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Original Research Article

Association of NLRP3 inflammasome and *Fretibacterium* species human oral taxon 360 in patients with periodontitis and gynaecological disorders: A correlative study

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Abstract

Background: The most prevalent endocrine diseases with multifactorial etiology are Polycystic Ovary Syndrome (PCOS) and Gestational Diabetes Mellitus (GDM). Chronic Periodontitis (CP) is an immunoinflammatory disease, studies suggest that CP is linked to PCOS and GDM. This study evaluated *Fretibacterium* HOT 360 and NLRP3 inflammasome levels in individuals with periodontitis and gynaecological diseases, particularly PCOS and GDM.

Materials and Methods: Totally 90 subjects were screened and they were categorised into Group 1 (N=45: Patients with gynaecological disorders and without periodontitis) and Group 2 (N=45: Patients with gynaecological disorders + Periodontitis). Saliva samples were collected to assess the levels of NLRP3 inflammasome and *Fretibacterium* HOT 360 using an ELISA and conventional PCR respectively. Periodontal parameters (Plaque score, PD, CAL, PISA) were evaluated for all the patients.

Results: The mean concentration of NLRP3 inflammasome and *Fretibacterium* HOT 360 was significantly higher in group 2 (Patients with gynaecological disorders and Periodontitis) than group 1 (Patients with gynaecological disorders and without periodontitis) at $p< 0.05$. In addition, NLRP3 inflammasome and *Fretibacterium* HOT 360 were positively correlated with all the periodontal parameters (Plaque score, PD, CAL, PISA) in both the groups with the higher value in group 2.

Conclusion: There is an association between chronic periodontitis and gynaecological disorders (PCOD and GDM) as demonstrated by significant levels of NLRP3 inflammasome and *Fretibacterium* HOT 360 levels in patients with periodontitis and gynaecological disorders.

Keywords: Chronic periodontitis, Polycystic ovary syndrome, Insulin resistance, NLRP3 inflammasome, *Fretibacterium.*

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1. Introduction

Periodontitis is a chronic inflammatory disease that involves the complex interactions between pathogenic periodontal microbiota and the host immune response, modulated by environmental and genetic factors.[1] It has been reported that 50% of Indian adults suffer from periodontal disease.[2] Due to its chronic inflammatory nature, CP is associated with a systemic state of oxidative stress, mitochondrial dysfunction and multiple systemic diseases. Diabetes mellitus is a traditional risk factor for CP and a bidirectional association between the two diseases has already been established. Also, CP has been associated with various insulin-resistance (IR) conditions, including rheumatoid arthritis, PCOS, and cardiovascular disease (CVD).[4] Numerous environmental factors, such as hormone levels, salivary pH, anaerobic conditions, and nutrition, might impact the oral microbiota.[5] Periodontitis has been associated with various risk factors that impact the spontaneous probability of conception, including age, bacterial vaginosis (BV), endometriosis (EM), PCOS, and obesity.[6] The most common plausible etiologic mechanism linking periodontal disease and many systemic diseases are chronic low-grade inflammation.[7,8]

Recent studies on the intricate molecular pathways behind the inflammatory response have given rise to the concept of the "inflammasome," a multiprotein oligomer molecule that controls inflammation in its early phases.[9] The NLRP3 (NOD-, LRR-, and pyrin domain-containing protein 3) complex is one of these inflammasomes and is essential to innate immunity and inflammatory processes. It is composed of a Nod-like receptor (NLR) that controls the expression of

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important proinflammatory cytokines, especially interleukin-18 (IL-18), and mediates the activation of protease enzymes (Caspase 1).[10] Periodontal inflammation is caused by the secretion of Interleukin-1β (IL-1β), a major pro-inflammatory cytokine[9] by host tissues and upregulation of NLRP3 inflammasome has also been reported.[11]

Polycystic ovarian syndrome (PCOS) is an endocrine disorder with multifactorial etiology.[12] The primary cause of infertility in PCOS is hyperandrogenism, which can activate the NLRP3 inflammasome and cause low-grade inflammation and the release of inflammatory mediators. Women with PCOS tend to have increased levels of androgen, oxidative stress, free fatty acid (FFA) and high-mobility group box 1 (HMGB1), molecules that serve as danger signals to activate the inflammasomes, especially the NLRP3 inflammasome pathway.[8]

