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Original Article

Biological markers and pulp pH values for the clinical prediction of suppurative pulpitis

Meng Jiang*, Haiqi Liu*

*Wuxi Stomatology Hospital, Wuxi 214000, Jiangsu, China. *Co-first authors.*

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Abstract: Objective: This study aims to explore the association between abnormal biomarkers in gingival crevicular fluid and pulp blood pH values and the occurrence of suppurative pulpitis. Methods: A retrospective analysis was conducted on the clinical data of 203 patients who visited Wuxi Stomatological Hospital from March 2024 to March 2025. According to clinical manifestations, the patients were divided into the chronic suppurative pulpitis group (61 cases), the acute suppurative pulpitis group (65 cases), and the orthodontic control group (77 cases). Logistic regression analysis was used to screen the risk factors for suppurative pulpitis, and a forest plot was drawn based on the analysis results. Additionally, a receiver operating characteristic (ROC) curve was plotted to evaluate the discriminatory power of the model. Moreover, scatter plots of the correlation between pulp blood pH values and various gingival crevicular fluid markers were drawn, and the predictive ability of each marker for abnormal pH values was evaluated using the ROC curve. Results: Pearson correlation analysis showed that IL-1β and CX3CR1 were negatively correlated with the pH value of dental pulp blood. CX3CR1 was positively correlated with the VAS pain score. Clustering heatmap showed that CX3CR1, IL-1β, TNF-α, PTX-3, and TREM-1 were highly clustered in the acute group. Multivariate Logistic regression analysis identified VAS pain score, IL-1β, and TNF-α as independent risk factors. ROC curve showed that VAS pain score, IL-1β, and TNF-α had good predictive abilities for suppurative pulpitis; IL-1β, TNF-α, CX3CR1 and ICAM-1 also showed good predictive abilities for abnormal pH values. Conclusion: VAS pain score, IL-1β, and TNF-α were identified as independent predictors of suppurative pulpitis.

Keywords: Suppurative pulpitis, biomarker, pulp blood, pH, independent predictors

Introduction

Suppurative pulpitis is a suppurative inflammatory lesion caused by the invasion and infection of pulp tissue by pathogenic microorganisms, and the pathological features include local abscess formation, inflammatory cell infiltration and tissue necrosis [1]. This disease usually occurs in the progressive stage of pulpitis. At the site of inflammation, white blood cells undergo necrosis, leading to the formation of a localized abscess in the pulp cavity; this development marks the severe stage of pulp lesions [2, 3]. Data released by the European Society of Endodontology indicates that approximately 50% of adults worldwide suffer from dental diseases caused by pulp infections, among which suppurative pulpitis ranks as one of the top three causes of non-traumatic dental visits [4, 5]. Suppurative pulpitis is usually accompanied by acute and severe pain that rarely recovers on its own. This increases the patient's suffering and economic burden, and affects the patient's everyday life and work situation [6].

Research has found that the changes in molecular levels in the tissues of suppurative pulpitis occur earlier than those in the cytology and histology of the dental pulp [7]. The precedence of such molecular events suggests that the detection of relevant biomarkers may hold potential value for predicting the direction and pace of disease progression. Some studies have also found that histological changes in the dental pulp precede the clinical onset of suppurative pulpitis [8]. Therefore, detecting molecular-level markers of pulpitis can reflect the status and progression of pulp lesions in patients, which is conducive to diagnosis and treatment [9, 10]. Based on the above situation, this study detected biomarkers in gingival crevicular fluid and measured pulp blood pH values in patients with

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acute or chronic suppurative pulpitis. These values were then compared with those from a control group of patients who needed tooth extraction for orthodontic reasons but had no clinical symptoms of pulpitis. The differences in expression levels and changes among the three groups were analyzed. Based on this, the association of biomarkers and pH value with the occurrence of suppurative pulpitis was explored to guide early clinical assessment and treatment, with the goal of preventing disease progression and reducing patient suffering.

Materials and methods

Study design

The sample size was estimated for the planned logistic regression model with 23 independent variables. Using a rule of thumb of 5-10 subjects per variable and a documented suppurative pulpitis incidence of 66.12% in Wuxi Stomatology Hospital, the initial sample size (N) was calculated as follows: $N=(23\times5)/0.6612\approx174$. Accounting for a potential 10% data loss, at least 192 samples are required.

Setting and participants

This study retrospectively included 235 patients who visited Wuxi Stomatological Hospital from March 2024 to March 2025 and met the inclusion criteria. After excluding 15 patients who did not complete the full treatment and 17 patients with incomplete medical records, a total of 203 valid cases were finally selected. Among them, 126 patients were diagnosed with suppurative pulpitis, including the acute group (n=65) and chronic group (n=61). Additionally, 77 patients who needed tooth extraction due to orthodontic requirements and had no clinical symptoms of pulpitis were included as the control group. The final sample size met the minimum sample size requirement for the study. The study protocol was approved by the Biomedical Ethics Review Committee of Wuxi Stomatological Hospital [Approval Number: 2025082901].

Diagnostic criteria: The diagnostic basis was the clinical guidelines released by the European Society of Endodontology in 2023 [4]. The clinical symptoms included sensitivity to hot and cold stimulation, positive temperature test, positive percussion pain. X-ray examination revealed low-density shadows at the root apex, abnormal expansion of the dental pulp cavity, dental pulp tissue lesions, and involvement of the periapical tissues. In addition, patients with acute suppurative pulpitis experience severe pain, spontaneous pain, referred pain, and significant nocturnal pain, which affects sleep. Cardinal features included bleeding on probing and hyperemic, edematous gingival papillae. These symptoms were accompanied by persistent pain, significant percussion sensitivity, and increased pain during chewing. Patients in the chronic group primarily experienced a dull pain, which is triggered by temperature changes, chewing pressure, or other stimuli, and lasts for a longer period [11].

Inclusion criteria: 1) Fulfillment of diagnostic criteria for acute or chronic suppurative pulpitis (acute or chronic group); 2) Treatment requirement of tooth extraction for orthodontic reasons without pulpitis symptoms (control group); 3) Management by the same medical team; 4) Availability of complete medical records and data. Exclusion criteria: 1) Recent use of antibiotics or immunosuppressants; 2) Other oral diseases such as oral ulcers and periodontitis; 3) Systemic inflammatory diseases; 4) Diabetes; 5) Abnormal coagulation function; 6) Incomplete medical records or data.

Determination of pulp biomarkers and pH

Gingival crevicular fluid: All patients' gingival crevicular fluid samples before treatment were collected by the conventional filter paper strip sampling method. Filter paper strips (Waterman Company, UK; specification: 2 mm×10 mm) were inserted into the bottom of the labial gingival sulcus and retained for 30 seconds, then placed in a centrifuge tube and shaken, after which the supernatant was collected. Procedures followed kit instructions.

