from 60.5 to 54.9 U/g in BII and from 71.7 to 52.2 U/g in BIII), and CAT activity increased (from 141.6 to 180.9 U/g in BII and from 130.2 to 173.7 U/g in BIII), with all changes being statistically significant ($P < 0.001$). While all dysplasia subgroups responded positively to antioxidant treatment, the magnitude of enzymatic changes varied, which may reflect differences in baseline oxidative stress or degree of dysplastic transformation.

Similarly, another study (35), investigated the efficacy of two topical treatments, Calendula officinalis gel and lycopene gel, for the treatment of tobacco-induced homogeneous leukoplakia in 60 patients. After one month of treatment, the results showed a significant reduction in lesion size in both treatment groups, with Group I (Calendula officinalis gel) showing a mean size reduction of 2.0 ± 1.0 cm and Group II (lycopene gel) showing a mean reduction of 1.57 ± 0.87 cm. However, there was no placebo arm in the study, and no significant histological or oxidative stress marker changes were reported.

These findings suggest that antioxidants, particularly in combination or topical application, may offer some benefit in the clinical management of oral leukoplakia. However, the absence of histological and oxidative stress marker

data in some studies highlights the need for further research to better understand the full scope of their therapeutic effects.

- Overall effectiveness: The studies by Kuriakose et al. (38) and Jain et al. (40) reported the highest rates of clinical lesion regression, with 67.5% and 74.19%, respectively, indicating a high effectiveness of the antioxidant treatments used in these trials.
- Histological improvement: The studies by Leunig et al. (41) and Patel et al. (36) showed the most significant histological improvements, with a complete absence of dysplasia in follow-up biopsies and 86% improvement in the latter, suggesting a strong preventive potential against malignant transformation.
- Oxidative stress markers: The studies by Jain et al. (40) and Rai et al. (43) demonstrated significant improvements in antioxidant profiles, with reductions in LPO, SOD, MDA, and 8-OHdG, and increases in CAT, vitamin C, and E, confirming the positive biochemical impact of antioxidant treatments.

Table 7. Synthesis of the Results of the Variables

Author (year)	Age	N	Intervention	Lesion size reduction	Histological improvement (%)	Oxidative stress markers	Comment
Leunig et al. (2000) (41)	34-63	**N= 6** patients with leukoplakia	Retinyl palmitate 300 × 103 IU/day for the first week 1.5 × 106 IU/day in the fifth week maintenance therapy with 150 × 103 IU/day	Not quantified	100% (no dysplasia in follow-up biopsies)	Not reported	Small sample size, effective histological repsonse monitored via fluorescence
Chiesa et al. (2005) (37)	>75	**N= 170** **GE** (n=84) **GC** (n=86)	**GE** :Fenretinide (4-HPR), 200 mg/d	Clinical reduction but Not quantified	Not reported	Not reported	Long term follow-up, significant prevention of

			GC : No treatment				recurrence and carcinoma
Lippman et al. (2006) (44)	26-84	**N= 35**	Fenretinide 200 mg/day for 3 months	34.3% partial response	Not reported	↑ Apoptosis (0.35% → 1.18%, *p* = 0.001)	9/12 responders relapsed within 9 months; better outcomes in prior responders to retinoids (70% vs. 20%, *p* = 0.015)
Rai, Balwant et al (2010) (43)	17-50	**N= 100** **Group leukoplakia** (n=25) **Group oral submucous**	Curcumin 1g caplets (900mg curcumin, 80mg desmethoxycurcumin, and 20mg bisdemethoxycurcumin)	**Group leukoplakia** : Significant improvement (p>0.05)	Not reported	Group leukoplakia: ↑ Vitamins C & E; ↓ MDA & 8-OHdG (p < 0.05)	Demonstrated antioxidant effect and clinical improvement in precancerous oral lesions

		fibrosis (n=25) **Group lichen planus** (n=25) **Group healthy** (n=25)					
Jain et al. (2011) (40)	-	**N= 50** **Group A** control :10 **Group B** experimental : 40 (subdivided by dysplasia	**Group A and B:** Antioxidants capsule Beta-carotene (10mg) ; Vitamin E acetate I.P. (25IU); Viatmin C acetate I.P. (100mg); Copper(1mg) ; Manganese	74.19% showed partial or complete regression at 1 yeas	None observed	Grupo B (I, II, II): Serum : ↓ LPO, ↓ SOD, ↑ CAT mucosa : ↓ LPO, ↓ SOD, ↑ CAT	No histological reversal ; enzymatic improvement

		grade : BI, BII, BII)	(1.5mg) ; Zinc (7.5mg) and Selenium (150 mcg)				
Patel et al. (2014) (36)	26-65	**N= 41** **Group A** (n=21) **Group B** (n=20)	**Group A:** Lycopene (3 mg), Vitamin E (200 I.U.), Selenium (100 mcg) x2/day **Group B:** placebo capsule x1/day	**Group A:** 9.85 ± 5.87 → 1.49 ± 3.08 cm² **Group B:** 7.58 ± 7.50 → 8.55 ± 7.69 cm²	**Group A:** 86% **Group B:** 20%	Not reported	Significant improvement in treated group
Kuriakose et al. (2016) (38)	26-74	**N= 223** **GE** (n=111) **GC** (n=112)	**GE**: Curcumin 3.6 g/day (oral) x 6 months **GC** : placebo	**GE** :67.5% **GC** :55.3%	Not significant (HR = 0.88, 95% CI: 0.45–1.71, p = 0.71)	Not reported	Significant and durable clinical repsonse with curcumin. No added benefit with prolonged treatment

Singh et al (2017) (35)	56-65	**N= 60** **Group I** (n=30) **Group II** (n=30)	**Group I:** Caléndula officinalis gel (2mg/g per day) x 1 month **Group II:** lycopene gel/day (2 mg/g per day) x 1 month	Mean difference: **Group I**: 2.0 ± 1.0 cm **Group II** : 1.57 ± 0.87 cm	Not reported	Not reported	No placebo arm
Kapoor, S et al. (2019) (42)	-	**N= 60** **Group A** OL (n=20) **Group B** OLP (n=20)	**Group A,B,C :** Oral Curcumin 400mg daily x 3 months	**Group A :** Mean size difference 2. 757 cm	Not reported	Not reported	Significant clinical response in leukoplakia group

		Group C OSMF (n=20)					
Fathima et al. (2022) (34)	20-60	**N= 20** **Group A** (n=10) **Group B** (n=10)	**Group A :** Topical Bleomycin + antioxidants **Group B :** Topical Curcumin gel + antioxidants	**Group A :** partial to complete resolution (p=0.01) **Group B:** not substantial	**Group B:** histopathological improvement of dysplasia (p=0.01)	Not reported	Bleomycin showed superior clinical resolution; curcumin improved dysplasia histologically
Ahmad et al. (2023) (39)	18-70	**N=300** **Grupo A**: (n=100)	**Group A :** Curcumin 500mg/day **Group B :** Lycopene 4mg/day	Clinical reduction but Not quantified	Not reported	Not reported	Combination therapy showed superior efficacy

		Grupo B: (n=100) **Grupo C**: (n=100)	**Group C:** Curcumin 500mg + lycopne 4mg				

GC= Group control, **GE**= group experimental, **LPO=** lipid peroxidation, **SOD=** Superoxide dismutase, **CAT=** Catalase, **MDA=** Malondialdhyde, **8-OHdG=** 8-hydroxy-2'-deoxyguanosine, **HR=** Hazard ratio, **CI=** Confidence interval, **OL=** Oral leukoplakia, **OLP=** Oral lichen planus, **OSMF=** Oral submucous fibrosis

10. DISCUSSION

In the present systematic review, an evaluation was carried out to assess the efficacy of antioxidant treatments such as curcumin, lycopene, vitamins and other nutraceuticals in improving the clinical, histopathological and biochemical parameters of oral leukoplakia in adult patients. A total of 11 clinical studies, including randomized controlled trials, cohort and controlled clinical trials, were included according to the predefined eligibility criteria. These antioxidant compounds are known for their anti-inflammatory, immunomodulatory, and free radical scavenging properties. Although the precise efficacy of antioxidant therapies in halting or reversing the progression of oral leuoplakia remains a subject of ongoing investigation, a growing body of evidence supports their potential benefits in the management of this potentially malignant disorder.

10.1. Evaluation of antioxidants on the clinical parameters of leukoplakia

Several studies have investigated the antioxidant effect of the selected compounds, and all of them shown a positive impact on the improvement of clinical parameters in oral leukoplakia.

One of the compounds thats demonstrated significant improvements in the clinical parameters of the disease is Lycopene. This compound is a natural carotenoid found mainly in tomatoes and other red fruits, know for ots strong antioxidant activity and its ability to neutralize reactive oxygen species (45). In the present review, three studies used lycopene-based treatments (35,36,39). All of them reported positive clinical outcomes in terms of lesión reduction. These effects may be attributed to Lycopene's ability to stabilize cell memebranes, inhibit cellular proliferation, and modulate gene expression related to oxidative stress and inflammation (46). In addition, some studies suggest that lycopene can enhance epithelial regeneration and inhibit angiogenesis (47,48), which could explain its therapeutic effects in precancerous oral lesions. Moreover, lycopene has been utilized in the management of other oral mucosal diseases, such as submucous fibrosis and oral lichen planus (OLP), demonstrating

significant improvements in clinical symtoms and mouth opening capacity (49–53). Beyond the oral cavity, lycopene has shown potential benefits in systemic conditions, including liver diseases and depression, attributed to its antioxidant and anti-inflammatory properties (54–56).

In the case of Curcumin, the clinical studies reviewed consistently demonstrated positive outcomes regarding lesion size reduction in patients with oral leukoplakia (34,38,39,42,43). This compound derived from Curcuma longa, has been used for centuries in traditional medicine for its anti-inflammatory and wound-healing properties (57,58). In the included trials, crcumin was administred either topically or orally, with results showing visible clinical improvement, expecially in terms of lesion regression and texture normalization. The beneficial clinical response may be explained by its local anti-inflammatory and epitelial restorative effects (59). Although the extent of improvement varied across studies, the overall evidence supports curcumin as a promishing antioxidant intervention for the clinical management of oral leukoplakia. Beyond leukoplakia, curcumin has been investigated for its therapeutic potential in other oral mucosal condictions. For instance, studies evaluated its efficacy in managing symptomatic OLP. One randomized controlled trial found that curcumin supplementation led to significant reduction in pain and lesion size in OLP patients (60). Curcumin's therapeutic application extends to systemic condictions as well. In the context of liver diseases, particularly non-alcoholic fatty liver disease, curcumin supplementation has been associated with significant decreases in hepatic fibrosis, indicating its potential anti-inflammatory effects on the liver (61). These findings collectivelly suggest that curcumin possesses multifaceted therapeutic properties, making it a promising candidate for managing various inflammatory and oxidative stress-related conditions.

Among the antioxidants evaluated for their effects on clinical parameters in oral leukoplakia, fenretinide has emerged as a promising therapeutic agent, particularly in cases resistant to conventional retinoid therapy. This synthetic retinoid analogue, derived from N-(4-hydroxyphenyl)retinamide, has been widely studied for its ability to modulate epithelial differentiation and inhibit cellular proliferation, making it particularly useful in treating premalignant oral lesions.

Clinically, its effects has been associated with the stabilizacion or reduction of lesion size and the prevention of new lesion development (62). Its oral administration over prolonged periods and the consistent obeservation of reduced lesion recurrence suggests a sustained clinical benefit. Together, these findings support the role of frenetinide as a clinically relevant antioxidant treatment capable of modifying the disease course in patients with high-risk or persistant leukoplakia (37,44). In addition to its applications i oral leukoplakia, fenretinide has also been investigated in other epithelial and systemic conditions. It has shown chemopreventive activity in breast cancer, particularly among high-risk premenopausal women, by inducing apoptosis and reducing serum IGF-1 levels (63). Furhermore, studies have demonstrated its efficacy in overian cancer, neuroblastoma, and prostate cancer, highlighting its potential in modulating cancer progression through both retinoid and non-retinoid pathways (64,65).

In the case of β-carotene and other mixed antioxidants (vitamin C, vitamin E, zinc, selenium, copper, and manganese), one of the studies included in this review did not assess histological or long-term outcomes but reported encouraging clinical improvement (40). The formulation likely exerted its impact through a combined antioxidant action, which helped mitigate oxidative damage associated with leukoplakia progression. β-carotene have been studied for their potential to reduce the risk of oral malignant transformation (66,67). β-carotene, as a provitamin A carotenoid, is known for its free-radical scavenging abilities and has demonstrated promising effects in OPMDs, incuding oral leukoplakia and oral submucous fibrosis (68). Like β-carotene, Vitamin C and E, individually and in comination, have also been widely investigated in the context of skin diseases such as photoaging, eczema and psoriasis due to their role in epitelial repair, anti-inflammatory effects and reduction of oxidative stress (69–71). These findings suggest that antioxidant combinations not only hold therapeutic potential in ral leukoplakia but may also contribute to the management of a broad spectrum of diseases driven by oxidative stress.

As for vitamin A, only one pilot study was included in this review (41), involving six patients treated with escalating doses of retinyl palmitate. Although this study lacked a control group and quantification of lesion size reduction, clinical

resolution was achieved in all cases, and follow-up biopsies confirmed complete disappearance of dysplasia. These findings suggest a potential role for high-dose vitamin A in reversing dysplastic changes in leukoplakia lesions, although the small sample size and absence of comparison groups limit generalizability. Beyond oral health, vitamin A has been utilized in the management of several other conditions. In dermatology, retinoids (vitamin A derivatives) are commonly prescribed for acne vulgaris and psoriasis due to their ability to normalize keratinization and reduce inflammation. In ophtalmology, vitamin A supplementation is critical in preventing and treating xerophtalmia, especially in population with dietary deficiencies. Additionally, vitamin A plays a rôle in immune function, and its supplementation has been shown to reduce morbidity and mortality in children with measles in areas where deficiency is prevalent (72,73).

Together, these findings support the notion that various antioxidant compounds particularly those derived from essential vitamins may serve as beneficial adjuvants in the management of oral leukoplakia. However, given the methodological limitations of some studies and the heterogeneity of antioxidant regimens used, further clinical trials with standardized protocols are needed to confirm their efficacy and safety.

10.2. Evaluation of Antioxidant effect on biochemical and oxidative stress markers in oral leukoplakia

Oral leukoplakia as a potentially malignant disorder where oxidative stress and chronic inflammation are believed to play a central role in its pathogenesis.

Among the antioxidant agent reviewed, curcumin has shown promising effects not only on clinical outcomes but also on biochemical markers. For instance, (43), observed that curcumin supplementation significantly increased serum levels of vitamins C and E while decreasing markers such as MDA and 8-OHdG, both of which are indicators of oxidative stress and DNA damage. These results are aligned with broader evidence suggesting that curcumin’s phenolic structure

confers strong free radical scavenging properties (74). Additionally, the same study observed elevated serum levels of vitamins C and E following treatment, indicating an enhancement of systemic antioxidant defenses. Such findings are particularly relevant considering that patients with potentially malignant disorders often exhibit reduced antioxidant capacity (75), which contributes to epithelial dysregulation and progression toward malignacy. Therefore, curcumin's dual capacity to lower oxidative stress markers and boost endogenous antioxidant vitamins supports its role as a systemic modulator in oral leukoplakia therapy. This is consistent with findings in other disease models. For example, curcumin has been shown to reduce oxidative stress markers in patients with metabolic syndrome like diabetes, by lowering MDA and increasing SOD and CAT activity, therby improving antioxidant status (76). Similarly, in a in vivo model of Alzheimer's disease, curcumin administration led to a reduction in lipid peroxidation and a significant restoration of antioxidant enzyme levels (77).

