

Potential of N-acetylcysteine in the management of low back pain: a scoping review of studies in humans and animal models

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Abstract

Low back pain (LBP) is a common type of pain that causes disability and impairs cognitive function. With over 80% of adults estimated to experience LBP during their lifetime, this type of pain not only has a significant impact on the individual, but also on public health systems and national economies. Unfortunately, there is no single standard of care for patients with LBP. N-acetylcysteine (NAC), which is used clinically to treat acetaminophen overdose, has recently been tested as a potential treatment for LBP. NAC is inexpensive and commercially available, and it has an established tolerance and safety profile. However, NAC's efficacy in LBP has not been established. This scoping review presents a summary of studies investigating the effects of NAC and the potential benefits in LBP treatment, and highlights its potential molecular mechanisms and side effects. A systematic literature search in Pubmed/MEDLINE, Embase, Scopus, Science Direct, Web of Science, Cinahl, and Lilacs databases was conducted. The PRISMA-ScR checklist was used to ensure integrity of the review. The scoping review protocol was registered in the Open Science Framework. No limit was set on study language and publication date. In total, 2357 articles were located, of which 16 were included. The studies show that NAC has potential for LBP treatment, but data are derived only from a few clinical trials and preclinical studies. Thus, there is much to learn and more clinical studies should be performed before NAC can be clinically recommended for the treatment of LBP.

Key words: Back pain; Chronic pain; Clinical studies; Preclinical studies; Reactive oxygen species; Mitogen-activated protein kinases

Introduction

Low back pain (LBP) is highly prevalent worldwide and covers a spectrum of different types of pain such as nociceptive, neuropathic, nociplastic, or non-specific (1). According to Urits et al. (2), LBP encompasses three distinct sources of pain: axial lumbosacral, radicular, and referred. The axial lumbosacral LBP is pain in the lumbar spine (L1-5) and sacral spine (S1 to the sacrococcygeal junction). Radicular LBP is leg pain that radiates into an extremity along a dermatomal distribution secondary to nerve or dorsal root ganglion irritation. Referred LBP is pain that spreads to a region distant from its source but along a non-dermatomal trajectory (2).

LBP contributes to disability (2) and impacts cognitive function (3). This type of pain has important implications for individuals, public health systems, and economies (4), since it is estimated that over 80% of adults will struggle with LBP during their lifetime (5). According to Otero-Ketterer et al. (4), direct healthcare costs with LBP are

comparable to those of cardiovascular disease, cancer, or mental health, while indirect costs are related to loss of work days. In addition, it is estimated that the incidence of LBP is likely to increase in low-income and middle-income countries in the next few decades (4).

LBP can be acute, subacute, and chronic (5). Acute LBP is defined as common, self-limiting, and lasting less than 4 weeks; subacute LBP lasts between 4 and 12 weeks and has increased risk of transitioning into chronic LBP; chronic LBP is defined as pain lasting more than 3 months (5). Regardless of the type of pain, a study reported that the point prevalence of LBP was 8.83% (95%CI: 7.04 to 8.64) in 2017, with 577 million people affected at any one time (6).

Acute and subacute LBP usually improve within 6 weeks and the average pain intensity is moderate (6). However, two-thirds of people with acute and subacute LBP still experience pain at 3 months and at 12 months (6).

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Received August 1, 2024 | Accepted January 31, 2025

This high number highlights the need for effective treatments for LBP. Unfortunately, a single standard of care for patients with LBP has not been established. For acute episodes, most guidelines of first line of care are advice, reassurance, and encouragement to engage in light physical activity; the pharmacological and physiotherapy interventions are available when second-line treatment is needed (6). For nonspecific LBP, there is some agreement on the use of non-steroidal anti-inflammatory drugs (NSAIDs) as first-line pharmacological therapy, and pharmacological and non-pharmacological approaches are added as needed (5). However, mild-moderate events have been treated with the use of heat wrap, muscle relaxants, NSAIDs, opioids, paracetamol, steroids, and inert treatment (6).

N-acetylcysteine (NAC) is a mucolytic agent clinically used to treat acetaminophen toxicity or overdose (7–10). NAC was included in the World Health Organization's list of essential medicines and is given orally, intravenously, or by inhalation (9). NAC has a favorable pharmacokinetic profile with few relevant drug-drug interactions, making it an ideal adjunct treatment to established therapies for complex diseases (8). In addition, NAC is commercially available and is considered to be an inexpensive medication (7–10).