Pulp blood: The affected tooth was extracted under local anesthesia, and a high-speed diamond drill with water-cooling was used to open the pulp access cavity. After the medullary cavity was penetrated, and bleeding was observed, the medullary blood was collected from the coronal medulla using a sterile No. 21 needle (Zarys, dicoNEX Co., Zabrze, Poland).

Table 1. Comparison of general information

Variables	Control group (n=77)	Suppurative pulpitis		F/χ^2	P
		Chronic group (n=61)	Acute group (n=65)		
Age (years, $\bar{x}\pm s$)	43.62±9.56	42.77±9.12	40.43±10.13	2.028	0.134
Gender (n, %)					
Male	33 (42.90)	34 (55.70)	35 (53.80)	1.376	0.255
Female	44 (57.10)	27 (44.30)	30 (46.20)		
BMI (kg/m^2, $\bar{x}\pm s$)	23.06±3.26	23.24±3.41	23.16±3.27	0.055	0.946
Education level (n, %)					
High school and below	40 (51.95)	29 (47.54)	28 (43.08)	0.552	0.577
College degree and above	37 (48.05)	32 (52.46)	37 (56.92)		
Alcohol habit (n, %)					
Yes	15 (19.48)	16 (26.23)	14 (21.54)	0.92	0.631
No	62 (80.52)	45 (73.77)	51 (78.46)		
Smoking habit (n, %)					
Yes	28 (36.36)	16 (26.23)	18 (27.69)	2.014	0.365
No	49 (63.64)	45 (73.77)	47 (72.31)		
FPG (mmol/L, $\bar{x}\pm s$)	8.44±1.97	8.38±1.90	8.53±2.03	0.086	0.918

Note: BMI, body mass index; FPG, fasting plasma glucose.

Clinical assessment

Visual Analogue Scale (VAS) pain score [12, 13] was used to evaluate the degree of pain before treatment in the three groups of patients. A 10-centimeter horizontal linear VAS measurement scale was used, with “0 points” and “10 points” marked at both ends. “0 points” represented “complete absence of pain”, while “10 points” indicated “the most intense pain imaginable”. During the assessment, the researcher explained the meaning and usage of the scale in detail to the participants, and the participants marked the corresponding position on the line based on their current perceived pain intensity. The researcher measured the centimeter distance between the marked point and the “0 points” end to convert it into a 0-10 point value, thereby transforming the subjective pain perception of the patients into quantifiable and comparable objective data, enabling an accurate assessment of their pain intensity.

Laboratory assessment

Biomarker detection of gingival crevicular fluid: The levels of Interleukin-1β (IL-1β), Tumor Necrosis Factor-alpha (TNF-α), Tissue Inhibitor of Metalloproteinases-1 (TIMP-1), Triggering Receptor Expressed on Myeloid Cells-1 (TREM-1), Pentraxin 3 (PTX3), Cluster of Differentiation 14 (CD14), Intercellular Adhesion Molecule-1 (ICAM-1), and High Mobility Group Box 1 (HMGB-1) were quantified using enzyme-linked immunosorbent assay with commercial kits from Shanghai Beyotime Biotechnology Co., Ltd. The level of C-X3-C Motif Chemokine Receptor 1 (CX3CR1) was measured using the kit from Xuanya Biotechnology Co., Ltd.

Detection of pulp blood pH value: Measurements were performed using the Horiba LAQUAtwin PH-33 device (Horiba Advanced Technology Co., Ltd., Kyoto, Japan).

Observation indicators

Potential factors associated with suppurative pulpitis were identified through literature review and consultation with dental specialists. The collected data were entered into an Excel spreadsheet and cross-verified by two independent researchers to ensure accuracy. Case data with abnormal or missing values were excluded. The following data were collected from all patients: 1) Primary indicators: pulpitis-related parameters (including VAS pain scale, pH value of pulp blood, spontaneous pain duration, and pulp vitality) and gingival crevicular fluid biomarkers (IL-1β, TNF-α, CX3CR1, TIMP-1, TREM-1, PTX3, CD14, HMGB-1and ICAM-1).

Table 2. Comparison of pulpitis-related parameters

Variables	Control group (n=77)	Suppurative pulpitis		F/χ^2	P
		Chronic group (n=61)	Acute group (n=65)		
WBC (×10^9/L, $\bar{x}$±s)	8.78±3.16	9.51±4.19	11.02±5.22*	5.079	0.007
CRP (mg/L, $\bar{x}$±s)	15.11±3.65	15.32±4.08	15.70±5.21	0.322	0.725
Site of the affected tooth (n, %)					
Incisor	26 (33.77)	15 (24.59)	15 (23.08)	3.668	0.453
Premolar	33 (42.86)	26 (42.62)	27 (41.54)		
Molar	18 (23.38)	20 (32.79)	23 (35.38)		
Depth of dental caries (mm, $\bar{x}$±s)	2.36±1.00	2.40±0.87	2.42±0.86	0.097	0.908
spontaneous pain duration (day, $\bar{x}$±s)	3.43±1.59	4.00±1.95*	4.23±1.79*	3.922	0.021
VAS pain scale (points, $\bar{x}$±s)	2.85±1.34	5.92±1.86*	6.70±2.29*	87.639	0.000
Pulp vitality test (n, %)					
Normal	75 (97.40)	55 (90.16)	48 (73.85)	20.722	0.000
Hypoactive	2 (2.60)	5 (8.20)	10 (15.38)*,#		
Unresponsive	0 (0.00)	1 (1.64)	7 (10.77)*,#		
Gingival crevicular fluid biomarkers					
IL-1β (ng/L, $\bar{x}$±s)	16.06±3.32	22.08±3.34*	32.77±5.41*,#	294.739	0.000
TNF-α (μg/L, $\bar{x}$±s)	20.50±4.05	26.55±4.52*	40.00±8.93*,#	181.531	0.000
TIMP-1 (μg/L, $\bar{x}$±s)	2.96±1.25	3.20±1.30	3.47±1.58	2.462	0.088
TREM-1 (ng/L, $\bar{x}$±s)	10.20±3.04	10.36±1.28	11.88±1.19*,#	12.930	0.000
PTX3 (μg/L, $\bar{x}$±s)	11.98±3.96	13.27±5.45	16.23±4.62*,#	15.126	0.000
CD14 (mg/L, $\bar{x}$±s)	2.77±0.59	2.92±0.54	2.93±0.39	2.166	0.117
CX3CR1 (μg/L, $\bar{x}$±s)	22.31±4.11	33.00±5.32*	53.58±7.58*,#	525.859	0.000
ICAM-1 (μg/L, $\bar{x}$±s)	234.15±55.42	296.20±23.89*	327.66±38.99*,#	88.776	0.000
HMGB-1 (μg/L, $\bar{x}$±s)	10.03±2.20	11.21±3.79	11.95±1.42*	9.874	0.000

Note: Compared with control group, *P<0.05; Compared with the chronic group, #P<0.05. WBC, white blood cell count; CRP, C-reactive protein; VAS pain scale, Visual Analogue Scale for pain; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha; TIMP-1, Tissue Inhibitor of Metalloproteinases-1; TREM-1, Triggering Receptor Expressed on Myeloid Cells-1; PTX3, Pentraxin 3; CD14, Cluster of Differentiation 14; ICAM-1, Intercellular Adhesion Molecule-1; HMGB-1, High Mobility Group Box 1; CX3CR1, C-X3-C Motif Chemokine Receptor 1.

