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Cold Plasma Treatment for *Candida* Biofilm on Resin Base Dentures: A Systematic Review

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Abstract

Introduction: *Candida* biofilm on resin base dentures is a common problem among denture wearers, leading to denture stomatitis and associated complications. Cold plasma treatment has been proposed as a novel and promising approach to eradicate *Candida* biofilm. This systematic review aims to evaluate the effectiveness of cold plasma treatment for *Candida* biofilm on resin base dentures.

Methods: This systematic review study was conducted following PRISMA guidelines. The main objective was to investigate whether cold plasma treatment could reduce the number of *Candida* cells in dentures compared to other disinfection methods or controls. In September 2024, an electronic search was performed without any limitation on the publication start date in PubMed, Web of Science, Google Scholar, Embase, and Scopus databases. English in vitro studies, focusing on acrylic denture bases using cold plasma treatment as the intervention, were included. The selected articles were assessed using the QUIN risk-of-bias tool for in vitro studies conducted in dentistry.

Results: Initially, 259 papers were identified, and 164 remained after removing duplicates. Following the screening of titles and abstracts, 21 papers remained. Ten articles were not related to dentures, and 11 studies were included. All of these articles demonstrated a medium risk of bias and were case-control in vitro studies. The evidence currently available suggests that cold plasma exhibits antimicrobial efficacy against denture candidiasis; however, its application is not without limitations.

Conclusion: Based on the findings of in vitro studies, cold plasma shows promise as an effective tool for disinfecting dentures. Notably, significant reductions in the *Candida* cell count can be achieved within a reasonable treatment duration, although the existing data present variable results.

Keywords: Candidiasis; Plasma gases; Anti-infective agents; Denture; Complete.

Introduction

Oral hygiene plays a major role in the care and longevity of complete dentures. There are various methods recommended for denture cleaning, including brushing, removing them at night, and using chemical solutions.[1] Regular cleaning prevents the accumulation of microorganisms, including *Candida* colonies, on the denture surface.[2] However, acrylic dentures have porous surfaces that provide an ideal environment for microorganisms and *Candida* colonies to thrive if not properly cared for on a daily basis. Failure to maintain proper hygiene can lead to rapid multiplication of these microorganisms, resulting in complications such as halitosis.[3]

Fungal infections caused by poor hygiene and improper dental prostheses are common in individuals wearing dentures. *Candida* species, particularly *Candida albicans*, are naturally present in the oral flora of most individuals.[4] Overnight use of dentures is a significant risk factor for fungal infections, as it creates an environment conducive to *Candida* colonization.[5] Candidiasis in dental prostheses is typically painless, but it presents with inflammation and erythema of the oral mucosa.[6] Treatment of candidiasis involves cleaning the denture by regularly brushing it and removing it from the mouth at night, followed by immersion in special cleaning solutions. It is important to note that due to the porosity of acrylic dentures, brushing and cleaning solutions alone may not effectively remove all microorganisms.[7]

Various techniques including disinfectants such as chlorhexidine, sodium hypochlorite, alkaline glutaraldehyde, povidone iodine, and microwave disinfection have been suggested for disinfecting dentures. Nevertheless, these methods can lead to complications such as discoloration, irritation of soft tissues, and compromised mechanical properties of the prosthesis.[8]

Cold plasma is a novel method for treating oral pathogens and has been investigated in multiple studies as an effective means of eliminating *Candida* infections on the tissue surface of removable dentures.[9] Cold plasma,

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also known as non-thermal plasma, is a plasma that is not in thermodynamic equilibrium and is considered a cutting-edge technology in the field of disinfection.[10,11]

A study in this area demonstrated that cold plasma significantly reduces the adhesion of fungi, including *C. albicans*, to the tissue surface of acrylic dentures.[12] However, another study showed that cold plasma exhibited weaker effectiveness in killing fungi compared to disinfectant sprays.[13] Additionally, it has been reported that exposure of the tissue surface of dentures to cold plasma enhances the response to antifungal drugs in cases of *Candida* infection.[14] Considering the number of studies conducted in this field, the objective of this systematic review was to evaluate the impact of cold plasma on acrylic dentures contaminated with *Candida*.

Methods

This systematic review study was conducted following PRISMA guidelines.[15] The study has been registered in the PROSPERO (CRD42024584940).

