Review Article

Gingival Crevicular Fluid Levels of Neurokinin A and Substance P in Patients with Symptomatic Irreversible Pulpitis: A Systematic Review and Meta-Analysis

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Aims and Objectives: The levels of neuropeptides neurokinin A (NKA) and substance P (SP) in the gingival crevicular fluid of patients with symptomatic irreversible pulpitis (SIP) were evaluated using a comprehensive review and meta-analysis. **Materials and Methods:** The data bases of Pubmed, Scopus, EBSCOhost, Science Direct, Proquest and Cochrane library databases were thoroughly searched. The quality of the study was evaluated using the Joanna Briggs Criteria. Twenty four studies were listed following a thorough search of full texts, abstracts, and removal of duplicates. Only two of these papers were eligible for inclusion in the meta-analysis. Since the results obtained were in mean and standard deviation, the levels of neuropeptides in the test and control groups were examined using the meta-analysis. **Results:** When compared with healthy teeth, NKA was not significantly elevated in GCF of individuals with SIP (P = 0.06; odd ratio = 1.34 [−0.05 to 2.74] at 95% confidence interval [CI]). Additionally, there was no evidence of an association between SP and SIP (P = 0.08; odds ratio = 0.84 [−0.10 to 1.77] at 95% CI). **Conclusion:** This systematic research demonstrated that in individuals with SIP, NKA, and SP are not substantially linked. However, the lack of study in this area makes it evident that additional research is needed, particularly in relation to pulpal disorders and NKA.

Keywords: *Gingival crevicular fluid, neurokinin A, painful teeth, substance P, symptomatic irreversible pulpits*

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Introduction

Clinicians have been compelled to rely on traditional diagnostic techniques due to the lack of valid diagnostic tools for pupal illness. The most frequent reason for orofacial discomfort is odontogenic pain brought on by pulpal inflammation.[1] However, to properly treat the patient's condition, an accurate diagnosis of the presenting disease state must be made due to the numerous complicating factors that could change the diagnosis and treatment plan. Therefore, determining if the pain has an odontogenic cause is the most important step. Particularly in the dental pulp, pain and inflammation are tightly related. The odontogenic pain known as symptomatic irreversible pulpitis (SIP) is one such condition that affects persons of all ages. When bacteria invade, the neural network is the first to be activated. Numerous studies have shown a connection between pulpal inflammation and neuropeptides made by sensory nerves.[2,3]

Neuropeptides have been known as chemical signals in the brain for the past 40 years. They can be defined as "small proteinaceous substances produced and

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released by neurons through the regulated secretory route and acting on neural substrates."[4] According to evidence, the tooth pulp contains three neuropeptides: substance P (SP), neurokinin A (NKA), and calcitonin gene-related peptide (CGRP).[5]

In addition, there is proof that inflamed pulp has higher quantities of these neuropeptides.[6] The neuropeptides have been demonstrated to enter the gingival crevicular fluid (GCF) and the periodontal ligament from the pulp through gateway channels such the apical foramen and accessory canals.[7,8] Due to their proximity to inflammatory foci, peripheral bodily fluids such as GCF and saliva are employed to detect indicators of acute and chronic inflammation. In individuals with migraine and cluster headaches, SP and CGRP have been found in their saliva, according to a study of the literature.[9] It has been previously reported that periodontal health can be determined by the detection of SP, NKA, and CGRP in GCF.[10-12] The importance of GCF as a source of biomarkers to detect periodontal diseases has been highlighted in a thorough review. They came to the conclusion that the GCF can accurately reflect the disease process in the periodontal apparatus because of its near proximity to the diseased site.[13] This systematic review carefully includes studies that collected GCF using periopaper despite the fact that there is a variance in the method of GCF collection. There has not yet been a review that examines the significance of two neuropeptides independently with regard to SIP.

Therefore, the main goal of this research was to investigate the null hypothesis that NKA and SP levels in patients with SIP do not change.

Materials and Methods

Study design

This systematic review and meta-analysis was carried out to explore the correlation of GCF levels of NKA and SP in teeth with systemic irreversible pulpitis. It was developed according to the preferred reporting items for systematic reviews and meta-analyses (PRISMA) guidelines.[14] After registering the protocol in PROSPERO (CRD42023390109), the study thoroughly evaluated full-text articles published from 1980 to 2023. The study was carried out between November 2022 and January 2023.