Lycopene has shown notable effects in reducing oxidative stress and improving the clinical outcomes in patients with oral leukoplakia. Jain's study (40), observed a significant reduction in MDA, a well-known biomarker of lipid peroxidation, indicating a decrease in oxidative damage following lycopene supplementation. MDA is widely regarded as a critical marker of oxidative stress, and its reduction suggests a protective effect against cellular damage (75), which is a central aspect of the pathogenesis of oral leukoplakia. Furthermore, the study by Jain also measured SOD and GPx, two essential antioxidant enzymes, and found a significant increase in both. Their elevation suggests that lycopene supplementation enhanced the systemic antioxidant defenses, providing further evidence for its therapeutic potential in oxidative stress-related conditions (78). The dual effects of lycopene lowering oxidative stress markers and reducing inflammation are particularly important in the context of potentially malignant disorders. Given that oral leukoplakia often presents with dysregulated epithelial cells and increased oxidative damage, Jain's findings support the therapeutic potential of lycopene as an adjunct to traditional treatments, promoting cellular regeneration and preventing malignant transformation. In vivo research on hypercholesterolemic rats revealed that lycopene supplementation decrease MDA levels and increases the activity of antioxidants enzymes such as SOD, CAT

and GPx indicating improved systemic redox status (79). A systemic review and meta-analysis of 13 randomized controlled trials concluded that lycopene supplementation significatly reduces DNA damage, as measured by comet assay tail length, renforcing its potential role in oxidative stress modulation (80).

Fenretinid has demonstrated notable effects in retinoid-resistant oral leukoplakia. For instance, a phase II clinical trial (44), in which fenretinide (200 mg/day for 3 months) significantly increased apoptosis levels in oral epithelium, from 0.35% at baseline to 1.18% post-treatment . Although this increase did not correlate with clinical response, it supports fenretinide's potential receptor-independent pro-apoptotic mechanism. This is consistent with preclinical findings showing that fenretinide induces apoptosis via mitochondrial pathways and ceramide accumulation (81). This pro-apoptotic effect of fenretinide is not limited to oral lesions. In vitro studies have also demonstrated its efficacy in other epithelial malignancies, such as small-cell lung cancer (SCLC) (82). These effects highlight the compound's potent ability to trigger programmed cell death even in aggressive neoplasms resistant to conventional retinoids. These findings support the broader potential of fenretinide as a therapeutic agent capable of modulating apoptosis pathways in various epithelial-derived malignancies, beyond the oral cavity.

Additionally, the results obtained in this analysis support the hypothesis that antioxidants such as curcumin, lycopene, fenretinide and other vitamins are effective in improving oral leukoplakia. These antioxidants may act synergistically or complementarily to reduce oxidative stress and inflammation, two key factors in the progression of leukoplakia towards oral cancer. Through their ability to regulate oxidative and anti-inflammatory biomarkers, these treatments can also improve the overall health of oral tissues, helping to prevent cellular damage and dysplasia. These findings are relevant since patients with oral leukoplakia often show a deficit in antioxidant capacity, which favors the progression of the disease toward malignant forms. Therefore, the use of these antioxidants may be a promising therapeutic strategy in the treatment of oral leukoplakia.

10.3. Limitations of the study

The current evidence regarding the impact of antioxidants such as curcumin, vitamin E, lycopene, and fenretinide in the treatment of oral leukoplakia is limited, with several significant challenges that need to be acknowledged. The available studies are scarce and often lack the robustness needed to draw firm conclusions. Among the most notable limitations are:

- **Limited clinical trials and observational studies**: There is a lack of comprehensive and specific research on these antioxidants, which makes it difficult to fully understand their real efficacy.
- **Small sample size and short duration**: Many studies have been conducted with a limited number of participants and over short periods, which may affect the generalization of the results.
- **Heterogeneity in study design and methodology**: There is considerable variability in how the studies were conducted, including differences in dosage, administration methods, participant selection criteria and the presence or absence of control or placebo groups.
- **Confounding factors and methodological adjustments**: Studies do not always adequately control for external factors that could influence the results, which raises concerns about the internal validity of the findings.

10.4. Futur research directions

To overcome the current limitations and advance knowledge regarding the efficacy of antioxidants in the treatment of oral leukoplakia, the following directions for future research are proposed:

• **Higher-quality and larger-scale studies**: Future research should employ more rigorous methodologies, larger sample sizes, and long-term follow-ups to provide more robust and generalizable results regarding the impact of antioxidants (such as curcumin, lycopene, vitamin E, and fenretinide) on oral leukoplakia.

• **Analysis of molecular and cellular mechanisms**: A deeper investigation into how antioxidants interact at the molecular and cellular levels with oral epithelial cells, specifically focusing on oxidative stress markers, apoptosis, and the modulation of gene expression, will help to clarify their therapeutic mechanisms.

• **Longitudinal studies to assess long-term efficacy**: Given the chronic nature of oral leukoplakia and its potential for malignant transformation, long-term studies are necessary to assess whether antioxidant treatments provide sustained benefits and reduce the risk of recurrence or progression to malignancy.

• **Evaluation of safety and adverse effects**: Future research should include more detailed assessments of potential side effects, toxicity, and interactions between antioxidants and other commonly used oral treatments, to ensure the safety of prolonged antioxidant therapy for oral leukoplakia.

• **Comparative studies**: Research comparing different antioxidant treatments (such as curcumin versus lycopene or vitamin E) and their effectiveness, both individually and in combination, will help optimize treatment strategies for oral leukoplakia.

By addressing these gaps, future studies can strengthen our understanding of the potential of antioxidants as a viable therapeutic option for managing oral leukoplakia and preventing its progression to oral cancer.

11. CONCLUSION

11.1. General conclusion

Antioxidants such as curcumin,lycopene, fenretinide and other vitamin have shown potential benefits in the treatment of oral leukoplakia. This suggests that antioxidants can help improve clinical outcomes by reducing oxidative stress and modulating inflammation in patients with oral leukoplakia.

11.2. Specific conclusions

1. The antioxidants studied significantly reduced key clinical parameters, including lesion size and the appearance of the lesions, as well as improving the clinical manifestations of oral leukoplakia. These results suggest that antioxidants may play a protective role in preventing the progression of oral leukoplakia and its malignant transformation.

2. Antioxidants also had a favorable impact on biochemical markers related to oxidative stress and inflammation, such as levels of MDA, 8-OHdG, and other biomarkers. This indicates that antioxidants can modulate oxidative damage and immune response in oral leukoplakia, potentially reducing the risk of further epithelial damage and cancer progression.

3. The use of antioxidants, particularly curcumin, vitamin, lycopene, and fenretinide, represents a promising therapeutic approach for the management of oral leukoplakia. However, further research is required to confirm their long-term efficacy and safety in clinical settings.

12. REFERENCES

1. Chaturvedi AK, Udaltsova N, Engels EA, Katzel JA, Yanik EL, Katki HA, et al. Oral Leukoplakia and Risk of Progression to Oral Cancer: A Population-Based Cohort Study. J Natl Cancer Inst. 1 oct 2020;112(10):1047-54.

2. Sciubba JJ. Oral precancer and cancer: etiology, clinical presentation, diagnosis, and management. Compend Contin Educ Dent Jamesburg NJ 1995. oct 2000;21(10A):892-8, 900-2; quiz 903.

3. Warnakulasuriya S. Prognostic biomarkers in oral leukoplakia. Oral Dis. nov 2019;25(8):2047.

4. Palmerín-Donoso A, Cantero-Macedo AM, Tejero-Mas M. [Oral leukoplakia]. Aten Primaria. janv 2020;52(1):59-60.

5. Cai X, Zhang J, Zhang H, Li T. Biomarkers of malignant transformation in oral leukoplakia: from bench to bedside. J Zhejiang Univ Sci B. 13 mai 2023;24(10):868-82.

6. Babiuch K, Bednarczyk A, Gawlik K, Pawlica-Gosiewska D, Kęsek B, Darczuk D, et al. Evaluation of enzymatic and non-enzymatic antioxidant status and biomarkers of oxidative stress in saliva of patients with oral squamous cell carcinoma and oral leukoplakia: a pilot study. Acta Odontol Scand. août 2019;77(6):408-18.

7. Villa A, Sonis S. Oral leukoplakia remains a challenging condition. Oral Dis. mars 2018;24(1-2):179-83.

8. Barros CC da S, Freitas R de A, Miguel MC da C, Dantas da Silveira ÉJ. DNA damage through oxidative stress is an important event in oral leukoplakia. Arch Oral Biol. mars 2022;135:105359.

9. Sies H. Oxidative stress: a concept in redox biology and medicine. Redox Biol. 2015;4:180-3.

10. Rubert A, Bagán L, Bagán JV. Retraction: Oral leukoplakia, a clinical-histopathological study in 412 patients. J Clin Exp Dent. mai 2021;13(5):e426-32.

11. Jomova K, Alomar SY, Alwasel SH, Nepovimova E, Kuca K, Valko M. Several lines of antioxidant defense against oxidative stress: antioxidant enzymes, nanomaterials with multiple enzyme-mimicking activities, and low-molecular-weight antioxidants. Arch Toxicol. mai 2024;98(5):1323-67.

12. Bhattarai BP, Singh AK, Singh RP, Chaulagain R, Søland TM, Hasséus B, et al. Recurrence in Oral Leukoplakia: A Systematic Review and Meta-analysis. J Dent Res. oct 2024;103(11):1066-75.

13. Rudrapal M, Khairnar SJ, Khan J, Dukhyil AB, Ansari MA, Alomary MN, et al. Dietary Polyphenols and Their Role in Oxidative Stress-Induced Human Diseases: Insights Into Protective Effects, Antioxidant Potentials and Mechanism(s) of Action. Front Pharmacol. 2022;13:806470.

14. Halliwell B. Oxidative stress and cancer: have we moved forward? Biochem J. 1 janv 2007;401(1):1-11.

15. Liguori I, Russo G, Curcio F, Bulli G, Aran L, Della-Morte D, et al. Oxidative stress, aging, and diseases. Clin Interv Aging. 2018;13:757-72.

16. Halliwell B. Understanding mechanisms of antioxidant action in health and disease. Nat Rev Mol Cell Biol. janv 2024;25(1):13-33.

17. McCord JM, Edeas MA. SOD, oxidative stress and human pathologies: a brief history and a future vision. Biomed Pharmacother Biomedecine Pharmacother. mai 2005;59(4):139-42.

18. Ursini F, Maiorino M, Brigelius-Flohé R, Aumann KD, Roveri A, Schomburg D, et al. Diversity of glutathione peroxidases. Methods Enzymol. 1995;252:38-53.

19. Brigelius-Flohé R, Kipp A. Glutathione peroxidases in different stages of carcinogenesis. Biochim Biophys Acta. nov 2009;1790(11):1555-68.

20. Forman HJ, Zhang H. Targeting oxidative stress in disease: promise and limitations of antioxidant therapy. Nat Rev Drug Discov. sept 2021;20(9):689-709.

21. Srivastava KC, Austin RD, Shrivastava D, Pranavadhyani G. Oxidant-antioxidant status in tissue samples of oral leukoplakia. Dent Res J. mars 2014;11(2):180-6.

22. Bose SC, Singh M, Vyas P, Singh M. Plasma zinc antioxidant vitamins, glutathione levels and total antioxidant activity in oral leukoplakia. Dent Res J. mars 2012;9(2):158-61.

23. Nagao T, Ikeda N, Warnakulasuriya S, Fukano H, Yuasa H, Yano M, et al. Serum antioxidant micronutrients and the risk of oral leukoplakia among Japanese. Oral Oncol. sept 2000;36(5):466-70.

24. Srivastava KC. Comparative Evaluation of Saliva's Oxidant-Antioxidant Status in Patients with Different Clinicopathological Types of Oral Leukoplakia. J Int Soc Prev Community Dent. août 2019;9(4):396-402.

25. Agrawal D, Gupta S, Agarwal D, Gupta OP, Agarwal M. Role of GSTM1 and GSTT1 polymorphism: susceptibility to oral submucous fibrosis in the North Indian population. Oncology. 2010;79(3-4):181-6.

26. Rai S, Sharma A, Ranjan V, Misra D, Panjwani S. Estimation of serum antioxidant enzymes in histopathological grades of oral leukoplakia, oral submucous fibrosis, and oral cancer: A clinicopathologic study. 2015;

27. Mampilly MO, Ravindran N, Parambil MS, Nilesh K, Jayagopalan P, Dhamali D. Assessment of Serum Selenium and Ceruloplasmin in Potentially Malignant Disorders and Oral Cancer. J Pharm Bioallied Sci. nov 2021;13(Suppl 2):S989-92.

28. Shah PH, Venkatesh R, More CB. Determination of role of ceruloplasmin in oral potentially malignant disorders and oral malignancy-A cross-sectional study. Oral Dis. nov 2017;23(8):1066-71.

29. Balasundaram N, Narayanan MA, Antony LSA, Kumar SR, Anandan S. Effectiveness of Fenugreek as an Adjuvant in the Management of Oral Potentially Malignant Disorders: A Randomized Controlled Trial. J Contemp Dent Pract. 1 oct 2024;25(10):921-9.

30. Mallery SR, Zwick JC, Pei P, Tong M, Larsen PE, Shumway BS, et al. Topical application of a bioadhesive black raspberry gel modulates gene expression and reduces cyclooxygenase 2 protein in human premalignant oral lesions. 2008;

31. Yang M, Chen X, Cheng C, Yan W, Guo R, Wang Y, et al. Cucurbitacin B induces ferroptosis in oral leukoplakia via the SLC7A11/mitochondrial oxidative stress pathway. Phytomedicine Int J Phytother Phytopharm. juill 2024;129:155548.

32. Ansari S, Charantimath S, Fernandes A, Malik JB, Panta P. Comparison of Effectiveness of Moringa Oleifera Leaves Extract Gel (2%) with Retino A (0.1%) Cream for Treatment of Oral Leukoplakia: Double Blinded Randomized Control Trial. Gulf J Oncolog. sept 2023;1(43):12-8.

33. Khaera SN, Zaizafun AF, Anisa AN, Mude AH. The effectiveness of watermelon rind extracts waste (citrullus lanatus l.) in removing free radical in oral leukoplakia patients. 2022

34. Fathima AIS, Manoharan GVGM. Comparison of Efficacy of Topical Application of Bleomycin with Adjuvant Antioxidants vs. Topical Curcumin Oral Gel with Adjuvant Antioxidants in the Treatment of Oral Leukoplakia. 2022

35. M S, A B. Comparison of effectiveness of Calendula officinalis extract gel with lycopene gel for treatment of tobacco-induced homogeneous leukoplakia: A randomized clinical trial. Int J Pharm Investig.

36. Patel JS, Umarji HR, Dhokar AA, Sapkal RB, Patel SG, Panda AK. Randomized controlled trial to evaluate the efficacy of oral lycopene in combination with vitamin E and selenium in the treatment of oral leukoplakia. J Indian Acad Oral Med Radiol. déc 2014;26(4):369.

37. Chiesa F, Tradati N, Grigolato R, Boracchi P, Biganzoli E, Crose N, et al. Randomized trial of fenretinide (4-HPR) to prevent recurrences, new localizations and carcinomas in patients operated on for oral leukoplakia: long-term results. Int J Cancer. 1 juill 2005;115(4):625-9.

38. Kuriakose MA, Ramdas K, Dey B, Iyer S, Rajan G, Elango KK, et al. A Randomized Double-Blind Placebo-Controlled Phase IIB Trial of Curcumin in Oral Leukoplakia. Cancer Prev Res Phila Pa. août 2016;9(8):683-91.

39. Ahmad T, Khan I, Sultana A, Rizvi MMA. A comparative study to evaluate the therapeutic effects of nutraceuticals in oral leukoplakia:- A randomized clinical trail. Natl J Maxillofac Surg. 2023;14(3):413-9.

