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RESEARCH

Assessment of cortisol as a neonatal pain biomarker in the application of non-pharmacological analgesia therapies: systematic review and meta-analysis

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Abstract

Background In the history of Neonatology, decades ago pain has been little studied because it was believed that newborns didn't have the capacity to experience pain. Nowadays, there is enough evidence for the existence of neonatal pain but its adequate treatment is an aspect that is continuously evolving. The objective of this study was to evaluate the effectiveness of non-pharmacological analgesia therapies used to alleviate pain in newborns by analysing neonatal cortisol levels as biological markers of pain.

Methods A systematic review and meta-analysis was performed following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA), and the Cochrane Collaboration Handbook. Searches were performed in databases such as PubMed, Web of Science, Scopus, CINAHL, Cochrane Library and Science Direct until the end of May 2024. The search identified 1075 articles, of which 10 studies met the inclusion criteria and had the necessary data to develop the meta-analysis. Furthermore, in each meta-analysis, subgroups were performed: non-pharmacological analgesia vs. placebo, and pre-post intervention by gestational age.

Results The meta-analysis found that breastfeeding exhibited a moderate effect size (SMD = -0.63; 95% CI = -1.07 to -0.19), sucrose showed a small effect (SMD = -0.15; 95% CI = -0.55 to 0.26), and skin to skin contact exhibited a large effect (SMD = -1.34; 95% CI = -2.21 to -0.46). Patients under 28 weeks have less post-intervention pain and showed a large effect (SMD = 1.44; 95% CI = 0.47 to 2.40), between 28 and 32 weeks they have more post-intervention pain and presented a small effect (SMD = -0.43; 95% CI = -0.86 to -0.0), and over 32 weeks they have an increased post-intervention pain and exhibited a large effect (SMD = -1.08; 95% CI = -1.65 to -0.51).

Conclusions Non-pharmacological therapies showed efficacy in pain reduction based on neonatal cortisol levels. Skin to skin contact is the most effective method to reduce pain from invasive procedures, such as heel pricks in preterm infants under 28 weeks. Breastfeeding also demonstrated to be an effective and safe alternative to use

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for pain relief and to reduce cortisol levels. However, the cortisol results indicate that sucrose was not effective in reducing neonatal pain.

Trial registration PROSPERO: CRD42023463831.

Keywords Cortisol, Neonatal pain, Newborn, Nursing care, Heel Prick, Analgesia, Breastfeeding, Skin to skin contact, Sucrose

Background

The International Association for the Study of Pain (IASP) defines acute pain as "an unpleasant somatic or visceral sensation caused by actual or potential tissue injury" [1, 2]. In the past, it was thought that infants lacked the ability to sense pain due to the incomplete myelination of their nervous system [3]. However, there is now sufficient evidence indicating that neonates do indeed experience pain, given that the neurophysiological and anatomical elements necessary for pain transmission develop prior to 24 weeks of gestational age. Conversely, the inhibitory mechanisms that respond to nociception and alleviate pain require several months to fully mature [3, 4]. Following their initial moments of life, infants are subjected to a series of invasive procedures, with heel pricks for screening endocrinometabolic conditions being the most common. Often these premature infants, classified as "preterm" need to be admitted to Neonatal Intensive Care Units (NICUs), where they may undergo up to 10–15 painful interventions daily [5, 6]. The World Health Organization (WHO), estimates that approximately 15 million preterm births occur annually [7]. On a national scale in 2023, data from the National Institute of Statistics (INE) of Spain, indicated that there were 19.879 preterm births [8]. This condition stands as one of the primary contributors to perinatal mortality and morbidity [9].

Research indicates that untreated moderate-intense pain can impact the neurodevelopment of newborns, leading to changes in motor skills, cognitive functions, and behavior, including increased heart rate and decreased oxygen saturation [10]. Consequently, it is imperative to alter clinical approaches and minimize stimuli to ensure the well-being of these infants. This shift in approach has encouraged the adoption of innovative care models like the Newborn Individualized Developmental Care and Assessment Program (NIDCAP) [11, 12]. One of the objectives of this approach is to evaluate the capacity of newborns to manage stress prior to, during, and after invasive procedures [13]. Furthermore, this approach sets forth a range of tactics, encompassing non-pharmacological interventions, to mitigate neonatal discomfort in minor invasive procedures. These interventions comprise breastfeeding, skin to skin contact, and oral administration of sucrose, among others [14, 15]. These measures have been introduced to avoid the potential adverse effects that medications may trigger in neonates, owing to disparities in pharmacokinetics and pharmacodynamics compared to adult patients. This leads to a slower elimination of drugs, resulting in a more pronounced and prolonged effect. Because of this, it's important to analyse whether non-pharmacological measures are really effective or not, in order to enhance their use [16, 17].

The incapacity of newborns to communicate their perception of pain has led to the development of tools for evaluating and assessing pain. These instruments rely on observing physiological and behavioural shifts that neonates undergo when experiencing pain [18]. Three of the most commonly employed validated scales are the Premature Infant Pain Profile (PIPP), used to assess acute and procedural pain in preterm newborns from 28 weeks of gestation, to full term infants of 40 weeks [19]. Similarly, the Neonatal Infant Pain Scale (NIPS), is extensively used to evaluating acute pain in preterm and full term infants [20]. And finally, the Neonatal Infant Acute Pain Assessment Scale (NIAPAS), used for assessing acute pain in preterm and full-term infants in the NICU [21].

Analyzing the interpretation of this scales is a great challenge since the signs and symptoms of pain can be confused with feelings of hunger or discomfort. They are even difficult to use in cases of newborns with neurological damage or under sedation infusions, due to the limited physical response [22].

That's why the use of biomarkers such as cortisol levels can be key for the detection and measurement of infant stress, improving pain relief attention and management [23]. Cortisol is a steroid hormone or glucocorticoid produced by the adrenal gland that is released in response to stress. Because it is liposoluble, it enters in saliva through the intracellular pathway, and its salivary concentration is not dependent on saliva flow, and closely approximates its free concentration in plasma [24, 25]. Until 5–6 months of age, circadian rhythms are not established, so the endogenous cortisol levels shown by newborns are independent and unrelated with the day/night cycle [26]. Neonatal cortisol levels in preterm newborns can range from 0.6 nmol/L to 52.1 nmol/L, with an average of 5.5 nmol/L [27]. Therefore, values higher than those considered normal would indicate the presence of pain [28].

As per the American Academy of Pediatrics (AAP), alleviating pain in newborns is an area that requires further attention [29]. Despite existing studies evaluating the effectiveness of employing non-pharmacological methods for pain relief, a consensus regarding their application has not yet been established, and there are few articles in the literature that evaluate neonatal pain using cortisol levels. The field of neonatal pain management is constantly evolving, for this reason, the aim of this study was to evaluate the effectiveness of non-pharmacological analgesia methods used to alleviate pain in newborns undergoing invasive procedures by analysing neonatal cortisol levels as biological markers of pain.

Methods

The present systematic review and meta-analysis was performed following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines [30], adhering to the suggestions outlined in the Cochrane Collaboration Handbook [31]. The International Prospective Register of Systematic Reviews (PROSPERO) registration number is CRD42023463831.