Research question

Thus, the question framed for the literature search was Does SIP lead to increased levels of NKA and SP in human GCF?

The PICO for the eligibility criteria was as follows:

(P): Human patients diagnosed with SIP and no systemic disease.

(I): Estimation of GCF levels for NKA and SP using a commercially available Enzyme-Linked Immunosorbent Assay (ELISA) kit.

(C): Healthy individuals with no systemic disorders and presence of no carious teeth.

(O): NKA and SP levels are raised in SIP.

Inclusion criteria

(a) Clinical human observational studies evaluating NKA levels and SP in human GCF
(b) Clinical observational studies where the test subjects were diagnosed with SIP
(c) Studies that used a commercially available ELISA kit to analyze the levels of NKA and SP

The publications with their titles and abstracts discovered during the electronic and manual searches were reviewed by two investigators (DA and AG). The articles that did not meet the criteria for inclusion were filtered out. All of the remaining articles were retrieved and thoroughly screened by two of the above-mentioned reviewers to achieve a consensus.

Exclusion criteria

(a) Animal studies.
(b) Studies not using GCF as the test sample
(c) Studies that did not analyze NKA and SP
(d) Clinical studies where the test subjects had a systemic disease.
(e) Narrative reviews.
(f) Studies that did a qualitative analysis

Information sources and search strategy

A broad-based search was implemented with individual keywords: "Neurokinin A," "gingival crevicular fluid" and "painful teeth." The precise and prescribed combination of accepted medical subject heading keywords with the Boolean operators "AND" and "OR" was carried out to obtain the search strategy finalized as

"neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH

Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields])).

Pubmed, Scopus, EBSCOhost, Science Direct, Proquest and Cochrane library databases were searched until January 2023 without restriction to language and year. The summary of search results in the databases is summarized in Table 1. The references of eligible studies were hand checked for additional studies by DA and AG. Google scholar and an OpenGrey search were performed for the gray literature as well. Wherever needed, the authors of eligible papers were contacted for further information related to the study or any

Table 1: Search strategy

Database	Search strategy (2022)	*n*
PubMed	"neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields]))	04
EBSCOhost	"neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields]))	03
Scopus	"neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields]))	02
Science Direct	"neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields]))	08
Proquest	neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields]))	21
Cochrane Library	neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields]))	02
Gray literature	"neurokinin a"[MeSH Terms] OR "neurokinin a"[All Fields]) AND ("substance p"[MeSH Terms] OR "substance p"[All Fields]) AND ("gingival crevicular fluid"[MeSH Terms] OR ("gingival"[All Fields] AND "crevicular"[All Fields] AND "fluid"[All Fields]) OR "gingival crevicular fluid"[All Fields]) AND (("pain"[MeSH Terms] OR "pain"[All Fields] OR "painful"[All Fields] OR "pains"[All Fields] OR "pain s"[All Fields] OR "painfulness"[All Fields]) AND ("teeth s"[All Fields] OR "teeths"[All Fields] OR "tooth"[MeSH Terms] OR "tooth"[All Fields] OR "teeth"[All Fields] OR "tooth s"[All Fields] OR "tooths"[All Fields]))	0
Total		40

Table 2: Characteristics of the study

Author, year, and Journal	Study design	Country	Total number of patients	Type of teeth included	Average VAS scale reading	Sample obtained	Method of sample collection	Biomarker evaluated	Diagnostic test for NKA and SP	NKA levels and control	SP levels and control	Conclusion
Heidari *et al.*,[16] 2017, J Dent (Tehran)	Case control observational study	Iran	40 children. (9 boys and 11 girls) % of male = 45 % of female = 55 Mean age = 6.80 + 1.47 years Control group: a primary molar in the other quadrant of the same jaw which was free of any carious lesion	Symptomatic irreversible pulpitis (SIP) in primary molar	VAS>5	GCF	Periopaper	Substance P	ELISA	2.23 ± 0.74 pg/mL Control group: 1.84 ± 0.38 pg/mL	2.23 + 0.69 pg/mL Control: 2.02 + 0.37 pg/mL	SP levels were not statistically significant compared to healthy controls ($P > 0.05$)