Gestational diabetes mellitus (GDM) refers to glucose intolerance with onset or first recognition during pregnancy.[13] The common feature in both the pathologies is dysregulation of the inflammatory response.[14] Increased production and overexpression of various inflammatory mediators, including matrix metalloproteases (MMP1, MMP9, and MMP13), receptor activator nuclear factor κB ligand (RANKL), prostaglandin E2 (PGE2), cytokines (IL1, IL6, and TNFα), and chemokines (IL8), have been linked to the pathogenic mechanism between diabetes and periodontal disease.[15-17] In addition, DM increases the overexpression of innate immune mediators including defensins and Toll-like receptors (TLRs) in gingival tissues, aggravating the severity of chronic periodontitis (CP).[18] The inflammasomes activated by metabolic damage and infection have also been proposed to be involved in the pathogenesis of periodontal disease and DM.[19]

Many environmental factors can impact the oral microbiota, such as pH, anaerobic conditions, diet, and hormone levels. The increased inflammatory response to the plaque microbiota in the periodontal tissues during pregnancy is mostly caused by female sex hormones. The development of systemic diseases, including diabetes mellitus, cardiovascular disease, and gynaecological disorders, is linked to the dysbiosis of the putative periodontal pathogens.[5] According to studies, unculturable bacteria linked to periodontitis may be a significant factor in the development of PLBW and the presence of periodontal infection during pregnancy.[20] *Fretibacterium sp.* HOT 360, a member of the phylum *synergistetes*, was chosen for the present study because of its increased prevalence in deep periodontal pockets.[21]

Since both the periodontal disease and gynaecological disorders (PCOS & GDM) have an increased inflammatory burden and are more prone to oral dysbiosis, this study evaluated the NLRP3 levels and *Fretibacterium* HOT 360 in patients with periodontal disease and gynaecological disorders (PCOS&GDM).

2. Materials and Methods

2.1. Study design

Patients diagnosed with polycystic ovary syndrome (PCOS) and gestation diabetes mellitus (GDM) reporting to the Department of Obstetrics and Gynaecology, Sri Ramachandra Medical College and Hospital were enrolled in this correlative study from August 2022 to November 2022. The study protocol was conducted with full accordance and approval of the Institutional Ethics Committee, Sri Ramachandra Institute of Higher Education & Research with the protocol number CSP/22/JUL/114/432. Following the Helsinki Declaration, written informed consent was obtained from each patient once they were informed about the study.

2.2. Inclusion criteria

1. Patients diagnosed with PCOS and GDM
2. Age group: 18-40 years
3. Patients with minimum of 20 existing natural teeth.

2.3. Exclusion criteria

1. Patients who had received any periodontal or antibacterial therapy within 3 months before the start of the study.
2. Pregnant women with systemic diseases other than GDM.
3. Anti-inflammatory or other type of medication taken in the last 30 days before the examination.
4. Smokers.

2.4. Sample size calculation

Using G* power software the optimal sample size was calculated. With α error of 0.05 and a power of 80% the minimum sample size was estimated to be 90 participants.[22]

Upon applying the above inclusion and exclusion criteria, the subjects were divided into two groups. Group 1 (Patients with gynaecological disorders and without periodontitis) and Group 2 (Patients with gynaecological disorders and Periodontitis).

Gynaecological disorders were diagnosed by a single trained gynaecologist. PCOS was diagnosed based on the 2003 Rotterdam Criteria.[23] GDM was diagnosed based on the Diabetes and Pregnancy Study Group of India (DIPSI) criteria.[24]

Periodontitis patients were screened by a single trained examiner using a standardized probing force of 0.75 N. The patients were classified as having Periodontitis based on 2012 definition by Eke et al.[25]

2.5. Periodontal assessment

A single trained examiner assessed the periodontal probing using a UNC 15 probe at the mesiobuccal, mid-buccal, distobuccal, mesiolingual, mid lingual and distolingual sites

of each tooth. The number of bleeding sites per tooth was also noted.

The probing depth and bleeding on probing values were used to calculate the periodontal inflamed surface area (PISA) scores for each patient by entering them into a previously obtained spreadsheet (http://www.parsprototo.info/docs/PISA-CAL.Xls).[26] Also, the level of oral hygiene was evaluated, using the O'Leary Index.