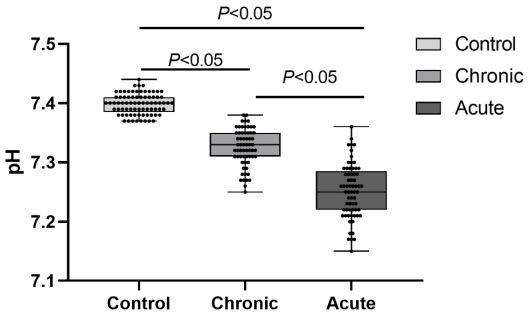

Figure 1. Box plots of pulp blood pH value in the three groups.

2) Secondary indicators: age, gender, BMI, education level, alcohol habit, smoking habit, fasting plasma glucose, white blood cell count, C-reactive protein, site of the affected tooth, and depth of dental caries.

Statistical analysis

All data were processed and analyzed using SPSS 26.0 statistical software. Count data are expressed as cases (rates) [n (%)] and analyzed using the χ^2 test. The Kolmogorov-Smirnov test was applied to assess normality. Normally distributed continuous variables are expressed as $\bar{x}$±s and compared using the independent samples t-test. Non-normally distributed data are presented as median (Q_{25}, Q_{75}) and analyzed using the Kruskal-Wallis H test. RStudio (R4.3.3) was used to draw: box line scatter plot of pulp blood pH value, scatter plot

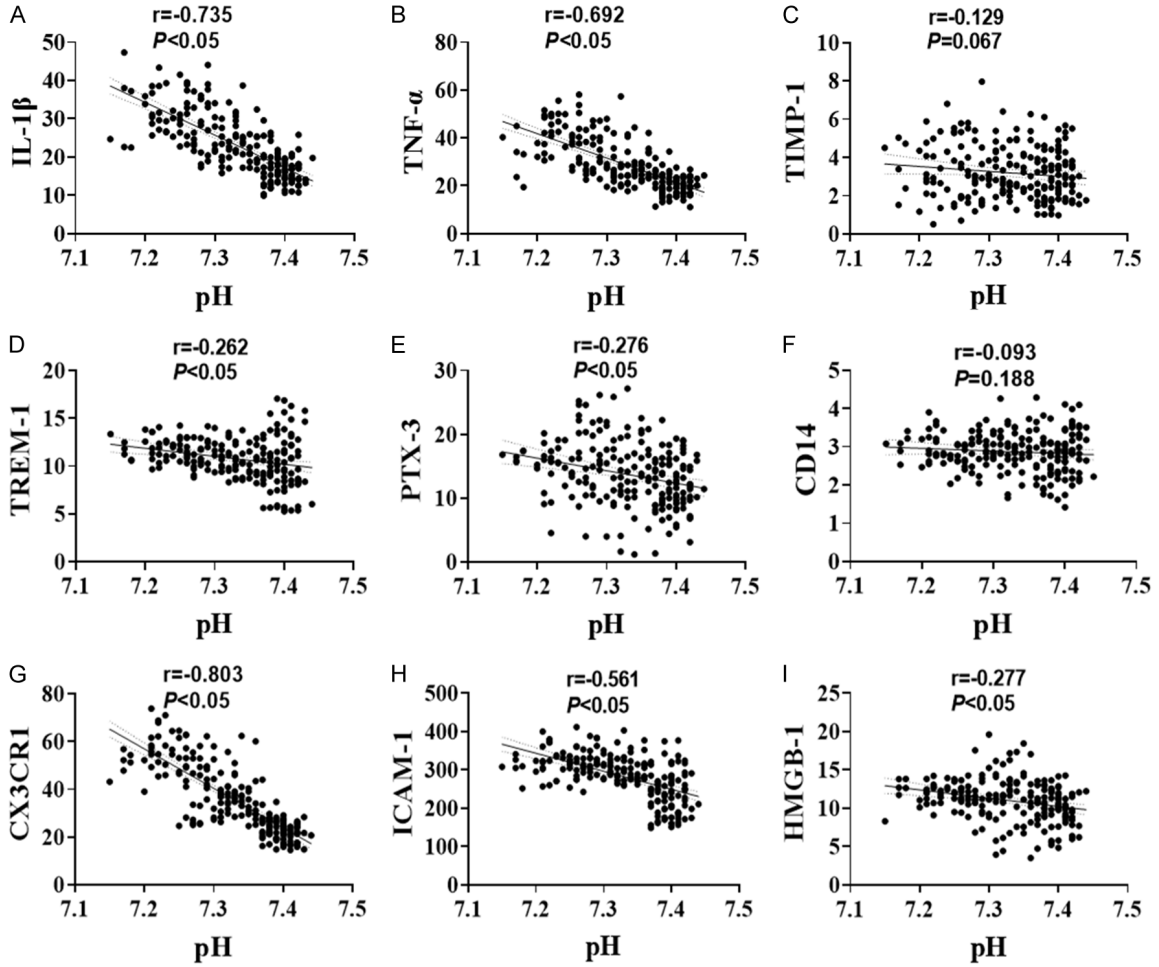

Figure 2. Scatter plots of the correlation between gingival crevicular fluid biomarkers and pulp blood pH value. Note: A. Scatter plot of the correlation between pulp blood pH value and IL-1β level in gingival crevicular fluid. B. Scatter plot of the correlation between pulp blood pH value and TNF-α level in gingival crevicular fluid. C. Scatter plot of the correlation between pulp blood pH value and TIMP-1 level in gingival crevicular fluid. D. Scatter plot of the correlation between pulp blood pH value and TREM-1 level in gingival crevicular fluid. E. Scatter plot of the correlation between pulp blood pH value and PTX3 level in gingival crevicular fluid. F. Scatter plot of the correlation between pulp blood pH value and CD14 level in gingival crevicular fluid. G. Scatter plot of the correlation between pulp blood pH value and CX3CR1 level in gingival crevicular fluid. H. Scatter plot of the correlation between pulp blood pH value and ICAM-1 level in gingival crevicular fluid. I. Scatter plot of the correlation between pulp blood pH value and HMGB-1 level in gingival crevicular fluid. IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha; TIMP-1, Tissue Inhibitor of Metalloproteinases-1; TREM-1, Triggering Receptor Expressed on Myeloid Cells-1; PTX3, Pentraxin 3; CD14, Cluster of Differentiation 14; ICAM-1, Intercellular Adhesion Molecule-1; HMGB-1, High Mobility Group Box 1; CX3CR1, C-X3-C Motif Chemokine Receptor 1.