Table 3: Characteristics of the study

Author, year, and Journal	Study design	Country	Total number of patients	Type of teeth included	Average VAS scale reading	Sample obtained	Method of sample collection	Biomarker evaluated	Diagnostic test for NKA and SP	NKA levels and control	SP levels and control	Conclusion
Heidari *et al.*,[15] 2017, Dent Res J (Isfahan)	Case control observational study	Iran	20 children (7 boys and 13 girls) % of female = 65% Mean age = 9.4 + 1.4 years Control: intact permanent first molar in the opposing quadrant of the same jaw	Symptomatic irreversible pulpitis (SIP) in permanent first molar	VAS>5	GCF	Periopaper	Substance P	ELISA	SIP group: 2.65 ± 0.56 pcgr/mL Control: 1.83 ± 0.65 pcgr/mL	SIP group: 2.65 + 0.56 pg/mL Control: 1.83 + 0.65 pg/mL	SP levels are significantly raised in SIP cases compared to healthy controls ($P < 0.05$)

Table 4: Risk of Bias assessment

Study	Were the groups comparable other than the presence of disease in cases or the absence of disease in controls?	Were cases and controls matched appropriately?	Were the same criteria used for identification of cases and controls?	Was exposure measured in a standard, valid and reliable way?	Was exposure measured in the same way for cases and controls?	Were confounding factors identified?	Were strategies to deal with confounding factors stated?	Were outcomes assessed in a standard, valid and reliable way for cases and controls?	Was the exposure period of interest long enough to be meaningful?	Was appropriate statistical analysis used?	Overall risk assessment
Heidari *et al.*[16] J Dent (Tehran), 2017	Yes	Yes	Yes	Yes	Yes	Unclear	Unclear	Yes	Yes	Yes	Moderate risk
Heidari *et al.*[15] Dent Res J (Isfahan), 2017	Yes	Yes	Yes	Yes	Yes	Unclear	Unclear	Yes	Yes	Yes	Moderate risk

other additional details. Any doubts or clarifications were resolved with a senior researcher (AS).

Study selection and data collection process

The finalized search strategy produced in PubMed was used to exhaustively search the three databases while keeping in mind the inclusion and exclusion criteria. Two reviewers (DA and AG), who collected the data and entered it into an Excel Sheet powered by Microsoft 365 Office, looked through each study two to three times. After all the data had been compiled, a third senior researcher (AS) with over 20 years of experience in systematic reviews and meta-analyses applied the inclusion and exclusion criteria. After carefully compiling the data, a comprehensive table was created for additional analysis [Tables 2 and 3]. Any differences of opinion were settled through consensus.

Risk of bias assessment

Data were collected and filtered by two examiners (DA and AG) in accordance with the stringent inclusion criteria. The risk of bias evaluation was carried out using the Joanna Briggs Institute Critical Appraisal Checklist (The Joanna Briggs Institute 2014). The likelihood of bias in cross-sectional and observational studies was evaluated based on 10 factors. Each topic was then discussed after separate application of the risk of bias for each study by two assessors (DA and AG). Any disagreements were settled by the third examiner (AS), who also reached the final consensus [Table 4].

Summary measures and synthesis of results

Continuous data from eligible studies were only included in the meta-analysis. The Inverse Variance method was applied to calculate the pooled odds ratio for the association between the levels of NKA and SP with SIP. All analyses were conducted using the RevMan 5.3 (Review Manager v.5.3; Copenhagen, Denmark: The Nordic Cochrane Centre, The Cochrane Collaboration 2014).

Results

The search conducted in three databases identified seven studies. After the elimination of duplicates, three studies were identified. These were further scanned for the eligibility and inclusion criteria which led to only two studies. The details are presented according to the updated PRISMA guidelines in Figure 1.

The references were managed with the EndNote Basic Software (Thomson, Reuters, New York, NY), and duplicates were removed accordingly.

Since there were two biomarkers assessed, the meta-analysis was conducted separately for each one. The