2.6. Sample collection for NLRP3 assessment:

1 ml of unstimulated saliva was collected from all the patients and stored at -80° Celsius. Participants were instructed to avoid food intake and oral hygiene procedures for at least 2 hours before saliva collection. The concentrations of NLRP3 were assessed by a specific Enzyme-linked immunosorbent assay (ELISA) kit according to the manufacturer's instructions. Plates were measured at 450nm by using a microplate reader. The concentrations of NLRP3 were calculated with the reader software in pg/ml.

2.7. Sample collection for bacterial detection

1 ml of unstimulated saliva was collected and centrifuged at 3000g for 10 minutes at room temperature to separate the debris samples. The obtained supernatants were stored at -20°C until extraction of genomic DNA.

2.8. DNA extraction

The DNA was extracted by simple boiling method, in which equal volume (200µl) of lysis buffer (10mmol/L Tris, 1.0mmol/L EDTA, 1% TritonX-100, pH 7.8) and samples were incubated at 56°C for 30 mins. Following incubation, the sample was boiled for 10 mins and centrifuged at 8000 rpm for 5 mins and 5µL of the supernatant was collected and immediately stored at -20°C until the assay.

2.9. Polymerase chain reaction amplification

Amplification was performed to amplify a 404-bp fragment of the 16s rRNA gene in a total volume of 25 µl consisting of 15.6 µl of sterile Milli-Q water, 2.5 µl of Taq Buffer, 1.0 µl of equal molar of four dinucleotide phosphates, 0.2 µl of each forward -5'GGAAACATTGACGACGCTG-3' and reverse primer - 5'-CTTAACCCAACATCTCACGAC-3', 0.5 µl of Taq polymerase enzyme and with the addition of 5 µl of template. PCR amplification was performed in Applied Biosystems Veriti 96-well thermocycler (Applied Biosystems, Foster City, CA). The cyclic conditions include initial denaturation of 95° C for 1 min and 36 cycles of denaturation at 95° C for 30s, annealing at 65°C for 1min and extension for 1min, followed by the final extension at 72°C for 2mins.

3.10. Amplicon detection

The amplified PCR products were determined using 2% agarose gel electrophoresis in which the agarose is stained with 0.2 µl of ethidium bromide (EtBr-0.5µg/ml) and subsequently, the products were visualized on 260nm wavelength of ultraviolet transilluminated in the gel documentation system (Bio-Rad Hercules, CA, USA). The appearance of the band at 404 bp was considered positive for the presence of *Fretibacterium* HOT 360. The sequence of primers of *Fretibacterium sp.* Human oral taxon 360 is as follows:

Primer F 5'-GGAAACATTGACGACGCTG-3'
Primer R 5'-CTTAACCCAACATCTCACGAC-3'

3.11. Statistical analysis

Statistical analysis was done using IBM SPSS (IBM Corp. Released 2011. IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY: IBM Corp). Mean and SD were used to summarize the data. Pearson's correlation was used in the correlation of NLRP3 with all the periodontal parameters. Independent samples t-test was used in intragroup comparison. A 'P' value of <0.05 will be considered as statistically significant.

3. Results

3.1. Sociodemographic and biochemical variables

All the demographic parameters (Age, BMI, FBS, PPBS) show a statistically significant difference in both the groups which are shown in **Table 1**. The mean age in Group 1 (Gynaecological disorders without periodontitis) and Group 2 (Gynaecological disorders with periodontitis) was 25.51±4.21 and 28.64±3.26 respectively.

Table 1: Mean values of the demographic variables in both groups

Parameters	G1-Without Periodontitis (N=45) Mean±SD	G2- With Periodontitis (N=45) Mean±SD	P-value
Age (Years)	25.51± 4.21	28.64±3.26	.000**
Body mass index (BMI)	27.98± 4.19	30.62±2.70	.001**
Fasting blood sugar (FBS) (mg/dl)	107.60±24.29	118.06±21.50	.033*
Post-prandial blood sugar (PPBS) (mg/dl)	146.33±55.39	168.60±41.26	.033*