Research Question

The main research question was formulated using the PICO (Population, Intervention, Comparison, Outcome) approach. In this case, the population (P) was dentures with *Candida* biofilms, the intervention (I) was cold plasma treatment, the comparison (C) was other methods of disinfection or control, and the outcome (O) was the change in the number of *Candida* cells. The aim of this study was to investigate whether cold plasma treatment could effectively reduce the amount of *Candida* cells in dentures compared to other disinfection methods or control groups.

Eligibility Criteria

The inclusion criteria for this study were as follows:

1. In vitro laboratory studies
2. Studies focusing on acrylic denture bases
3. Studies using cold plasma treatment as the intervention
4. English articles

Exclusion Criteria

1. reprints
2. Letters to the editor and correspondences
3. Quasi-experimental studies
4. Commentaries and conference abstracts
5. Articles without full-text availability

Search Strategy

In September 2024, an electronic search of five databases (PubMed, Web of Science, Google Scholar, Embase, and Scopus) was conducted without any restriction on the publication start date. A combination of free and Medical Subject Heading (MeSH) terms was used to collect data. The keywords used included "acrylic denture," "poly methyl methacrylate," "resin denture," "denture base," "plasma gas," "non-thermal plasma," "cold plasma," "atmospheric plasma," "air plasma," "low-temperature plasma," and "cold atmospheric pressure plasma." The exact search strategy is available in Supplementary file 1. The reference lists of included articles were also reviewed to find additional relevant studies. Duplicate references were removed and managed using EndNote Basic software.

Study Selection

Citations were organized and screened using EndNote X20 software. Duplicates were removed, and titles were reviewed to exclude irrelevant citations. The next step involved screening abstracts. The remaining articles were assessed based on the inclusion and exclusion criteria by examining the full text. Two researchers (KK and EN) independently conducted the screening process and consulted a third reviewer (HE) in case of disagreements.

Assessment of the Risk of Bias

The QUIN tool, designed for evaluating in vitro studies of dentistry, was used by two independent reviewers (KK and EN) to appraise the selected articles and assess the risk of bias.[16] Discrepancies were resolved through discussions with a third reviewer (HE). The QUIN tool consists of 12 criteria with scoring and grading options to evaluate the quality of in vitro studies. Each criterion was scored as adequately specified (2 points), inadequately specified (1 point), not specified (0 points), or not applicable. The scores were summed to obtain a total score, which was then used to grade the studies as high ($<50\%$), medium (50% to 70%), or low ($>70\%$) risk of bias. Studies with a high risk of bias, including those without a control group, were excluded from the study.

Data Extraction Process

A customized form in Microsoft Excel was used to extract and organize data from the included studies. The form included details such as study ID (first author's last name and publication date), country of origin, samples, sample size, *Candida* type, sample groups, device, plasma mode, distance, results, and risk of bias.

Results

Search Results

Initially, 259 papers were identified, and after removing duplicates, 164 studies remained for assessment. Following the screening of titles and abstracts, 143 papers were excluded, leaving 21 full-text articles for review by two independent reviewers (Figure 1). According to the predefined inclusion and exclusion criteria, 10 articles were found to be unrelated to dentures, resulting in a final inclusion of 11 studies.

Assessment of Risk of Bias

All of the included articles had a low risk of bias, with final scores of either 80 to 90. The results of the bias assessment are presented in Table 1.

Characteristics of the Studies

All the included studies were case-control in vitro studies. The samples used in all articles were disk-shaped resins, except for one study that utilized dentures.[13] *Candida albicans* was studied in all of the articles, except for one study that focused on *Candida glabrata*.[19] The incubation conditions, device used, plasma mode, and distance between the cold plasma and samples are presented in Table 2.

Results of Cold Plasma Treatment

The studies reviewed the efficacy of cold plasma treatment in reducing *Candida* biofilm on resin-based dentures. Ten case-control in vitro studies were included in the analysis. Of these, *C. albicans* was studied in nine studies, while one study focused on *C. glabrata*. Overall, 60% of the studies found that cold plasma treatment was successful in reducing the amount of C. albicans biofilm on denture surfaces. However, one study reported that SEPTOPRINT® was more effective than cold plasma treatment.[13] Another study found no significant difference in *C. albicans* adherence between the experimental (treated with cold plasma) and the control group, regardless of whether saliva was present or not.[18] Regarding *C. glabrata*, one study showed that cold plasma treatment in the absence of saliva resulted in lower adherence compared to the control group. However, after exposure to saliva, there was no significant difference in *C. glabrata* adherence between the experimental and control groups.[19] It is worth noting that one study reported lower amounts of *C. albicans* in the group receiving no treatment compared to the experimental groups.[17] In addition to CFUs, the studies also assessed clinical parameters such as denture stomatitis severity, microbial adhesion, and surface roughness.