40. Jain RK, Singh GB, Singh AP, Goel RK, Aryya NC, Jha SK. Role of measurement of antioxidant enzymes in evaluation of antioxidant therapy in tobacco abusers with oral leukoplakia. Indian J Otolaryngol Head Neck Surg Off Publ Assoc Otolaryngol India. oct 2011;63(4):336-42.

41. Leunig A, Betz CS, Baumgartner R, Grevers G, Issing WJ. Initial experience in the treatment of oral leukoplakia with high-dose vitamin A and follow-up 5-aminolevulinic acid induced protoporphyrin IX fluorescence. Eur Arch Oto-Rhino-Laryngol Off J Eur Fed Oto-Rhino-Laryngol Soc EUFOS Affil Ger Soc Oto-Rhino-Laryngol - Head Neck Surg. 2000;257(6):327-31.

42. Kapoor S, Arora P. Effect of curcumin in management of potentially malignant disorders-A comparative study. 2019.

43. Rai B, Kaur J, Jacobs R, Singh J. Possible action mechanism for curcumin in pre-cancerous lesions based on serum and salivary markers of oxidative stress. J Oral Sci. juin 2010;52(2):251-6.

44. Lippman SM, Lee JJ, Martin JW, El-Naggar AK, Xu X, Shin DM, et al. Fenretinide activity in retinoid-resistant oral leukoplakia. Clin Cancer Res Off J Am Assoc Cancer Res. 15 mai 2006;12(10):3109-14.

45. Khan UM, Sevindik M, Zarrabi A, Nami M, Ozdemir B, Kaplan DN, et al. Lycopene: Food Sources, Biological Activities, and Human Health Benefits. Oxid Med Cell Longev. 19 nov 2021;2021:2713511.

46. Marzocco S, Singla RK, Capasso A. Multifaceted Effects of Lycopene: A Boulevard to the Multitarget-Based Treatment for Cancer. Molecules. janv 2021;26(17):5333.

47. Wang R, Lu X, Yu R. Lycopene Inhibits Epithelial–Mesenchymal Transition and Promotes Apoptosis in Oral Cancer via PI3K/AKT/m-TOR Signal Pathway . Drug Des Devel Ther. 24 juin 2020;14:2461-71.

48. Tao A, Wang X, Li C. Effect of Lycopene on Oral Squamous Cell Carcinoma Cell Growth by Inhibiting IGF1 Pathway. Cancer Manag Res. 26 janv 2021;13:723-32.

49. Ahmad T, Khan I, Rizvi MM, Saalim M, Manzoor N, Sultana A. An overview of effect of lycopene and curcumin in oral leukoplakia and oral submucous fibrosis. Natl J Maxillofac Surg. 2021;12(3):316-23.

50. Guo J, Xie H, Wu H, Liang M. Efficacy of Lycopene in the Treatment of Oral Submucous Fibrosis: A Meta-analysis of Randomized Controlled Trials. J Evid-Based Dent Pract. déc 2020;20(4):101471.

51. Al-Maweri SA, Halboub E, Al-Qadhi G, Al-Wesabi M, Al-Sharani HM, Parveen S, et al. Efficacy of lycopene for management of oral potentially malignant disorders: A systematic review and meta-analysis. Oral Surg Oral Med Oral Pathol Oral Radiol. janv 2023;135(1):79-95.

52. Bao J, Chen C, Yan J, Wen Y, Bian J, Xu M, et al. Antioxidant therapy for patients with oral lichen planus: A systematic review and meta-analysis. Front Pharmacol [Internet]. 10 nov 2022.

53. Saawarn N, Shashikanth MC, Saawarn S, Jirge V, Chaitanya NC, Pinakapani R. Lycopene in the management of oral lichen planus: a placebo-controlled study. Indian J Dent Res Off Publ Indian Soc Dent Res. 2011;22(5):639-43.

54. Ibrahim IM, Althagafy HS, Abd-alhameed EK, Al-Thubiani WS, Hassanein EHM. Promising hepatoprotective effects of lycopene in different liver diseases. Life Sci. 1 déc 2022;310:121131.

55. Niu K, Guo H, Kakizaki M, Cui Y, Ohmori-Matsuda K, Guan L, et al. A tomato-rich diet is related to depressive symptoms among an elderly population

aged 70 years and over: a population-based, cross-sectional analysis. J Affect Disord. 10 janv 2013;144(1-2):165-70.

56. Stice CP, Xia H, Wang XD. Tomato lycopene prevention of alcoholic fatty liver disease and hepatocellular carcinoma development. Chronic Dis Transl Med. 1 déc 2018;4(4):211-24.

57. Akbik D, Ghadiri M, Chrzanowski W, Rohanizadeh R. Curcumin as a wound healing agent. Life Sci. 22 oct 2014;116(1):1-7.

58. Tejada S, Manayi A, Daglia M, Nabavi SF, Sureda A, Hajheydari Z, et al. Wound Healing Effects of Curcumin: A Short Review. Curr Pharm Biotechnol. 2016;17(11):1002-7.

59. Bahrami A, Majeed M, Sahebkar A. Curcumin: a potent agent to reverse epithelial-to-mesenchymal transition. Cell Oncol Dordr Neth. août 2019;42(4):405-21.

60. Khaitan T, Vishal null, Kabiraj A, Sinha DK, Ranjan R, Singh R. Curcuma Longa in the Treatment of Symptomatic oral lichen planus: A non-randomized controlled trial. Indian J Dermatol. 2022;67(4):478.

61. Saadati S, Sadeghi A, Mansour A, Yari Z, Poustchi H, Hedayati M, et al. Curcumin and inflammation in non-alcoholic fatty liver disease: a randomized, placebo controlled clinical trial. BMC Gastroenterol. 25 juill 2019;19(1):133.

62. Wang D, Pei P, Shea F, Spinney R, Chang A, Lahann J, et al. Growth modulatory effects of fenretinide encompass keratinocyte terminal differentiation: a favorable outcome for oral squamous cell carcinoma chemoprevention. Carcinogenesis. 1 juin 2024;45(6):436-49.

63. Veronesi U, De Palo G, Marubini E, Costa A, Formelli F, Mariani L, et al. Randomized trial of fenretinide to prevent second breast malignancy in women with early breast cancer. J Natl Cancer Inst. 3 nov 1999;91(21):1847-56.

64. Lovat PE, Ranalli M, Annichiarrico-Petruzzelli M, Bernassola F, Piacentini M, Malcolm AJ, et al. Effector mechanisms of fenretinide-induced apoptosis in neuroblastoma. Exp Cell Res. 10 oct 2000;260(1):50-60.

65. Salata GC, Malagó ID, Carvalho Dartora VFM, Marçal Pessoa AF, Fantini MC de A, Costa SKP, et al. Microemulsion for Prolonged Release of Fenretinide in the Mammary Tissue and Prevention of Breast Cancer Development. Mol Pharm. 6 sept 2021;18(9):3401-17.

66. Garewal HS, Katz RV, Meyskens F, Pitcock J, Morse D, Friedman S, et al. β-Carotene Produces Sustained Remissions in Patients With Oral Leukoplakia: Results of a Multicenter Prospective Trial. Arch Otolaryngol Neck Surg. 1 déc 1999;125(12):1305-10.

67. Shalaby RH, Fathi Y, Elsaadany B. Efficacy and safety of beta carotones in treatment of oral leukoplakia: systematic review and meta-analysis. J Oral Res. 31 déc 2019;8(6):488-98.

68. Garewal H. Potential role of β-carotene in prevention of oral cancer. Am J Clin Nutr. 1 janv 1991;53(1):294S-297S.

69. Stahl W, Sies H. Carotenoids and flavonoids contribute to nutritional protection against skin damage from sunlight. Mol Biotechnol. sept 2007;37(1):26-30.

70. Fernández-García E. Skin protection against UV light by dietary antioxidants. Food Funct. sept 2014;5(9):1994-2003.

71. Thawabteh AM, Jibreen A, Karaman D, Thawabteh A, Karaman R. Skin Pigmentation Types, Causes and Treatment-A Review. Mol Basel Switz. 18 juin 2023;28(12):4839.

72. Sherwin JC, Reacher MH, Dean WH, Ngondi J. Epidemiology of vitamin A deficiency and xerophthalmia in at-risk populations. Trans R Soc Trop Med Hyg. avr 2012;106(4):205-14.

73. McLaren DS. Vitamin A deficiency disorders. J Indian Med Assoc. août 1999;97(8):320-3.

74. Shehzad A, Wahid F, Lee YS. Curcumin in cancer chemoprevention: molecular targets, pharmacokinetics, bioavailability, and clinical trials. Arch Pharm (Weinheim). sept 2010;343(9):489-99.

75. Jelic MD, Mandic AD, Maricic SM, Srdjenovic BU. Oxidative stress and its role in cancer. J Cancer Res Ther. 2021;17(1):22-8.

76. Panahi Y, Khalili N, Sahebi E, Namazi S, Reiner Ž, Majeed M, et al. Curcuminoids modify lipid profile in type 2 diabetes mellitus: A randomized controlled trial. Complement Ther Med. août 2017;33:1-5.

77. Shao S, Ye X, Su W, Wang Y. Curcumin alleviates Alzheimer's disease by inhibiting inflammatory response, oxidative stress and activating the AMPK pathway. J Chem Neuroanat. 1 déc 2023;134:102363.

78. Kara Ö, Baş H, Pandır D. Furan toxicity on testes and protective role of lycopene in diabetic rats. J Turk Ger Gynecol Assoc. 2016;17(4):191-6.

79. Albrahim T. Lycopene Modulates Oxidative Stress and Inflammation in Hypercholesterolemic Rats. Pharmaceuticals. 17 nov 2022;15(11):1420.

80. Chen J, Song Y, Zhang L. Effect of lycopene supplementation on oxidative stress: an exploratory systematic review and meta-analysis of randomized controlled trials. J Med Food. mai 2013;16(5):361-74.

81. Hail N, Kim HJ, Lotan R. Mechanisms of fenretinide-induced apoptosis. Apoptosis Int J Program Cell Death. oct 2006;11(10):1677-94.

82. Lotan R. Retinoids and Apoptosis: Implications for Cancer Chemoprevention and Therapy. JNCI J Natl Cancer Inst. 15 nov 1995;87(22):1655-7.

13. ANNEXES

Table 1. Database Search Formula.

Database	Search Formula	Number of articles	Date
Medline (PubMed)	(("oral leukoplakia"[All Fields] OR ("leucoplakias"[All Fields] OR "leukoplakia"[MeSH Terms] OR "leukoplakia"[All Fields] OR "leucoplakia"[All Fields] OR "leukoplakias"[All Fields])) AND ("antioxidant s"[All Fields] OR "antioxidants"[Pharmacological Action] OR "antioxidants"[MeSH Terms] OR "antioxidants"[All Fields] OR "antioxidant"[All Fields] OR "antioxidating"[All Fields] OR "antioxidation"[All Fields] OR "antioxidative"[All Fields] OR "antioxidatively"[All Fields] OR "antioxidatives"[All Fields] OR "antioxidizing"[All Fields] OR "antioxidant therapy"[All Fields] OR "antioxidant treatment"[All Fields])) AND (2000:2025[pdat])	87	10/02/2025
Web of Science	("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment") (Topic) and Review Article (Exclude – Document Types) and 1988 or 1989 or 1991 or 1992 or 1993 or 1994 or 1995 or 1997 or 1998 or 1999 (Exclude – Publication Years)	60	10/02/2025

SCOPUS	TITLE-ABS-KEY (("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment")) AND PUBYEAR > 1999 AND PUBYEAR < 2025 AND (LIMIT-TO (DOCTYPE , "ar")) AND (LIMIT-TO (LANGUAGE , "English"))	89	10/02/2025

Table 2 :Prisma 2020 checklist

Section and topic	Item n.°	Checklist item	Location where item is reported
TITLE			
Título	1	Identify the report as a systematic review	p.1
ABSTRACT			
Abstract	2	See the PRISMA 2020 for Abstracts checklist	p.1
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of existing knowledge.	p.7-13
Objectives	4	Provide an explicit statement of the objective(s) or question(s) the review addresses.	p.17
METODS			

Eligibility criteria	5	Specify the inclusion and exclusion criteria for the review and how studies were grouped for the syntheses.	p.19
Information sources	6	Specify all databases, registers, websites, organisations, reference lists and other sources searched or consulted to identify studies. Specify the date when each source was last searched or consulted.	p.21
Search strategy	7	Present the full search strategies for all databases, registers and websites, including any filters and limits used.	p.21
Selection process	8	Specify the methods used to decide whether a study met the inclusion criteria of the review, including how many reviewers screened each record and each report retrieved, whether they worked independently, and if applicable, details of automation tools used in the process.	p.22
Data collection process	9	Specify the methods used to collect data from reports, including how many reviewers collected data from each report, whether they worked independently, any processes for obtaining or confirming data from study investigators, and if applicable, details of automation tools used in the process.	p.23
Data items	10a	List and define all outcomes for which data were sought. Specify whether all results that were compatible with each outcome domain in each study were sought (e.g. for all measures, time points, analyses), and if not, the methods used to decide which results to collect.	24
	10b	List and define all other variables for which data were sought (e.g. participant and intervention characteristics, funding sources). Describe any assumptions made about any missing or unclear information.	24

Study risk of bias assessment	11	Specify the methods used to assess risk of bias in the included studies, including details of the tool(s) used, how many reviewers assessed each study and whether they worked independently, and if applicable, details of automation tools used in the process.	24-26
Effect measures	12	Specify for each outcome the effect measure(s) (e.g. risk ratio, mean difference) used in the synthesis or presentation of results.	26
Synthesis methods	13a	Describe the processes used to decide which studies were eligible for each synthesis (e.g. tabulating the study intervention characteristics and comparing against the planned groups for each synthesis (item #5)).	26
	13b	Describe any methods required to prepare the data for presentation or synthesis, such as handling of missing summary statistics, or data conversions.	
	13c	Describe any methods used to tabulate or visually display results of individual studies and syntheses.	
	13d	Describe any methods used to synthesize results and provide a rationale for the choice(s). If meta-analysis was performed, describe the model(s), method(s) to identify the presence and extent of statistical heterogeneity, and software package(s) used.	
	13e	Describe any methods used to explore possible causes of heterogeneity among study results (e.g. subgroup analysis, meta-regression).	
	13f	Describe any sensitivity analyses conducted to assess robustness of the synthesized results.	

Reporting bias assessment	14	Describe any methods used to assess risk of bias due to missing results in a synthesis (arising from reporting biases).	35-36
Certainty assessment	15	Describe any methods used to assess certainty (or confidence) in the body of evidence for an outcome.	35-36
Results			
Study selection	16a	Describe the results of the search and selection process, from the number of records identified in the search to the number of studies included in the review, ideally using a flow diagram.	27
	16b	Cite studies that might appear to meet the inclusion criteria, but which were excluded, and explain why they were excluded.	28-29
Study characteristics	17	Cite each included study and present its characteristics.	30-31
Risk of bias in studies	18	Present assessments of risk of bias for each included study.	32-34
Results of individual studies	19	For all outcomes, present, for each study: (a) summary statistics for each group (where appropriate) and (b) an effect estimate and its precision (e.g. confidence/credible interval), ideally using structured tables or plots.	35-46
Results of syntheses	20a	For each synthesis, briefly summarise the characteristics and risk of bias among contributing studies.	35-46

	20b	Present results of all statistical syntheses conducted. If meta-analysis was done, present for each the summary estimate and its precision (e.g. confidence/credible interval) and measures of statistical heterogeneity. If comparing groups, describe the direction of the effect.	
	20c	Present results of all investigations of possible causes of heterogeneity among study results.	
	20d	Present results of all sensitivity analyses conducted to assess the robustness of the synthesized results.	
Reporting biases	21	Present assessments of risk of bias due to missing results (arising from reporting biases) for each synthesis assessed.	53
Certainty of evidence	22	Present assessments of certainty (or confidence) in the body of evidence for each outcome assessed.	53-54
DISCUSSION			
Discussion	23a	Provide a general interpretation of the results in the context of other evidence.	47-49
	23b	Discuss any limitations of the evidence included in the review.	50-52
	23c	Discuss any limitations of the review processes used.	53
	23d	Discuss implications of the results for practice, policy, and future research.	53
OTHER INFORMATION			

Registration and protocol	24a	Provide registration information for the review, including register name and registration number, or state that the review was not registered.	18
	24b	Indicate where the review protocol can be accessed, or state that a protocol was not prepared.	-
	24c	Describe and explain any amendments to information provided at registration or in the protocol.	-
Support	25	Describe sources of financial or non-financial support for the review, and the role of the funders or sponsors in the review.	-
Competing interests	26	Declare any competing interests of review authors.	-
Availability of data, code and other materials	27	Report which of the following are publicly available and where they can be found: template data collection forms; data extracted from included studies; data used for all analyses; analytic code; any other materials used in the review.	-

Declaration of artificial intelligence (AI) use in the development of the TFG :

During the preparation of this work, an AI technology was used to support syntax correction and ensure linguistic accuracy.