of correlation between pulp blood pH value and each biomarker, scatter plot of correlation between biomarker and VAS score, heat map of biomarker level clustering, and forest plot of logistic regression analysis. Multivariate Logistic regression was used to analyze the independent predictors of suppurative pulpitis and the influence of various gingival crevicular fluid biomarkers on the pH value of pulp blood. $P<0.05$ was considered statistically significant.

Results

Comparison of general information data

There were no significant differences in general information among the three groups of patients (all $P>0.05$). See **Table 1**.

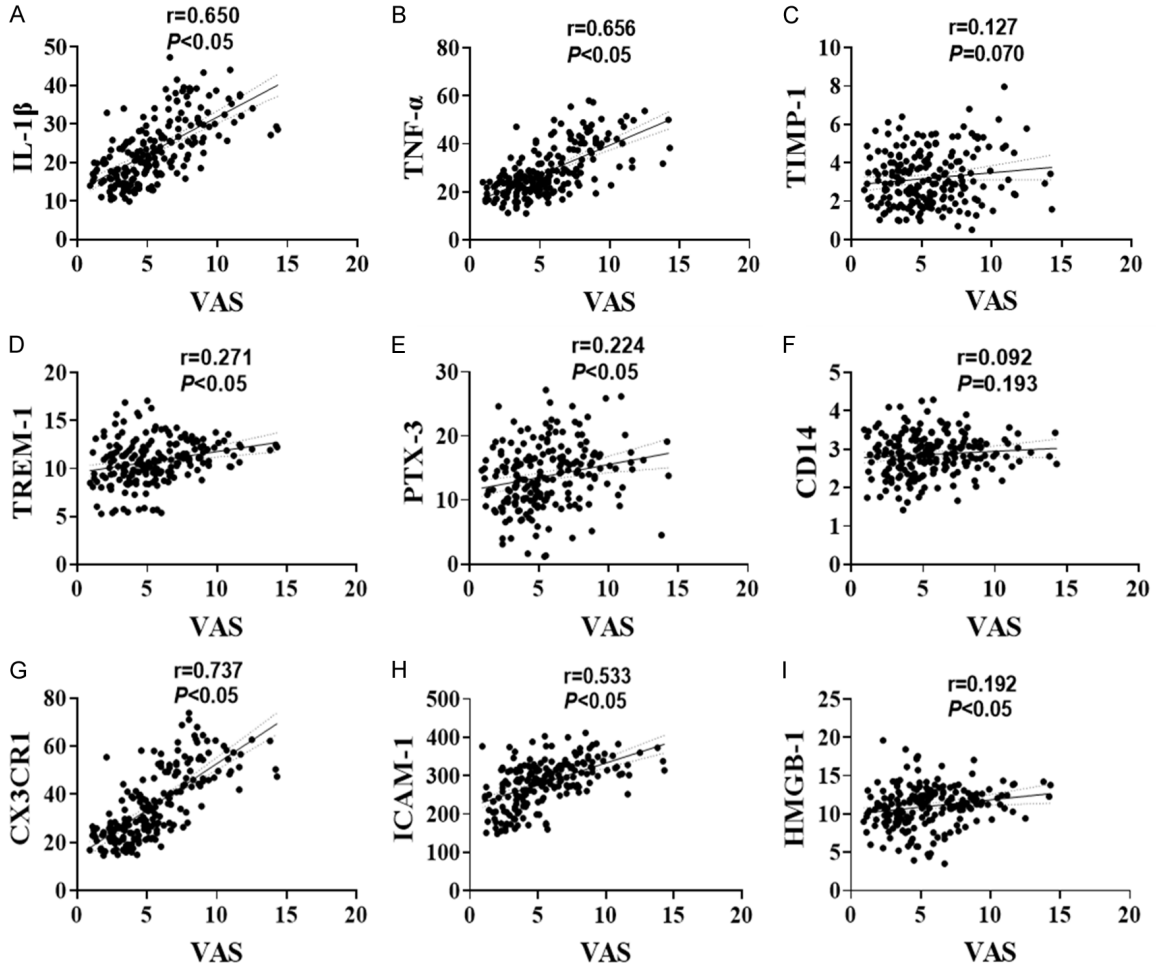

Figure 3. Scatter plots of the correlation between gingival crevicular fluid biomarkers and VAS pain scale. Note: A. Scatter plot of the correlation between IL-1β levels in gingival crevicular fluid and the VAS pain score of patients. B. Scatter plot of the correlation between TNF-α levels in gingival crevicular fluid and the VAS pain score of patients. C. Scatter plot of the correlation between TIMP-1 levels in gingival crevicular fluid and the VAS pain score of patients. D. Scatter plot of the correlation between TREM-1 levels in gingival crevicular fluid and the VAS pain score of patients. E. Scatter plot of the correlation between PTX3 levels in gingival crevicular fluid and the VAS pain score of patients. F. Scatter plot of the correlation between CD14 levels in gingival crevicular fluid and the VAS pain score of patients. G. Scatter plot of the correlation between CX3CR1 levels in gingival crevicular fluid and the VAS pain score of patients. H. Scatter plot of the correlation between ICAM-1 levels in gingival crevicular fluid and the VAS pain score of patients. I. Scatter plot of the correlation between HMGB-1 levels in gingival crevicular fluid and the VAS pain score of patients. VAS pain scale, Visual Analogue Scale for pain; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha; TIMP-1, Tissue Inhibitor of Metalloproteinases-1; TREM-1, Triggering Receptor Expressed on Myeloid Cells-1; PTX3, Pentraxin 3; CD14, Cluster of Differentiation 14; ICAM-1, Intercellular Adhesion Molecule-1; HMGB-1, High Mobility Group Box 1; CX3CR1, C-X3-C Motif Chemokine Receptor 1.