Discussion

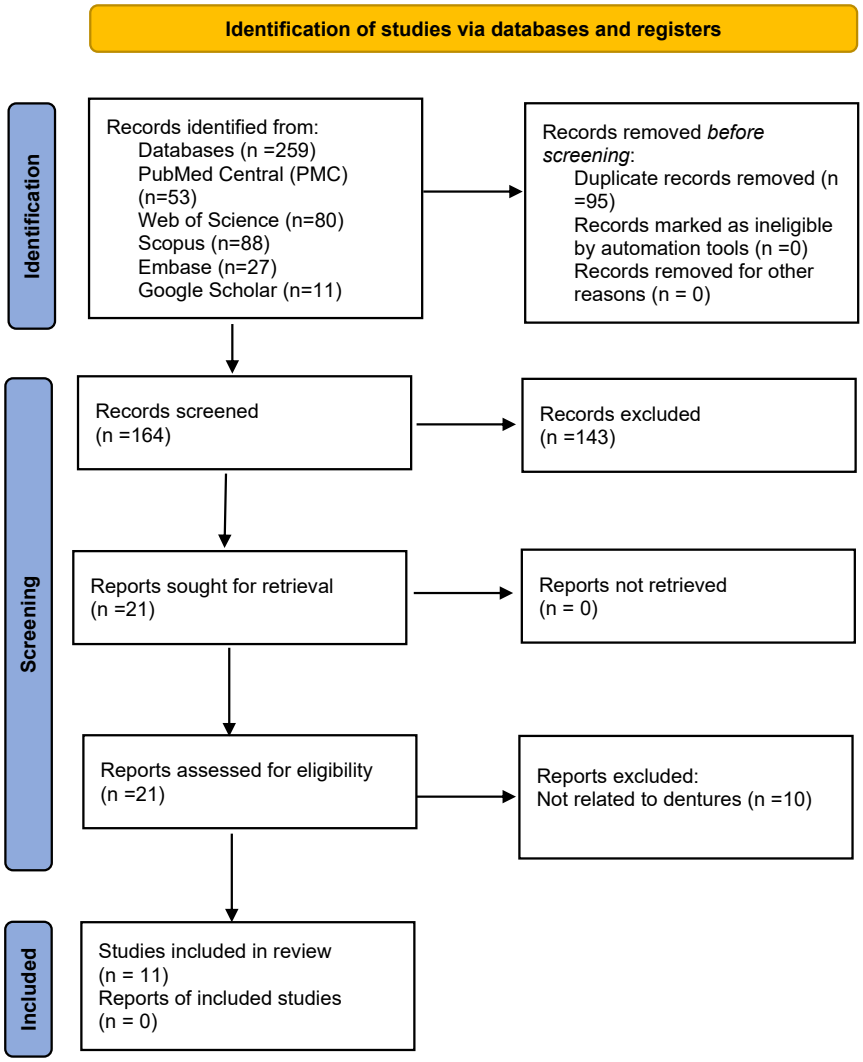

Figure 1. The PRISMA Flowchart of the Selection Process of Systematic Review

Table 1. Risk of Bias Assessment

First Author	Clearly Stated Aims/ Objectives	Detailed Explanation of Sample Size Calculation	Detailed Explanation of The Sampling Technique	Details of the Comparison Group	A Detailed Explanation of the Methodology	Operator Details	Randomization	Method Of Measurement of Outcome	Outcome Assessor Details	Blinding	Statistical Analysis	Presentation of Results	Final Score	Risk of Bias
Asnaashari[13]	2	0	NA	2	2	2	1	2	2	NA	2	2	85	Low
Nagay[10]	2	0	NA	2	2	2	1	2	2	NA	2	2	85	Low
Yildirim[17]	2	0	NA	2	2	2	1	2	2	NA	2	2	85	Low
Zamperini[18]	2	0	NA	2	2	2	0	2	2	NA	2	2	80	Low
Zamperini,[19] 2013	2	0	NA	2	2	2	0	2	2	NA	2	2	80	Low
Zamperini,[20] 2010	2	0	NA	2	2	2	0	2	2	NA	2	2	80	Low
Matthes [21]	2	0	NA	2	2	2	0	2	2	NA	2	2	80	Low
Pan [12]	2	0	NA	2	2	2	0	2	2	NA	2	2	80	Low
Qian [22]	2	0	NA	2	2	2	1	2	2	NA	2	2	85	Low
Wang,[14] 2013	2	0	NA	2	2	2	0	2	2	NA	2	2	80	Low
Wang,[23] 2016	2	0	NA	2	2	2	2	2	2	NA	2	2	90	Low