Tool : Chatgpt 4

Function : used for spelling and grammar adjustments, as well as reforing sentence structure to entrance readability.

Prompt used :

« Can you assess this sentence is properly structured and easy to understand »

« Can you ensure that this paragraph is grammatically correct and well-structured »

« Can u correct the syntax of this paragraph »

EFICACIA DEL TRATAMIENTO ANTIOXIDANTE EN EL MANEJO DE LA LEUCOPLASIA ORAL: REVISIÓN SISTEMÁTICA

Titulo corto: Tratamiento antioxidante en leucoplasia oral

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Resumen

Introducción: La leucoplasia oral es el trastorno potencialmente maligno más frecuente de la cavidad oral. Su desarrollo y posible transformación maligna se han relacionado con el estrés oxidativo. En este contexto, los tratamientos antioxidantes han sido propuestos como una estrategia terapéutica para contrarrestar el daño oxidativo y prevenir la progresión de las lesiones.

Objetivos: Evaluar la eficacia clínica y bioquímica de los tratamientos antioxidantes en pacientes con leucoplasia oral.

Material y método: Se realizó una revisión sistemática siguiendo las directrices PRISMA. Se efectuó una búsqueda electrónica en las bases de datos PubMed, Scopus y Web of Science de estudios publicados entre 2000 y 2025. Se incluyeron ensayos clínicos aleatorizados, estudios clínicos controlados y estudios observacionales que evaluaran el uso de antioxidantes tópicos o sistémicos en pacientes con leucoplasia oral.

Resultados: Se incluyeron 11 estudios con un total de 695 pacientes. Los antioxidantes más investigados fueron el licopeno, la curcumina, la vitamina A y el fenretinida. La mayoría de los estudios reportaron mejoras clínicas significativas en cuanto a reducción del tamaño o regresión de la lesión. A nivel bioquímico, se observó una disminución del estrés oxidativo evidenciada por la reducción de los niveles de LPO, MDA y 8-OHdG, así como un aumento de las enzimas antioxidantes como la catalasa (CAT), vitaminas C y E. Algunos estudios mostraron una tasa elevada de apoptosis en el epitelio alterado. Los resultados sobre la superóxido dismutasa (SOD) fueron variables. La heterogeneidad en los diseños, dosis y duración del tratamiento limitó la comparación directa entre estudios.

Discusión: A pesar de las limitaciones metodológicas, los tratamientos antioxidantes parecen ofrecer beneficios tanto clínicos como bioquímicos en la gestión de la leucoplasia oral. Se requieren ensayos clínicos adicionales, estandarizados y de alta calidad para confirmar su eficacia y su papel en la prevención de la transformación maligna.

Palabras clave: Oral leukoplakia, Leukoplakia, Antioxidants, Antioxidant therap Antioxidant treatment

Introducción

La leucoplasia oral (LO) es el trastorno potencialmente maligno más común de la mucosa oral. Clínicamente, se presenta como una placa blanca persistente que no puede eliminarse mediante raspado y que no puede caracterizarse como ninguna otra lesión definible. La LO se considera una condición premaligna debido a su potencial de transformación maligna en carcinoma oral de células escamosas (COCE), un tipo de cáncer oral que representa una proporción significativa de los casos de cáncer a nivel mundial. Se estima que aproximadamente entre el 5 % y el 15 % de los individuos con LO pueden experimentar transformación maligna, lo que resalta la importancia de una detección precoz y un manejo adecuado de este trastorno (1,2).

La patogénesis de la LO está estrechamente relacionada con el estrés oxidativo, que resulta de un desequilibrio entre la producción de especies reactivas de oxígeno (ROS) y los mecanismos de defensa antioxidante del organismo. El estrés oxidativo contribuye al daño del ADN, la peroxidación lipídica y la desregulación de vías de señalización implicadas en la carcinogénesis (3-4).
En los últimos años, ha aumentado el interés por el uso terapéutico de antioxidantes, tanto naturales como sintéticos, debido a su capacidad para neutralizar el estrés oxidativo, modular la respuesta inflamatoria y, potencialmente, revertir o detener la progresión de las lesiones de LO (1,5,6). Compuestos como el licopeno, la curcumina, los retinoides y la vitamina E han mostrado resultados prometedores en estudios preclínicos y clínicos. Sin embargo, la eficacia clínica de estos tratamientos antioxidantes sigue siendo incierta debido a la heterogeneidad en el diseño de los estudios, las dosis utilizadas y los criterios de evaluación.

El objetivo de esta revisión sistemática fue evaluar la eficacia del tratamiento antioxidante en el manejo de la leucoplasia oral, tanto en términos de resultados clínicos (reducción del tamaño de la lesión, regresión, mejoría del aspecto clínico) como de resultados bioquímicos (modulación del estrés oxidativo o de biomarcadores inflamatorios). Esta revisión sigue las directrices PRISMA e

incluye estudios clínicos y ensayos controlados aleatorizados publicados entre los años 2000 y 2025.

Materiales y Metodos

Se realizó una revisión sistemática siguiendo las directrices PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses), que proporcionan un marco estructurado para la elaboración transparente y reproducible de revisiones sistemáticas y metaanálisis.

Pregunta PICO:

El objetivo de esta revisión es responder a una pregunta formulada mediante el método PICO

- P (Población): Pacientes adultos diagnosticados con leucoplasia oral.
- I (Intervención): Tratamientos antioxidantes (por ejemplo, vitamina A, cúrcuma, entre otros), administrados por vía tópica o sistémica.
- C (Comparación): Placebo, grupo control (sin tratamiento) o tratamiento convencional (vigilancia activa, cirugía, corticosteroides). También se consideraron comparaciones entre diferentes tratamientos antioxidantes.
- O (Resultados):
    - O1: Reducción del tamaño de las lesiones de leucoplasia oral.
    - O2: Prevención de la progresión a formas más graves como displasia o cáncer.
    - O3: Mejora de los síntomas clínicos (dolor, inflamación, dificultad para comer o hablar).
    - O4: Cambios en los biomarcadores de estrés oxidativo (niveles en saliva o sangre).

Criterios de inclusión

Se utilizaron los siguientes criterios para determinar los estudios elegibles para esta revisión sistemática:

- Diseño del estudio: Ensayos clínicos controlados aleatorizados (ECA), estudios controlados y estudios observacionales.
- Idiomas: Estudios publicados en inglés, español y francés.
- Población: Pacientes con leucoplasia oral.
- Sujetos: Estudios realizados en humanos.

- Intervención: Tratamientos antioxidantes (vitamina A, cúrcuma, té verde, vitamina C, vitamina E) para la leucoplasia oral, administrados por vía tópica o sistémica.
- Comparación: Placebo, grupo control (sin tratamiento) o tratamientos convencionales (vigilancia activa, cirugía, corticosteroides).
- Comparaciones entre diferentes antioxidantes.
- Resultados:
    - Reducción del tamaño de la lesión.
    - Prevención de la transformación maligna (progresión a displasia o cáncer).
    - Mejoría clínica (alivio del dolor, reducción de la inflamación, mejora funcional).
    - Reducción del estrés oxidativo medida mediante biomarcadores salivales o sanguíneos.
- Fecha de publicación: Estudios publicados entre 2000 y 2025.

Criterios de exclusión

Se excluyeron los estudios según los siguientes criterios:

- Estudios con pacientes con otras condiciones orales (ej. cáncer oral).
- Estudios sin tratamiento antioxidante.
- Estudios sin grupo de comparación o sin resultados medibles relacionados con la regresión de la lesión.
- Estudios con baja calidad metodológica, con alto riesgo de sesgo o datos insuficientemente reportados.
- Estudios con menos de 4 pacientes (n < 4).
- Estudios publicados en idiomas que no fueran inglés, español o francés.

Fuentes de información y estrategia de búsqueda:

Se utilizaron las siguientes bases de datos para identificar estudios relevantes: PubMed, Web of Science y SCOPUS. (("oral leukoplakia"[All Fields] OR ("leucoplakias"[All Fields] OR "leukoplakia"[MeSH Terms] OR "leukoplakia"[All Fields] OR "leucoplakia"[All Fields] OR "leukoplakias"[All Fields])) AND ("antioxidant s"[All Fields] OR "antioxidants"[Pharmacological Action] OR "antioxidants"[MeSH Terms] OR "antioxidants"[All Fields] OR "antioxidant"[All

Fields] OR "antioxidating"[All Fields] OR "antioxidation"[All Fields] OR "antioxidative"[All Fields] OR "antioxidatively"[All Fields] OR "antioxidatives"[All Fields] OR "antioxidizing"[All Fields] OR "antioxidant therapy"[All Fields] OR "antioxidant treatment"[All Fields])) AND (2000:2025[pdat]), ("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment") (Topic) and Review Article (Exclude – Document Types) and 1988 or 1989 or 1991 or 1992 or 1993 or 1994 or 1995 or 1997 or 1998 or 1999 (Exclude – Publication Years), TITLE-ABS-KEY (("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment")) AND PUBYEAR > 1999 AND PUBYEAR < 2025 AND (LIMIT-TO (DOCTYPE , "ar")) AND (LIMIT-TO (LANGUAGE , "English"))

Proceso de selección de estudios:

La selección se realizó en dos etapas:

Etapa 1: Se evaluaron títulos y resúmenes para descartar estudios que no cumplían los criterios preliminares (ej. población no adecuada, ausencia de tratamiento antioxidante). Se documentaron las razones de exclusión.

Etapa 2: Se revisaron los textos completos de los estudios preseleccionados para confirmar su elegibilidad. Se aplicaron criterios adicionales, como calidad metodológica e idioma. Se excluyeron estudios que no cumplían los criterios o cuyo texto completo no estaba disponible.

Extracción de datos:

Se extrajeron datos relevantes como las características de los estudios, tamaño de muestra y dosis de antioxidantes. También se recogieron resultados clínicos (reducción del tamaño de la lesión) y bioquímicos (biomarcadores de estrés oxidativo e inflamación).

Evaluación de la calidad:

Se utilizó la herramienta Cochrane Risk of Bias para evaluar la calidad metodológica y el riesgo de sesgo. Esta herramienta valora aspectos como la aleatorización, el cegamiento y el manejo de datos faltantes. Cada estudio fue clasificado como de bajo, alto o riesgo de sesgo incierto

Síntesis de los datos:

Se emplearon métodos cualitativos y cuantitativos para sintetizar los datos. Los resultados clínicos se resumieron de manera narrativa, agrupando los estudios según el tipo de antioxidante y sus resultados. Se elaboró un diagrama de flujo PRISMA para ilustrar el proceso de selección y una tabla para resumir las características de los estudios incluidos.

Resultados

Selección de estudios

El proceso de búsqueda inicial identificó un total de 244 artículos: de PubMed (n=87), Scopus (n=89), Web of Science (n=60) y búsqueda manual (n=8). Como resultado, se conservaron 25 artículos para la revisión del texto completo. En total, se incluyeron 11 estudios en la síntesis cualitativa. La Figura 1 presenta el diagrama de flujo PRISMA que ilustra este proceso de selección. Además, los detalles de los artículos excluidos se encuentran en la Tabla 1.

Análisis de las características de los estudios incluidos

De los 11 estudios incluidos en esta revisión, 5 evaluaron la curcumina como principal antioxidante (7-11), 3 se centraron en el licopeno (12,13,11), y 2 en el fenretinide (14,15). Un estudio también incluyó combinaciones de antioxidantes o análisis enzimáticos de antioxidantes (16) y 1 en la vitamina A a dosis altas (17).). Se incluyeron un total de 365 pacientes: 140 recibieron curcumina, 97 licopeno, 52 fenretinide, 36 vitamina A, y el resto recibió otras terapias antioxidantes. La duración de los tratamientos varió de 1 a 12 meses. Los resultados primarios evaluados fueron la resolución clínica, la reducción del tamaño de la lesión, los cambios histopatológicos y los marcadores bioquímicos del estrés oxidativo (por ejemplo, MDA, 8-OHdG). Algunos estudios también examinaron resultados a largo plazo, como la recurrencia y la transformación maligna.

Evaluación de la calidad metodológica

La Tabla 2 presenta la evaluación de calidad de los ECA y fueron evaluados con la herramienta Cochrane, y los estudios no aleatorizados con la escala NOS

(tabla 3 y 4). La mayoría de los ECA presentaron riesgo moderado de sesgo, principalmente por cegamiento incompleto o falta de información sobre la aleatorización. Los estudios observacionales mostraron sesgo moderado debido a la selección de cohortes o factores de confusión. Los estudios sin grupo control tuvieron mayor sesgo, especialmente por falta de comparabilidad. En general, la calidad metodológica fue variable, aunque la mayoría de los estudios apoyan la eficacia de los antioxidantes para la leucoplasia oral.

Síntesis de resultados

El análisis reveló dos ejes principales de evaluación del tratamiento antioxidante en la leucoplasia oral (tabla 5):

1. Eficacia clínica de los antioxidantes

La mayoría de los ensayos clínicos controlados reportaron una reducción significativa en el tamaño de las lesiones y una notable mejora en su apariencia clínica tras el tratamiento con antioxidantes, en comparación con placebo o terapias convencionales. Por ejemplo, la curcumina, utilizada sola o en combinación con otros antioxidantes, demostró una efectividad notable, con altas tasas de regresión completa o parcial de las lesiones en los estudios (7-11). De igual forma, el licopeno resultó eficaz, con mejoras clínicas observadas a pocas semanas del inicio del tratamiento, como se informó en estudios como los de (12,13,11). La combinación de múltiples antioxidantes (vitamina E y selenio) también produjo resultados favorables, sugiriendo un posible efecto sinérgico (16). Otros compuestos, como el fenretinide (un derivado de la vitamina A), mostraron una eficacia más variable, a veces limitada a la prevención de recurrencias o a subgrupos específicos de pacientes (10,14).

2. Impacto sobre biomarcadores

A nivel bioquímico, varios estudios evaluaron el efecto de los antioxidantes sobre los marcadores de estrés oxidativo (como GPx, SOD y especies reactivas de oxígeno). En general, los resultados indican un aumento de los niveles endógenos de antioxidantes y una disminución de los marcadores proinflamatorios tras el tratamiento. Por ejemplo, (16) reportó una disminución significativa en los niveles de LPO y SOD tras la administración de β-caroteno y una combinación de otros antioxidantes (vitamina E, selenio...). En otro estudio

(9), se observó un aumento de las vitaminas C y E, y una disminución del MDA y 8-OHdG. Además, un estudio que involucró el fenretinide (15) mostró algunas mejoras en los biomarcadores y un aumento en la apoptosis, aunque los efectos fueron más modestos, lo que sugiere que la eficacia antioxidante puede depender del compuesto específico, la duración del tratamiento o el perfil del paciente.