Data sources

A systematic review of the literature in PubMed, Cochrane Library, Web of Science, Scopus, CINHAL, and Science Direct databases was conducted up from the beginning to the end of May 2024. Following an assessment of non-pharmacological treatment modalities, the search criteria encompassed the subsequent terms: (“analgesia” OR “breastfeeding” OR “skin to skin contact” OR “sucrose”) AND (“cortisol” OR “neonatal pain”) AND (“nursing care” OR “heel prick”). Additionally, references cited in the chosen articles were scrutinized for potential inclusion in this review. Two authors independently determined whether studies fulfilled the inclusion criteria through examination of titles, abstracts, keywords and the full text. A third author was asked if there was a disagreement on any criteria in any article.

Study selection

The studies were chosen based on the following inclusion criteria, studies which include: (1) infants with a minimum gestational age of 28 weeks, regardless of sex or ethnic group; (2) infants with a weight exceeding 1 kg; (3) infants with an Apgar test score higher than 5 in the initial 5 min; (4) an evaluation of neonatal pain using cortisol levels; (5) a comparison of three non-pharmacological analgesia methods and their effect pre and post intervention; (6) a comparison of cortisol levels according to weeks of gestational age; and (7) a design as randomized controlled trial (RCT) or quasi-experimental design. Studies were excluded on the basis of the following, when: (1) involving infants with congenital anomalies; (2) included pain assessment without using cortisol levels; or (3) were ineligible by design as narrative reviews, observational studies, letters to the editor, or case reports.

Data extraction and quality assessment

The selected studies provided the following data: author information and year or publication, country where the intervention took place, sample details (size, gestational age, birth weight, Apgar test scores, and pain assessment); and intervention characteristics (type and dose). The interventions were categorized as follows based on their characteristics: breastfeeding, skin to skin contact and oral sucrose, because these are the three most commonly used techniques in neonatal units, because of their simplicity, safety and ease of administration. The risk of bias (ROB) in randomized controlled trials was evaluated using the Cochrane RoB2 tool. This instrument includes the evaluation of five areas: randomization process, deviations from intended interventions, missing data on outcomes, measurement of outcomes, and selection of reported outcomes [32].

The overall ROB for individual trials was classified as “low risk” if the study received a low risk in all key domains; “some concern” if any area was assessed as having some concern; and “high risk” if at least one area was rated as high risk, or multiple areas were rated as some concern. Additionally, the ROB in non-randomized studies was evaluated using the ROBINS-I tool, which includes seven areas: confounding, selection of participants, classification of interventions, deviations from intended interventions, missing data on outcomes, measurement of outcomes, and selection of reported outcomes [33]. The assessment of potential bias was categorized as “low risk” if the study received low rating across all domains, “moderate risk” if it received moderate ratings across all domains, “serious risk” if it was deemed significant in at least one domain, and “critical risk” if it was considered critical in at least one domain. This process was independently performed by two authors, they reviewed each publication to confirm its adherence to the specified inclusion criteria. A third author was asked if there was a disagreement on any criteria in any article. Decisions regarding which data had to be incorporated in the systematic review and meta-analysis were reached through collective deliberation.

Data synthesis and analysis

Only 10 studies met the inclusion criteria and had the necessary data to develop the meta-analysis. Furthermore, in each meta-analysis, subgroups were performed: non-pharmacological analgesia vs. placebo, and pre-post intervention by gestational age. The meta-analysis and statistical analyses were performed using Review Manager Software (RevMan, Version 5.3). Standardized mean differences (SMDs) and a 95% CI were used for the data analysis. The heterogeneity among trials was measured using the I^2 statistic. I^2 values of 25%, 50%, and 75% represent small, moderate, and large degrees of heterogeneity. According to Cohen guidelines, SMDs of 0.2 represent small effect, values around 0.5 medium effect, and values higher than 0.8 large effect size [34]. The significance criterion was established at $P < 0.05$. The asymmetry was evaluated using funnel plots for indicating the possible risk of publication of small studies [35].

Results

Study selection

The search identified 1075 articles, which was reduced to 346 after the duplicates were removed. After the full review process, the studies were included in the systematic review and meta-analysis [36–45]. Figure 1 shows the flow diagram of the study selection process.

Study characteristics

This systematic review and meta-analysis focused on the non-pharmacological treatment of neonatal pain and its efficacy measured by cortisol levels included 10 studies: 1 on breastfeeding [36], 2 on oral sucrose [37, 38], and 4 on skin to skin contact [39–42]. On the other hand, 3 articles studied the differences in cortisol levels according to the gestational age of the patients [43–45]. The total number of participants was 521, with gestational ages ranging from 28 to 39 weeks. The studies were carried out on three continents (4 in America, 4 in Europe, and 2 in Asia). Some studies compared the effect of non-pharmacological therapies such as breastfeeding, skin to skin contact, and oral sucrose with standard nursing care. The characteristics of all included studies are presented in Table 1.