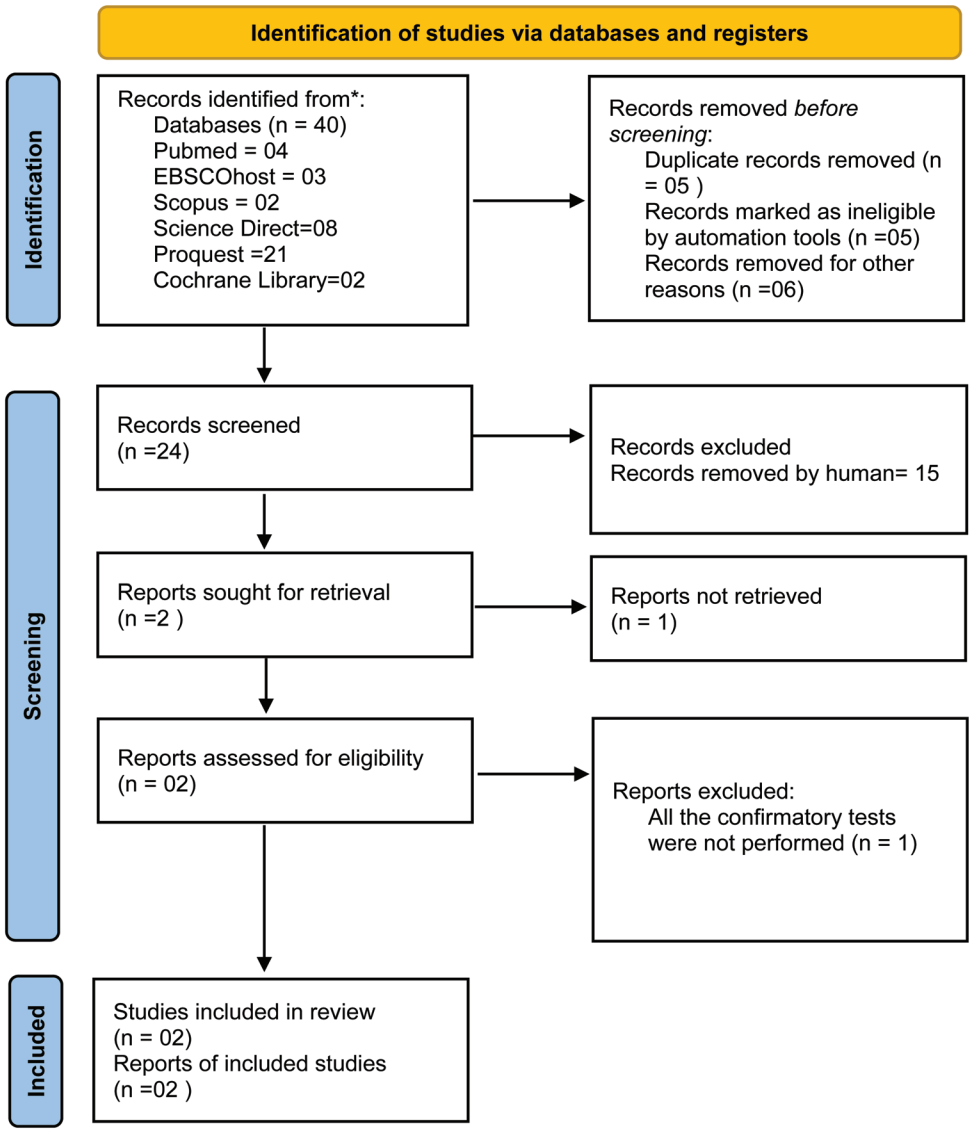

Figure 1: preferred reporting items for systematic reviews and meta-analyses flow chart

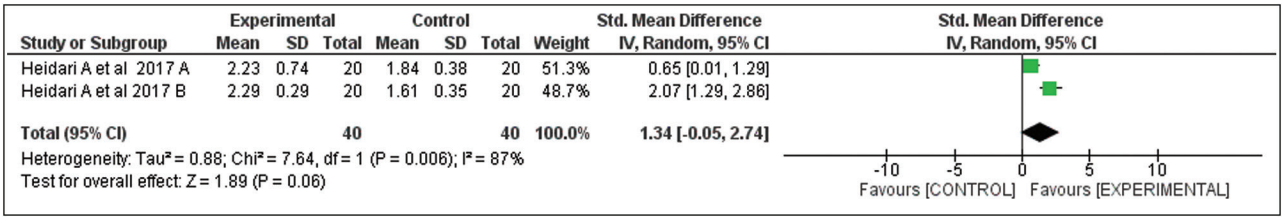

Figure 2: NKA Forest plot

meta-analysis of the NKA biomarker presented a pooled odd ratio of 1.34 (95% confidence interval, −0.05 to 2.74). The forest plot [Figure 2] shows the diamond in the experimental group, which is SIP but it was statistically insignificant (P = 0.06). The low risk of bias was depicted in the funnel plot [Figure 3]. The analysis for SP presented a Random Effects Inverse Variance of 0.84 (−0.10 to 1.77). The forest plot [Figure 4] here as well shows the diamond in the experimental group but the analysis is statistically insignificant (P = 0.08). The funnel plot revealed the low risk of bias for SP as well [Figure 5]. Furthermore, there is heterogenicity in both studies with an I^2 of 87% for NKA and 75% for SP.