Discusión

Evaluación de los antioxidantes sobre los parámetros clínicos de la leucoplasia oral

Los antioxidantes han mostrado efectos beneficiosos en la reducción de lesiones clínicas en el tratamiento de la leucoplasia oral. Entre los compuestos evaluados, el licopeno, un carotenoide natural, ha destacado especialmente, con varios estudios que informan reducciones significativas de las lesiones. Sus efectos se atribuyen a su capacidad para estabilizar las membranas celulares, inhibir la proliferación celular y modular genes relacionados con el estrés oxidativo y la inflamación (11,12,13). Además, el licopeno ha demostrado beneficios en el tratamiento de otras afecciones orales y sistémicas, como la fibrosis submucosa y enfermedades hepáticas (18-22).

La curcumina, derivada de *Curcuma longa*, también ha mostrado resultados positivos, especialmente en la reducción del tamaño de las lesiones y la mejora de la textura del tejido. Sus efectos beneficiosos se relacionan con sus propiedades antiinflamatorias y regeneradoras del epitelio (7,8,10,11). Este compuesto también se ha estudiado por su potencial terapéutico en afecciones como el liquen plano oral y enfermedades hepáticas (23-28).

El fenretinide, un análogo sintético de los retinoides, ha mostrado resultados prometedores en casos resistentes a terapias convencionales. Se ha asociado con la estabilización y reducción de las lesiones mediante la modulación de la diferenciación epitelial y la inhibición de la proliferación celular, lo que sugiere un beneficio clínico sostenido (14,15). Este compuesto también ha demostrado efectos en el cáncer de mama y otras afecciones epiteliales (29,30).

Compuestos como el β-caroteno, la vitamina C, la vitamina E y otras combinaciones antioxidantes han mostrado resultados prometedores en la reducción del estrés oxidativo, aunque los estudios sobre estas formulaciones no siempre evaluaron resultados histológicos ni a largo plazo (16, 31-33).

En conclusión, los antioxidantes, especialmente el licopeno, la curcumina y el fenretinide, muestran un importante potencial terapéutico en el manejo de la leucoplasia oral. Sin embargo, se requieren ensayos clínicos más rigurosos para confirmar su eficacia y seguridad a largo plazo

Evaluación del efecto antioxidante sobre los marcadores bioquímicos y de estrés oxidativo en la leucoplasia oral

La leucoplasia oral es un trastorno potencialmente maligno en el que el estrés oxidativo y la inflamación crónica juegan un papel central en su patogénesis. Entre los antioxidantes evaluados, la curcumina ha mostrado efectos significativos tanto a nivel clínico como bioquímico. Los estudios encontraron que la suplementación con curcumina aumentó los niveles séricos de vitaminas C y E, al tiempo que disminuyó los marcadores de estrés oxidativo como MDA y 8-OHdG. Estos hallazgos sugieren que las propiedades antioxidantes de la curcumina pueden reducir el daño oxidativo y mejorar las defensas antioxidantes sistémicas, lo cual es crucial en pacientes con trastornos potencialmente malignos que a menudo presentan una capacidad antioxidante reducida (9).

El licopeno también demostró efectos significativos en la reducción del estrés oxidativo y en la mejora de los resultados clínicos. Un estudio observó una disminución de los niveles de MDA y un aumento de las enzimas antioxidantes como SOD y GPx, lo que indica que el licopeno mejora las defensas antioxidantes sistémicas (16). La doble acción del licopeno en la reducción del estrés oxidativo y la inflamación respalda su papel en la regeneración celular y en la prevención de la transformación maligna en la leucoplasia oral.

El fenretinide, especialmente en casos resistentes a los retinoides, ha mostrado efectos prometedores al aumentar la apoptosis en células epiteliales orales. Aunque la respuesta clínica no se correlacionó directamente con los niveles de apoptosis, la capacidad del fenretinide para inducir apoptosis a través de vías mitocondriales y la acumulación de ceramidas apoya su potencial terapéutico en

casos de leucoplasia oral no respondientes (15). Además, su capacidad para reducir los niveles séricos de retinol indica su actividad biológica.

En conclusión, los antioxidantes como la curcumina, el licopeno y el fenretinide, junto con las vitaminas, desempeñan un papel crucial en la reducción del estrés oxidativo y la inflamación en la leucoplasia oral. Estos antioxidantes pueden actuar de manera sinérgica para mejorar la salud del tejido oral y prevenir la progresión hacia el cáncer oral, ofreciendo una estrategia terapéutica prometedora para el manejo de esta enfermedad.

Conclusión

Antioxidantes como la curcumina, el licopeno, el fenretinide y otras vitaminas han mostrado beneficios potenciales en el tratamiento de la leucoplasia oral. Esto sugiere que los antioxidantes pueden contribuir a mejorar los resultados clínicos al reducir el estrés oxidativo y modular la inflamación en pacientes con leucoplasia oral.

Bibliografia

(1) Chaturvedi AK, Udaltsova N, Engels EA, Katzel JA, Yanik EL, Katki HA, et al. Oral Leukoplakia and Risk of Progression to Oral Cancer: A Population-Based Cohort Study. *J Natl Cancer Inst*. 2020 Oct 1;112(10):1047–54.

(2) Sciubba JJ. Oral precancer and cancer: etiology, clinical presentation, diagnosis, and management. *Compend Contin Educ Dent*. 2000 Oct;21(10A):892–8, 900–2; quiz 903.

(3) Babiuch K, Bednarczyk A, Gawlik K, Pawlica-Gosiewska D, Kęsek B, Darczuk D, et al. Evaluation of enzymatic and non-enzymatic antioxidant status and biomarkers of oxidative stress in saliva of patients with oral squamous cell carcinoma and oral leukoplakia: a pilot study. *Acta Odontol Scand*. 2019 Aug;77(6):408–18.

(4) Sies H. Oxidative stress: a concept in redox biology and medicine. *Redox Biol*. 2015;4:180–3.

(5) Jomova K, Alomar SY, Alwasel SH, Nepovimova E, Kuca K, Valko M. Several lines of antioxidant defense against oxidative stress: antioxidant enzymes, nanomaterials with multiple enzyme-mimicking activities, and low-molecular-weight antioxidants. *Arch Toxicol*. 2024 May;98(5):1323–67.

(6) Rudrapal M, Khairnar SJ, Khan J, Dukhyil AB, Ansari MA, Alomary MN, et al. Dietary Polyphenols and Their Role in Oxidative Stress-Induced Human Diseases: Insights Into Protective Effects, Antioxidant Potentials and Mechanism(s) of Action. *Front Pharmacol*. 2022;13:806470.

(7) Fathima AIS, Manoharan GVGM. Comparison of Efficacy of Topical Application of Bleomycin with Adjuvant Antioxidants vs. Topical Curcumin Oral Gel with Adjuvant Antioxidants in the Treatment of Oral Leukoplakia. *J Indian Acad Oral Med Radiol*. 2022.

(8) Kapoor S, Arora P. Effect of curcumin in management of potentially malignant disorders - A comparative study. *J Indian Acad Oral Med Radiol*. 2019.

(9) Rai B, Kaur J, Jacobs R, Singh J. Possible action mechanism for curcumin in pre-cancerous lesions based on serum and salivary markers of oxidative stress. *J Oral Sci*. 2010 Jun;52(2):251–6.

(10) Kuriakose MA, Ramdas K, Dey B, Iyer S, Rajan G, Elango KK, et al. A Randomized Double-Blind Placebo-Controlled Phase IIB Trial of Curcumin in Oral Leukoplakia. *Cancer Prev Res*. 2016 Aug;9(8):683–91.

(11) Ahmad T, Khan I, Sultana A, Rizvi MMA. A comparative study to evaluate the therapeutic effects of nutraceuticals in oral leukoplakia: A randomized clinical trial. *Natl J Maxillofac Surg*. 2023;14(3):413–9.

(12) Patel JS, Umarji HR, Dhokar AA, Sapkal RB, Patel SG, Panda AK. Randomized controlled trial to evaluate the efficacy of oral lycopene in combination with vitamin E and selenium in the treatment of oral leukoplakia. *J Indian Acad Oral Med Radiol*. 2014 Dec;26(4):369.

(13) M S, A B. Comparison of effectiveness of *Calendula officinalis* extract gel with lycopene gel for treatment of tobacco-induced homogeneous leukoplakia: A randomized clinical trial. *Int J Pharm Investig*. 2017 Jun;7

(14) Chiesa F, Tradati N, Grigolato R, Boracchi P, Biganzoli E, Crose N, et al. Randomized trial of fenretinide (4-HPR) to prevent recurrences, new localizations and carcinomas in patients operated on for oral leukoplakia: long-term results. *Int J Cancer*. 2005 Jul 1;115(4):625–9.

(15) Lippman SM, Lee JJ, Martin JW, El-Naggar AK, Xu X, Shin DM, et al. Fenretinide activity in retinoid-resistant oral leukoplakia. *Clin Cancer Res*. 2006 May 15;12(10):3109–14.

(16) Jain RK, Singh GB, Singh AP, Goel RK, Aryya NC, Jha SK. Role of measurement of antioxidant enzymes in evaluation of antioxidant therapy in tobacco abusers with oral leukoplakia. *Indian J Otolaryngol Head Neck Surg*. 2011 Oct;63(4):336–42.

(17) Leunig A, Betz CS, Baumgartner R, Grevers G, Issing WJ. Initial experience in the treatment of oral leukoplakia with high-dose vitamin A and follow-up 5-aminolevulinic acid induced protoporphyrin IX fluorescence. Eur Arch Oto-Rhino-Laryngol Off J Eur Fed Oto-Rhino-Laryngol Soc EUFOS Affil Ger Soc Oto-Rhino-Laryngol - Head Neck Surg. 2000;257(6):327-31.

(18) Ahmad T, Khan I, Rizvi MM, Saalim M, Manzoor N, Sultana A. An overview of effect of lycopene and curcumin in oral leukoplakia and oral submucous fibrosis. *Natl J Maxillofac Surg*. 2021;12(3):316–2

(19) Guo J, Xie H, Wu H, Liang M. Efficacy of Lycopene in the Treatment of Oral Submucous Fibrosis: A Meta-analysis of Randomized Controlled Trials. *J Evid Based Dent Pract*. 2020 Dec;20(4):101471.

(20) Al-Maweri SA, Halboub E, Al-Qadhi G, Al-Wesabi M, Al-Sharani HM, Parveen S, et al. Efficacy of lycopene for management of oral potentially malignant disorders: A systematic review and meta-analysis. *Oral Surg Oral Med Oral Pathol Oral Radiol*. 2023 Jan;135(1):79–95.

(21) Bao J, Chen C, Yan J, Wen Y, Bian J, Xu M, et al. Antioxidant therapy for patients with oral lichen planus: A systematic review and meta-analysis. *Front Pharmacol*. 2022 Nov 10;13.

(22) Saawarn N, Shashikanth MC, Saawarn S, Jirge V, Chaitanya NC, Pinakapani R. Lycopene in the management of oral lichen planus: a placebo-controlled study. *Indian J Dent Res*. 2011;22(5):639–43.

(23) Akbik D, Ghadiri M, Chrzanowski W, Rohanizadeh R. Curcumin as a wound healing agent. *Life Sci*. 2014 Oct 22;116(1):1–7.

(24) Tejada S, Manayi A, Daglia M, Nabavi SF, Sureda A, Hajheydari Z, et al. Wound Healing Effects of Curcumin: A Short Review. *Curr Pharm Biotechnol*. 2016;17(11):1002–7.

(25) Bahrami A, Majeed M, Sahebkar A. Curcumin: a potent agent to reverse epithelial-to-mesenchymal transition. *Cell Oncol*. 2019 Aug;42(4):405–21.

(26) Khaitan T, Vishal, Kabiraj A, Sinha DK, Ranjan R, Singh R. *Curcuma longa* in the Treatment of Symptomatic Oral Lichen Planus: A non-randomized controlled trial. *Indian J Dermatol*. 2022;67(4):478.

(27) Saadati S, Sadeghi A, Mansour A, Yari Z, Poustchi H, Hedayati M, et al. Curcumin and inflammation in non-alcoholic fatty liver disease: a randomized, placebo controlled clinical trial. *BMC Gastroenterol*. 2019 Jul 25;19(1):133.

(28) Curcumin and inflammation in non-alcoholic fatty liver disease: a randomized, placebo controlled clinical trial. *BMC Gastroenterology*

(29) Veronesi U, De Palo G, Marubini E, Costa A, Formelli F, Mariani L, et al. Randomized trial of fenretinide to prevent second breast malignancy in women with early breast cancer. *J Natl Cancer Inst*. 1999 Nov 3;91(21):1847–56.

(30) Lovat PE, Ranalli M, Annichiarrico-Petruzzelli M, Bernassola F, Piacentini M, Malcolm AJ, et al. Effector mechanisms of fenretinide-induced apoptosis in neuroblastoma. *Exp Cell Res*. 2000 Oct 10;260(1):50–60.

(31) Garewal HS, Katz RV, Meyskens F, Pitcock J, Morse D, Friedman S, et al. β-Carotene Produces Sustained Remissions in Patients With Oral Leukoplakia: Results of a Multicenter Prospective Trial. *Arch Otolaryngol Head Neck Surg*. 1999 Dec 1;125(12):1305–10.

(32) Shalaby RH, Fathi Y, Elsaadany B. Efficacy and safety of beta carotenes in treatment of oral leukoplakia: systematic review and meta-analysis. *J Oral Res*. 2019 Dec 31;8(6):488–98

(33) Garewal H. Potential role of β-carotene in prevention of oral cancer. *Am J Clin Nutr*.