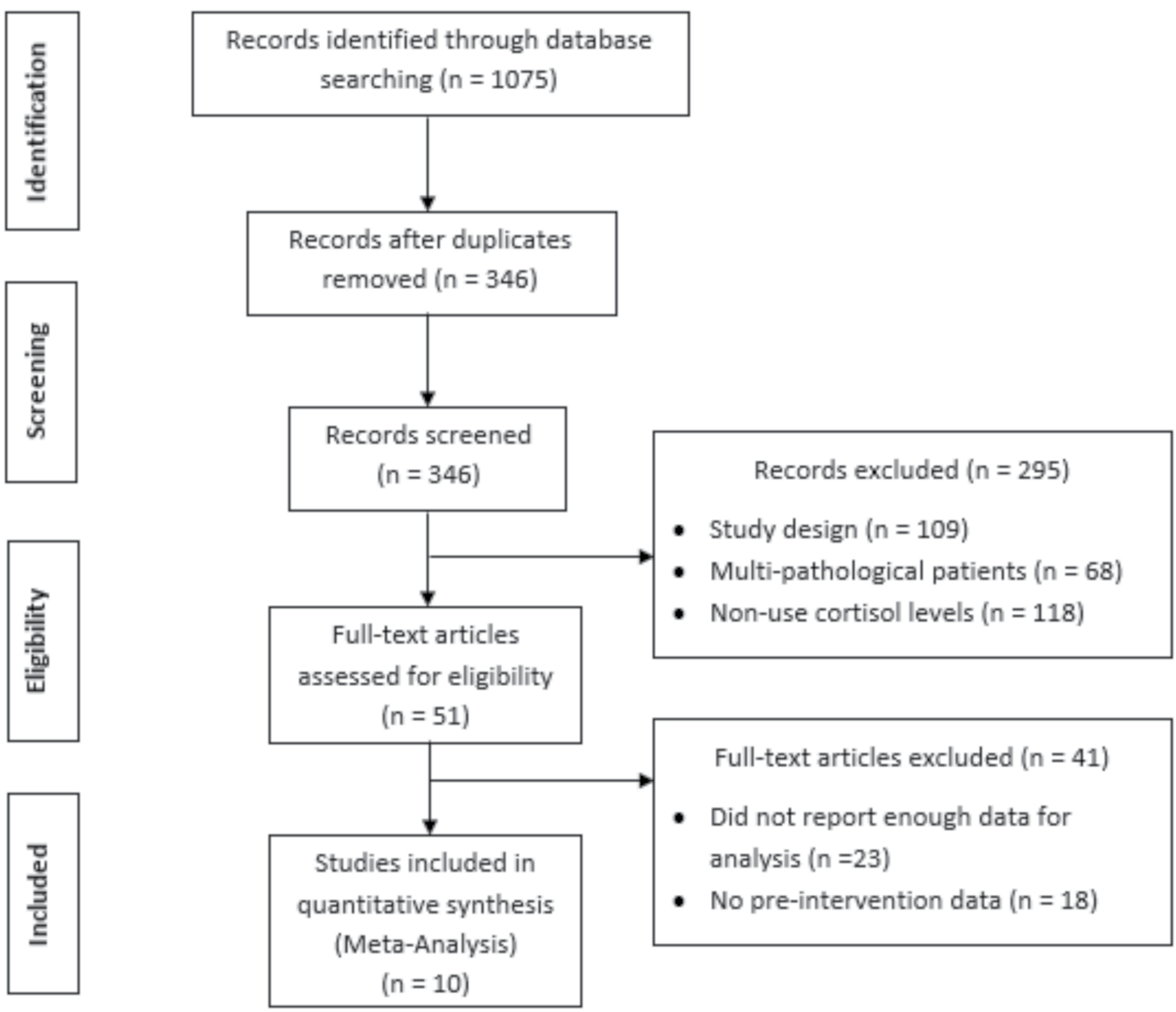

Fig. 1 Flow diagram of the study selection process

Table 1 Characteristics of included studies on neonatal pain and non-pharmacological analgesia

Author and year	Country	Sample characteristics					Intervention characteristics		
		n (girls)	Gestational age (weeks)	Birth weight (kg)	Apgar test 5′	Pain measurement	Intervention	Characteristics	Dose
Tasci et al., 2020 [36]	Turkey	IG: 42 (NA) CG: 42 (NA)	IG: 38–42 CG: 38–42	IG: 2.5-4 CG: 2.5-4	IG: >8 CG: >8	Cortisol levels and NIPS scale	IG: Breast milk odor CG: Formula milk odor	Filter paper impregnated with milk near the nose 3 min before, removed 9 min after heel-prick. Cortisol levels and NIPS scale were measured before, during and 2 min after puncture.	2 ml breast milk 2 ml formula milk
Boyer et al., 2004 [37]	Canada	IG: 27 (NA) CG: 30 (NA)	IG: 28.2 CG: 28.1	IG: NA CG: NA	IG: NA CG: NA	Cortisol levels	IG: 24% Sucrose CG: Sterile water	Sucrose or sterile water was administered before heel-prick. Cortisol levels were measured before and 30 min after heel-prick.	Three doses of 0.1 ml of 24% sucrose or sterile water.
Orellana et al., 2019 [38]	Spain	IG: 20 (NA) CG: 17 (NA)	IG: 35 CG: 34.6	IG: 2.005 CG: 2.050	IG: NA CG: NA	Cortisol levels and NIPS scale	IG: 24% Sucrose + non-nutritive sucking CG: Sterile water	Sucrose or sterile water was administered 2 min before heel-prick. Cortisol levels were measured 10 min before and 1 min after the puncture. NIPS scale were measured before, during, and 10 min after puncture.	0.5 ml of 24% sucrose or sterile water.
Rosemary et al., 2009 [39]	USA	IG1: 14 (9) IG2: 16 (6) CG: 10 (7)	IG1: 38.9 IG2: 39.2 CG: 38.3	IG1: 3.284 IG2: 3.306 CG: 3.222	IG1: 9 IG2: 9 CG: 8.9	Cortisol levels	IG1: SSC IG2: ATVV CG: Standard nursing care	Cortisol samples were collected before, following and 10 min post intervention.	SSC or ATVV 15 min before heel-prick.
Takahashi et al., 2011 [40]	Japan	IG: 61 CG: 18	IG: 39.8 CG: 39.7	IG: 3.056 CG: 3.152	IG: 9.5 CG: 9.4	Cortisol levels	IG: SSC for more than 60 min CG: SSC for less than 60 min	Cortisol levels were measured pre intervention, 60 min and 120 min post intervention.	SSC more or less than 60 min
Morelius et al., 2015 [41]	Sweden	IG: 18 (7) CG: 19 (13)	IG: 34 CG: 34	IG: 2.468 CG: 2.512	IG: NA CG: NA	Cortisol levels	IG: SSC CG: Standard nursing care	Cortisol levels were collected before and 30 min after heel-prick.	SSC pre and 30 min after heel-prick.
Qiu et al., 2017 [42]	China	IG: 30 (17) CG: 32 (17)	IG: 34.30 CG: 33.33	IG: 1.93 CG: 2.00	IG: 9.64 CG: 9.40	Cortisol levels and PIPP scale	IG: SSC CG: Standard nursing care	Cortisol levels were measured before and after puncture.	SSC pre and after heel-prick.
Haley et al., 2006 [43]	Canada	IG1: 22 (11) IG2: 17 (5)	IG1: < 28 IG2: > 32	IG1: 1.346 IG2: 3.412	IG1: NA IG2: NA	Cortisol levels	IG1 and IG2: Standard nursing care	Cortisol samples were collected before and 20 min after the heel-prick.	
D'Agata et al., 2019 [44]	USA	IG1: 32 (10) IG2: 43 (25)	IG1: < 28 IG2: 28–32	IG1: 0.956 IG2: 1.205	IG1: NA IG2: NA	Cortisol levels	IG1 and IG2: Standard nursing care	Cortisol levels were measured pre-intervention and post-intervention.	
Nelson et al., 2001 [45]	Sweden	IG: 11 (7)	IG: > 32	IG: 3.450	IG: > 7	Cortisol levels	IG: Standard nursing care	The baseline sample of cortisol was taken just before heel-prick, and the other sample 30 min after heel-prick.	

Abbreviations: skin to skin contact (SSC), Auditory-tactile-visual-vestibular stimulation (ATVV), intervention group (IG), control group (CG), not available (NA), neonatal infant pain scale (NIPS), premature infant pain profile (PIPP)

Risk of Bias

As per the RoB2 assessment in RCTs, only the 14,3% of the studies exhibited a high risk, while 28,6% displayed a moderate risk, and finally the 57,1% indicated a low risk. The details of the included trials are shown in Supplementary Fig. 1 and Supplementary Table 1. In RCTs, the ROBINS-I scale showed that 60% of the articles had a low risk, 20% displayed a moderate risk, 10% serious risk, and another 10% critical risk. Supplementary Fig. 2 presents the details of ROBINS-I scale.

Effects of non-pharmacological therapies on cortisol levels

The meta-analysis found that non-pharmacological therapies presented a large effect (SMD = -0.89; 95% CI = -1.49 to -0.28; n = 388; z = 2.88; P = 0.004) on neonatal pain with high heterogeneity (I^2 = 86%) when compared with control group or standard nursing care (Fig. 2).

The subgroup breastfeeding exhibited a moderate effect size (SMD = -0.63; 95% CI = -1.07 to -0.19; n = 84; z = 2.81), significant differences (P = 0.005). Sucrose vs. control group showed a small effect size (SMD = -0.15; 95% CI = -0.55 to 0.26; n = 95; z = 0.70); and non-significant differences (P = 0.48), between experimental and control group, and there is no heterogeneity (I^2 = 0%). Skin to skin contact exhibited a large effect size (SMD = -1.34; 95% CI = -2.21 to -0.46; n = 209; z = 2.99), significant differences (P = 0.003), and high heterogeneity (I^2 = 86%). The funnel plot presented a potential publication bias (Supplementary Fig. 3).