**Highly significant ($p \leq 0.01$); *Statistically significant ($p \leq 0.05$)

Table 2: Mean values of the periodontal parameters in both groups

Parameters	G1-Without Periodontitis (N=45) Mean±SD	G2-With Periodontitis (N=45) Mean±SD	P-Value
Plaque score	21.98±3.04	36.75±4.96	.000**
Probing depth (PD)	1.47± 0.15	3.43±0.32	.005**
Clinical attachment loss (CAL)	0	5.31±0.30	.040**
Periodontal epithelial surface area (PESA)(mm²)	799.88±53.43	1216.61±176.64	.000**
PISA (Periodontal inflamed surface area (mm²)	319.74±59.60	597.24±118.05	.000**

**Highly significant (p ≤0.01); *Statistically significant (p≤0.05)

3.2. Mean values of periodontal parameters in both the groups

The mean probing depth in Group 1 (Gynaecological disorders without periodontitis) and Group 2 (Gynaecological disorders with periodontitis) was 1.47±0.15 and 2.43±0.32 respectively. The mean CAL was 0 and 1.31±0.3 in group 1 and group 2 respectively. The mean PISA was 319.74±59.60 and 597.24±118.05 respectively. All the periodontal parameters (Plaque score, PD, CAL, PESA, PISA) show a statistically significant difference in both the groups which are shown in **Table 2**.

3.3. Mean values of NLRP3 and Fretibacterium HOT 360 in both the groups

The Mean values of NLRP3 in Group 1 (Gynaecological disorders without periodontitis) and Group 2 (Gynaecological disorders with periodontitis) were 0.054 ± .008 and 0.081 ± .034 respectively. The mean value of *Fretibacterium* HOT 360 was 0.244±.434 and 0.533±.504 in group 1 and group 2 respectively. Both NLRP3 and *Fretibacterium* HOT 360 showed a statistically significant difference in both the groups which are shown in (**Table 3**).

Table 3: Mean values of NLRP3 and *Fretibacterium* HOT 360 in both groups

	G1-Without Periodontitis (N=45) Mean±SD	G2- With Periodontitis (N=45) Mean±SD	P-value
NLRP3	.054±.008	.081±.034	.000**
Fretibacterium HOT 360	.244±.434	.533±.504	.005*

**Highly significant (p ≤0.01); *Statistically significant (p≤0.05)

3.4. Correlation of NLRP3 with plaque score, probing depth and PISA

The correlation of NLRP3 with plaque score, probing depth and PISA in both groups was done using Pearson coefficient analysis. A significant positive correlation was found between NLRP3 and plaque score in group 2 whereas a negative correlation was found in group 1 which is shown in (**Figure 1**) (**Table 4**).

Table 4: Correlation of NLRP3 with periodontal parameters in both the groups

Parameters	Group 1		Group 2	
	Pearson correlation	P value	Pearson correlation	P value
Plaque score	.077	.614	.448	.001**
Probing depth	.663	.000**	.582	.000**
PISA	.131	.393	.590	.000**