Adequately Specified (Score = 2), Inadequately Specified (Score = 1), Not Specified (Score = 0), NA: Not applicable.
(> 70% = low risk of bias, 50% to 70% = medium risk of bias, and < 50% = high risk of bias) .
Final score = total score × 100/ 2 × number of criteria applicable.

Denture stomatitis is a common oral disease in the elderly, primarily caused by host-related factors and an imbalance in oral microflora resulting from wearing dentures.[24] The key focus in treating denture stomatitis is to eliminate the pathogens that have colonized on the dentures.[20] Therefore, it is essential to use a safe disinfection protocol that does not harm the integrity of acrylic-based dentures.

Candida albicans is a prominent fungal species associated with biofilm formation in denture wearers. Treatment failures often occur due to resistance to antifungal agents, and patient compliance. Therefore, it is important to investigate more effective strategies for controlling biofilm formation and providing better treatment for denture stomatitis patients.[14]

Cold plasma is produced through a high-voltage discharge within a vacuum chamber, producing reactive oxygen and nitrogen species.[25] These reactive species interact with microbial cells, damaging their DNA and ultimately leading to inactivation and death.[26] The low-temperature plasma jet can be precisely controlled to maintain low electrical power, ensuring minimal heat generation and making it suitable for delicate surfaces like dentures.

Surface reactions resulting from cold plasma treatment are influenced by various factors, such as the characteristics of the substrate, the plasma generator, the type of working gas, reaction settings (including power, pressure, working distance, and duration), as well as the design of the reactor.[27] After plasma treatment, changes in the chemical microenvironment of the surface are observed, such as a decrease in the C/O proportion and the formation of new chemical bonds, such as C-OH and -CF3 groups. The decrease in the C/O ratio suggests that the resin surface undergoes oxidation, resulting in the formation of new C-OH bonds. The presence of C-OH bonds has been shown to improve hydrophilicity, which can help reduce the adhesion of C. albicans to denture base materials. This suggests that the formation of new C-OH bonds through cold plasma treatment may be a positive alternative for reducing denture stomatitis.[12]

The use of cold atmospheric plasma jets has shown effectiveness in inactivating microorganisms, including *Candida*, in liquid solutions or as biofilms.[28] The exact mechanisms by which plasma inactivates the microorganisms are still not fully understood.[29] Atmospheric plasma consists of a complex mixture of charged particles, reactive atoms, and molecules (such as atomic oxygen, ozone, hydroxyl groups, and nitrogen oxides), and ultraviolet (UV) radiation. Each of these can lead to bacterial deactivation through various means, including electrostatic disruption, physical membrane etching, interference with transport within cells, DNA breakage, and damage to intracellular proteins. While UV radiation is not the major inactivation factor in cold atmospheric plasma jets, reactive oxygen species are believed to play a key role in microbial inactivation.[30]

Asnaashari et al stated that all disinfection methods resulted in a significant decrease in microbial colonies; however, ASEPTOPRINT® spray, an alcohol-based impression disinfectant containing ethanol, 1-propanol, and quaternary ammonium compounds, demonstrated greater antimicrobial effectiveness than cold plasma.[13] Wang et al found that the biofilm could be completely eliminated within eight minutes in all plasma groups.