Differences in cortisol levels according to weeks of gestational age

The meta-analysis found that differences in weeks of gestational age presented a small effect (SMD = 0.07; 95% CI = -1.00 to 1.14; n = 250; z = 0.13; P = 0.90) on cortisol levels with high heterogeneity (I^2 = 93%), when compared pre and post-intervention groups (Fig. 3).

Patients under 28 weeks have less post-intervention pain and showed a large effect (SMD = 1.44; 95% CI = 0.47 to 2.40; n = 108; z = 2.92), significant differences (P = 0.004), and high heterogeneity (I^2 = 78%).

Patients between 28 and 32 weeks have more post-intervention pain and presented a small effect (SMD = -0.43; 95% CI = -0.86 to -0.0; n = 86; z = 1.98), non-significant differences (P = 0.05). Finally, patients over 32 weeks have an increased post-intervention pain and exhibited a large effect (SMD = -1.08; 95% CI = -1.65 to -0.51; n = 56; z = 3.74), significant differences (P = 0.0002), and there is no heterogeneity (I^2 = 0%). The funnel plot presented a potential publication bias (Supplementary Fig. 4).

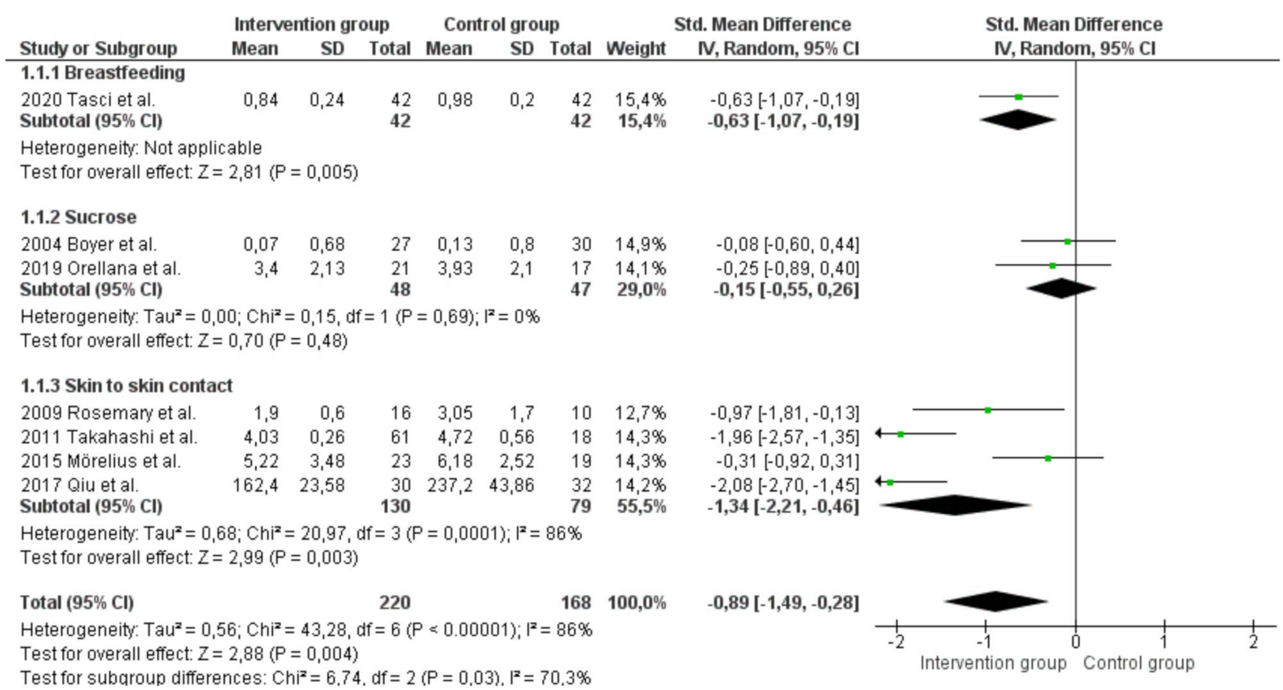

Fig. 2 Effects of non-pharmacological therapies on cortisol levels in neonatal patients. The forest plot illustrates the overall effects on pain between experimental and control groups. Squares represent the standardized (Std.) mean difference (SMD) for each trial. Diamonds represent the pooled SMD across trials

Study or Subgroup	Pre-intervention Mean	SD	Total	Post-intervention Mean	SD	Total	Weight	Std. Mean Difference IV, Random, 95% CI
1.2.1 < 28 weeks								
2006 Haley et al.a	0,3	0,1	22	0,1	0,1	22	19,8%	1,96 [1,23, 2,69]
2019 D'Agata et al.a	7,98	1,22	32	6,6	1,55	32	20,7%	0,98 [0,46, 1,50]
Subtotal (95% CI)			**54**			**54**	**40,5%**	**1,44 [0,47, 2,40]**
Heterogeneity: Tau² = 0,38; Chi² = 4,65, df = 1 (P = 0,03); I² = 78%								
Test for overall effect: Z = 2,92 (P = 0,004)								
1.2.2 28-32 weeks								
2019 D'Agata et al.b	4,48	1,21	43	5,77	4	43	21,0%	-0,43 [-0,86, -0,00]
Subtotal (95% CI)			**43**			**43**	**21,0%**	**-0,43 [-0,86, -0,00]**
Heterogeneity: Not applicable								
Test for overall effect: Z = 1,98 (P = 0,05)								
1.2.3 > 32 weeks								
2001 Nelson et al.	11,22	3,56	11	18,33	6,8	11	18,7%	-1,26 [-2,19, -0,33]
2006 Haley et al.b	0,2	0,1	17	0,3	0,1	17	19,8%	-0,98 [-1,69, -0,26]
Subtotal (95% CI)			**28**			**28**	**38,5%**	**-1,08 [-1,65, -0,51]**
Heterogeneity: Tau² = 0,00; Chi² = 0,22, df = 1 (P = 0,64); I² = 0%								
Test for overall effect: Z = 3,74 (P = 0,0002)								
Total (95% CI)			**125**			**125**	**100,0%**	**0,07 [-1,00, 1,14]**
Heterogeneity: Tau² = 1,36; Chi² = 58,73, df = 4 (P < 0.00001); I² = 93%								
Test for overall effect: Z = 0,13 (P = 0,90)								
Test for subgroup differences: Chi² = 19,44, df = 2 (P < 0.0001), I² = 89,7%								

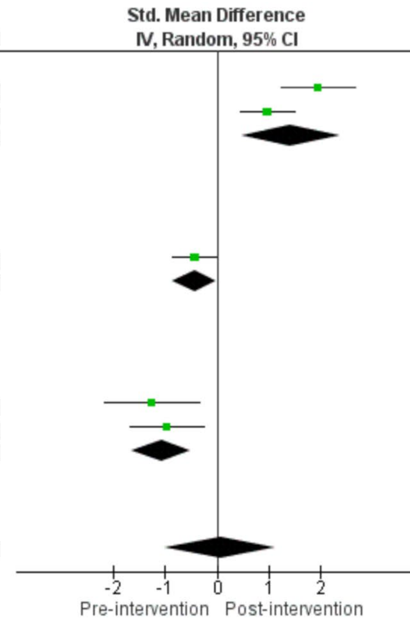

Fig. 3 Differences in cortisol levels according to weeks of gestational age. The forest plot illustrates the overall effects on pre and post-intervention pain. Squares represent the standardized (Std.) mean difference (SMD) for each trial. Diamonds represent the pooled SMD across trials