**Highly significant (p ≤0.01); *Statistically significant (p≤0.05)

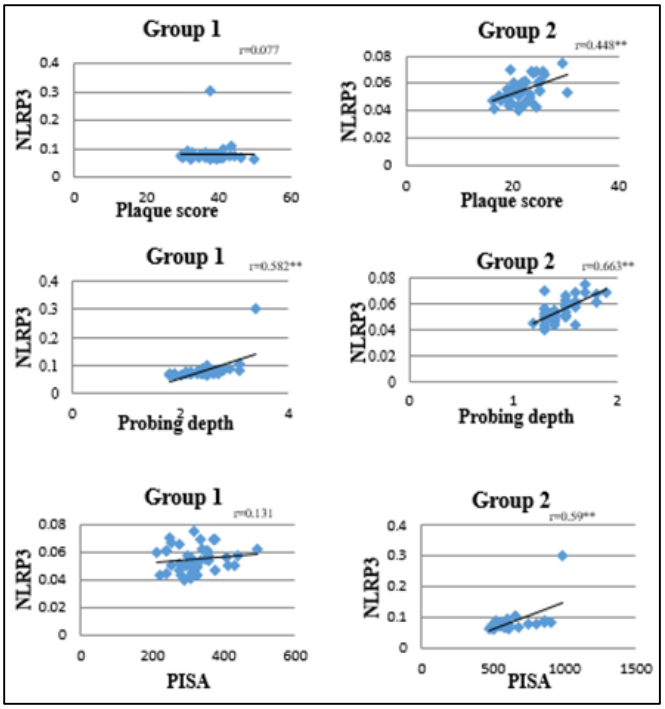

Figure 1: Correlation of NLRP3 with plaque score, probing depth, and PISA

3.5. Comparison of periodontal parameters based on the presence of Fretibacterium HOT 360 in group 1 and group 2

A significant positive correlation was found between the presence of *Fretibacterium* HOT 360 and all the periodontal parameters (Plaque score, PD, PESA, PISA) in both groups with a higher value in group 2 (**Table 5**).

Table 5: Comparison of periodontal parameters based on the presence of *Fretibacterium* HOT 360 in group 1 and group 2

Periodontal parameters	***Fretibacterium* Hot 360**		
	Group 1 (N=11) Mean±SD	**Group 2 (N=21) Mean±SD**	**P value**
Plaque score	23.50±2.66	38.0±4.74	.034*
Probing depth	1.63±0.13	2.62±0.28	.052*
PESA (mm^2)	857.39±37.73	1316±161.55	.004**
PISA (mm^2)	312.50±56.11	656.32±132.64	.007**

**Highly significant ($p \leq 0.01$); *Statistically significant ($p \leq 0.05$)

4. Discussion

Periodontitis is a chronic inflammatory disease characterised by a complex interplay between bacteria and the inflammatory response of the human immune system, that leads to changes in the homeostasis of connective tissue and bone. It is currently understood that several host-related factors influence the immunological and inflammatory responses, that play an essential role in the pathogenesis of periodontitis.[27]

The activation of the NLRP3 inflammasome results in the activation of caspase 1, which subsequently promotes the production of mature IL-1β and IL-18 from proIL-1β and proIL-18, respectively.[28] Interleukin-1β (IL-1β), a significant pro-inflammatory cytokine secreted by host tissues, is the major source of periodontal disease.[21] An upregulation of NLRP3 inflammasome has been reported in periodontal disease.[9] Studies have reported that these cytokines play a considerably significant part in the controlling of ovarian steroidogenesis, and maturation of ovarian follicles in addition to other reproductive events along with significant escalation of IL-18 was observed in PCOS women. It has already been shown that there is a bidirectional association between diabetes mellitus (DM) and periodontitis. Specifically, patients with DM are more likely to develop periodontitis, and periodontal inflammation impairs glycaemic control and may exacerbate the clinical course of DM.[16,29] Interleukin (IL-1β and IL-18), regulated by NLRP3 inflammasome, are important inflammatory cytokines in the initiation of maternal IR during GDM.[21]

Studies have reported that unculturable periodontitis-associated bacteria may play an important role in periodontal inflammation during pregnancy and subsequent PLBW.[5] In this study, *Fretibacterium sp.* HOT 360 which belongs to phylum synergistetes was selected because of its prevalence in deep periodontal pockets.[21]

Since both the periodontal diseases and gynaecological disorders (PCOS&GDM) have an increased inflammatory burden and are more prone to oral dysbiosis, this study evaluated the NLRP3 levels and *Fretibacterium* HOT 360 in patients with periodontal disease and gynaecological disorders (PCOS&GDM). The results of the present study demonstrated that the levels of NLRP3 were elevated in group 2 (Patients with periodontitis and gynaecological disorders (PCOS & GDM)) than in group 1 (Patients with gynaecological disorders (PCOS&GDM) without periodontitis) which could possibly due to the increased inflammatory burden, since both the diseases are chronic inflammatory in nature.