Comparison of pulpitis-related parameters

The pulpitis-related parameters among the three groups of patients are shown in **Table 2**. The duration of spontaneous pain, VAS pain scale, IL-1β, TNF-α, CX3CR1 and ICAM-1 in the chronic group were higher than those in the control group (all $P<0.05$). The WBC, duration of spontaneous pain, VAS pain scale, the proportion of sluggish and unresponsive responses in pulp vitality tests, IL-1β, TNF-α, TREM-1, PTX3, CX3CR1, ICAM-1, and HMGB-1 in the acute group were higher than those in the control group (all $P<0.05$). The proportion of blunted and unresponsive pulp vitality, IL-1β, TNF-α, TREM-1, PTX3, CX3CR1 and ICAM-1 in acute group were higher than those in chronic group (all $P<0.05$).

Figure 4. Clustering heat maps of gingival crevicular fluid biomarkers. Note: IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha; TIMP-1, Tissue Inhibitor of Metalloproteinases-1; TREM-1, Triggering Receptor Expressed on Myeloid Cells-1; PTX3, Pentraxin 3; CD14, Cluster of Differentiation 14; ICAM-1, Intercellular Adhesion Molecule-1; HMGB-1, High Mobility Group Box 1; CX3CR1, C-X3-C Motif Chemokine Receptor 1.

Comparison of pulp blood pH values

The pulp blood pH values among three groups of patients are shown in **Figure 1**. The pH value of pulp blood in the acute group (7.25±0.05) was lower than that in the chronic group (7.32±0.03) and the control group (7.40±0.02), and the pH value of pulp blood in the chronic group was lower than that in the orthodontic control group (all $P<0.05$).

Correlation analysis of gingival crevicular fluid biomarkers and pulp blood pH value

The levels of IL-1β (r=-0.735) and CX3CR1 (r=-0.803) in the gingival crevicular fluid of the patients were negatively correlated with the pH value of dental pulp blood (both $P<0.05$). TNF-α (r=-0.692), TREM-1 (r=-0.262), PTX3 (r=-0.276), ICAM-1 (r=-0.561), and HMGB-1 (r=-0.277) showed a weak negative correlation with the pH value of pulp blood (both $P<0.05$). CD14 (r=-0.093, P=0.188) and TIMP-1 (r=-0.129, P=0.067) showed no correlation with the pH value of pulp blood. See **Figure 2**.

Correlation analysis of gingival crevicular fluid biomarkers and pain degree

CX3CR1 (r=0.737) in the gingival crevicular fluid of patients was positively correlated with the VAS pain scales ($P<0.05$). IL-1β (r=0.650), TNF-α (r=0.656) TREM-1 (r=0.271), PTX3 (r=0.224), ICAM-1 (r=0.533), and HMGB-1 (r=0.192) showed a weak positive correlation with the VAS pain scales (all $P<0.05$). TIMP-1 (r=0.127, P=0.070), and CD14 (r=0.092, 0.193) showed no correlation with the VAS pain scales. See **Figure 3**.

Analysis of gingival crevicular fluid biomarker levels

The biomarker levels among the three groups of patients showed obvious clustering characteristics. Compared with the chronic group and the control group, CX3CR1, IL-1β, TNF-α, PTX-3, and TREM-1 were highly clustered in the acute group. See **Figure 4**.

Univariate logistic regression analysis of predictors for suppurative pulpitis

Taking whether suppurative pulpitis occurred as the dependent variable (assignment information: no suppurative pulpitis occurred =0, suppurative pulpitis occurred =1), all indicators were included as independent variables for univariate logistic regression analysis. The results showed that WBC (odds ratio [OR]=1.090, 95% confidence interval [CI]: 1.016-1.169), VAS pain scales (OR=3.806,

Table 3. Univariate Logistic regression analysis of predictors for suppurative pulpitis

Characteristics	B	S.E	Wald χ^2	*P*	*OR*	95% *CI*
Age (years)	-0.022	0.015	2.161	0.142	0.978	0.949-1.007
Gender	0.479	0.292	2.694	0.101	1.614	0.911-2.859
BMI (kg/m^2)	-0.012	0.046	0.067	0.796	0.988	0.902-1.082
Education level	-0.269	0.290	0.861	0.353	0.764	0.433-1.349
Alcohol habit	0.256	0.356	0.518	0.472	1.292	0.643-2.594
Smoking habit	0.436	0.310	1.970	0.160	1.546	0.841-2.842
FPG (mmol/L)	0.003	0.074	0.002	0.968	1.003	0.868-1.160
WBC (×10^9/L)	0.086	0.036	5.712	0.017	1.090	1.016-1.169
CRP (mg/L)	0.022	0.034	0.417	0.518	1.022	0.957-1.092
Site of the affected tooth						
Incisor			3.519	0.172		
Premolar	-0.728	0.388	3.516	0.061	0.483	0.226-1.033
Molar	-0.397	0.281	1.232	0.267	0.672	0.333-1.355
Depth of dental caries (mm)	0.062	0.158	0.154	0.694	1.064	0.780-1.452
Spontaneous pain duration (days)	0.224	0.085	6.894	0.009	1.251	1.058-1.478
VAS pain scale (points)	1.337	0.193	48.205	0.000	3.806	2.610-5.551
Pulp vitality test						
Normal			4.887	0.087		
Hypoactive	-20.886	14210.440	0.000	0.999	0.000	0.000
Unresponsive	-19.203	14210.440	0.000	0.999	0.000	0.000
IL-1β (ng/L)	0.545	0.083	43.567	0.000	1.725	1.467-2.028
TNF-α (μg/L)	0.358	0.057	39.487	0.000	1.430	1.279-1.599
TIMP-1 (μg/L)	0.207	0.109	3.610	0.057	1.230	0.993-1.523
TREM-1 (ng/L)	0.198	0.070	8.141	0.004	1.219	1.064-1.398
PTX3 (μg/L)	0.125	0.033	14.310	0.000	1.133	1.062-1.209
CD14 (mg/L)	0.581	0.284	4.199	0.040	1.788	1.026-3.116
CX3CR1 (μg/L)	0.569	0.109	27.344	0.000	1.767	1.427-2.187
ICAM-1 (μg/L)	0.038	0.005	48.761	0.000	1.039	1.028-1.050
HMGB-1 (μg/L)	0.232	0.061	14.556	0.000	1.261	1.119-1.420

Note: S.E, Standard Error; OR, odds ratio; 95% CI, 95% confidence interval; BMI, body mass index; FPG, fasting plasma glucose; WBC, white blood cell count; CRP, C-reactive protein; VAS pain scale, Visual Analogue Scale for Pain; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha; TIMP-1, Tissue Inhibitor of Metalloproteinases-1; TREM-1, Triggering Receptor Expressed on Myeloid Cells-1; PTX3, Pentraxin 3; CD14, Cluster of Differentiation 14; ICAM-1, Intercellular Adhesion Molecule-1; HMGB-1, High Mobility Group Box 1; CX3CR1, C-X3-C Motif Chemokine Receptor 1.