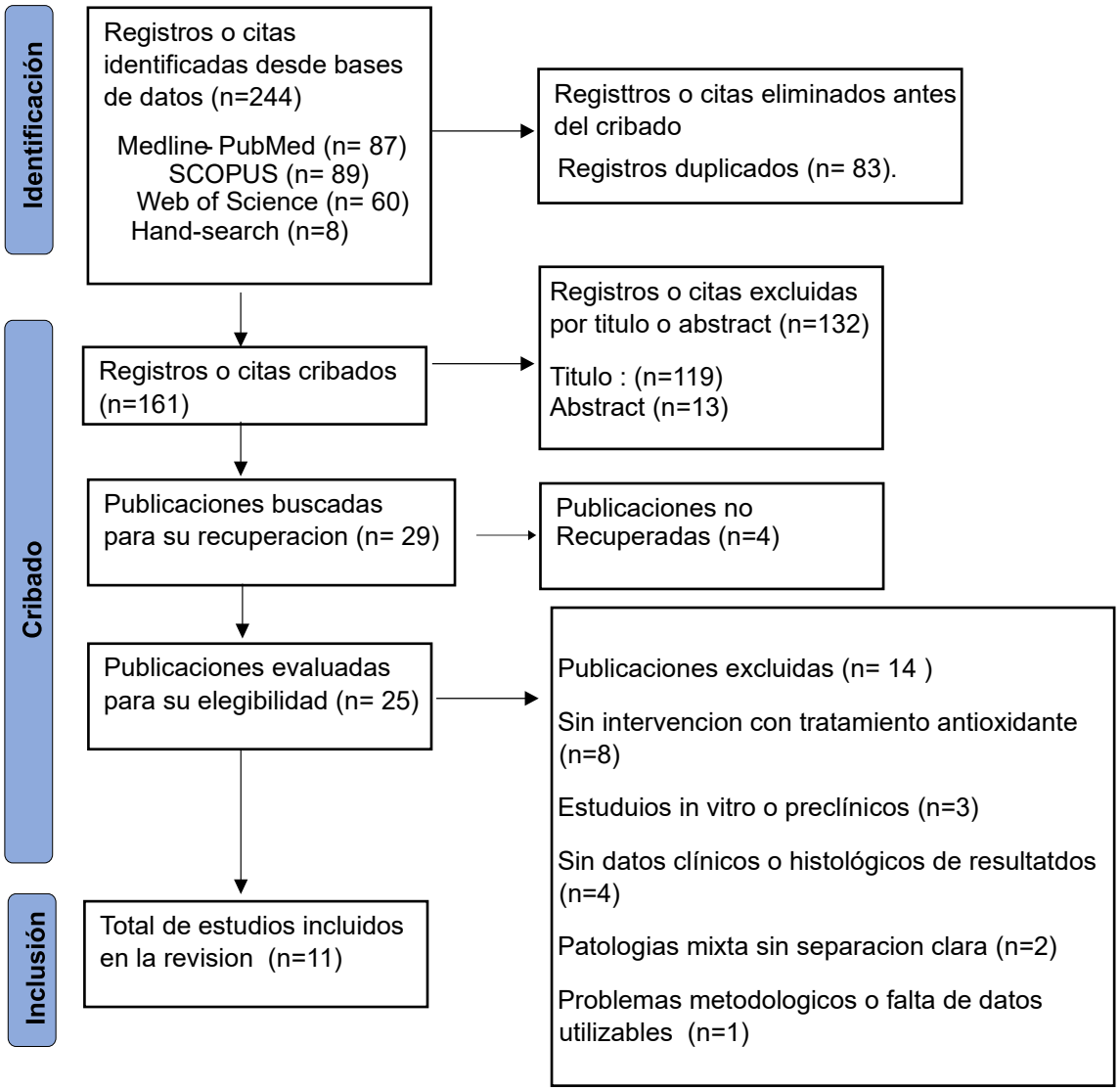

Fig. 1. Diagrama de flujo de búsqueda y proceso de selección de títulos durante la revisión sistemática

Tabla 1: *Caracteristicas de los estudios incluidos*

Variable / Caracteristica	Licopeno (n=3)	Curcuma (n=5)	Vitamina A (n=1)	Fenretinida (n=2)	Otros (n=2)	Total (n=11)
Tipo de estudio : ECA	3	2	0	1	0	6
Tipo de estudio : prospectivo no aleatorizado	0	1	1	0	2	4
Tipo de estudio: Fase II / Piloto	0	0	0	1	0	1
Tamano de muestra : (min–max)	41-300	25–300	6	35–170	10-50	6-300
Reduccion del tamano de la lesión	3	3	1	2	2	11
Mejoria histologica	2	2	1	1	2	4
Marcadores de estres oxidativo	1	2	0	0	1	3
Recidiva/ Nuevas lesiones	0	0	0	1	0	1

Tabla 2 : *Evaluación del riesgo de sesgo para los estudios aleatorizados según el Manual Cochrane.*

	Generar secuencia aleatorizada (sesgo seleccion)	Ocultacion de la asignacion sesgo selección	Cegamiento particpantes y personal (sesgo deteccion)	Cegamiento evaluacion de resultados (sesgo	Seguimientoy exclusiones (sesgo desercion)	Description selectiva (sesgo notificacion)	Otros sesgos
Fathimaet al. (2022) (7)	?	−	?	?	?	?	+
Manisha Singh et al. (2017) (13)	+	+	+	+	?	+	+
Patel et al. (2014) (12)	+	+	+	+	?	+	?
Chiesa et al. (2005) (14)	+	+	?	?	?	?	?
Kuriakose et al. (2016) (10)	+	+	+	+	+	+	+
Ahmad et al. (2023) (11)	+	?	−	?	?	+	?

***Tabla 3**: Evaluación del riesgo de sesgo de los estudios observacionales no aleatorizados utilizando la escala de Newcastle-Ottawa – estudios observacionales con grupo control no aleatorizado.*

	Definicion de los casos	Representatividad	Selecion de los controles	Definicion de los controles	Comparabilidad (factor mas importante)	Comparabilidad (cualquier otra variable)	Comprobacion de la exposicion	Mismo metedo para ambos grupos	Tasa de abandonos	Total
Jain et al. (2011) (16)	☆	☆	-	☆	-	-	☆	☆	☆	6

***Tabla 4**: Evaluación del riesgo de sesgo de los estudios observacionales no aleatorizados utilizando la escala de Newcastle-Ottawa – estudios de cohortes observacionales sin grupo control.*

	Representatividad cohorte	Seleccion cohorte no expuesta	Comprobacion exposicion	Demonstracion no presencia variable interes al inicio	Comparabilidad (factor mas importante)	Comparabilidad (otros factores)	Medicion resultados	Suficiente seguimiento	Tasa de abandonos	Total
Leunig et al. (2000) (17)	☆	-	☆	☆	-	-	☆	☆	☆	6
Kapoor et al. (2019) (8)	☆	-	☆	☆	-	-	☆	☆	☆	6
Rai et al (2010) (9)	☆	-	☆	☆	-	-	☆	☆	☆	6
Lippman et al. (2006) (15)	☆		☆	☆			☆	☆	☆	6

***Tabla 5** : Sintesis de los resultados de las variables*

Autor (año)	**Edad**	**N**	**Intervencion**	**Reduccion del tamaño**	**Mejora histologica (%)**	**Marcadores de estres oxidativo**	**Comentario**
Leunig et al. (2000) (17)	34-63	**N= 6** pacientes con leucoplasia	Retinil palmitato 300 × 103 IU/día durante la primera semana 1.5 × 106 IU/dia en la quinta semana Terapia de mantenimiento con 150 × 103 IU/day	No cuantificado	100% (sin displasia en biopsias de seguimiento)	No reportado	Tamaño muestra pequeño, respuesta histológica efectiva monitorizada por fluorescencia

Chiesa et al. (2005) (14)	>75	**N= 170** **GE** (n=84) **GC** (n=86)	**GE** :Fenretinida (4-HPR), 200 mg/d **GC** : sin tratamiento	Reduccion clinica pero no cuantificada	No reportado	No reportado	Seguimiento a largo plazo, prevencion significativa de recurrencia y carcinoma
Lippman et al. (2006) (15)	26-84	**N= 35**	Fenretinida 200 mg/dia durante 3 meses	34.3% respuesta parcial	No reportado	↑ Apoptosis (0.35% → 1.18%, *p* = 0.001)	9/12 respondedores recayeron en 9 meses; mejores resultados en respondedores previos a retinoides (70% vs. 20%, p = 0.015)
Rai et al (2010) (9)	17-50	**N= 100** **Groupo leucoplasia** (n=25)	Curcumina capsula 1g (900mg curcumina, 80mg desmetoxacurcumina, 20mg	**Groupo leucoplasia** : mejora significativa (p>0.05)	No reportado	Groupo leucoplasia: ↑ Vitamina C & E; ↓ MDA &	Efecto antioxidante y mejora clínica en lesiones orales precancerosas

		Groupo fibrosis submucosa oral (n=25) **Groupo liquen plano** (n=25) **Groupo sano** (n=25)	bisdemetoxicurcumina)			8-OHdG ($p < 0.05$)	
Jain et al. (2011) (16)	-	**N= 50** **Groupo A** control :10 **Groupo B** experimental : 40 (subdivido por grado de	**Groupo A and B :** capsula antioxidante Beta-caroteno (10mg) ; Vitamina E acetato I.P. (25IU); Viatmina C acetato I.P. (100mg);	74.19% mostró regresión parcial o completa a 1 año	No se observo	Grupo B (I, II, II): Suero : ↓ LPO, ↓ SOD, ↑ CAT	Sin reversión histológica; mejora enzimática

		displasia : BI, BII, BII)	Cobre(1mg) ; Manganeso (1.5mg) ; Zinc (7.5mg) and Selenio (150 mcg)			mucosa : ↓ LPO, ↓ SOD, ↑ CAT	
Patel et al. (2014) (12)	26-65	**N= 41** **Groupo A** (n=21) **Group B** (n=20)	**Groupo A:** Licopeno (3 mg), Vitamina E (200 I.U.), Selenio (100 mcg) x2/day **Groupo B:** capsula placebo x1/day	**Groupo A:** 9.85 ± 5.87 → 1.49 ± 3.08 cm² **Groupo B:** 7.58 ± 7.50 → 8.55 ± 7.69 cm²	**Groupo A:** 86% **Groupo B:** 20%	No reportado	Mejora significativa en el grupo tratadoa
Kuriakose et al. (2016) (10)	26-74	**N= 223** **GE** (n=111) **GC** (n=112)	**GE**: Curcumina 3.6 g/dia (oral) x 6 months **GC** : placebo	**GE** :67.5% **GC** :55.3%	No significativo (HR = 0.88, 95% CI: 0.45–1.71, p = 0.71)	No reportado	Respuesta clínica significativa y duradera con curcumina. Sin beneficio adicional

							con tratamiento prolongado
Manisha Singh et al (2017) (13)	56-65	**N= 60** **Groupo I** (n=30) **Groupo II** (n=30)	**Groupo I:** Gel de calendula officinalis(2mg/g al dia) x 1 mes **Groupo II:** licopeno gel/dia (2 mg/g al dia) x 1 mes	Diferencia media **Groupo I**: 2.0 ± 1.0 cm **Groupo II** : 1.57 ± 0.87 cm	No reportado	No reportado	Sin grupo placebo
Kapoor, S et al. (2019) (8)	-	**N= 60** **Groupo A** OL (n=20) **Groupo B** OLP (n=20)	**Groupo A,B,C :** Curcumina oral 400mg dia x 3 meses	**Groupo A :** diferencia media de tamaño 2.757 cm	No reportado	No reportado	Respuesta clínica significativa en el grupo de leucoplasia

		Groupo C OSMF (n=20)					
Fathima et al. (2022) (7)	20-60	**N= 20** **Groupo A** (n=10) **Groupo B** (n=10)	**Groupo A :** Bleomicina tópica + antioxidantes **Groupo B :** Gel de curcumina topica + antioxidantes	**Groupo A :** resolucion parcial o completa (p=0.01) **Group B:** not substantial	**Groupo B:** mejora histopatologica de la displasia (p=0.01)	No reportado	Bleomicina mostró mejor resolución clínica; la curcumina mejoró histológicamente la displasia
Ahmad et al. (2023) (11)	18-70	**N=300** **Grupo A**: (n=100)	**Groupo A :** Curcumina 500mg/day **Group B :** Licopeno 4mg/day	Reduccion clinica pero no cuantificada	No reportado	No reportado	La terapia combinada mostro eficacia superior

		Grupo B: (n=100) **Grupo C**: (n=100)	**Groupo C:** Curcumina 500mg + licopeno 4mg				

EFFICACY OF ANTIOXIDANT TREATMENT IN THE MANAGEMENT OF ORAL LEUKOPLAKIA : SYSTEMATIC REVIEW

Running title: Antioxidant treatment in oral leukoplakia

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Abstract

Introduction: Oral leukoplakia is the most common potentially malignant disorder of the oral cavity. Its development and potential malignant transformation have been associated with oxidative stress. In this context, antioxidant treatments have been proposed as a therapeutic strategy to counteract oxidative damage and prevent lesion progression.

Objectives: To evaluate the clinical and biochemical efficacy of antioxidant treatments in patients with oral leukoplakia, by analyzing lesion regression, size reduction, and improvements in oxidative stress-related biomarkers

Material and Methods : A systematic review was conducted following the PRISMA guidelines. An electronic search was performed in the PubMed, Scopus, and Web of Science databases for studies published between 2000 and 2025. Randomized controlled trials, controlled clinical studies, and observational studies assessing the use of topical or systemic antioxidants in patients with oral leukoplakia were included.

Results: A total of 11 studies involving 695 patients were included. The most studied antioxidants were lycopene, curcumin, vitamin E, and fenretinide. Most studies reported significant clinical improvements in terms of lesion size reduction or regression. On a biochemical level, a decrease in oxidative stress was observed, evidenced by reductions in LPO, MDA, and 8-OHdG levels, as well as increases in antioxidant enzymes such as catalase (CAT) and levels of vitamins C and E. Some studies also reported higher apoptosis rates in altered epithelium. Results concerning superoxide dismutase (SOD) were variable. The heterogeneity in study design, dosage, and treatment duration limited direct comparison between studies.

Discussion: Despite methodological limitations, antioxidant treatments appear to provide both clinical and biochemical benefits in the management of oral leukoplakia. Further high-quality, standardized clinical trials are needed to confirm their efficacy and role in preventing malignant transformation.

Keywords: Oral leukoplakia, Leukoplakia, Antioxidants, Antioxidant therapy, Antioxidant treatment

Introduction

Oral leukoplakia (OL) is the most common potentially malignant disorder of the oral mucosa. Clinically, it appears as a persistent white patch that cannot be rubbed off and cannot be characterized as any other definable lesion. OL is considered a premalignant condition due to its potential for malignant transformation into oral squamous cell carcinoma (OSCC), a type of oral cancer that accounts for a significant proportion of cancers worldwide. Studies estimate that approximately 5–15% of individuals with OL may experience malignant transformation, which underscores the importance of early detection and appropriate management of this disorder (1,2).

The pathogenesis of OL is closely associated with oxidative stress, which results from an imbalance between the production of reactive oxygen species (ROS) and the antioxidant defense mechanisms of the body. Oxidative stress contributes to DNA damage, lipid peroxidation, and dysregulation of signaling pathways involved in carcinogenesis. (3,4)

In recent years, there has been increasing interest in the therapeutic use of antioxidants, both natural and synthetic, for their ability to neutralize oxidative stress, modulate inflammatory responses, and potentially reverse or halt the progression of OL lesions.(1,5,6) Compounds such as lycopene, curcumin, retinoids, and vitamin A have shown promise in preclinical and clinical studies. However, the clinical efficacy of antioxidant treatments remains inconclusive due to variations in study design, dosages, and outcome measures

The aim of this systematic review was to evaluate the efficacy of antioxidant therapy in the management of oral leukoplakia, both in terms of clinical outcomes (lesion size reduction, regression, improvement in clinical appearance) and biochemical outcomes (modulation of oxidative stress or inflammatory biomarkers). This review follows PRISMA guidelines and includes clinical and randomized controlled studies published between 2000 and 2025

Material and Methods

A systematic review was conducted following the PRISMA (Preferred Reporting Items for Systematic reviews and Meta-Analyses) guidelines, which provide a structured framework for the transparent and reproducible reporting of systematic reviews and meta-analyses.

PICO question:

The objective of this review is to answer a question formulated using the PICO method:

- P (Population): Adult patients diagnosed with oral leukoplakia.
- I (Intervention): Antioxidant treatments (e.g., vitamin A, curcumin, among others) administered topically or systemically.
- C (Comparison): Placebo, control group (no treatment), or conventional treatment (active surveillance, surgery, corticosteroids). Comparison between different antioxidant treatments.
- O (Outcomes):
    - O1: Reduction in the size of oral leukoplakia lesions.
    - O2: Prevention of progression to more severe forms, such as dysplasia or cancer.
    - O3: Improvement in clinical symptoms (pain, inflammation, difficulty eating or speaking).
    - O4: Changes in biomarkers of oxidative stress (levels in saliva or blood).

Inclusion Criteria

The following criteria were used to determine the studies eligible for inclusion in this systematic review.

- Study Design: Randomized controlled trials (RCTs), controlled studies, and observational studies

- Studies published in English, Spanish, and French.
- Population : Patients with oral leukoplakia.
- Studies conducted on human subjects.
- Intervention : Antioxidant treatments (vitamin A, curcumin, green tea, vitamin C, vitamin E) for oral leukoplakia, administered either topically or systemically.
- Comparison : Placebo, control group (no treatment), or conventional treatments (active surveillance, surgery, corticosteroids).
- Studies comparing different antioxidant treatments.
- Outcome :
    - Reduction in lesion size,
    - Prevention of malignant transformation, including progression to dysplasia or cancer.
    - Improvement in clinical symptoms such as pain relief, reduction of inflammation, and functional benefits (easier eating or speaking).
    - Biological markers of oxidative stress reduction, such as changes in salivary or blood biomarkers.
- Publication date : Studies published between 2000 and 2025.