Discussion

This systematic review and meta-analysis evaluate the effectiveness of non-pharmacological analgesia methods, used to alleviate pain in newborns undergoing invasive procedures by analysing neonatal cortisol levels as biological markers of pain. Our results suggest that the application of breastfeeding as a non-pharmacological intervention significantly reduces neonatal pain measured by cortisol levels, after heel lance. Similarly, the use of skin to skin contact has a positive and significant effect on lowering neonatal cortisol levels. However, the cortisol data show that administration of sucrose prior to heel prick was not effective in reducing pain. Specifically, our findings indicate that skin to skin contact has even more effect than breastfeeding on lowering cortisol levels, and thus, on neonatal pain. According to gestational age, newborns under 28 weeks have lower cortisol levels and less post-intervention pain, so non-pharmacological therapies are effective in this group of patients. But, on the other side, patients between 28 and 32 weeks have higher cortisol levels after procedures compared to pre-intervention levels. Likewise, newborns over 32 weeks experience more pain following interventions, indicating that the non-pharmacological methods studied are not effective in reducing pain in this group.

The effect of cortisol has been studied as the most commonly selected biomarker to measure both acute and chronic stress responses, but in the pediatric population it is an area that needs further research and development [46, 47]. Cortisol is a glucocorticoid hormone produced in the adrenal cortex, its synthesis begins around 28–30 weeks of gestation, and it is regulated by the hypothalamic-pituitary-adrenal axis (HPA). For this reason, cortisol levels in preterm infants are lower than in term infants, and data from our meta-analysis refute this statement [48–50]. Specifically, salivary cortisol is considered a feasible option for assessing HPA axis activity, due to the non-invasive method of sample collection, which would be ideal to implement in neonatal units [51]. The analysis explores how cortisol levels can predict the presence of stress in newborns and how these levels can decrease when non-pharmacological analgesia therapies are employed [52, 53].

Stress is a generic term that includes various symptoms such as pain, agitation or irritability [54]. Infants born preterm have impaired stress regulation, based on cortisol production values considered normal. Preterm babies spend their first weeks of life in the NICU receiving critical care [55]. Stress related to this unit has been associated with cortisol levels that are dysregulated in preterm infants in response to a stressor [56]. Research has found associations between exposure to neonatal pain and neonatal stress due to the number of invasive procedures during the NICU stay [55].

Preterm infants are more sensitive to pain and stress, which cannot be distinguished in neonates with pain assessment tools, hence the term 'pain/stress' is used [57]. In addition, higher cortisol levels were associated with greater exposure to pain/stress. It is important to note that infants exposed to more pain/stress from invasive procedures showed elevated cortisol levels prior to cortisol testing [55]. Multiple factors may contribute to these higher cortisol levels in preterm infants including cumulative stress related to multiple procedures [58]. It is ethically problematic to apply pain or a specific stimulus to young children for research purposes. The lack of standardised protocols for the assessment of stress reactivity has hindered the reproducibility and replicability of infant physiological stress studies [55].

In a systematic review the main selected articles established that cortisol is a reliable indicator of physiological stress. Furthermore, variations in cortisol throughout the day may be an indicator of a state of perceived high stress [54]. Cortisol reactivity in infancy has been assessed in various experimental settings including physical stressors (e.g. blood draw). The results of one meta-analysis emphasize that strong cortisol responses to painful stimuli have been observed [59].

The published studies indicate that newborns who received breastfeeding showed a quicker stabilization after heel lance [60–62]. This is due to the high lactose content in breast milk and essential amino acids like L-tryptophan, which promote the release of internal pain-relieving opioids including beta-endorphins. These substances are involved in reducing the transmission of pain to the nervous system [63, 64]. In addition, after ingestion of breast milk, cholecystokinin (CKK), a neuropeptide that induces a calming effect by causing drowsiness, is released [65, 66]. These mechanisms explain the ability of breastfeeding to relieve pain and to be one of the most widely used approaches with the highest level of evidence [67, 68]. The results of our meta-analysis are consistent with these investigations, showing that breastfeeding is one of the most effective therapies for reducing pain in newborns, undergoing invasive procedures such as heel prick.

Regarding the influence of the skin to skin contact, research suggests that groups in which this method has been implemented achieved lower cortisol levels during and after heel prick [69, 70]. Our outcomes align with these investigations, as skin to skin contact has been shown to have a positive and significant impact on reducing cortisol levels and neonatal pain. The way skin to skin contact generates analgesic effect is through direct mother-infant interaction combined with maternal smell, bonding and heartbeat [71, 72]. These processes stimulate the release of hormones such as oxytocin in the newborn, which decreases sympathetic nervous system function, and correlates with a calm and restful state, that ultimately improves hemodynamic stability [73].

Finally, concerning to the use of oral sucrose, there are contradictions in published evidence. There is a stream of articles recommending its use because the sweet taste of sucrose causes the release of endorphins and serotonins, producing and analgesic effect that lasts between 5 and 10 min [74, 75]. These publications conclude that sucrose is successful in reducing the manifestations of neonatal pain [76]. Conversely, other studies suggest that this non-pharmacological therapy does not seem to reduce heel stick pain compared to breastfeeding and skin to skin contact [77–80]. Data for our meta-analysis also indicate that there is no difference in cortisol levels between the control and intervention groups. In addition, questions regarding the most effective dose and concentration per day, as well as possible long-term negative impact of sucrose use, remain unresolved. Therefore, the effectiveness of using this method is not conclusive yet [81].

Non-pharmacological interventions to reduce stress in newborns may be brain-protective. During weeks to months in the NICU, very preterm infants are exposed to a high number of life-saving skin-breaking procedures and interventions, as well as routine handling that elicit behavioral, physiological and hormonal responses [57].

Nursing has the capacity to be at the forefront of care, being a driver of change for innovation in healthcare [82]. The use of new methods to measure and reduce pain aims to humanise nursing care, by encouraging the involvement of parents as primary caregivers [83, 84]. The union of the family with the care team is essential to achieve the maximum wellbeing, and communication is the basis for providing adequate care [85]. Nursing care in these units involves combining aspects derived from new technologies with the human dimension, due to the direct and continuous contact established with patient and her family [86]. Care centred only on pathological and procedural aspects would not take into account the newborn as a biopsychosocial being. The nursing team occupies a privileged place in neonatal units, as it is able to minimise the impact of this experience [87].

Study limitations

An important limitation is the small number of published articles evaluating cortisol levels and non-pharmacological analgesia therapies in neonatal patients. The high heterogeneity of the reviewed studies can be explained by the inclusion of studies in which participants had different sample sizes and gestational ages. Additionally, discrepancies in the type of intervention and dose administered may have influenced the overall consistency of the results. Only studies published in English were included in this meta-analysis. Also, we only included randomized controlled trials, and quasi-experimental studies. So our results are limited to the studies included in this review. Therefore, additional research is imperative to bolster the confidence needed to implement non-pharmacological analgesia therapies in neonatal units.