Table 4. Multivariate Logistic regression of predictive factors for suppurative pulpitis

Characteristics	B	S.E	Wald χ^2	*P*	*OR*	95% *CI*
VAS pain score (points)	1.498	0.366	16.789	0.000	4.473	2.185-9.158
IL-1β (ng/L)	0.564	0.143	15.531	0.000	1.757	1.328-2.326
TNF-α (μg/L)	0.300	0.096	9.791	0.002	1.349	1.118-1.628

Note: S.E, Standard Error; OR, odds ratio; 95% CI, 95% confidence interval; VAS pain scale, Visual Analogue Scale for pain; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha.

95% CI: 2.610-5.551), IL-1β (OR=1.725, 95% CI: 1.467-2.028), TNF-α (OR=1.430, 95% CI: 1.279-1.599), TREM-1 (OR=1.219, 95% CI: 1.064-1.398), PTX3 (OR=1.133, 95% CI: 1.062-1.209), CD14 (OR=1.788, 95% CI: 1.026-3.116), CX3CR1 (OR=1.767, 95% CI: 1.427-2.187), ICAM-1 (OR=1.039, 95% CI: 1.028-1.050), HMGB-1 (OR=1.261, 95% CI: 1.119-

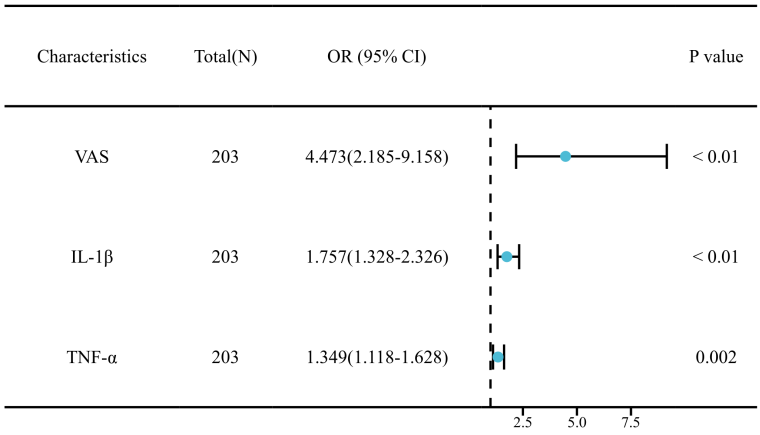

Figure 5. Forest map of influencing factors for suppurative pulpitis. Note: OR, odds ratio; CI, confidence interval; VAS, Visual Analogue Scale; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha.

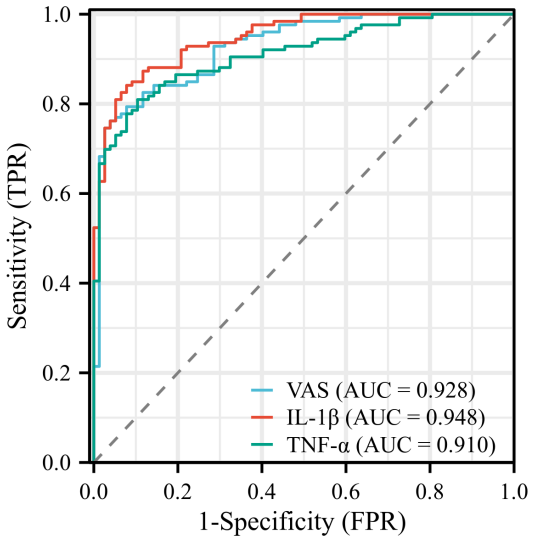

Figure 6. ROC curve of the independent predictive factor model for suppurative pulpitis. Note: ROC, receiver operating characteristic; TPR, true positive rate; FPR, false positive rate; VAS, Visual Analogue Scale; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha.

1.420) were related to the occurrence of purulent pulpitis (all $P<0.05$). See **Table 3**.

Multivariate logistic regression analysis of predictors for suppurative pulpitis

Taking whether suppurative pulpitis occurred as the dependent variable (assignment information: no suppurative pulpitis occurred =0, suppurative pulpitis occurred =1), indicators identified through inter-group difference analysis as having significant differences were entered into the model as independent variables. After screening the independent variables by the forward method, binary logistic regression analysis was conducted. The results showed that VAS pain scales (OR=4.473, 95% CI: 2.185-9.158), IL-1β (OR=1.757, 95% CI: 1.328-2.326), TNF-α (OR=1.349, 95% CI: 1.118-1.628) were independent predictors for suppurative pulpitis in patients (all $P<0.05$). See **Table 4**. The results of the forest plot are shown in **Figure 5**. It displayed that VAS pain score, IL-1β, and TNF-α were all positively correlated influencing factors and did not intersect with the invalid line (all $P<0.05$).

Validation of the prediction model for suppurative pulpitis

The ROC curve results are shown in **Figure 6** and **Table 5**. VAS pain score (AUC=0.928), IL-1β (AUC=0.948), and TNF-α (AUC=0.910) exhibited good predictive ability for suppurative pulpitis.

The value assessment of gingival crevicular fluid biomarkers for the predictive model of pulp blood pH value

The results of evaluating the predictive ability of gingival crevicular fluid biomarkers for pulp blood pH value by ROC curve are shown in **Figure 7** and **Table 6**. According to the pH value conditions of patients in each group, the normal pH value range was set at 7.37-7.44, and the abnormal pH value range was set at 7.15-7.36. Taking the pH value of pulp blood as the state variable (normal or abnormal) and the levels of each gingival crevicular fluid biomarker as the detection variable, the ROC curve was drawn. The results showed that IL-1β (AUC=0.928), TNF-α (AUC=0.905), CX3CR1 (AUC=0.966), and ICAM-1 (AUC=0.867) had good predictive abilities for abnormal pH values.

Table 5. The predictive value of independent predictors for suppurative pulpitis

Characteristics	Optimal cutoff value	AUC	95% *CI*	Sensitivity	Specificity	*J*
VAS pain score (points)	4.842	0.928	0.893-0.963	0.762	0.961	0.723
IL-1β (ng/L)	20.609	0.948	0.921-0.975	0.841	0.922	0.763
TNF-α (μg/L)	24.378	0.910	0.872-0.949	0.810	0.896	0.706

Note: AUC, area under the curve; CI, confidence interval; VAS pain scale, Visual Analogue Scale for pain; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha.

