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Cold Atmospheric Pressure Plasma: A New Approach for Management of Periodontitis and Peri-implantitis

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Abstract

Background and Aim: Plasma is commonly known as the fourth state of matter. Cold atmospheric pressure plasma (CAPP) is a new, simple, and non-invasive approach that has become popular in various fields. Due to its potential antibacterial properties, it has become a widely used disinfectant in clinical science. It has even been employed for therapeutic purposes, such as cancer treatment and wound healing. This review aims to introduce CAPP as a new approach for management of periodontitis and peri-implantitis.

Materials and Methods: A search was conducted in Scopus, PubMed and Web of Science databases for articles published between 2000 and 2023 using the keywords: "plasma gases", "non-thermal atmospheric pressure plasma", "cold atmospheric pressure plasma", "cold plasma", "periodontal", "periodontitis" and "peri-implantitis".

Results: The results showed that CAPP is effective for reducing the bacterial load and inflammation, modification of implant surfaces, promoting tissue regeneration, and improvement of clinical parameters. The use of CAPP in periodontal therapy and management of peri-implantitis is still in its early stages but studies have shown promising results.

Conclusion: CAPP appears to be a promising adjuvant therapy for periodontitis and management of peri-implantitis. However, further clinical studies are needed to fully explore its potential and determine its effectiveness for this purpose.

Key Words: Peri-implantitis; Periodontitis; Plasma Gases

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Introduction

Periodontitis is a prevalent chronic inflammatory disease, and a major cause of tooth loss. It leads to progressive destruction of tooth-supporting structures, gingival bleeding, pocket formation, and bone loss, if left untreated [1, 2]. Although periodontitis is a multifactorial disease, biofilm accumulation on tooth surfaces has been identified as an essential contributing factor in development and progression of periodontitis [3]. A study on samples of subgingival plaque from patients with periodontitis identified three major pathogenic bacteria namely *Tannerella forsythia*, *Treponema denticola*, and *Porphyromonas gingivalis (P. gingivalis)* that are often found together and are significantly associated with severe periodontitis. These bacteria are referred to as the "red complex" bacteria and are known as a significant cause of periodontal disease [4].

Scaling and root planing (SRP) is considered as the gold standard treatment for periodontitis as it aims to remove calculus and necrotic cementum to prevent recolonization of bacteria and formation of supragingival biofilm. However, it is not effective in removing all periodontal pathogens and accessing deep periodontal pockets [5-7]. Therefore, several adjunctive therapies such as antibiotic therapy and photodynamic therapy are used to improve the clinical outcomes, which have shown promising results. Antibiotics can effectively eliminate periodontal pathogens, but development of bacterial resistance is a major concern in this adjunctive treatment [8].

Peri-implantitis is an infectious disease that affects the soft tissue and bone surrounding osseointegrated implants, causing inflammation and eventual bone loss [9]. The main risk factors for peri-implantitis are poor oral hygiene, dental plaque, smoking, and periodontitis [10]. Prevention is the key to minimize the risk of peri-implantitis. Treatment options include non-surgical debridement, surgical intervention, implant surface decontamination with laser, air-powder abrasion technique, and use of antibiotics and antimicrobial agents such as chlorhexidine [9].

Cold atmospheric pressure plasma (CAPP) is another recently introduced adjunctive therapy. Plasma is the fourth state of matter, which was first identified by William Crooks in 1879. It contains free electrons, ions, and active radicals caused by excitation of gases such as argon, helium, oxygen, and nitrogen through microwaves, radiofrequency, or electrical fields [11, 12]. The plasma technology is categorized as thermal and non-thermal based on the temperature component. In thermal plasma, atoms and heavy particles are both available at the same temperature. In non-thermal plasma, also known as the CAPP, electrons are hotter than ions and neutral components [12, 13]. In brief, CAPP generates reactive species of oxygen or nitrogen, UV radiation, ions, and charged particles that can affect the target directly or indirectly by plasma-activated media [14-16].

There are different systems and gases used to produce CAPP, depending on the specific application and desired characteristics of plasma. Some common systems include atmospheric pressure plasma jet (APPJ), dielectric barrier discharge (DBD) and plasma needle.

The APPJ system produces plasma by creating a stream of ions and radicals that are ejected from a nozzle at atmospheric pressure. The DBD system uses two electrodes separated by a dielectric material. A high voltage is applied to the electrodes, creating an electrical discharge that ionizes the gas between them and generates plasma [17]. A plasma needle is a variation of the APPJ system that uses a needle-shaped electrode to generate highly focused plasma for medical and biological applications [18].