Conclusions

The non-pharmacological therapies examined showed that skin to skin contact is the most effective method to reduce pain measured by cortisol levels, from invasive procedures such as heel pricks in preterm infants under 28 weeks. Breastfeeding also demonstrated to be an effective and safe alternative to use for pain relief and to reduce cortisol levels. However, the cortisol data indicate that sucrose was not effective in reducing neonatal pain. These measures highlight the importance of integrating proactive approaches to mitigate acute pain derived from standard nursing care. The use of these therapies may lead to a reduction in the use of drugs in neonatal units, avoiding possible side effects, as they are considered a more natural alternative to the conventional pharmacological paradigm. In terms of research, this meta-analysis accentuates the identification of gaps in existing knowledge and underlines the need for further investigations to validate the safety and effectiveness of the suggested interventions.

Abbreviations

IASP	International Association for the Study of Pain
NICUs	Neonatal Intensive Care Units
WHO	World Health Organization
INE	National Institute of Statistics
NIDCAP	Newborn Individualized Developmental Care and Assessment Program
PIPP	Premature Infant Pain Profile
NIPS	Neonatal Infant Pain Scale
NIAPAS	Neonatal Infant Acute Pain Assessment Scale
AAP	American Academy of Pediatrics
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
RCT	Randomized Controlled Trial
ROB	Risk of Bias
ROBINS-I	Risk of Bias in Non-Randomized Studies
SMDs	Standardized Mean Differences
HPA	Hypothalamic-Pituitary-Adrenal axis
CKK	Cholecystokinin

Supplementary Information

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Supplementary Material 1

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Author contributions
Conceptualization I.G.-V and S.G.-C; methodology I.G.-V, J.S.-I and P.P.C; formal analysis I.G.-V, J.S.-I, C.A.-B and S.G.-C; investigation I.G.-V, J.S.-I, P.P.C and S.G.-C; resources B.Y.-A, S.H.-I, and F.P.C; data curation I.G.-V, J.S.-I, B.Y.-A and S.G.-C; writing—original draft preparation I.G.-V, J.S.-I and S.G.-C; writing—review and editing I.G.-V, J.S.-I, P.P.C, B.Y.-A and S.G.-C; visualization A.C.P and J.M.P-P; supervision P.P.C, S.H.-I, F.P.C and S.G.-C; project administration P.P.C, S.H.-I, F.P.C and S.G.-C. All authors reviewed the manuscript.

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Declarations

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References

1. Raja SN, Carr DB, Cohen M, Finnerup NB, Flor H, Gibson S, et al. The revised international association for the study of pain definition of pain: concepts, challenges, and compromises. Pain. 2020;161(9):1976–82.
2. Treede RD. The international association for the study of pain definition of pain: as valid in 2018 as in 1979, but in need of regularly updated footnotes. Pain Rep. 2018;3(2):e643.
3. Gursul D, Hartley C, Slater R. Nociception and the neonatal brain. Semin Fetal Neonatal Med. 2019;24(4):101–16.
4. Johnston C. Neonatal pain: A journey spanning three decades. Paediatr Neonatal Pain. 2020;2(2):33–9.
5. Fernández MGE, González-Pacheco N, Sánchez-Redondo MD, Cernada M, Martín A, Pérez-Muñuzuri A et al. Sedoanalgesia En Las unidades neonatales. 2021; 95 (2): 1–11.
6. Carbajal R, Eriksson M, Courtois E, Boyle E, Avila-Alvarez A, Andersen RD, et al. Sedation and analgesia practices in neonatal intensive care units (EUROPAIN): results from a prospective cohort study. Lancet Respir Med. 2015;3(10):796–812.
7. Ohuma EO, Moller AB, Bradley E, Chakwera S, Hussain-Alkhateeb L, Lewin A, et al. National, regional, and global estimates of preterm birth in 2020, with trends from 2010: a systematic analysis. Lancet. 2023;402(10409):1261–71.
8. Instituto Nacional de Estadística. Nacimientos por Tipo de Parto, Tiempo de Gestación y Grupo de Edad de la Madre; Instituto Nacional de Estadística: Madrid, Spain, 2023.
9. Shiff I, Bucsea O, Pillai Riddell R. Psychosocial and Neurobiological vulnerabilities of the hospitalized preterm infant and relevant non-pharmacological pain mitigation strategies. Front Pediatr. 2021;25(9):568755.
10. McPherson C, Miller SP, El-Dib M, Massaro AN, Inder TE. The influence of pain, agitation, and their management on the immature brain. Pediatr Res. 2020;88(2):168–75.
11. Sheldon RE. Developmental care for preemies and their families: one neonatologist's journey toward NIDCAP practice. Neo Rev. 2017;18:568–75.
12. Harrillo-Acevedo FD, Rico-Becerra JI, López-Martínez A. The philosophy of the developmental centered care of the premature infant (NIDCAP): A literature review. Enferm Glob. 2017;16(48):577–89.