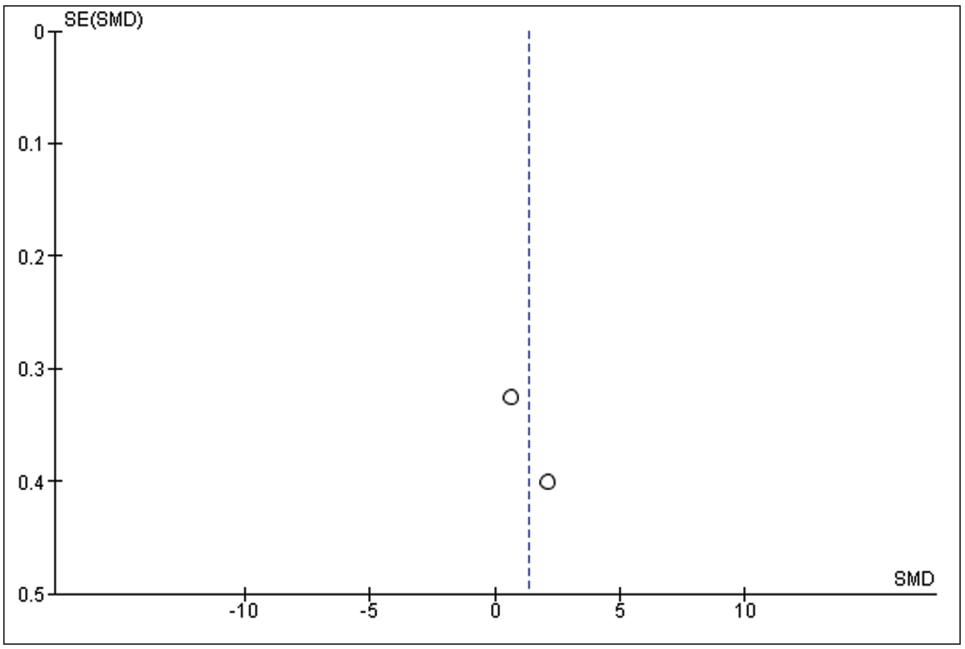

Figure 3: NKA Funnel plot

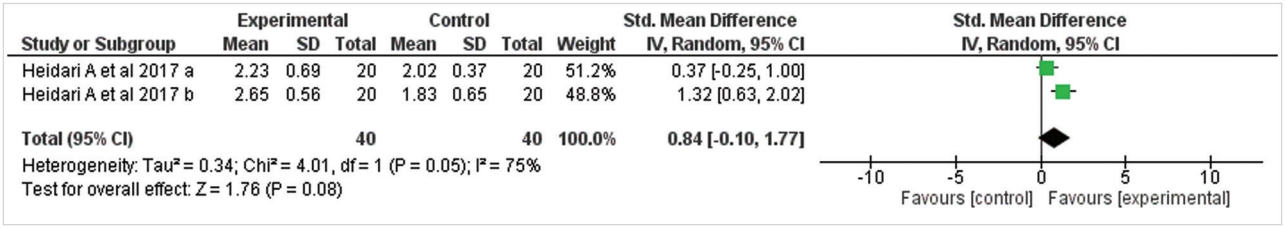

Figure 4: SP Forest plot

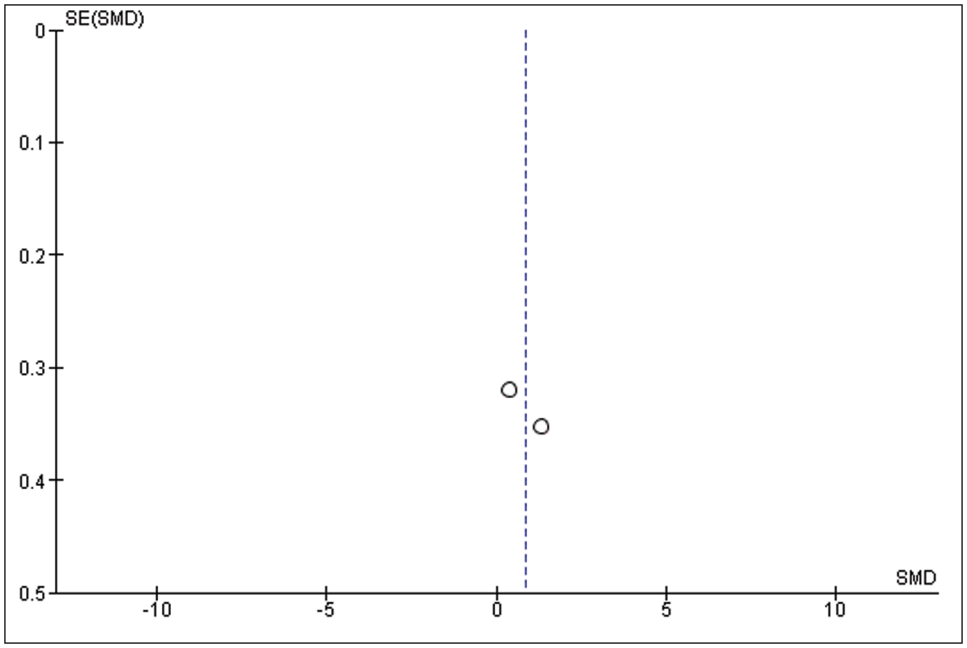

Figure 5: SP Funnel plot

GCF sample collection

After drying the gingiva for 15 s and isolating the gingiva around the involved tooth, a gamma sterilized no. 30 period paper was inserted 1 mm of the buccal surface of the gingival sulcus for 30 s. Sampling was done between 9 AM and 11 AM to avoid the confounding variable of the circadian rhythm. The paper strips were then placed in the Eppendorf tubes containing 30 µL of phosphate-buffered saline. The samples were stored at −70°C till the ELISA test was done.

Confirmatory tests

A commercially available ELISA was used in both the included studies, and the procedure was carried out according to the manufacturer's instructions.

The ELISA indicated that there is a significant increase in NKA in the GCF of patients diagnosed with SIP but no significant difference was found in the levels of SP.

Study characteristics of clinical studies included in the meta-analysis

Heidari *et al*.[15] conducted a cross-sectional study to evaluate the levels of SP, and NKA in GCF of painful human teeth and compared the levels to healthy teeth. Children diagnosed with SIP (n = 20) in the permanent first molar were screened through the strict inclusion and exclusion criteria and were to subjected to GCF sampling where a gamma-sterilized paper number 30 was used. A similar technique was used for healthy individuals (n = 20) as well. The NKA and SP levels were analyzed in a commercially available SP kit. The levels of NKA were significantly raised in painful teeth (2.29 ± 0.29 pg/mL) compared with healthy teeth (1.61 ± 0.35 pg/mL) ($P < 0.05$). Similarly, the levels of SP were significantly raised in painful teeth (2.65 ± 0.56 pg/mL) compared with healthy teeth (1.83 ± 0.65 pg/mL) ($P < 0.05$).