These are just a few examples of the many different systems used to generate cold atmospheric plasma. The choice of system depends on the intended application and the specific properties of the plasma that are required.

CAPP is utilized in dentistry for various purposes, including sterilization of dental equipment, disinfection of root canals, tooth bleaching, composite curing, improvement of adhesive properties, decontamination of implant surfaces, and treatment of oral candidiasis [16, 19]. CAPP was found to be more effective than photodynamic therapy and diode laser in elimination of Enterococcus faecalis during root canal therapy [20]. The objective of this study was to conduct a comprehensive review of the literature on the application of CAPP in periodontology.

Materials and Methods

For this review article, a search was conducted for relevant articles published between 2000 and 2023 in journals indexed in PubMed, Scopus, or Web of Science databases. The searched keywords were "plasma gases", "non-thermal atmospheric pressure plasma", "cold atmospheric pressure plasma", "cold plasma", "periodontal", "periodontitis" and "peri-implantitis". Studies were initially selected based on keywords and title. Next, all articles were assessed, and studies investigating the efficacy of CAPP in the periodontal field were included. Original articles and experimental studies published in English were included, while review articles were excluded. Also, the reference lists of the selected articles were examined for relative studies to be included. Finally, 23 studies were included.

Results and Discussion

Antibacterial effects of CAPP:

Four studies investigated the effectiveness of CAPP against *P. gingivalis* biofilm and found that CAPP was able to significantly reduce the number of viable cells in *P. gingivalis* biofilm. Its effect was directly proportional to the exposure time [21-24].

Jungbauer et al. [25] investigated the effects of CAPP on 12 bacterial strains, including the red complex bacteria. The results indicated that after 120 seconds of exposure to planktonic bacteria, a significant reduction was seen in all species except *Treponema denticola*.

Anti-inflammatory activities of CAPP:

Studies have shown that CAPP can increase the expression of anti-inflammatory cytokines such as interleukin (IL)-10 and transforming growth factor (TGF)-β1, and promote the production of collagen type I and matrix metalloproteinase-1, which can facilitate periodontal wound healing and tissue regeneration [26-30]. Additionally, it may reduce the expression of pro-inflammatory cytokines such as tumor necrosis factor-α, IL-8, IL-6, and IL-1β [31-34].

Histological effects of CAPP:

Kwon et al. [30] conducted a study on the effects of a CAPP jet on cellular activity of human gingival fibroblasts (HGF) and found high cell viability. However, following exposure to CAPP, no significant alternation in proliferation or attachment of cells was observed. In another study, Seo et al. [32] investigated the effect of microwave-pulsed CAPP on HGFs and demonstrated that cell viability was unaffected, but after 5 minutes of exposure to CAPP, cell proliferation increased.

Evidence shows that enhanced wettability of dentin or bone substitutes improves the ability of osteoblasts to spread [35]. Koban et al. [35] demonstrated that CAPP treatment resulted in enhanced wettability and superior spreading of osteoblasts on dentin surfaces. However, Matthes et al. [36] reported that CAPP treatment did not improve the spreading of osteoblasts on the implant surface.

When CAPP was used as an adjunct to SRP in rats with ligature-induced periodontitis, a remarkable decrease in the number of osteoclasts, lower expression of RANKL, and reduced alveolar bone loss were observed [31].

Clinical effectiveness of CAPP:

Küçük et al. [37] conducted a clinical study on the effectiveness of CAPP in treatment of patients with periodontitis. The results demonstrated that CAPP was more effective than SRP alone in reducing periodontal pocket depth and improving clinical attachment gain. Furthermore, CAPP was found to be effective in reducing the bacterial load of subgingival red complex bacteria and improving the bleeding on probing (BOP), plaque index (PI) and gingival index (GI). Overall, these findings suggest that CAPP is a promising treatment option for patients with periodontitis.