13. Meesters NJ, Simons SHP, Van Rosmalen J, Holsti L, Reiss IKM, Van Dijk M. Acute pain assessment in prematurely born infants below 29 weeks: A long way to go. Clin J Pain. 2019;35(12):975–82.
14. Maxwell LG, Fraga MV, Malavolta CP. Assessment of pain in the newborn: an update. Clin Perinatol. 2019;46(4):693–707.
15. Weng Y, Zhang J, Chen Z. Effect of non-pharmacological interventions on pain in preterm infants in the neonatal intensive care unit: a network meta-analysis of randomized controlled trials. BMC Pediatr. 2024;24(1):9.
16. McPherson C, Smith JR. Analgesia in the premature neonate: walking the tightrope requires a great team. J Perinat Neonatal Nurs. 2017;31(2):99–100.
17. Claassen K, Thelen K, Coboeken K, Gaub T, Lippert J, Allegaert K, et al. Development of a Physiologically-Based Pharmacokinetic model for preterm neonates: evaluation with in vivo data. Curr Pharm Des. 2015;21(39):5688–98.
18. Olsson E, Ahl H, Bengtsson K, Vejayaram DN, Norman E, Bruschettini M, Eriksson M. The use and reporting of neonatal pain scales: a systematic review of randomized trials. Pain. 2020;162(2):353–60.
19. Gibbins S, Stevens BJ, Yamada J, Dionne K, Campbell-Yeo M, Lee G, et al. Validation of the premature infant pain Profile-Revised (PIPP-R). Early Hum Dev. 2014;90(4):189–93.
20. Hudson-Barr D, Capper-Michel B, Lambert S, Palermo TM, Morbeto K, Lombardo S. Validation of the pain assessment in neonates (PAIN) scale with the neonatal infant pain scale (NIPS). Neonatal Netw. 2002;21(6):15–21.
21. Pölkki T, Korhonen A, Axelin A, Saarela T, Laukkala H. Development and preliminary validation of the neonatal infant acute pain assessment scale (NIAPAS). Int J Nurs Stud. 2014;51(12):1585–94.
22. Glenzel L, do Nascimento Oliveira P, Marchi BS, Ceccon RF, Moran CA. Validity and reliability of pain and behavioral scales for preterm infants: A systematic review. Pain Manag Nurs. 2023;24(5):84–96.
23. Ten Barge JA, Baudat M, Meesters NJ, Kindt A, Joosten EA, Reiss IKM, et al. Biomarkers for assessing pain and pain relief in the neonatal intensive care unit. Front Pain Res. 2024;15(5):1343551.
24. Herrington CJ, Olomu IN, Geller SM. Salivary cortisol as indicators of pain in preterm infants: a pilot study. Clin Nurs Res. 2004;13(1):53–68.
25. Olszewska M, Pointinger-Tomasik S, Kwinta P. Assessment of salivary cortisol concentrations for procedural pain monitoring in newborns. J Perinat Med. 2023;51(4):564–72.
26. Dionysakopoulou C, Lianou L, Boutopoulou B, Giannakopoulou M, Vlachioti E, Koumpagioti D, et al. The role of substance P, neurokinin A, neuropeptide Y, and cortisol in assessing neonatal pain. Neonatal Netw. 2023;42(2):65–71.
27. Forclaz MV, Moratto E, Pennisi A, Falco S, Olsen G, Rodríguez P, et al. Salivary and serum cortisol levels in newborn infants. Arch Argent Pediatr. 2017;115(3):262–6.
28. Grunau RE, Weinberg J, Whitfield MF. Neonatal procedural pain and preterm infant cortisol response to novelty at 8 months. Pediatrics. 2004;114(1):77–84.
29. Lim Y, Godambe S. Prevention and management of procedural pain in the neonate: an update. Am Acad Pediatr 2016 Arch Dis Child Educ Pract Ed. 2017;102(5):254–6.
30. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffman TC, Mulrow CD, Shamseer L, Tetzlaff JM, Akl EA, Brennan SE, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. BMJ. 2021;372(29):71.
31. Cumpston M, Li T, Page MJ, Chandler J, Welch VA, Higgins JP, Thomas J. Updated guidance for trusted systematic reviews: A new edition of the Cochrane handbook for systematic reviews of interventions. Cochrane Database Syst Rev. 2019;10(10):ED000142.
32. Sterne JA, Savovic J, Page MJ, Elbers RG, Blencowe NS, Boutron I, et al. RoB 2: A revised tool for assessing risk of bias in randomized trials. BMJ. 2019;366:48–98.
33. Sterne JA, Hernán MA, Reeves BC, Savovic J, Berkman ND, Viswanathan M, et al. ROBINS-I: A tool for assessing risk of bias in non-randomized studies of interventions. BMJ. 2016;355:19–49.
34. Lachenbruch PA, Cohen J. Statistical power analysis for the behavioural sciences. J Am Stat Assoc. 1989;84:1096. 2nd ed.
35. Sterne JA, Egger M, Smith GD. Systematic reviews in health care: investigating and dealing with publication and other biases in meta-analysis. BMJ. 2001;323:101–5.
36. Tasci B, Kuzlu Ayyildiz T. The calming effect of maternal breast milk odor on term infant: A randomized controlled trial. Breastfeed Med. 2020;15(11):724–30.
37. Boyer K, Johnston C, Walker CD, Filion F, Sherrard A. Does sucrose analgesia promote physiologic stability in preterm neonates? Biol Neonate. 2004;85(1):26–31.
38. Orellana GF, Ortiz ES, Ibáñez AB, Albuixech RC, Camino ML, Betegón AA. Evaluation of oral sucrose effectiveness by saliva cortisol determination and the NIPS scale for pain assessment in newborns. Nure Inv. 2019;16(98):1–14.
39. White-Traut RC, Schwertz D, McFarlin B, Kogan J. Salivary cortisol and behavioral state responses of healthy newborn infants to tactile-only and multisensory interventions. J Obstet Gynecol Neonatal Nurs. 2009;38(1):22–34.
40. Takahashi Y, Tamakoshi K, Matsushima M, Kawabe T. Comparison of salivary cortisol, heart rate, and oxygen saturation between early skin-to-skin contact with different initiation and duration times in healthy, full-term infants. Early Hum Dev. 2011;87(3):151–7.
41. Mörelius E, Örtenstrand A, Theodorsson E, Frostell A. A randomised trial of continuous skin-to-skin contact after preterm birth and the effects on salivary cortisol, parental stress, depression, and breastfeeding. Early Hum Dev. 2015;91(1):63–70.
42. Qiu J, Jiang YF, Li F, Tong QH, Rong H, Cheng R. Effect of combined music and touch intervention on pain response and β-endorphin and cortisol concentrations in late preterm infants. BMC Pediatr. 2017;17(1):38.
43. Haley DW, Weinberg J, Grunau RE. Cortisol, contingency learning, and memory in preterm and full-term infants. Psychoneuroendocrinology. 2006;31(1):108–17.
44. D'Agata AL, Roberts MB, Ashmeade T, Dutra SVO, Kane B, Groer MW. Novel method of measuring chronic stress for preterm infants: skin cortisol. Psychoneuroendocrinology. 2019;102:204–11.
45. Nelson N, Arbring K, Theodorsson E. Neonatal salivary cortisol in response to heelstick: method modifications enable analysis of low concentrations and small sample volumes. Scand J Clin Lab Invest. 2001;61(4):287–91.
46. Olszewska M, Kwinta P. The influence of repeated pain exposure on morning salivary cortisol in term and preterm neonates. Pediatr Endocrinol Diabetes Metab. 2023;29(1):4–9.
47. Gibbins S, Stevens B, Beyene J, Chan PC, Bagg M, Asztalos E. Pain behaviours in extremely low gestational age infants. Early Hum Dev. 2008;84(7):451–8.
48. Davis EP, Granger DA. Developmental differences in infant salivary alpha-amylase and cortisol responses to stress. Psychoneuroendocrinology. 2009;34(6):795–804.
49. Jansen J, Beijers R, Riksen-Walraven M, de Weerth C. Cortisol reactivity in young infants. Psychoneuroendocrinology. 2010;35(3):329–38.
50. Provenzi L, Giorda R, Fumagalli M, Brambilla M, Mosca F, Borgatti R, et al. Telomere length and salivary cortisol stress reactivity in very preterm infants. Early Hum Dev. 2019;129:1–4.
51. Provenzi L, Giusti L, Fumagalli M, Tasca H, Ciceri F, Menozzi G, et al. Pain-related stress in the neonatal intensive care unit and salivary cortisol reactivity to socio-emotional stress in 3-month-old very preterm infants. Psychoneuroendocrinology. 2016;72:161–5.
52. Roué JM, Rioualen S, Gendras J, Misery L, Gouillou M, Sizun J. Multi-modal pain assessment: are near-infrared spectroscopy, skin conductance, salivary cortisol, physiologic parameters, and neonatal facial coding system interrelated during venepuncture in healthy, term neonates? J Pain Res. 2018;11(11):2257–67.
53. Turpeinen U, Hämäläinen E. Determination of cortisol in serum, saliva and urine. Best Pract Res Clin Endocrinol Metab. 2013;27(6):795–801.
54. Aguilar Cordero MJ, Sánchez López AM, Mur Villar N, García García I, Rodríguez López MA, Ortegón Piñero A, et al. Cortisol salival Como indicador de estrés fisiológico En Niños y Adultos: revisión Sistemática. Nutr Hosp. 2014;29(5):960–8.
55. McLean MA, Nakajima L, Chau CMY, Weinberg J, Synnes AR, Miller SP, et al. Cortisol levels are related to neonatal pain exposure in children born very preterm at age 18 months in two independent cohorts. Paediatr Neonatal Pain. 2023;5(3):86–95.
56. Ginnell L, O'Carroll S, Ledsham V, Jiménez Sánchez L, Stoye DQ, Sullivan G, et al. Emotion regulation and cortisol response to the still-face procedure in preterm and full-term infants. Psychoneuroendocrinology. 2022;141:105760.
57. Grunau RE. Neonatal pain in very preterm infants: Long-term effects on brain, neurodevelopment and pain reactivity. Rambam Maimonides Med J. 2013;4(4):e0025.
58. Grunau RE, Haley DW, Whitfield MF, Weinberg J, Yu W, Thiessen P. Altered basal cortisol levels at 3, 6, 8 and 18 months in infants born at extremely low gestational age. J Pediatr. 2007;150(2):151–6.
59. Puhakka IJA, Peltola MJ. Salivary cortisol reactivity to psychological stressors in infancy: A meta-analysis. Psychoneuroendocrinology. 2020;115:104603.
60. Lan HY, Yang L, Lin CH, Hsieh KH, Chang YC, Yin T. Breastmilk as a multisensory intervention for relieving pain during newborn screening procedures: A randomized control trial. Int J Environ Res Public Health. 2021;18(24):123–30.