Studies have demonstrated that NAC is a precursor of the antioxidant glutathione and modulates redox reactions (8–10). However, NAC has other actions. NAC appears to modulate the homeostasis of the neurotransmitters glutamate/dopamine, in addition to exhibiting antiviral and anti-inflammatory activity; NAC also appears to promote protein/DNA stabilization (8,10).

Improved knowledge of NAC's mechanisms of action has expanded its clinical applications. In particular, NAC has been evaluated for the treatment of chronic pain (8–11). A previous review analyzing a variety of pain conditions such as sickle cell disease, complex regional pain syndrome type-1/reflex sympathetic dystrophy, rheumatoid arthritis, painful diabetic neuropathy, pelvic pain/endometriosis, and chronic neuropathic pain reported that there is still insufficient evidence of NAC's analgesic efficacy in treating some chronic pain conditions (11). A previous review highlighted the protective role of glutathione and its precursor NAC in osteoarthritis (12), but there is no review on LBP and NAC currently. Although NAC is considered a safe substance, there are still doubts as to whether or not it has side effects (8–10). A previous review indicated that side effects tend to increase with increasing NAC dosage, but the authors commented that this observation required further investigation (13). Also, an association was observed between long-term use of oral NAC and higher risk of knee osteoarthritis, although the authors reported that a dose-response relationship could not be determined (7). Thus, the purpose of this scoping review was to present a summary of studies investigating NAC's effects in LBP management and the

potential benefits it may provide for LBP treatment. The potential molecular mechanisms by which NAC may relieve LBP and the adverse effects of NAC are also discussed.

Material and Methods

We followed the Preferred Reporting Items for Systematic Review and Meta-Analyses extension for Scoping Reviews (PRISMA-ScR) guidelines in the study process (14). The scoping review protocol was registered in the Open Science Framework (doi: 10.17605/OSF.IO/HVD4C) and was conducted using the following five steps: construction of the research question, identification of relevant studies, selection of studies, extraction of data, and synthesis of results.

Elaboration of the research question

According to the PICO (Patient, Intervention, Comparison, and Outcome) strategy, the question in this review was: what are the effects of NAC treatment in LBP management?

Identification of relevant studies

Descriptors used in the review process were selected by consulting the Health Sciences Descriptors (HSDs) and MeSH terms, generating a specific search strategy for each selected database. The selected databases were PubMed/MEDLINE, Embase, Scopus, Science Direct, Web of Science, Cinahl, and Lilacs.

Eligibility criteria

Articles were included if they i) reported the effect of NAC in LBP management, regardless of whether the study involved patients, experimental animal models, or cell culture, ii) reported findings of an empirical study, without any sample size requirement, iii) involved a randomized trial, and iv) were published in a peer-reviewed journal.

Articles were excluded if they i) lacked full text and ii) were specific types of manuscripts (meeting summaries, editorials, and letters to the editor).

No limit was set on study language and publication date. The final searches were completed on March 20, 2024.

Selection of studies

All records were imported into a review management system. Subsequently, the studies selected were fed into "Covidence", a web-based systematic review software package (Veritas Health Innovation), to detect duplicate records, which were excluded. Two researchers (G.S. and L.M.F.) independently carried out the article selection process to create a bibliographic database, which included the following details for each publication: authors, title, journal, year, DOI (Digital Object Identifier), and

abstract. From this initial selection, a set of studies was obtained and the title and abstract of each study was analyzed in order to assess whether or not the study met the objective of the review. The researchers then verified agreements and disagreements regarding the selected studies, and the selected studies were read in full to avoid excluding important studies for the review. If the evaluators disagreed on whether or not to include a particular study, a third evaluator was consulted (W.A.P or M.L.G).

Extraction of data

The extracted items in all studies were: authors, year, pain condition, gender, age, treatment with NAC (dose, administration route, dosing interval), and results. The Kappa coefficient was utilized to assess the interrater reliability.