CAPP in peri-implantitis:

Koban et al. [38] conducted a study to compare the antimicrobial efficacy of CAPP and chlorhexidine against dental biofilm on titanium discs. The results showed that CAPP was more effective than CHX in treating single- and multispecies dental biofilms on titanium discs in vitro. In another study by Preissner et al. [39], the bactericidal efficiency of CAPP on microrough titanium dental implants was investigated. The method involved seeding titanium implants with bacteria and subjecting them to CAPP treatment. The results showed a significant reduction in the bacterial count on implants after CAPP treatment. Shi et al. [40] evaluated the effects of CAPP in treatment of ligature-induced peri-implantitis in beagle dogs. Micro-computed tomography and histological analysis of block biopsies revealed a significantly higher bone level in the CAP group than in the control group 3 months after treatment. The counts of *P. gingivalis* and *Tannerella forsythia* were significantly lower than those of the control group and baseline values. In another study, Zhou et al. [34] assessed the effect of adjunctive use of CAPP for surgical mechanical debridement on peri-implantitis in beagle dogs. After 3 months of intervention, the plasma group experienced a noteworthy enhancement in clinical parameters and bone height when compared to the control group. Furthermore, IL-1β and IL-17 levels decreased in the plasma group in contrast to the control group, while IL-6 levels remained unchanged. Studies evaluating the effect of CAPP on *P. gingivalis* biofilm cultured on titanium discs reported a significant reduction in bacterial count in the CAPP group [41, 42].

Jungbauer et al. [25] evaluated the effects of CAPP on 12 bacterial species, after culturing biofilms on dentin and titanium specimens. The results indicated a decrease in metabolic activity of the biofilm following 120 seconds of irradiation.

Yang et al. [43] investigated helium CAPP as a surface modification to treat zirconia implant abutments and evaluated its efficacy in reducing oral bacterial adhesion and proliferation. After the treatment, the chemical composition of the zirconia surface was altered while the surface topography remained the same. This resulted in decreased bacterial growth and biofilm formation. As the treatment time increased, the zirconia abutment demonstrated more effective inhibition of bacteria. A study by Jeong et al. [33] demonstrated that CAPP treatment of titanium surfaces improved cellular morphology, cell viability, and protein absorption of soft tissue.

Canullo et al. [44] studied the effect of argon CAPP pre-treatment of healing abutments on the peri-implant microbiome and soft tissue integration of 30 periodontally stable patients. The results showed that the CAPP treated group had lower plaque accumulation and inflammation, as well as lower PI and BOP. Microbial analysis indicated that the majority of late colonizers were present in the control group, while the CAPP group showed a greater prevalence of early colonizers, indicating less advanced plaque formation.

Based on the reported favorable results, CAPP could be an effective method for treatment and prevention of peri-implantitis. A summary of reviewed articles is presented in Table 1.

Table 1. Summary of the reviewed studies

Reference/ Year	Specimens	Main Outcome	Device setting	Gas	Time of exposure	Result
Hong et al, 2022 [21]	*P. gingivalis* biofilm	Antibacterial effect	Plasma brush Parameters: • Voltage: 3 kV • Power: 10W • Gas flow rate: 3000 standard cubic centimeter per minute (sccm)	• Argon • Argon + 1% Oxygen	1, 2, and 5 min	– Reduction in bacterial count
De Morais Gouvea Lima et al, 2022 [22]	• Planktonic growth of *P. gingivalis* • *P. gingivalis* and *Streptococcus gordonii* Dual Species Biofilms	Antibacterial effect	Plasma jet Parameters: • Voltage: 10 kV • Power: 0.6W • Gas flow rate: 1 L/minute	Helium	1, 3, 5, and 7 min	– A significant reduction in planktonic growth was observed in proportion to the length of exposure time – Inhibition of *P. gingivalis*