Table 2. The Characteristics of the Included Studies

First Author, Year	Objective	Methods & Material		Groups	Result
		Specimens	Type and Settings of Cold Plasma		
Asnaashari,[13] 2019	Comparison of the antimicrobial effect of cold plasma and ASEPTOPRINT® on disinfecting dentures	30 maxillary complete dentures as *C. albicans* source.	The pulsed plasma reactor was used. Cold plasma was generated by high voltage discharge in a vacuum chamber with a dielectric barrier discharge mode.	Control (ASEPTOPRINT® spray) Cold plasma for 30 seconds Cold plasma for 60 seconds	SEPTOPRINT® spray showed more antimicrobial effects compared to cold plasma.
Nagay,[10] 2020	The effect of non-thermal plasma (SF6- and HMDSO-based) on soft liner on its surface and physicochemical properties, and fungal colonization	75 disk-shaped resinous liner material as *C. albicans* source.	SF6: gas: SF6 (100%); Base pressure 2.2 (Pa) Time: 10 minutes HMDSO: gas: O2 (100%) Time: 10 minutes HMDSO (65.2 %) + Ar (34.8 %) Time: 1 minute 50 seconds Base pressure 2.6 Pa	No surface treatment (control) SF 6-based HMDSO-based	higher C. albicans counts for control compared with SF6 ($P=0.025$) and HMDSO ($P=0.036$)/Both treatments were effective to reduce *C. Albicans* adhesion
Yildirim,[17] 2005	The effect of the treatment of acrylic resin surfaces with glow-discharge plasma, and candidial adherence to these surfaces.	102 flat circular acrylic resin as *C. albicans* source.	Radio frequency glow discharge unit (RF 300–13Æ56 SEREN IPS) was used for 15 minutes.	Control Oxygen 50 w Oxygen 100 w	Yeast cells retained on modified surfaces in higher numbers than on controls, more yeast cells were counted on $O_2$100 group than on the O_2 50 group.
Zamperini,[18] 2011	Comparison of different surface modifications with plasma treatments on the adherence of *C. albicans* to denture bases.	180 disk shaped denture base acrylic resin as *C. albicans* source.	argon atmosphere at 50 W and 70 W; atmospheric air at 130 W; argon atmosphere, followed by plasma treatment in a sulphur hexafluoride atmosphere, both performed at 70 W. The plasma exposure time was 5 minutes. For groups Ar/50 W, Ar /O2 70 W and AAt /130 W, the plasma parameters were chosen based on the degree of surface hydrophobicity.	Control argon atmosphere at 50 W Ar /O2 atmosphere at 70 W atmospheric air at 130 W argon atmosphere, followed by plasma in a sulphur hexafluoride atmosphere performed at 70 W	The adherence of *C. albicans* was not significantly reduced by plasma treatments when compared with the control, irrespective of the presence or absence of saliva and surface roughness.
Zamperini,[19] 2013	Comparison of two glow-discharge plasma treatments to modify the surface chemistry of a denture base acrylic resin to reduce the *C. glabrata* adhesion	54 disk shaped denture base acrylic resin as *C. glabrata* source	Plasma treatments were by radiofrequency power (13.56 MHz) to two parallel plate electrodes fitted inside a homemade stainless steel vacuum chamber.	Control argon atmosphere at 50 W atmospheric air at 130 W	The Ar/50 W plasma treatment reduced the adhesion of *C. glabrata* to the denture base resin compared to control.
Zamperini,[20] 2010	Comparison of different plasma treatments to modify a denture base acrylic resin to reduce the *C. albicans* adhesion	180 acrylic resin disks as *C. albicans* source.	argon atmosphere at 50 W (group Ar/50 W); argon/oxygen atmosphere at 70 W (group ArO2/70 W); atmospheric air at 130 W (AAt/130 W); argon atmosphere, followed by plasma treatment in a sulphur hexafluoride atmosphere, both performed at 70 W (group ArSF6/70 W). The plasma exposure time was 5 minutes. For groups Ar/50 W, ArO2/70 W and AAt/130 W, the plasma parameters were chosen based on the degree of surface hydrophobicity.	Control argon atmosphere at 50 W argon/oxygen atmosphere at 70 W atmospheric air at 130 W (AAt/130 W) argon atmosphere, followed by plasma in a sulphur hexafluoride atmosphere at 70 W (group ArSF6/70 W)	The adherence of *C. albicans* was significantly reduced by ArO2/70 W and ArSF6/70 W plasma treatments when compared to the control group, regardless of the presence or absence of saliva and surface roughness.
Matthes,[21] 2015	Antimicrobial potential of plasma with argon or argon plus 1 % oxygen as the working gas on *C. albicans* biofilms grown for 2, 7, or 16 days on polymethylmethacrylate discs.	20 PMMA discs *C. albicans*	A volume dielectric barrier discharge plasma source was used. High voltage (40 kHz, 10 kV) was applied between the two electrodes to generate the plasma. Pure Ar was applied as the working gas for maintaining the discharge and flowed through the system at a rate of 50 standard cm^3 per minute. Treatment time was 10 minutes per disc side.	Ar plasma and Ar + 1 % O2 plasma for 1, 5, or 10 min Ar gas CHX NaOCl CHX + Ar plasma NaOCl + Ar plasma	The argon plasma treatment showed a high anticandidal effect superior to CHX which increased with increasing treatment time. No clinically relevant effect was shown against *C. albicans* biofilms. No increased antimicrobial effects after the combined treatment with CHX or NaOCl solutions and plasma.