Results

The database search identified a total of 2357 articles, 300 of which were duplicated. The title and abstract of the remaining articles (2057) were independently evaluated by two researchers (G.S. and L.M.F.). A total of 2032 articles were excluded and 25 articles required a full text screening. After screening, 9 articles were excluded and 16 articles that met the inclusion criteria were included in this scoping review (15–30). The process for selecting and including publications is shown in Figure 1. The Kappa coefficient was (0.96, 95%CI: 0.91; 1.0, $P < 0.001$) for title and abstract screening and (0.78, 95%CI: 0.54; 0.98, $P < 0.001$) for the extraction process. No further disagreements were found in the following stages.

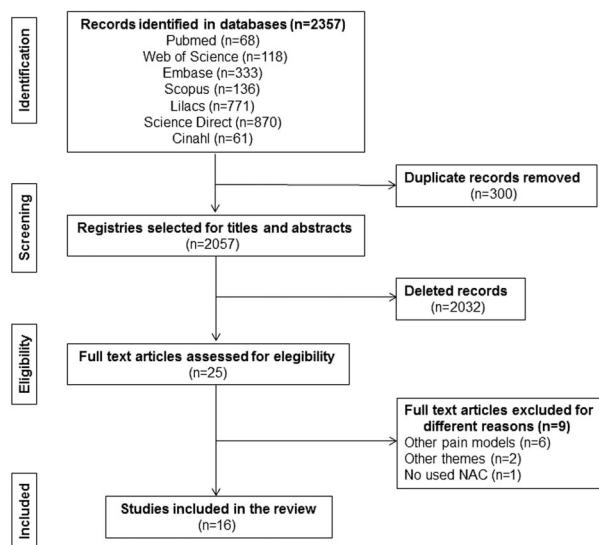


Figure 1. PRISMA flow chart of study selection.

All the selected studies were published between 2002 and 2023 (Figure 2A). The largest number of publications was from 2019 ($n=3$) and 2023 ($n=2$). The studies included in this scoping review were published in 12 different journals (Figure 2B). The highest number of publications was found for Osteoarthritis Cartilage ($n=3$), Spine ($n=2$), and Oxid Med Cell Longev ($n=2$). In the selected studies, the first authors were from Iran ($n=1$), Japan ($n=3$), USA ($n=3$), China ($n=3$), Germany ($n=2$), Turkey ($n=1$), Spain ($n=1$), United Kingdom ($n=1$), and France ($n=1$). Osteoarthritis was the most common condition.

Only three studies administered NAC to patients (Table 1). The patients were of both sexes and aged from 40 to 67 years. NAC dose and treatment schedule differed across studies. Oral NAC was administered at a dose of 600 mg twice a day for 8 weeks (15). Intraoperative NAC was used at a dose of 150 mg/kg (16). Intra-articular injection of NAC 3 mL (300 mg/3 mL Asist ampoule) was administered as a single shot (23). Pain was effectively reduced in all these studies. However, this effect was assessed at different times. Oral NAC reduced VAS (Visual Analogue Scale) scores and Oswestry Disability Index scores at weeks 2 and 4 of the treatment, but a significant reduction was found at week 4 to week 8 (15). Intraoperative NAC decreased opioid consumption and pain scores measured using VAS and numeric rating scale (16). Opioid consumption was reduced by 19.3% at 12 h, by 20% at 18 and 36 h, and by 22–24% after adjusting intraoperative opioid consumption at all times.

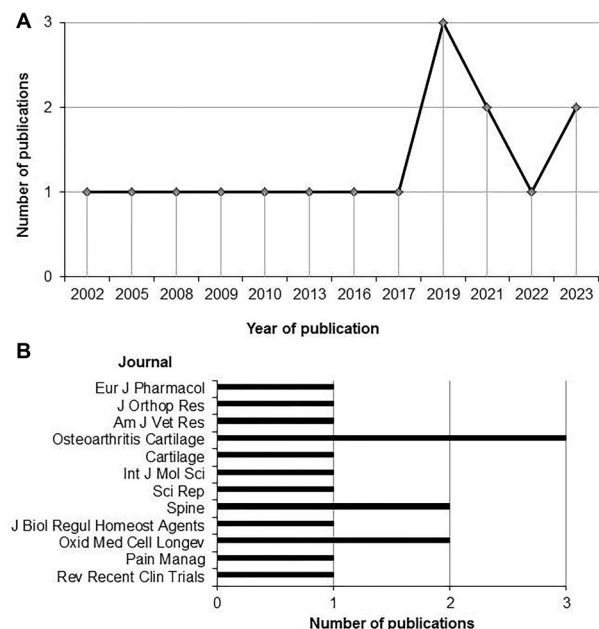


Figure 2. Number of publications per year (A) and journal (B) of the 16 studies included in the review.