61. Benoit B, Newman A, Martin-Misener R, Latimer M, Campbell-Yeo M. The influence of breastfeeding on cortical and bio-behavioural indicators of procedural pain in newborns: findings of a randomized controlled trial. Early Hum Dev. 2021;154:105–308.
62. Aydin D, Inal S. Effects of breastfeeding and heel warming on pain levels during heel stick in neonates. Int J Nurs Pract. 2019;25(3):127–34.
63. Wu HP, Yin T, Hsieh KH, Lan HY, Feng RC, Chang YC, et al. Integration of different sensory interventions from mother's breast milk for preterm infant pain during peripheral venipuncture procedures: A prospective randomized controlled trial. J Nurs Scholarsh. 2020;52(1):75–84.
64. Avcin E, Kucukoglu S. The effect of breastfeeding, Kangaroo care, and facilitated tucking positioning in reducing the pain during heel stick in neonates. J Pediatr Nurs. 2021;61:410–6.
65. Velumula PK, Elbakoush F, Tabb C 2nd, Farooqi A, Lulic-Botica M, Jani S, Natarajan G, Bajaj M. Breast milk vs 24% sucrose for procedural pain relief in preterm neonates: a non-inferiority randomized controlled trial. J Perinatol. 2022;42(7):914–9.
66. Bembich S, Cont G, Causin E, Paviotti G, Marzari P, Demarini S. Infant analgesia with a combination of breast milk, glucose, or maternal holding. Pediatrics. 2018;142:16–34.
67. Harrison D, Reszel J, Bueno M, Sampson M, Shah VS, Taddio A, Larocque C, Turner L. Breastfeeding for procedural pain in infants beyond the neonatal period. Cochrane Database Syst Rev. 2016;10:112–48.
68. Shah PS, Torgalkar R, Shah VS. Breastfeeding or breast milk for procedural pain in neonates. Cochrane Database Syst Rev. 2023;8(8):CD004950.
69. Pavlyshyn H, Sarapuk I. Skin-to-skin contact an effective intervention on pain and stress reduction in preterm infants. Front Pediatr. 2023;22(11):1148946.
70. Olsson E, Ahlsén G, Eriksson M. Skin-to-skin contact reduces near-infrared spectroscopy pain responses in premature infants during blood sampling. Acta Paediatr. 2016;105(4):376–80.
71. Johnston C, Campbell-Yeo M, Disher T, Benoit B, Fernandes A, Streiner D et al. Skin-to-skin care for procedural pain in neonates. Cochrane Database Syst Rev. 2017;2(2):Cd008435.
72. Nimbalkar S, Shukla VV, Chauhan V, Phatak AG, Patel D, Chapla A, et al. Blinded randomized crossover trial: Skin-to-skin care vs. sucrose for preterm neonatal pain. J Perinatol. 2020;40(6):896–901.
73. Pavlyshyn H, Sarapuk I, Horishna I, Slyva V, Skubenko N. Skin-to-skin contact to support preterm infants and reduce NICU-related stress. Int J Dev Neurosci. 2022;82(7):639–45.
74. Tutag Lehr V, Cortez J, Grever W, Cepeda E, Thomas R, Aranda JV. Randomized placebo-controlled trial of sucrose analgesia on neonatal skin blood flow and pain response during heel lance. Clin J Pain. 2015;31(5):451–8.
75. Gao H, Gao H, Xu G, Li M, Du S, Li F, et al. Efficacy and safety of repeated oral sucrose for repeated procedural pain in neonates: A systematic review. Int J Nurs Stud. 2016;62:118–25.
76. Oliveira DJD, Medeiros KS, Sarmento ACA, Oliveira FJD, Costa APF, Souza NL, et al. Use of glucose for pain management in premature neonates: a systematic review and meta-analysis protocol. BMJ Open. 2021;11(12):e052901.
77. Yamada J, Bueno M, Santos L, Haliburton S, Campbell-Yeo M, Stevens B. Sucrose analgesia for heel-lance procedures in neonates. Cochrane Database Syst Rev. 2023;8(8):CD014806.
78. Chang J, Filoteo L, Nasr AS. Comparing the analgesic effects of 4 non-pharmacologic interventions on term newborns undergoing heel Lance: A randomized controlled trial. J Perinat Neonatal Nurs. 2020;34(4):338–45.
79. Lago P, Cavicchiolo ME, Mion T, Dal Cengio V, Allegro A, Daverio M, et al. Repeating a dose of sucrose for heel Prick procedure in preterms is not effective in reducing pain: A randomised controlled trial. Eur J Pediatr. 2020;179(2):293–301.
80. Banga S, Datta V, Rehan HS, Bhakhri BK. Effect of sucrose analgesia, for repeated painful procedures, on short-term neurobehavioral outcome of preterm neonates: A randomized controlled trial. J Trop Pediatr. 2016;62(2):101–6.
81. Stevens B, Yamada J, Campbell-Yeo M, Gibbins S, Harrison D, Dionne K, et al. The minimally effective dose of sucrose for procedural pain relief in neonates: a randomized controlled trial. BMC Pediatr. 2018;18(1):85–93.
82. Gómez-Cantarino S, García-Valdivieso I, Dios-Aguado M, Yáñez-Araque B, Molina Gallego B, Moncunill-Martínez E. Nursing perspective of the humanized care of the neonate and family: A systematic review. Children. 2021;8(1):35.
83. Mangat AK, Oei JL, Chen K, Quah-Smith I, Schmözler GM. A review of non-pharmacological treatments for pain management in newborn infants. Children. 2018;5(10):130.
84. Guo W, Liu X, Zhou X, Wu T, Sun J. Efficacy and safety of combined non-pharmacological interventions for repeated procedural pain in preterm neonates: A systematic review of randomized controlled trials. Int J Nurs Stud. 2019;102:103471.
85. Hatfield LA, Murphy N, Karp K, Polomano RC. A systematic review of behavioural and environmental interventions for procedural pain management in preterm infants. J Pediatr Nurs. 2019;44:22–30.
86. Gómez-Cantarino S, García-Valdivieso I, Moncunill-Martínez E, Yáñez-Araque B, Ugarte Gurrutxaga MI. Developing a family-centered care model in the neonatal intensive care unit (NICU): A new vision to manage healthcare. Int J Environ Res Public Health. 2020;17(19):7197.
87. Avila-Alvarez A, Carbajal R, Courtois E, Pertega-Diaz S, Anand KJ, Muñiz-Garcia J. Clinical assessment of pain in Spanish neonatal intensive care units. Pediatr (Barc). 2016;85(4):181–8.

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