Table 2. Continued.

First Author, Year	Objective	Methods & Material		Groups	Result
		Specimens	Type and Settings of Cold Plasma		
Pan,[12] 2015	The effects of cold plasma treatment on the physical properties of heat-polymerized acrylic resin and early *Candida* albicans adhesion.	36 PMMA denture base resin samples in cylinder shape as *C. albicans* source.	A copper foil, served as a single electrode, was connected to a 10-kHz sinusoidal high-voltage source with an 18-kV peak-to-peak voltage. Premixed argon and oxygen (98% Ar and 2% O2 per volume) was used as the working gas and passed through the Teflon tube at a flow rate of 300 L/h. Treatment time varied from 90 seconds to 8.5 minutes based on specimen's size.	Control Ar/O2 plasma	Ar/O2 plasma treatment significantly reduced early *Candida* albicans adherence
Qian,[22] 2015	Comparison of different atmospheric pressure cold plasma treatment time on surface physical and chemical properties of denture base acrylic resin, and its effect on *C. albicans* adherence.	45 disks of acrylic resin denture base material as *C. albicans* source	A copper foil is connected to a 10 kHz sinusoidal high voltage source with an 18 kV peak-to-peak voltage. Ar/O2 (98% Ar and 2% O2 per volume) at a flow rate of 5 L/min is used as working gas.	Control plasma treated for 30 seconds 60 seconds 90 seconds 120 seconds	plasma treatment groups showed significantly lower *C. albicans* adherence than the control group. The lowest adherence of *C. albicans* was detected on 90 s group.
Wang,[14] 2013	Effect of cold plasma on oral *Candida albicans* biofilm on denture stomatitis.	24 denture base resin as *C. albicans* source	A direct current power supply was used to generate a negative high voltage. Premixed argon and oxygen (98% Ar and 2% O2 per volume, Ar/O2) or premixed argon and oxygen and nitrogen (88% Ar, 2% O2 and 10% N2 per volume, Ar/O2/N2) were used as working gas. The voltage was 0.4-0.6 kV and the current was 30-35 mA. In the Alternating current cold plasma jet. copper foil was connected to a 10-kHz sinusoidal high-voltage source with an 18-kV peak-to-peak voltage. The duration was 0, 1, 2, 4, 6, 8 min for each group.	Control Argon and oxygen Argon and oxygen and nitrogen	Strains treated with plasma showed a higher sensitivity to antifungal drugs when compared with control groups. Plasma alone or assisted with drug therapy can provide a valuable tool for *Candida albicans* infection.
Wang,[23] 2016	The effect of Ar/O2 cold plasma treatment on the Candida *albicans* biofilms formed on the PMMA denture base resin and their drug susceptibility .	PMMA denture base resin cylinders as *C. albicans* source	The plasma was driven by a 10-kHz sinusoidal alternative voltage source with 18 kV (peak-to-peak). Ar and O2 mixture (Ar 98%, O2 2%) was used as working gas with the treatment time fixed at 2 min.	Control Plasma treatment for 1min 2 min 4 min 6 min 8 min	Excellent inactivation was confirmed after 8 min cold Ar/O2 (2 %) plasma treatment, while no significant influences on PMMA surface roughness is observed.

SF6, sulfur hexafluoride; HMDSO, hexamethyldisiloxane; CFU = colony-forming units; PMMA, polymethylmethacrylate; Ar, argon.