Heidari *et al*.[16] conducted an observational study where the levels of NKA and GCF were studied in children with SIP (n = 20) in a primary molar and healthy control intact primary molar of the opposite quadrant in the same jaw. The mean NKA levels were significantly higher in painful teeth were 2.23 ± 0.74 pg/mL compared with healthy individuals (1.84 ± 0.38 pg/mL) ($P < 0.05$). The levels of SP were, however, not significantly higher in painful teeth (2.23 ± 0.69 pg/mL) compared with healthy individuals (2.20 ± 0.37 pg/mL), $P > 0.05$.

Discussion

The present study's findings show that there is no statistically significant difference between the NKA and SP levels in the GCF of teeth with SIP and healthy teeth. We also wish to point out that NKA was significantly greater in both investigations' results compared with SP, which was nonsignificant in the second permanent molar study. These contradict with two studies that were ineligible for the review because of a different analysis method.

Awadeh *et al*.[5] quantitively analyzed the SP, NKA, and CGRP in GCF patients with painful teeth compared with healthy controls. Their study could not be part of our meta-analysis as they used the radioimmunoassay to assess the level of the biomarkers. Their results led them to conclude that pulp inflammation results in significant neuropeptide elevation. Dincer *et al*.[17] conducted a qualitative analysis on different biomarkers in GCF of patients with SIP and concluded that SP and NKA were significantly raised in SIP.

We postulate that this is the benefit of a meta-analysis where similar studies, although very limited in this case, are grouped and analyzed for the result.