Table 1. Summary of findings from the clinical trials included in the review.

Authors (year) (country)	Pain condition	Number of patients	NAC regimen and dose	Main effects of NAC
Heidari et al. (2023) (Iran)	Acute lumbar radiculopathy associated with disc herniation, with low back pain only and low back pain + leg pain	62	NAC 600 mg twice a day simultaneously with Naproxen 500 twice a day for eight weeks.	NAC decreased VAS scores (means; 95%CI) Baseline (6.34; 6.02,6.67) Week 2 (5.82; 5.58,6.07) Week 4 (5.28; 5.06,5.50) Week 8 (3.96; 3.58,4.35) NAC decreased Oswestry Disability Index scores (means; 95%CI) Baseline (61.56; 59.21,63.92) Week 2 (53.75; 50.68,56.82) Week 4 (53.90; 51.62,56.18) Week 8 (38.75; 36.15,42.34)
Wilson et al. (2023) (USA)	Elective posterior spine surgery	30	NAC (150 mg/kg) or placebo infusion initiated after general anesthetic induction and infused over 60 min. Postoperative opioid consumption in intravenous morphine milligram equivalents was estimated from a linear mixed model of opioid consumption over time including main effects for treatment group, postoperative time, the interaction between group and post-operative time, and a random subject effect.	NAC decreased opioid consumption by around 20% in 6-h postoperative intervals up to 48 h. Time (min) for first opioid (median, 95% CI) was 25.5 (6.0,84.0) and 16.0 (5.0, 33.0) for NAC and placebo groups, respectively. Similar PACU VAS scores, PACU length of stay, and hospital length of stay were found in NAC and placebo groups. Patient reported postoperative pain (using numeric rating scale) on average 1.1 units lower in the NAC treated group compared with placebo (mean difference in pain scores for placebo vs NAC (95%CI): -1.07 (-2.2,0.07)).
Ozcamdalli et al. (2017) (Turkey)	Knee osteoarthritis	10	Intra-articular injections of 3 mL of NAC (300 mg/3 mL) as a single shot.	NAC decreased VAS and improved WOMAC scores (mean $\pm$ SD). VAS before (7.4 ± 0.6) vs after NAC (5.2 ± 0.6) injection. Total WOMAC scores before (44.3 ± 5.1) vs after NAC (28.1 ± 3.7) injection. NAC decreased C-reactive protein and MMP-3 levels. NAC reduced total oxidant status without change in total antioxidant concentration. NAC reduced chondroitin-6-sulfate and cross-linked C-terminal telopeptide of type 2 collagen levels.

NAC: N-acetylcysteine; VAS: Visual Analogue Scale; WOMAC: Western Ontario and McMaster Universities Arthritis Index; MMP-3: matrix metalloproteinase-3; PACU: Postoperative Anesthesia Care Unit; CI: confidence interval.

Intra-articular NAC injection decreased VAS scores and improved total Western Ontario and McMaster Universities Arthritis Index (WOMAC) score and WOMAC domains of stiffness and physical function, which were measured 6 weeks after the injection (23). This study also demonstrated that NAC reduced C-reactive protein level and

oxidant status and improved functional status of patients with osteoarthritis without significant changes in total antioxidant concentration.

Most of the studies included in this scoping review were research articles (n=13) using experimental animal models (n=7) (Table 2) and cell culture (n=6) (Table 3).

Table 2. Summary of findings from included studies using animal experimental models.