Exclusion Criteria

- Studies were excluded based on the following criteria:
- Studies involving patients with other oral conditions (e.g., oral cancer).
- Studies without antioxidant treatment.
- Studies lacking a comparison group or measurable outcomes related to lesion regression.
- Studies with low methodological quality, including those with significant risk of bias or insufficient data reporting.
- Studies with less than 4 patients. ($n<4$)
- Studies published in languages that are inaccessible for analysis (languages other than English, Spanish, or French).

Sources of information and search strategy:

The following databases were used to identify relevant studies: PubMed, Web of Science, and SCOPUS. (("oral leukoplakia"[All Fields] OR ("leucoplakias"[All Fields] OR "leukoplakia"[MeSH Terms] OR "leukoplakia"[All Fields] OR "leucoplakia"[All Fields] OR "leukoplakias"[All Fields])) AND ("antioxidant s"[All Fields] OR "antioxidants"[Pharmacological Action] OR "antioxidants"[MeSH Terms] OR "antioxidants"[All Fields] OR "antioxidant"[All Fields] OR "antioxidating"[All Fields] OR "antioxidation"[All Fields] OR "antioxidative"[All Fields] OR "antioxidatively"[All Fields] OR "antioxidatives"[All Fields] OR "antioxidizing"[All Fields] OR "antioxidant therapy"[All Fields] OR "antioxidant treatment"[All Fields])) AND (2000:2025[pdat]), ("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment") (Topic) and Review Article (Exclude – Document Types) and 1988 or 1989 or 1991 or 1992 or 1993 or 1994 or 1995 or 1997 or 1998 or 1999 (Exclude – Publication Years), TITLE-ABS-KEY (("oral leukoplakia" OR leukoplakia) AND (antioxidants OR "antioxidant therapy" OR "antioxidant treatment")) AND PUBYEAR > 1999 AND PUBYEAR < 2025 AND (LIMIT-TO (DOCTYPE , "ar")) AND (LIMIT-TO (LANGUAGE , "English"))

Study selection process:

The study selection was performed in two stages:

Stage 1: Titles and abstracts were screened for relevance. Studies not meeting the preliminary inclusion criteria (e.g., wrong population, no antioxidant therapy) were excluded. The reasons for exclusion were documented.

Stage 2: Full-text articles of the pre-selected studies were reviewed to confirm their eligibility. Additional exclusion criteria, such as methodological quality and language restrictions, were applied. Studies not meeting the criteria or whose full text was not accessible were excluded.

Data mining:

Relevant data, including study characteristics, patient sample sizes, and antioxidant dosage, were extracted from the selected studies. Data on clinical outcomes, such as lesion size reduction, and biochemical markers (e.g., oxidative stress markers, inflammatory cytokines) were also collected.

Quality assessment:

The Cochrane Risk of Bias Tool was used to assess the methodological quality and risk of bias in the included studies. This tool evaluates aspects such as randomization, blinding, and handling of missing data. Each study was rated as having a low, high, or unclear risk of bias.

Data synthesis:

Data were synthesized using a combination of qualitative and quantitative methods. For clinical outcomes, results from the different studies were summarized in a narrative format, and where applicable. The studies were grouped according to the type of antioxidant and their outcomes. A flowchart following the PRISMA statement was created to illustrate the study selection process, and a table was produced to summarize the characteristics of the included studies.

Results

Study Selection

The initial search process identified a total of 244 articles: from PubMed (n=87), Scopus (n=89), Web of Science (n=60) and hand-search (n=8). As a result, 25 articles were retained for full-text review. In total, 11 studies were included in the qualitative synthesis. Figure 1 presents the PRISMA flow diagram illustrating this selection process. Additionally, details of the excluded articles are provided in Table 1.

Analysis of the characteristics of the reviewed studies

Of the 11 studies included in this review, 5 assessed curcumin as the main antioxidant (7-11), 3 focused on lycopene (12,13,11), 2 on fenretinide (14,15). One studies also included combinations of antioxidants or enzyme-based antioxidant analysis (16), and 1 on high-dose vitamin A (17). A total of 365 patients were included: 140 received curcumin, 97 lycopene, 52 fenretinide, 36 vitamin A, and the remaining received other antioxidant therapies. Treatment durations ranged from 1 to 12 months. The primary outcomes assessed were clinical resolution, lesion size reduction, histopathological changes, and biochemical markers of oxidative stress (e.g., MDA, 8-OHdG). Some studies also examined long-term outcomes such as recurrence and malignant transformation.

Evaluation of methodological quality

Table 2 presents the quality assessment of the RCTs, the Cochrane Risk of Bias tool was used, while non-randomized and uncontrolled studies were assessed with the NOS (table 3 and 4). Most RCTs showed moderate risk of bias, mainly due to incomplete blinding or lack of detail on randomization. Observational studies generally presented moderate bias due to cohort selection and confounding factors. Studies without control groups had higher bias, especially due to lack of comparability and external influences. These results show variability in methodological quality across studies, though most support the effectiveness of antioxidant therapies in managing oral leukoplakia.

Synthesis of results

The analysis of the included studies revealed two main axes of evaluation regarding antioxidant treatments in oral leukoplakia: clinical improvement of lesions and modulation of oxidative stress or inflammatory biomarkers (Table 5).

Clinical efficacy of antioxidants

Most randomized controlled trials reported a significant reduction in lesion size and a notable improvement in clinical appearance following antioxidant treatment, compared to placebo or conventional therapies. For instance,

curcumin, used alone or in combination with other antioxidants, demonstrated marked effectiveness, with high rates of complete or partial lesion regression in the studies (7-11). Similarly, lycopene, proved effective, with clinical improvement observed within a few weeks of treatment, as reported in studies such as those by (12,13,11). The combination of multiple antioxidants (vitamin E, and selenium) also yielded favorable outcomes, suggesting a possible synergistic effect (16). Other compounds, like fenretinide (a vitamin A derivative), showed more variable efficacy, sometimes limited to recurrence prevention or specific subgroups of patients (10,14).

Impact on biomarkers

Biochemically, several studies evaluated the effect of antioxidants on oxidative stress markers (such as GPx, SOD, and reactive oxygen species). Overall, results indicate an increase in endogenous antioxidant levels and a reduction in pro-inflammatory markers after treatment. For example, (16) reported a significant disminution in LPO and SOD levels following administration of B-carotene and a mix of other antioxidant (Vitamin E, selenium…). In another study (9), observed augmentation of Vitamins C & E and disminution of MDA & 8-OHdG. Furthermore a study involving fenretinide (15) demonstrated some improvements in biomarkers and increase in apoptosis, although the effects were more modest, suggesting that antioxidant efficacy may depend on the specific molecule, treatment duration, or patient profile.

Discusion

Evaluation of antioxidants on the clinical parameters of leukoplakia

Antioxidants have shown beneficial effects in reducing clinical lesions in the treatment of oral leukoplakia. Among the compounds evaluated, lycopene, a natural carotenoid, has particularly stood out, with several studies reporting significant lesion reduction. Its effects are attributed to its ability to stabilize cell membranes, inhibit cell proliferation, and modulate genes related to oxidative stress and inflammation (11,12,13)). Additionally, lycopene has demonstrated

benefits in managing other oral and systemic conditions, such as submucous fibrosis and liver diseases (18-22).

Curcumin, derived from *Curcuma longa*, has also shown positive outcomes, especially in reducing lesion size and improving tissue texture. The beneficial effects are linked to its anti-inflammatory and epithelial restorative properties (7,8,10,11). This compound has also been studied for its potential in managing conditions like oral lichen planus and liver diseases, highlighting its therapeutic potential (23-28).

Fenretinide, a synthetic retinoid analogue, has shown particular promise in cases resistant to conventional therapies. It has been associated with lesion stabilization and reduction by modulating epithelial differentiation and inhibiting cell proliferation, suggesting a sustained clinical benefit (14,15). This compound has also demonstrated effects in breast cancer and other epithelial conditions (29,30).

Compounds such as β-carotene, vitamin C, vitamin E, and other mixed antioxidants have shown promising results in reducing oxidative stress, although studies on these formulations have not always assessed histological or long-term outcomes (16, 31-33).

In conclusion, antioxidants, particularly lycopene, curcumin, and fenretinide, show significant therapeutic potential in managing oral leukoplakia. However, more rigorous clinical trials are needed to confirm their long-term efficacy and safety.

Evaluation of Antioxidant effect on biochemical and oxidative stress markers in oral leukoplakia

Oral leukoplakia is a potentially malignant disorder where oxidative stress and chronic inflammation play a central role in its pathogenesis. Among the antioxidants evaluated, curcumin has shown significant effects on both clinical outcomes and biochemical markers. Studies found that curcumin supplementation increased serum levels of vitamins C and E while decreasing markers of oxidative stress such as MDA and 8-OHdG. These findings suggest that curcumin’s antioxidant properties can reduce oxidative damage and enhance

systemic antioxidant defenses, which is crucial in patients with potentially malignant disorders that often have reduced antioxidant capacity (9).

Lycopene also demonstrated significant effects in reducing oxidative stress and improving clinical outcomes. One study observed a reduction in MDA levels and an increase in antioxidant enzymes like SOD and GPx, indicating that lycopene enhances systemic antioxidant defenses (16). The dual action of lycopene in lowering oxidative stress markers and inflammation supports its role in promoting cellular regeneration and preventing malignant transformation in oral leukoplakia.

Fenretinide, particularly in retinoid-resistant cases, has shown promising effects by increasing apoptosis in oral epithelial cells. Though clinical response did not directly correlate with apoptosis levels, fenretinide's ability to induce apoptosis via mitochondrial pathways and ceramide accumulation supports its therapeutic potential in unresponsive oral leukoplakia (15). Additionally, its ability to reduce serum retinol levels further indicates its biological activity.

In conclusion, antioxidant such as curcumin, lycopene, and fenretinide, alongside vitamins, play crucial roles in reducing oxidative stress and inflammation in oral leukoplakia. These antioxidants may act synergistically to improve oral tissue health and prevent progression toward oral cancer, offering a promising therapeutic strategy for managing the disease.

Conclusion

Antioxidants such as curcumin, lycopene, fenretinide and other Vitamin have shown potential benefits in the treatment of oral leukoplakia. This suggests that antioxidants can help improve clinical outcomes by reducing oxidative stress and modulating inflammation in patients with oral leukoplakia.

Bibliography

(1) Chaturvedi AK, Udaltsova N, Engels EA, Katzel JA, Yanik EL, Katki HA, et al. Oral Leukoplakia and Risk of Progression to Oral Cancer: A Population-Based Cohort Study. *J Natl Cancer Inst*. 2020 Oct 1;112(10):1047–54.

(2) Sciubba JJ. Oral precancer and cancer: etiology, clinical presentation, diagnosis, and management. *Compend Contin Educ Dent*. 2000 Oct;21(10A):892–8, 900–2; quiz 903.

(3) Babiuch K, Bednarczyk A, Gawlik K, Pawlica-Gosiewska D, Kęsek B, Darczuk D, et al. Evaluation of enzymatic and non-enzymatic antioxidant status and biomarkers of oxidative stress in saliva of patients with oral squamous cell carcinoma and oral leukoplakia: a pilot study. *Acta Odontol Scand*. 2019 Aug;77(6):408–18.

(4) Sies H. Oxidative stress: a concept in redox biology and medicine. *Redox Biol*. 2015;4:180–3.

(5) Jomova K, Alomar SY, Alwasel SH, Nepovimova E, Kuca K, Valko M. Several lines of antioxidant defense against oxidative stress: antioxidant enzymes, nanomaterials with multiple enzyme-mimicking activities, and low-molecular-weight antioxidants. *Arch Toxicol*. 2024 May;98(5):1323–67.

(6) Rudrapal M, Khairnar SJ, Khan J, Dukhyil AB, Ansari MA, Alomary MN, et al. Dietary Polyphenols and Their Role in Oxidative Stress-Induced Human Diseases: Insights Into Protective Effects, Antioxidant Potentials and Mechanism(s) of Action. *Front Pharmacol*. 2022;13:806470.

(7) Fathima AIS, Manoharan GVGM. Comparison of Efficacy of Topical Application of Bleomycin with Adjuvant Antioxidants vs. Topical Curcumin Oral Gel with Adjuvant Antioxidants in the Treatment of Oral Leukoplakia. *J Indian Acad Oral Med Radiol*. 2022. Available from: https://www.scopus.com/inward/record.uri?eid=2-s2.0-85146477914

(8) Kapoor S, Arora P. Effect of curcumin in management of potentially malignant disorders - A comparative study. *J Indian Acad Oral Med Radiol*. 2019.

(9) Rai B, Kaur J, Jacobs R, Singh J. Possible action mechanism for curcumin in pre-cancerous lesions based on serum and salivary markers of oxidative stress. *J Oral Sci*. 2010 Jun;52(2):251–6.

(10) Kuriakose MA, Ramdas K, Dey B, Iyer S, Rajan G, Elango KK, et al. A Randomized Double-Blind Placebo-Controlled Phase IIB Trial of Curcumin in Oral Leukoplakia. *Cancer Prev Res*. 2016 Aug;9(8):683–91.

(11) Ahmad T, Khan I, Sultana A, Rizvi MMA. A comparative study to evaluate the therapeutic effects of nutraceuticals in oral leukoplakia: A randomized clinical trial. *Natl J Maxillofac Surg*. 2023;14(3):413–9.

(12) Patel JS, Umarji HR, Dhokar AA, Sapkal RB, Patel SG, Panda AK. Randomized controlled trial to evaluate the efficacy of oral lycopene in combination with vitamin E and selenium in the treatment of oral leukoplakia. *J Indian Acad Oral Med Radiol*. 2014 Dec;26(4):369.

(13) M S, A B. Comparison of effectiveness of *Calendula officinalis* extract gel with lycopene gel for treatment of tobacco-induced homogeneous leukoplakia: A randomized clinical trial. *Int J Pharm Investig*. 2017 Jun;7

(14) Chiesa F, Tradati N, Grigolato R, Boracchi P, Biganzoli E, Crose N, et al. Randomized trial of fenretinide (4-HPR) to prevent recurrences, new localizations and carcinomas in patients operated on for oral leukoplakia: long-term results. *Int J Cancer*. 2005 Jul 1;115(4):625–9.

(15) Lippman SM, Lee JJ, Martin JW, El-Naggar AK, Xu X, Shin DM, et al. Fenretinide activity in retinoid-resistant oral leukoplakia. *Clin Cancer Res*. 2006 May 15;12(10):3109–14.

(16) Jain RK, Singh GB, Singh AP, Goel RK, Aryya NC, Jha SK. Role of measurement of antioxidant enzymes in evaluation of antioxidant therapy in tobacco abusers with oral leukoplakia. *Indian J Otolaryngol Head Neck Surg*. 2011 Oct;63(4):336–42.

(17) Leunig A, Betz CS, Baumgartner R, Grevers G, Issing WJ. Initial experience in the treatment of oral leukoplakia with high-dose vitamin A and follow-up 5-aminolevulinic acid induced protoporphyrin IX fluorescence. Eur Arch Oto-Rhino-Laryngol Off J Eur Fed Oto-Rhino-Laryngol Soc EUFOS Affil Ger Soc Oto-Rhino-Laryngol - Head Neck Surg. 2000;257(6):327-31.