						and *S. gordonii* dual species biofilm growth
Liu et al, 2011 [23]	*P. gingivalis biofilm*	Antibacterial effect	Plasma jet Parameters: • Voltage: 8 kV • Frequency: 10 kHz • Pulse width: 200 ns to dc	Helium + Oxygen	5 min	– Almost all bacteria were killed
Mahasneh et al, 2011 [24]	*P. gingivalis* biofilm	Antibacterial effect	Plasma pencil Parameters: • Voltage: 8 kV • Frequency: 5 kHz • Pulse width: 500 ns • Gas flow rate: 5 L/minute	Helium	5, 7, 9, and 11 min	– The CAPP exposure demonstrated significant inactivation of bacteria that depended on the exposure time
Jungbauer et al, 2022 [25]	• Suspension of 11 bacterial strains, including Red complex bacteria • Twelve bacterial strains (including Red complex bacteria) on titanium and dentin surfaces	Antibacterial effect and Biofilm reduction	Piezoelectric direct discharge (PDD) Parameters: • Frequency: 50 kHz • Power: 8 W	Ambient Air	10, 30, 60, and 120 s	– In suspension, all species showed a significant reduction except for Treponema denticola – A reduction in metabolic activity of the biofilms on titanium and dentin sample specimens after being exposed to CAPP for 120 seconds
Kleineidam et al, 2019 [27]	Human periodontal ligament cells	Anti-inflammatory	Plasma jet Parameters: Voltage: 3–5 kV	Ambient Air	30, 60, and 120 s	– Expression of the MMP-1 and wound healing rate were significantly enhanced.
Arndt et al, 2015 [28]	Human skin keratinocytes	Anti-inflammatory	Microwave plasma system Parameters: Microwave: 2.45 GHz Power: 80 W Gas flow rate: 4 L/minute	Argon	2 min	– ß-defensins, TGF-ß1, TGF-ß2, and IL-8 expressions were induced
Arndt et al, 2013 [29]	• HGFs • Mouse skin model	Anti-inflammatory	Microwave plasma system Parameters: Microwave: 2.45 GHz Power: 80 W Gas flow rate: 4 L/minute	Argon	2 min	– Induced the expression of TGF-ß1, TGF-ß2, MCP-1, IL-6, IL-8, alpha-SMA, and collagen type I
Kwon et al/ 2016 [30]	• HGFs	• Anti-inflammatory • improve cellular viability	Plasma jet Parameters: • Voltage: 2.2 kV • Gas flow rate: 1 L/minute	Ambient Air	1, 2, and 4 min	– Increased in expression of TGF-ß and vascular endothelial growth factor was observed – Higher cellular viability was observed after the exposure to CAPP, while

						no significant difference in cellular attachment and proliferation was observed
Zhang et al, 2018 [31]	Rats with ligature-induced periodontitis	• Anti-inflammatory	Plasma brush Parameters: • Power: 5 W to 15 W • Gas flow rate: 3000 sccm	Argon + 1% Oxygen	2 min	– The expression of IL-10 increased, while the expression of IL-1β and TNF-α decreased – After CAPP treatment, there was a significant decline in osteoclast count, reduced RANKL expression, and decreased loss of alveolar bone
Seo et al/ 2017 [32]	HGFs	• Anti-inflammatory • improve cellular proliferation	Microwave plasma system Parameters: Microwave: 30 MHz Power: 40 W Gas flow rate: 2 L/minute	Argon	1, 3, 5, and 10 min	– The CAPP treatment resulted in a decrease in the expression of TNF-α in lipopolysaccharide-stimulated HGFs compared to untreated cells – Cellular viability was unaffected; however, cellular proliferation was increased
Jeong et al/ 2018 [33]	• HOKs • HGFs	• Anti-inflammatory • improvement in soft tissue cells characteristics	Plasma jet Parameters: • Voltage: 2.24 kV • Power: 2.4 W • Gas flow rate: 5 L/minute	Ambient Air	10 min	– After exposure to CAPP, a decrease in the expression of IL-1β, IL-6, and IL-8 was observed in inflamed cells except for IL-8 in inflamed HOKs – Improvement in viability and morphology of cells, and protein absorption.
Zhou et al, 2022 [34]	Beagle dogs with ligature-induced peri-implantitis	• Anti-inflammatory • Treatment of peri-implantitis • Improvement in clinical parameters	Plasma jet Parameters: • Frequency: 8 kHz • Gas flow rate: 2 L/minute	Helium + 0.1% Oxygen	45 s each side of the implant (buccal, lingual, mesial, distal)	– Reduced expression of IL-1β and IL-17 – Probing depth, sulcus bleeding index, and bone height were significantly improved
Koban et al, 2011 [35]	• Human dentin discs • Hydroxyapatite • Mammoth ivory	• Improve wettability • Improve osteoblasts spreading	Plasma jet • Gas flow rate: 5 L/minute	• Argon • Argon + 0.2% Oxygen • Argon + 1% Oxygen	10, 30, 60, and 120 s	– Enhanced wettability and superior spreading of osteoblasts
Matthes et al/ 2017 [36]	Rough, Biofilm covered titanium discs	Improve wettability	Plasma jet Parameters: • 1 MHz • Voltage: 2–6 kV • Power: 3.5 W • Gas flow rate: 5	Argon	300 and 720 s	– Osteoblast spreading was not improved by a combination of erythritol powder with CAPP – The water contact angle decreased after CAPP