Multiple studies are being conducted globally to reach a conclusion or to respond to a specific research issue. For a final, decisive declaration, these findings must be combined. Therefore, "Evidence-based medicine" can be described as "the systematic, quantitative, preferentially experimental approach to obtaining and using medical information" in the entry. One such quantitative study that compiles information from related studies to reach a clinically focused result is meta-analysis. Meta-analysis is described by Glass[18] as "The statistical analysis of a large collection of analysis results from individual studies for the purpose of integrating the findings." The current study's goal was to demonstrate any correlation, if any, between the GCF levels of NKA and SP in SIP. We believe that this is the first comprehensive review and meta-analysis to assess the relationship between two biomarkers and SIP.

In this modern era of patient-oriented treatment, there is increasing research to develop a targeted and disease-specific diagnosis that will enhance patient comfort. In this context, several studies have unveiled the potential of biomarkers in saliva, which could monitor the disease progression.[19-21] But there is an increasing shift in trend toward GCF as a medium for biomarkers.

A wide range of inflammatory mediators are released by different cells during inflammation and, thus, are responsible for tissue destruction.[22] Neuropeptides are one of the largest classes of pro-inflammatory mediators released from the nerve fiber terminals.

SP and NKA are two of the three neuropeptides released from the stimulated nerve endings that are responsible for intrapulpal pressure increase, which results in pain. As has been already pointed out that there is an evidence of the presence of NKA and SP in the GCF, the GCF can serve as the best media to detect the levels of these biomarkers.

The studies included in the meta-analysis showed heterogeneity of 87% for NKA and 75% for SP. This could be attributed to the nature of the patients chosen, the type of teeth in the experimental group, the sampling technique, the confirmatory test process, and the analysis of results.

Different studies used different methods to measure the inflammatory mediators, but ELISA stands out as the most reliable and effective method.[16] Hence, it was paramount to include only those studies that used ELISA and had completed a quantitative analysis. As mentioned earlier, Dincer *et al*.[17] also used ELISA, which shows to prove that ELISA is a safe and reliable method to evaluate the biomarker levels.

A pilot study conducted by Arslan *et al*.[23] reported the effects of endodontic treatment on salivary levels of CGRP and SP. Their observations led them to establish a positive correlation between pain and increased levels of CGRP and SP. These levels considerably decreased after endodontic treatment. Here, the peripheral fluid analyzed was saliva but it is being mentioned here to emphasize the importance of calibrating these neuropeptides with respect to SIP. A commercially available ELISA kit was used here as well to analyze the results, and it revealed higher values of CGRP and SP in saliva before endodontic treatment. However, we would want to emphasize that GCF as a media would be more specific than saliva, and hence, this present review was designed to include only those studies which tested the levels of neuropeptides in GCF with a commercially available ELISA kit.

The authors realize that the number of studies may be a hurdle in arriving at a conclusion, but through this review, we would want to further highlight that more research needs to be carried out especially randomized control trials, which could enable one to arrive at more conclusive data. To the best of our knowledge, no review and meta-analysis have analyzed the importance of two neuropeptides in SIP.

Conclusion

Hence, we would want to conclude that although the individual studies included in this review showed significant results for the NKA neuropeptide, the overall meta-analysis was nonsignificant. This highlights the importance of such reviews even if there are only two studies that fit the inclusion criteria. Increased sample size and the conduct of more studies would have probably brought about a significant analysis.

Clinicians worldwide are still relying on the guidelines laid down by the American Association of Endodontics, and it is well known that an accurate correlation between the clinical and actual status of the pulp is never achieved. It is time to shift toward the so-called "Molecular Diagnostic Markers (MDM)," which can enable one to accurately predict the condition of the disease and further plan out the treatment strategy.

With this review, we would want to emphasize the need to conduct more studies that would investigate other biomarkers and establish the best biomarker to be evaluated in SIP.

Registration

This systematic review and meta-analysis has been registered in PROSPERO (CRD42023390109).

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Nil.

Conflicts of interest

None to declare.

Author contribution

DA: concept, design, supervision, writing, and critical review. AS and AG: concept, design, supervision, analysis and/or interpretation, writing, and critical review.

Ethical policy and institutional review board statement

Not Applicable.

Patient declaration of consent

Not applicable.

Data availability statement

The data that support the study results are available from the author Dr. Dax Abraham at daxabraham.sds@mrei.ac.in

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