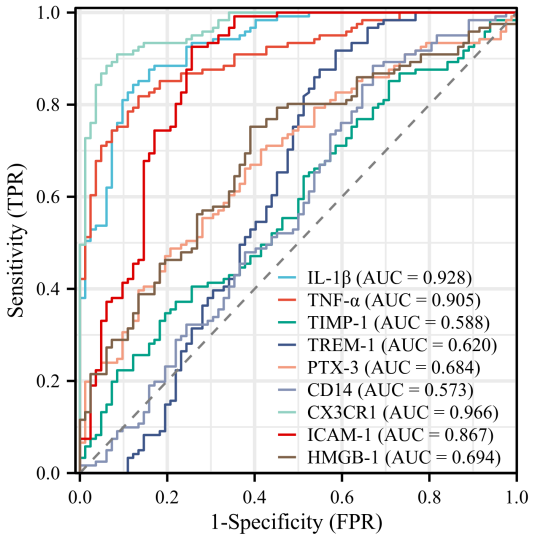

Figure 7. ROC curve of gingival crevicular fluid markers for predicting abnormal pulp pH. Note: ROC, receiver operating characteristic; TPR, true positive rate; FPR, false positive rate; AUC, area under the curve; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha; TIMP-1, Tissue Inhibitor of Metalloproteinases-1; TREM-1, Triggering Receptor Expressed on Myeloid Cells-1; PTX3, Pentraxin 3; CD14, Cluster of Differentiation 14; ICAM-1, Intercellular Adhesion Molecule-1; HMGB-1, High Mobility Group Box 1; CX3CR1, C-X3-C Motif Chemokine Receptor 1.

Discussion

Suppurative pulpitis is an inflammatory reaction in the pulp cavity caused by pathogenic bacteria infection and cold or hot stimulation during eating [14]. Once suppurative pulpitis occurs, a series of subtle but significant changes will take place in the pulp cavity. The most notable of these is the alteration in the levels of various inflammatory factors. These inflammatory factors are like the “commanders” and “messengers” in the inflammatory response. In the pulp cavity, excessive expression of inflammatory factors may indirectly damage nearby healthy tissues, thus aggravating the severity of the disease [14]. The occurrence of suppurative pulpitis is accompanied by changes in the levels of various inflammatory factors, which play a crucial regulatory role in the progression of the disease [15]. The results of this study indicate that the levels of TNF-α and IL-1β in the gingival crevicular fluid of patients, as well as the VAS pain score are independent predictors for suppurative pulpitis. This finding may serve as a reference for the early identification of suppurative pulpitis, assessment of disease severity, and formulation of treatment plans.

In purulent pulpitis, tumor necrosis factor-α and interleukin-1β activate the nuclear factor κ-B and filament activation protein kinase signaling pathway. This activation will eventually lead to the destruction and disorder of normal skin structure, and will also promote the production of matrix metalloproteinase [16]. In addition, IL-1β can activate neutrophil protease, thus promoting the formation of local abscess [16, 17]. This study found that the levels of IL-1β and TNF-α in the gingival crevicular fluid of patients with suppurative pulpitis were significantly higher than those in the control group, and their pain VAS scores were also markedly elevated. These findings are consistent with the research results reported [18, 19]. This may be because TNF-α triggers an inflammatory mediator reaction in the stimulated pulp tissue. In purulent pulpitis, IL-1β not only activates inflammatory reactions with TNF-α, but also activates neutrophil protease and promotes the formation of pulp tissue abscess, which is consistent with the research results of Ghosh et al. [20]. This also explains why the VAS pain scores of patients with suppurative pulpitis is elevated, which is consistent with the research findings of Gopalsamy et al [21]. In conclusion, the VAS pain score is a valid diagnostic measure for suppurative pulpitis, and it is evident that VAS is an essential tool for comprehending the intricate pathological mechanisms of diseases rather than just a symptom indication.

Table 6. The predictive value of gingival crevicular fluid markers for abnormal pH of dental pulp tissue

Characteristics	Optimal cutoff value	AUC	95% *CI*	Sensitivity	Specificity	*J*
IL-1β (ng/L)	20.609	0.928	0.894-0.963	0.843	0.878	0.721
TNF-α (μg/L)	24.378	0.905	0.866-0.945	0.818	0.866	0.684
TIMP-1 (μg/L)	3.826	0.588	0.509-0.667	0.372	0.780	0.152
TREM-1 (ng/L)	9.300	0.620	0.532-0.707	0.917	0.415	0.332
PTX3 (μg/L)	12.495	0.684	0.611-0.758	0.702	0.585	0.288
CD14 (mg/L)	2.442	0.573	0.489-0.656	0.884	0.329	0.214
CX3CR1 (μg/L)	28.154	0.966	0.945-0.987	0.909	0.915	0.824
ICAM-1 (μg/L)	270.590	0.867	0.810-0.923	0.926	0.744	0.670
HMGB-1 (μg/L)	10.675	0.694	0.621-0.767	0.752	0.610	0.362

Note: AUC, area under the curve; CI, confidence interval; IL-1β, Interleukin-1β; TNF-α, Tumor Necrosis Factor-alpha; TIMP-1, Tissue Inhibitor of Metalloproteinases-1; TREM-1, Triggering Receptor Expressed on Myeloid Cells-1; PTX3, Pentraxin 3; CD14, Cluster of Differentiation 14; ICAM-1, Intercellular Adhesion Molecule-1; HMGB-1, High Mobility Group Box 1; CX3CR1, C-X3-C Motif Chemokine Receptor 1.

This study examined the relationship between specific biomarkers and the pH value of dental pulp blood. The results showed that IL-1β and CX3CR1 are negatively correlated with the pH of pulp blood. The increase in extracellular accumulation of lactic acid generated by glycolysis and the reduction of the pH value of dental pulp blood may be the reason why these indicators are negatively related to the pH value. Further study of the relationship between these biomarkers and VAS pain score, found that CX3CR1 has a good correlation with VAS score. This observation may be related to the role of CX3CR1 in the recruitment of inflammatory cells: CX3CR1 promotes the migration of inflammatory cells to the painful site. With the accumulation of local inflammatory cells, the inflammatory reaction is aggravated, usually accompanied by the enhancement of pain perception [22]. Therefore, the VAS score of pain is positively correlated with CX3CR1. Through logical regression, TNF-α, IL-1β and pain VAS levels were found to be independent predictors of disease occurrence. These results highlight the potential clinical benefits of tracking TNF-α and IL-1β levels, which are conducive to the early detection and intervention of suppurative cases. These results suggest that clinical dentists need to strengthen the monitoring of TNF-α and IL-1β levels in patients with pulpitis, so as to diagnose and intervene in suppurative pulpitis as early as possible.