The results of the present study were in accordance with the study by Mitra DK et al., who compared the NLRP3 levels in the saliva of subjects with chronic periodontitis and healthy controls and found that increased levels of NLRP3 were seen in subjects suffering from chronic periodontitis.[31] In 2022 Zhou F et al. investigated the relationship between NLRP3 inflammasome and high-risk reproductive disorders and reported that NLRP3 inflammasome is associated with various high-risk reproductive disorders which are in accordance with our findings.[8] Zeng et al. investigated changes in the infiltration of leucocyte subpopulation in the decidual and uterine tissues of the rat model. They explored the role of activation of uterine NLRP3 inflammasomes in pre-eclampsia (PE) and concluded that uterine NLRP3 inflammasome activation and increased macrophage infiltration contribute to an LPS-induced inflammatory response and PE-like symptoms.[32]

With regard to *Fretibacterium* HOT 360, in the present study, their mean concentration was significantly higher in group 2 (Patients with periodontitis and gynaecological disorders) than in patients with gynaecological disorders and without periodontitis at $p<0.05$. In group 1 the presence of Fretibacterium HOT 360 was significantly positively correlated with periodontal parameters (PD and PESA), whereas the plaque score and PISA did not show a positive association with the presence of *Fretibacterium* HOT 360. In group 2, a significant positive association was found between the presence of *Fretibacterium* HOT 360 and periodontal parameters (PD, CAL, PESA, PISA) whereas the plaque score did not show a positive association with the presence of *Fretibacterium* HOT 360. The elevated levels of *Fretibacterium* HOT 360 in group 2 (Patients with periodontitis and gynaecological disorders (PCOS&GDM)) than in group 1 (patients with gynaecological disorders without periodontitis) could be due to elevated levels of uncultivable bacteria in periodontitis as demonstrated by Khemwong et al. who found that the presence of Fretibacterium sp. HOT 360 was significantly higher in the group with periodontitis than in the healthy group.[21]

The results of the present study are consistent with the study by Balan et al., who revealed a correlation between

gingival bleeding and the amounts of *P. gingivalis*, *T. denticola*, *Fretibacterium* sp., and *P. intermedia* in saliva and plaque samples from pregnant women.[33] The results of our findings are in accordance with Oliveira et al., who assessed the prevalence of new periodontal species in subjects with periodontal health (PH), generalized aggressive periodontitis (GAgP) and generalized chronic periodontitis (GChP). They concluded that in subjects with GChP and GAgP *Bacteroidales sp.* HOT 274, *Desulfobulbus* sp. HOT 041, *Fretibacterium sp.* HOT 360 and *Fretibacterium sp.* HOT 362 were found than in than in PH subjects.[35] The present study findings suggest that *Fretibacterium sp.* HOT 360 may be periodontal disease-related bacteria, and associated with gynaecological disorders. Since *Fretibacterium sp.* HOT 360 could not be cultured, its characteristics and virulent factors are still unknown.

To the best of our knowledge, this is the first study assessing *Fretibacterium* HOT 360 and NLRP3 in patients with gynaecological disorders and periodontal disease. Our findings showed that patients with periodontitis and gynaecological disorders had elevated levels of the NLRP3 inflammasome and Fretibacterium sp. HOT 360, suggesting that periodontitis could further increase the inflammatory burden in patients with gynaecological disorders. Hence, an appropriate diagnosis and intervention should be carried out in those patients to prevent periodontal breakdown and also to reduce the systemic inflammatory burden.

However, this study has certain limitations. Studies with larger sample sizes and a more diverse group of participants in terms of severity are needed to improve the reliability of our results. A second limitation is that real-time PCR could have been used to quantify the bacteria. Future studies can also plan for therapeutic intervention to prevent periodontal breakdown and also to reduce the systemic inflammatory burden in patients with periodontal disease and gynaecological disorders.

5. Conclusion

Within the limitations of this study, it can be concluded that there is a definitive association between chronic periodontitis and gynaecological disorders (PCOD&GDM) as demonstrated by the significant levels of NLRP3 inflammasome and *Fretibacterium* HOT 360 levels in patients with periodontitis and gynaecological disorders.

6. Authors Contributions

Madhumathi Devaraj did the acquisition and analysis and drafted the work. Madhumathi Devaraj and Muthukumar Santhanakrishnan interpreted the data. Sri Vidhya Marimuthu did the editing and drafting work. All authors read and approved the final manuscript.

7. Source of Funding

None.

8. Conflict of Interest

None.