			L/minute			treatment
Küçük et al/ 2020 [37]	Patients who are systemically healthy and have periodontitis	Improvement in clinical parameters	Plasma jet Parameters: • Max power: 5 W • Frequency: 420 Hz–1220 Hz • Pulse width: 5–10 µs	Ambient Air	2.5 min	– Improvement in BOP, PI, PD, and clinical attachment level gain – Reduction in the count of supra gingival red complex bacteria

Koban et al, 2011[38]	Titanium discs	Biofilm reduction	Three different devices include:
			• Plasma jet
			  • Gas flow rate: 5 L/minute
			• Volume DBD Parameters:
			  • Frequency: 40 kHz
			  • Voltage: 10 kV
			  • Gas flow rate: 0.05 L/minute
			• Hollow electrodes DBD parameters:
			  • Frequency: 37.6 kHz
			  • Voltage: 8.4 kV
			  • Gas flow rate: 1 L/minute
				• Argon
				Argon + 1% Oxygen
					1, 2, 5, and 10 min	– CAPP was proven to be significantly more effective than CHX against S. mutans and saliva biofilms on titanium discs

Preissner et al, 2016 [39]	Microrough titanium dental implants	Bactericidal efficacy in dental implants	Plasma jet Parameters: • Gas flow rate: 4.3 L/minute	Argon	60 and 120 s	– A significant reduction in the bacterial count was observed following the treatment (compared to diode laser 980nm and 1% sodium chloride) – There was no significant difference observed between 120-second and 60-second CAPP exposures.
Shi et al, 2015 [40]	Beagle dogs with ligature-induced peri-implantitis	Treatment of peri-implantitis • Bactericidal efficacy in dental implants • improvement in clinical parameters	Plasma needle Parameters: • Voltage: 8 kV • Frequency: 8 kHz • Pulse width: 1600 ns • Gas flow rate: 2 L/minute	Helium + 0.1% Oxygen	3 min	– A higher level of bone was observed after 3 months of treatment in the CAPP-treated group compared to the control group – A significant reduction in the count of P. gingivalis and Tannerella forsythia was observed – Reduction in sulcus bleeding index and PD

Carreiro et al, 2019 [41]	• Titanium discs • Gingival epithelium	• Biofilm reduction • improvement in soft tissue cells characteristics	Plasma jet Parameters: • Power: 8 W • Voltage: 220 V • Frequency: 50,60 Hz • Gas flow rate: 5 L/minute	Argon	1 and 3 min	– The CAPP treatment was found to reduce the p. gingivalis count in comparison to the control, though it was not as effective as the 0.2% chlorhexidine treatment – in gingival epithelium, high viability and low cytotoxicity were – observed
Lee et al, 2019 [42]	Titanium discs	Biofilm reduction	Plasma jet Parameters: • Voltage: 7 kV • Frequency: 10 kHz • Gas flow rate: 5 L/minute	Helium	3 and 5 min	– CAPP treatment can effectively reduce bacterial count on sandblasted and acid-etched titanium discs.
Yang et al, 2020 [43]	Zirconia implant abutments	• Biofilm reduction and inhibition of bacterial growth • Surface modification approach	Dielectric barrier discharge Parameters: • Voltage: 2.85 kV • Frequency: 17 kHz • Gas flow rate: 13.5 L/minute	Helium	30, 60, and 90 s	– A significant reduction in bacterial growth and biofilm formation following CAPP application – Decrease in contact angle – The chemistry of the zirconia surface has been altered, while the topography remains the same
Canullo et al, 2023 [44]	Systemically healthy patients who received smooth surface and rough ultrathin threaded micro-surface abutments	• Improvement in clinical parameters	Parameters: • Power: 75 W Pressure: 10 MPa	Argon	12 min	– Reduction in plaque accumulation, PI, inflammation, and BOP – The control group was dominated by late colonizers, while the CAPP treated group had a higher prevalence of early colonizers

*CHX: Chlorhexidine, PD: Pocket depth, PI: Plaque index, GI: Gingival index, BOP: Bleeding on probing, HGFs: Human gingival fibroblasts, HOKs: Human oral keratinocytes

Conclusion

CAPP is a novel therapeutic approach that can be effectively used as an adjunct to conventional mechanical debridement therapy for periodontitis, and has also been found to be effective in management of peri-implantitis. This study showed promising results for CAPP in reducing inflammation, eliminating bacteria, and improving clinical parameters; however, further clinical studies are required to determine the long-term safety and efficacy of CAPP in treating periodontitis.

Conflict of interests and financial disclosure

The authors declare no conflict of interests.

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