This study has certain limitations. This study is a single-center retrospective study. The sample source is relatively concentrated, which may lead to selection bias, and the sample size is limited, so the generalizability of the conclusions needs to be further verified through multi-center studies. Currently, the detection of gingival crevicular fluid biomarkers and pH values is limited by the sensitivity of existing technologies, and some indicators may not have been fully identified. In the future, it is necessary to conduct multi-center prospective studies combined with multi-omics analysis methods to screen for more accurate combined biomarkers, in order to further improve the prediction system and enhance diagnostic efficacy.

Conclusion

This study has confirmed that the VAS score for pain, as well as the levels of TNF-α and IL-1β, can serve as independent predictors for the occurrence of suppurative pulpitis, providing an effective reference for early clinical diagnosis and timely intervention.

Disclosure of conflict of interest

None.

Address correspondence to: Meng Jiang, Wuxi Stomatology Hospital, Wuxi 214000, Jiangsu, China. E-mail: 15006173636@163.com

References

[1] Shao L, Wang Q, Chen B and Zheng Y. The Roles and molecular mechanisms of HIF-1α in pulpitis. J Dent Res 2025; 104: 715-724.

[2] Ricucci D, Siqueira JF Jr, Abdelsayed RA, Lio SG and Rôças IN. Bacterial invasion of pulp blood vessels in teeth with symptomatic irreversible pulpitis. J Endod 2021; 47: 1854-1864.

[3] Karrar RN, Craig SG, Duncan HF, Abushouk SA, Elfiel SY, Lundy FT, Clarke M and El-Karim IA; Reliability Assessment Group. Clinical validation of a proposed diagnostic classification for pulpitis. Int Endod J 2025; 58: 1158-1171.
[4] Duncan HF, Kirkevang LL, Peters OA, El-Karim I, Krastl G, Del Fabbro M, Chong BS, Galler KM, Segura-Egea JJ and Kebschull M; ESE Workshop Participants and Methodological Consultant. Treatment of pulpal and apical disease: the European society of endodontology (ESE) S3-level clinical practice guideline. Int Endod J 2023; 56 Suppl 3: 238-295.
[5] Guo K, Xu X, Gao J, Zhang Y, Wang Y, Zhuang Y, Zhu Y, Zhou Z, Chen X, Zhang Z and Wei W. Study on pulp metabolism of patients with pulpitis using ultra-performance liquid chromatography coupled with Orbitrap mass spectrometry. Clin Chim Acta 2024; 558: 117894.
[6] Wang M, Xia T and Wang Y. Nd: YAG laser irradiation in the treatment of live pulp preservation in pediatric cariogenic pulpitis. Technol Health Care 2025; 33: 537-544.
[7] Gu F, Huang D, Li R, Peng L, Huan T, Ye K, Bian Z and Yin W. Roles of pyroptosis in the progression of pulpitis and apical periodontitis. J Inflamm Res 2025; 18: 3361-3375.
[8] Careddu R and Duncan HF. A prospective clinical study investigating the effectiveness of partial pulpotomy after relating preoperative symptoms to a new and established classification of pulpitis. Int Endod J 2021; 54: 2156-2172.
[9] Chen J, Xu H, Xia K, Cheng S and Zhang Q. Resolvin E1 accelerates pulp repair by regulating inflammation and stimulating dentin regeneration in dental pulp stem cells. Stem Cell Res Ther 2021; 12: 75.
[10] Shamszadeh S, Eghbal MJ and Asgary S. Advancing dentin-pulp regeneration: clinical perspectives and insights from stem/progenitor cell transplantation (part II). Am J Stem Cells 2024; 13: 132-142.
[11] Al-Helou N. Contemporary endodontics for children and adolescents. British Dental Journal 2023; 235: 21.
[12] Qu M, Zhao J, Zhang Y, Xu Z, Ma C and Cui H. Utilizing the visual analogue scale (VAS) to monitor and manage pain in post-operative skin wounds after thoracic surgery. Int Wound J 2024; 21: e14503.
[13] Bulzan M, Cavalu S, Voită-Mekeres F and Hozan CT. Assessment of pain intensity after total hip arthroplasty using the Visual Analogue Scale (VAS). J Med Life 2024; 17: 1049-1053.
[14] Shetty P, Shetty S, Rai P, Kumar BK and Bhat R. Role of oral microbiota in irreversible pulpitis - current strategies and future perspectives. Acta Microbiol Immunol Hung 2023; 70: 177-186.
[15] Wang J, Qiao J, Ma L, Li X, Wei C, Tian X and Liu K. Identification of the characteristics of infiltrating immune cells in pulpitis and its potential molecular regulation mechanism by bioinformatics method. BMC Oral Health 2023; 23: 287.
[16] Lowe MM, Naik HB, Clancy S, Pauli M, Smith KM, Bi Y, Dunstan R, Gudjonsson JE, Paul M, Harris H, Kim E, Shin US, Ahn R, Liao W, Hansen SL and Rosenblum MD. Immunopathogenesis of hidradenitis suppurativa and response to anti-TNF-α therapy. JCI Insight 2022; 7: e165502.
[17] Sabat R, Alavi A, Wolk K, Wortsman X, McGrath B, Garg A and Szepietowski JC. Hidradenitis suppurativa. Lancet 2025; 405: 420-438.
[18] Yan Z, Dai J, Wang J, Feng Q, Wang Y, Han T and Wu C. RNF167-mediated ubiquitination of Tollip inhibits TNF-α-triggered NF-κB and MAPK activation. FASEB J 2023; 37: e23089.
[19] Wang Y and Thorlacius H. Mast cell-derived tumour necrosis factor-alpha mediates macrophage inflammatory protein-2-induced recruitment of neutrophils in mice. Br J Pharmacol 2005; 145: 1062-1068.
[20] Ghosh R and Bishayi B. Neutralization of TLR2 in combination with either TNF-α or IL-1β antibody reduces the severity of septic arthritis through STAT3/mTOR signalling in lymphocytes. Cell Immunol 2024; 405-406: 104878.
[21] Gopalsamy B, Farouk AAO, Tengku Mohamad TAS, Sulaiman MR and Perimal EK. Antiallodynic and antihyperalgesic activities of zerumbone via the suppression of IL-1β, IL-6, and TNF-α in a mouse model of neuropathic pain. J Pain Res 2017; 10: 2605-2619.
[22] Liu N, Zhang GX, Zhu CH, Lan XB, Tian MM, Zheng P, Peng XD, Li YX and Yu JQ. Antinociceptive and neuroprotective effect of echinacoside on peripheral neuropathic pain in mice through inhibiting P2X7R/FKN/CX3CR1 pathway. Biomed Pharmacother 2023; 168: 115675.