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References

1. Hajishengallis G. Periodontitis: from microbial immune subversion to systemic inflammation. *Nat Rev Immunol*. 2015;15(1):30–44.
2. Janakiram C, Mehta A, Venkitachalam R. Prevalence of periodontal disease among adults in India: A systematic review and meta-analysis. *J Oral Biol Craniofacial Res.* 2020;10(4):800–6.
3. Abdulkareem AA, Al-Taweel FB, Al-Sharqi AJB, Gul SS, Sha A, Chapple ILC. Current concepts in the pathogenesis of periodontitis: from symbiosis to dysbiosis. *J Oral Microbiol.* 2023;15(1):2197779.
4. Bhuyan R, Bhuyan SK, Mohanty JN, Das S, Juliana N, Juliana IF. Periodontitis and Its Inflammatory Changes Linked to Various Systemic Diseases: A Review of its Underlying Mechanisms. *Biomedicines.* 2022;10(10):2659.
5. Ye C, Xia Z, Tang J, Khemwong T, Kapila Y, Kuraji R, et al. Unculturable and culturable periodontal-related bacteria are associated with periodontal inflammation during pregnancy and with preterm low birth weight delivery. *Sci Rep.* 2020;10(1):15807.
6. Machado V, Lopes J, Patrão M, Botelho J, Proença L, Mendes JJ. Validity of the association between periodontitis and female infertility conditions: a concise review. *Reproduction*. 2020;160(3):R41–54.
7. Oppermann RV, Weidlich P, Musskopf ML. Periodontal disease and systemic complications. *Braz Oral Res.* 2012;26:39–47.
8. Zhou F, Li C, Zhang SY. NLRP3 inflammasome: a new therapeutic target for high-risk reproductive disorders? *Chin Med J (Engl).* 2021;134(01):20–7.
9. Aral K, Milward MR, Kapila Y, Berdeli A, Cooper PR. Inflammasomes and their regulation in periodontal disease: a review. *J Periodontal Res*. 2020;55(4):473–87.
10. Li Q, Wang H, Liu L, Weng Y, Xu S, Li L, et al. Suppression of the NLRP3 Inflammasome through Activation of the Transient Receptor Potential Channel Melastatin 2 Promotes Osteogenesis in Tooth Extraction Sockets of Periodontitis. *Am J Pathol.* 2023;193(2):213–32.
11. Zhao Y, Quan Y, Lei T, Fan L, Ge X, Hu S. The Role of Inflammasome NLPR3 in the Development and Therapy of Periodontitis. *Int J Med Sci.* 2022;19(10):1603–14.
12. Stein Sr IF. The Stein–Leventhal syndrome: a curable form of sterility. *N Engl J Med.* 1958;259(9):420–3.
13. Zhou Y, Qi H, Yin N. Adaptations and alterations of maternal microbiota: From physiology to pathology. *Med Microecol*. 2021;9:100045.
14. García-Hernández AL, Muñoz-Saavedra ÁE, González-Alva P, Moreno-Fierros L, Llamosas-Hernández FE, Cifuentes-Mendiola SE, et al. Upregulation of proteins of the NLRP3 inflammasome in patients with periodontitis and uncontrolled type 2 diabetes. *Oral Dis.* 2019;25(2):596–608.
15. Nassar H, Kantarci A, Van Dyke TE. Diabetic periodontitis: a model for activated innate immunity and impaired resolution of inflammation. *Periodontol 2000*. 2007;43:233.
16. Preshaw PM, Alba AL, Herrera D, Jepsen S, Konstantinidis A, Makrilakis K, et al. Periodontitis and diabetes: a two-way relationship. *Diabetologia.* 2012;55:21–31.
17. Grover HS, Luthra S. Molecular mechanisms involved in the bidirectional relationship between diabetes mellitus and periodontal disease. *J Indian Soc Periodontol.* 2013;17(3):292.
18. Rojo-Botello NR, García-Hernández AL, Moreno-Fierros L. Expression of toll-like receptors 2, 4 and 9 is increased in gingival