(18) Ahmad T, Khan I, Rizvi MM, Saalim M, Manzoor N, Sultana A. An overview of effect of lycopene and curcumin in oral leukoplakia and oral submucous fibrosis. *Natl J Maxillofac Surg*. 2021;12(3):316–2

(19) Guo J, Xie H, Wu H, Liang M. Efficacy of Lycopene in the Treatment of Oral Submucous Fibrosis: A Meta-analysis of Randomized Controlled Trials. *J Evid Based Dent Pract*. 2020 Dec;20(4):101471.

(20) Al-Maweri SA, Halboub E, Al-Qadhi G, Al-Wesabi M, Al-Sharani HM, Parveen S, et al. Efficacy of lycopene for management of oral potentially malignant disorders: A systematic review and meta-analysis. *Oral Surg Oral Med Oral Pathol Oral Radiol*. 2023 Jan;135(1):79–95.

(21) Bao J, Chen C, Yan J, Wen Y, Bian J, Xu M, et al. Antioxidant therapy for patients with oral lichen planus: A systematic review and meta-analysis. *Front Pharmacol*. 2022 Nov 10;13. Available from: https://www.frontiersin.org/articles/10.3389/fphar.2022.1030893/full

(22) Saawarn N, Shashikanth MC, Saawarn S, Jirge V, Chaitanya NC, Pinakapani R. Lycopene in the management of oral lichen planus: a placebo-controlled study. *Indian J Dent Res*. 2011;22(5):639–43.

(23) Akbik D, Ghadiri M, Chrzanowski W, Rohanizadeh R. Curcumin as a wound healing agent. *Life Sci*. 2014 Oct 22;116(1):1–7.

(24) Tejada S, Manayi A, Daglia M, Nabavi SF, Sureda A, Hajheydari Z, et al. Wound Healing Effects of Curcumin: A Short Review. *Curr Pharm Biotechnol*. 2016;17(11):1002–7.

(25) Bahrami A, Majeed M, Sahebkar A. Curcumin: a potent agent to reverse epithelial-to-mesenchymal transition. *Cell Oncol*. 2019 Aug;42(4):405–21.

(26) Khaitan T, Vishal, Kabiraj A, Sinha DK, Ranjan R, Singh R. *Curcuma longa* in the Treatment of Symptomatic Oral Lichen Planus: A non-randomized controlled trial. *Indian J Dermatol*. 2022;67(4):478.

(27) Saadati S, Sadeghi A, Mansour A, Yari Z, Poustchi H, Hedayati M, et al. Curcumin and inflammation in non-alcoholic fatty liver disease: a randomized, placebo controlled clinical trial. *BMC Gastroenterol*. 2019 Jul 25;19(1):133.

(28) Curcumin and inflammation in non-alcoholic fatty liver disease: a randomized, placebo controlled clinical trial. *BMC Gastroenterology*

(29) Veronesi U, De Palo G, Marubini E, Costa A, Formelli F, Mariani L, et al. Randomized trial of fenretinide to prevent second breast malignancy

in women with early breast cancer. *J Natl Cancer Inst*. 1999 Nov 3;91(21):1847–56.

(30) Lovat PE, Ranalli M, Annichiarrico-Petruzzelli M, Bernassola F, Piacentini M, Malcolm AJ, et al. Effector mechanisms of fenretinide-induced apoptosis in neuroblastoma. *Exp Cell Res*. 2000 Oct 10;260(1):50–60.

(31) Garewal HS, Katz RV, Meyskens F, Pitcock J, Morse D, Friedman S, et al. β-Carotene Produces Sustained Remissions in Patients With Oral Leukoplakia: Results of a Multicenter Prospective Trial. *Arch Otolaryngol Head Neck Surg*. 1999 Dec 1;125(12):1305–10.

(32) Shalaby RH, Fathi Y, Elsaadany B. Efficacy and safety of beta carotenes in treatment of oral leukoplakia: systematic review and meta-analysis. *J Oral Res*. 2019 Dec 31;8(6):488–98.

(33) Garewal H. Potential role of β-carotene in prevention of oral cancer. *Am J Clin Nutr*.

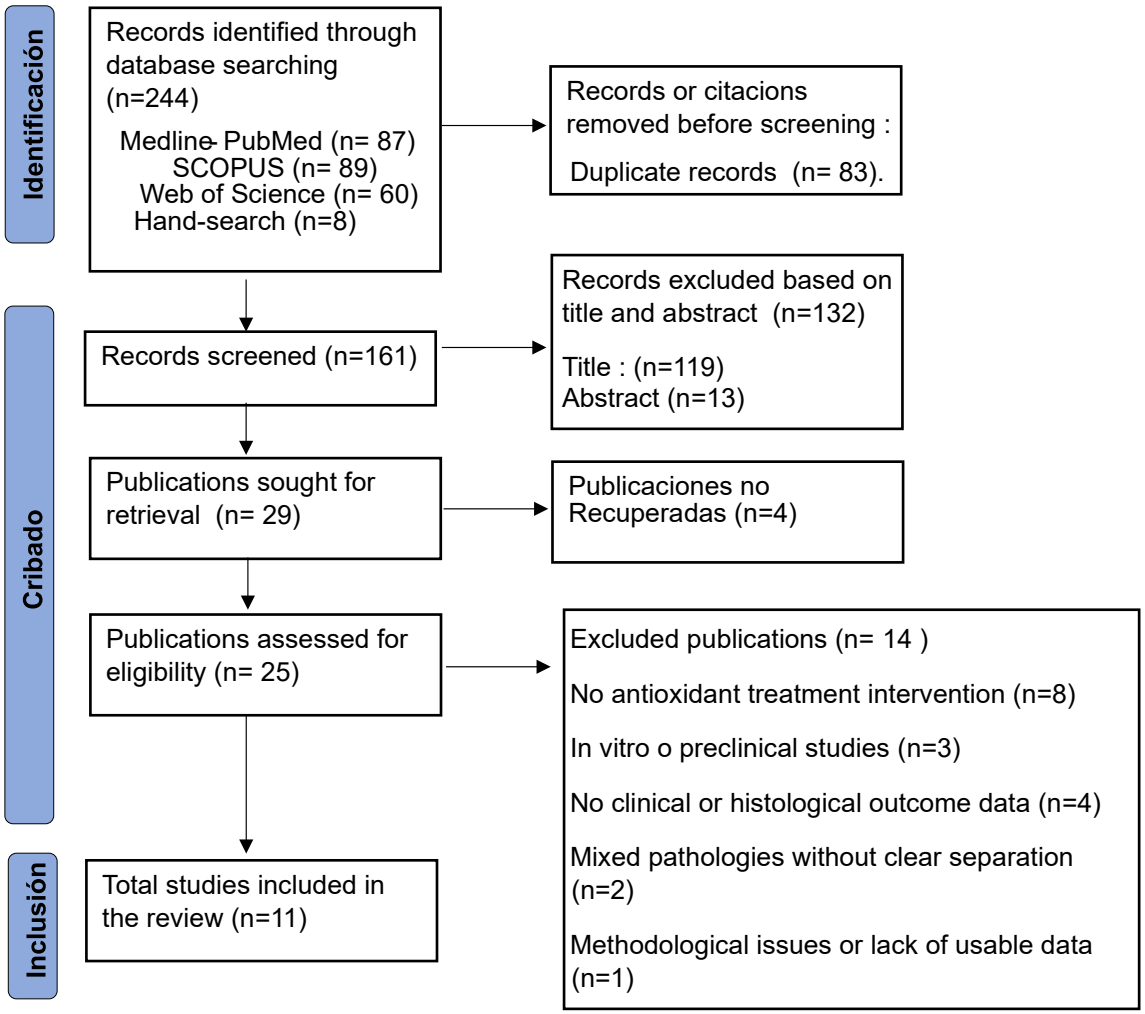

***Fig. 1**. Flow diagram of the search and selection process of titles during the systematic review.*

***Table 1:** Characteristics of the included studies*

Variable / Characteristic	Lycopene (n=3)	Curcumin (n=5)	Vitamin A (n=1)	Fenretinide (n=2)	Others (n=2)	Total (n=11)
Type of study: RCT	3	2	0	1	0	6
Type of study: Non-	0	1	1	0	2	4

Variable / Characteristic	Lycopene (n=3)	Curcumin (n=5)	Vitamin A (n=1)	Fenretinide (n=2)	Others (n=2)	Total (n=11)
randomized prospective						
Type of study: Phase II / Pilot	0	0	0	1	0	1
Sample size (min–max)	41-300	25–300	6	35–170	10-50	6-300
Lesion size reduction	3	3	1	2	2	11
Histological improvement	2	2	1	1	2	4
Oxidative stress markers	1	2	0	0	1	3
Adverse events / Safety	0	0	0	1	0	1
Recurrence / New lesions	0	0	0	1	0	1

Table 2 *Risk of Bias Assessment for Randomized Studies According to the Cochrane Handbook.*

	Generation of randomized sequence (election bias)	Allocation concealment (selection bias)	Binding of participants and personnel (detection bias)	Blinding of outcome assessment (detection bias)	Follow-up and exclusions (attrition bias)	Selective reporting (reporting bias)	Other bias
Fathimaet al. (2022) (7)	?	−	?	?	?	?	+
Manisha Singh et al. (2017) (13)	+	+	+	+	?	+	+
Patel et al. (2014) (12)	+	+	+	+	?	+	?
Chiesa et al. (2005) (14)	+	+	?	?	?	?	?
Kuriakose et al. (2016) (10)	+	+	+	+	+	+	+
Ahmad et al. (2023) (11)	+	?	−	?	?	+	?

Table 3 *: Risk of bias assessment of non-randomized observational studies using the Newcastle-Ottawa scale – observational studies with a non-randomized control group*

	Case definition adequate	Representtiveness of cases	Selection of controls	Definition of controls	Comparability (main factor)	Comparability (other factors)	Ascertainment of exposure	Same method of ascertainment cases and	Drop-out rate	Total
Jain et al. (2011) (16)	☆	☆	-	☆	-	-	☆	☆	☆	6

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Table 4*: Risk of bias assessment of non-randomized observational studies using the Newcastle-Ottawa scale – observational cohort studies without a control group.*

	Represetativeness of exposed cohort	Selection of the non-exposed cohort	Ascertainment of exposure	Demonstrationthat outcome of interest not present at start	Comparability of cohort based on design or analysis	Comparability of cohort based on any additional factors	Assessment of outcome	Sufficient follow-up	Drop-out rate	Total
Leunig et al. (2000) (17)	☆	-	☆	☆	-	-	☆	☆	☆	6
Kapoor, S et al. (2019) (8)	☆	-	☆	☆	-	-	☆	☆	☆	6
Rai, Balwant et al (2010) (9)	☆	-	☆	☆	-	-	☆	☆	☆	6
Lippman et al. (2006) (15)	☆		☆	☆			☆	☆	☆	6

Table 5 *: Synthesis of the Results of the Variables*

Author (year)	**Age**	**N**	**Intervention**	**Lesion size reduction**	**Histological improvement (%)**	**Oxidative stress markers**	**Comment**
Leunig et al. (2000) (17)	34-63	**N= 6** patients with leukoplakia	Retinyl palmitate 300 × 103 IU/day for the first week 1.5 × 106 IU/day in the fifth week maintenance therapy with 150 × 103 IU/day	Not quantified	100% (no dysplasia in follow-up biopsies)	Not reported	Small sample size, effective histological repsonse monitored via fluorescence
Chiesa et al. (2005) (14)	>75	**N= 170** **GE** (n=84) **GC** (n=86)	**GE** :Fenretinide (4-HPR), 200 mg/d	Clinical reduction but Not quantified	Not reported	Not reported	Long term follow-up, significant prevention of

			GC : No treatment				recurrence and carcinoma
Lippman et al. (2006) (15)	26-84	**N= 35**	Fenretinide 200 mg/day for 3 months	34.3% partial response	Not reported	↑ Apoptosis (0.35% → 1.18%, *p* = 0.001)	9/12 responders relapsed within 9 months; better outcomes in prior responders to retinoids (70% vs. 20%, *p* = 0.015)
Rai, Balwant et al (2010) (9)	17-50	**N= 100** **Group leukoplakia** (n=25) **Group oral submucous**	Curcumin 1g caplets (900mg curcumin, 80mg desmethoxycurcumin, and 20mg bisdemethoxycurcumin)	**Group leukoplakia** : Significant improvement (p>0.05)	Not reported	Group leukoplakia: ↑ Vitamins C & E; ↓ MDA & 8-OHdG (p < 0.05)	Demonstrated antioxidant effect and clinical improvement in precancerous oral lesions

		fibrosis (n=25) **Group lichen planus** (n=25) **Group healthy** (n=25)					
Jain et al. (2011) (16)	-	**N= 50** **Group A** control :10 **Group B** experimental : 40 (subdivided by dysplasia	**Group A and B :** Antioxidants capsule Beta-carotene (10mg) ; Vitamin E acetate I.P. (25IU); Viatmin C acetate I.P. (100mg); Copper(1mg) ; Manganese	74.19% showed partial or complete regression at 1 yeas	None observed	Grupo B (I, II, II): Serum : ↓ LPO, ↓ SOD, ↑ CAT mucosa : ↓ LPO, ↓ SOD, ↑ CAT	No histological reversal ; enzymatic improvement

		grade : BI, BII, BII)	(1.5mg) ; Zinc (7.5mg) and Selenium (150 mcg)				
Patel et al. (2014) (12)	26-65	**N= 41** **Group A** (n=21) **Group B** (n=20)	**Group A:** Lycopene (3 mg), Vitamin E (200 I.U.), Selenium (100 mcg) x2/day **Group B:** placebo capsule x1/day	**Group A:** 9.85 ± 5.87 → 1.49 ± 3.08 cm² **Group B:** 7.58 ± 7.50 → 8.55 ± 7.69 cm²	**Group A:** 86% **Group B:** 20%	Not reported	Significant improvement in treated group
Kuriakose et al. (2016) (10)	26-74	**N= 223** **GE** (n=111) **GC** (n=112)	**GE**: Curcumin 3.6 g/day (oral) x 6 months **GC** : placebo	**GE** :67.5% **GC** :55.3%	Not significant (HR = 0.88, 95% CI: 0.45–1.71, p = 0.71)	Not reported	Significant and durable clinical repsonse with curcumin. No added benefit with prolonged treatment

Manisha Singh et al (2017) (13)	56-65	**N= 60** **Group I** (n=30) **Group II** (n=30)	**Group I:** Caléndula officinalis gel (2mg/g per day) x 1 month **Group II:** lycopene gel/day (2 mg/g per day) x 1 month	Mean difference: **Group I**: 2.0 ± 1.0 cm **Group II** : 1.57 ± 0.87 cm	Not reported	Not reported	No placebo arm
Kapoor, S et al. (2019) (8)	-	**N= 60** **Group A** OL (n=20) **Group B** OLP (n=20)	**Group A,B,C :** Oral Curcumin 400mg daily x 3 months	**Group A :** Mean size difference 2. 757 cm	Not reported	Not reported	Significant clinical response in leukoplakia group

		Group C OSMF (n=20)					
Fathima et al. (2022) (7)	20-60	**N= 20** **Group A** (n=10) **Group B** (n=10)	**Group A :** Topical Bleomycin + antioxidants **Group B :** Topical Curcumin gel + antioxidants	**Group A :** partial to complete resolution (p=0.01) **Group B:** not substantial	**Group B:** histopathological improvement of dysplasia (p=0.01)	Not reported	Bleomycin showed superior clinical resolution; curcumin improved dysplasia histologically
Ahmad et al. (2023) (11)	18-70	**N=300** **Grup A**: (n=100)	**Group A :** Curcumin 500mg/day **Group B :** Lycopene 4mg/day	Clinical reduction but Not quantified	Not reported	Not reported	Combination therapy showed superior efficacy

		Grup B: (n=100) **Grup C**: (n=100)	**Group C:** Curcumin 500mg + lycopne 4mg				