The intracellular protein concentration in the treated groups significantly decreased with prolonged treatment, indicating that the sterilization effect was closely linked to protein loss.[14] Additionally, strains exposed to plasma exhibited higher sensitivity to antifungal medications compared to the control groups. Matthes et al concluded that utilizing cold plasma, whether alone or in combination with antiseptics, represents a promising approach for daily antifungal treatment on dentures provided that the biofilms are no more than two days old.[21]

Qian et al stated that cold plasma treatment has the potential to enhance the surface wettability of denture base acrylic resin and decrease the adhesion of *C. albicans*.[22] Wang et al concluded that biofilms of *C. albicans*, approximately 100 μm in thickness, were completely inactivated after eight minutes of plasma treatment. Scanning electron microscopy showed alterations in the fungi's morphology. Following one minute of plasma treatment, the sessile minimum inhibitory concentration of the biofilm for amphotericin B and fluconazole showed a significant reduction.[14]

Zamperini et al showed that surfaces treated with Ar/50 W may effectively decrease the adhesion of *C. glabrata* to denture base resins.[19] Zamperini et al showed that the adhesion of *C. albicans* was significantly lower after exposure to ArO_2/70 W and $ArSF_6$/70 W plasmas compared to the control group, regardless of surface roughness and saliva.[20]

Preventing the initial adhesion of *Candida* species to denture surfaces is critical for controlling the infection. Cold plasma treatment can alter the surface characteristics of denture materials, such as acrylic resins commonly used in dentures. By increasing surface hydrophilicity or altering surface roughness, Cold plasma can reduce the affinity of *Candida* cells for the surface, thereby preventing biofilm formation. Studies utilizing atomic force microscopy and contact angle measurements have shown that cold plasma-treated surfaces exhibit reduced

microbial adhesion, which could translate to fewer infections and easier cleaning for denture wearers.

The efficacy of cold plasma treatment in reducing *Candida* biofilm on denture surfaces seems to vary across different studies. Some studies have reported positive results, showing that cold plasma treatment can effectively decrease *C. albicans* biofilm. However, other studies have found no significant difference in *C. albicans* adherence after cold plasma treatment compared to control groups. Furthermore, there may be variations in the types of cold plasma devices used, treatment parameters, and evaluation methods among different studies, which could contribute to the discrepancies in the findings. Overall, cold plasma treatment shows promise as a means of treating and preventing denture-related candidiasis. It has antimicrobial effects, including damage to cell membranes, protein oxidation, and DNA damage, without the high temperatures associated with thermal plasmas. Additionally, cold plasma can penetrate and disrupt biofilms, reducing both the biofilm biomass and the cells' resistance to antifungal agents. However, further research is needed to fully understand the underlying mechanisms and optimize the application of cold plasma in denture stomatitis treatment.

Limitations

The limited number of studies limited the overall strength and generalizability of the findings. The included studies were heterogeneous in terms of study design and treatment protocols. This heterogeneity may introduce bias and make it difficult to draw definitive conclusions and categorize the results based on each plasma type and setting. Additionally, the long-term effects and safety of cold plasma treatment for *Candida* biofilm on dentures remain unclear due to the limited follow-up periods in the included studies. This review has primarily focused on laboratory or clinical outcomes, without considering patient-reported outcomes such as quality of life or patient satisfaction. All studies did not mention all factors like sample size and distance of cold plasma and samples.

Conclusion

The available evidence from in-vitro studies suggests that cold plasma treatment holds promise as a potential therapeutic option for managing *Candida* biofilm on denture surfaces. Further research is needed to clarify its effectiveness, optimize treatment parameters, standardize protocols, and investigate the long-term effects of cold plasma treatment on denture stomatitis patients.

Authors' Contribution

Conceptualization: Hosein Eslami, Katayoun Katebi.
Data curation: Elham Nazari.
Formal analysis: Katayoun Katebi.
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Project administration: Hosein Eslami.
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Writing–original draft: Hosein Eslami, Vahid Fakhrzadeh, Elham Nazari, Katayoun Katebi, Mohammad Alinejad.
Writing–review & editing: Hosein Eslami, Vahid Fakhrzadeh, Elham Nazari, Katayoun Katebi, Mohammad Alinejad.

Competing Interests

None.

Ethical Approval

This study was approved by regional ethics committee (IR.TBZMED.REC.1403.931).

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Supplementary Files

Supplementary file 1. The exact search strategy.

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