tissue from patients with type 2 diabetes and chronic periodontitis. *J Periodontal Res.* 2012;47(1):62–73.
19. Huang X, Yang X, Ni J, Xie B, Liu Y, Xuan D, et al. Hyperglucose contributes to periodontitis: involvement of the NLRP3 pathway by engaging the innate immunity of oral gingival epithelium. *J Periodontol.* 2015;86(2):327–35.
20. Pérez-Chaparro PJ, Gonçalves C, Figueiredo LC, Faveri M, Lobão E, Tamashiro N, et al. Newly identified pathogens associated with periodontitis: a systematic review. *J Dent Res.* 2014;93(9):846–58.
21. Khemwong T, Kobayashi H, Ikeda Y, Matsuura T, Sudo T, Kano C, et al. Fretibacterium sp. human oral taxon 360 is a novel biomarker for periodontitis screening in the Japanese population. *PLoS One*. 2019;14(6):e0218266.
22. Surlin P, Lazar L, Sincar C, Gheorghe DN, Popescu DM, Boldeanu VM, et al. NLRP3 inflammasome expression in gingival crevicular fluid of patients with periodontitis and chronic hepatitis C. *Mediators Inflamm.* 2021;2021:6917919.
23. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome. *Fertil Steril.* 2004;81(1):19–25.
24. Rawat D, Zangmo R, Chowdhury SR, Yadav AK, Sharma KA, Singh N, et al. Diabetes in Pregnancy Study Group India (DIPSI) and WHO (1999) diagnostic criteria for GDM: A meta-analysis. *Diabetes Metab Syndr Clin Res Rev.* 2022;16(10):102622.
25. Eke PI, Page RC, Wei L, Thornton-Evans G, Genco RJ. Update of the case definitions for population-based surveillance of periodontitis. *J Periodontol.* 2012;83(12):1449–54.
26. Nesse W, Abbas F, Van Der Ploeg I, Spijkervet FKL, Dijkstra PU, Vissink A. Periodontal inflamed surface area: quantifying inflammatory burden. *J Clin Periodontol.* 2008;35(8):668–73.
27. Chandramohan N. Estimaton of NLRP3 Inflammasome & Interleukin-1β Levels in Gingival Crevicular Fluid of Chronic Periodontitis Patients Before & After Scaling and Root Planing–An Interventional Study. Rajiv Gandhi University of Health Sciences (India); 2018.
28. kochar kaur K, Gnk A. Open Access Journal of Gynecology Committed to Create Value for Researchers An Update on the Innovative Part of NLRP3 Inflammasome Regarding Newer Strategies for Treatment of Reproductive Conditions Possessing Greater Risk: A Systematic Review An Update o. *Open Access J Gynecol*. 2022;7:1–13.
29. Holmstrup P, Damgaard C, Olsen I, Klinge B, Flyvbjerg A, Nielsen CH, et al. Comorbidity of periodontal disease: two sides of the same coin? An introduction for the clinician. *J Oral Microbiol.* 2017;9(1):1332710.
30. Isola G, Polizzi A, Santonocito S, Alibrandi A, Williams RC. Periodontitis activates the NLRP3 inflammasome in serum and saliva. *J Periodontol.* 2022;93(1):135–45.
31. Mitra DK, Chavan RR, Prithyani SS, Kandawalla SA, Shah RA, Rodrigues SV. Comparative evaluation of the levels of nod-like receptor family pyrin domain-containing protein (NLRP) 3 in saliva of subjects with chronic periodontitis and healthy controls. *J Indian Soc Periodontol.* 2022;26(3):230.
32. Zeng H, Han X, Zhu Z, Yu S, Mei S, Cheng X, et al. Increased uterine NLRP3 inflammasome and leucocyte infiltration in a rat model of preeclampsia. *Am J Reprod Immunol*. 2021;86(6):e13493.
33. Balan P, Chong YS, Umashankar S, Swarup S, Loke WM, Lopez V, et al. Keystone species in pregnancy gingivitis: a snapshot of oral microbiome during pregnancy and postpartum period. *Front Microbiol*. 2018;9:2360.
34. Abu Fanas S, Brigi C, Varma SR, Desai V, Senok A, D'souza J. The prevalence of novel periodontal pathogens and bacterial complexes in Stage II generalized periodontitis based on 16S rRNA next generation sequencing. *J Appl Oral Sci.* 2021;29:e20200787.
35. Oliveira R, Fermiano D, Feres M, Figueiredo LC, Teles FRF, Soares GMS, et al. Levels of candidate periodontal pathogens in subgingival biofilm. *J Dent Res*. 2016;95(6):711–8.
36. Abusleme L, Dupuy AK, Dutzan N, Silva N, Burleson JA, Strausbaugh LD, et al. The subgingival microbiome in health and periodontitis and its relationship with community biomass and inflammation. *ISME J*. 2013;7(5):1016–25.

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