Authors (year) (country)	Low back pain model	Gender	NAC regimen and dose	Main effects of NAC
Che et al. (2022) (China)	IL-1 β -induced nucleus pulposus cell degeneration	Old mice	Dissolved in drinking water for six months (150 mg/kg per day)	Upregulated polyamine oxidase in human degenerated disc samples. Stabilized polyamine metabolism and suppressed oxidative stress in mice.
Zheng et al. (2021) (China)	Puncture of intervertebral disc	Male rats	Injection into intervertebral disc (1 mM)	Decreased LBP (assessed by von Frey test and acetone test) in rats. Reduced ROS level parallel to decrease in pain of rats.
Kinoshita et al. (2019) (Japan)	Sarcopenia induced by axotomy of the sciatic nerve	Male rats	Dissolved in drinking water (1 g/L) for 4 weeks	Prevented MAPKs and apoptosis in C2C12 cells. Prevented amyotrophy and fatty degeneration after axotomy in rats.
Kaneko et al. (2019) (Japan)	Osteoarthritis induced by mechanical stress	Male rats	Dissolved in distilled water (5 g/L) for 5 or 8 weeks	Blocked ROS accumulation and inhibited TNF α and MMP13 expressions in chondrocyte cultures. Blocked ROS accumulation and inhibited MMP13 expression in rats. Prevented the down-regulation of type II collagen and apoptosis in rats.
Riegger et al. (2019) (Germany)	Cartilage trauma	Female rabbits	Intra-articular injection (5 mg)	Reduced trauma-induced hypocellularity and MMP-13 expression. Reduced cluster formation. Increased type II collagen carboxy propeptide and proteoglycan staining intensity.
Nakagawa et al. (2010) (Japan)	Osteoarthritis induced by transection of anterior cruciate ligament	Male rats	Injection into articular cavity (5 mg) every 3 to 4 days for 8 weeks	Prevented nitric oxide-induced apoptosis and ROS overproduction in chondrocytes. Prevented p53 up-regulation and caspase-3 activation in chondrocytes. Prevented cartilage destruction and chondrocyte apoptosis in rats.
Baltzer et al. (2008) (USA)	Animals with intervertebral disc disease	Male and female dogs	Intravenous injection (50 mg/kg)	No effect on neurologic outcome and urinary 15F _{2t} isoprostane excretion.

NAC: N-acetylcysteine; LBP: low back pain; IVD: intervertebral disc; ROS: reactive oxygen species; MAPKs: mitogen-activated protein kinases; TNF α : tumor necrosis factor alpha; MMP-13: matrix metalloprotease-13.

The experimental animal models used old mice (17), young, mature, and old male rats (18,20,21,26), female rabbits (22), and male and female dogs (28). NAC dose and time of treatment differed across the studies. NAC was administered in drinking water at the doses of 150 mg/kg per day for six months (17), 1 g/L for 4 weeks (20), and 5 g/L for 5 or 8 weeks (21). The administrations were intervertebral disc injection (1 mM) (18), intra-articular injection (5 mg) (22), injections into articular cavity every 3 to 4 days for 8 weeks (26), and intravenous injection before hemilaminectomy (28). In an experimental animal LBP model, NAC relieved pain (18). NAC also prevented osteoarthritis development and progression (21). However, NAC had no effect on deep pain sensation in dogs with intervertebral disc disease (28). Pain behavior was assessed using von Frey and acetone tests (18) and neurologic outcome including deep pain sensation (28). In all studies, NAC reduced reactive oxygen species (ROS)

levels (17,18,21,26), inhibited pro-inflammatory cytokines (21), prevented mitogen-activated protein kinases (MAPKs) changes and apoptosis (20,26), and rescued cell viability and extracellular matrix components (21,22,26). However, NAC had no effect on urinary 15F_{2t} isoprostane excretion (28), a biomarker of ROS activity.

In the cell culture studies, most studies (n=4) used human chondrocytes (19,24,27,30). One study also used synoviocytes from patients (27). Two studies used cells from experimental animal models (25,29). In these studies, cells were cartilage specimens obtained from the stifle joints of healthy adult mixed breed dogs (25) and chondrocytes from the articular cartilage of the femoral head of 3-, 6-, and 15–18-month-old rats (30). Cell culture studies demonstrated that NAC decreased ROS accumulation and increased antioxidant activity (19,25,29), diminished pro-inflammatory factors (27), and rescued cell viability and extracellular matrix components (19,24,27).

Table 3. Summary of findings from included studies using cell culture.

Authors (year) (country)	Cell types	Main effects of NAC
Li et al. (2021) (China)	Human chondrocytes submitted to cyclic tensile stress	Rescued cell viability and decreased ROS accumulation. Partly rescued main extracellular matrix components and F-actin polymerization.
Riegger et al. (2016) (Germany)	Human full-thickness cartilage explants subjected to a definite impact trauma	Increased cell viability and reduced apoptosis. Suppressed trauma-induced gene expression of extracellular matrix-destructive enzymes.
Dycus et al. (2013) (USA)	Cartilage specimens obtained from the stifle joints of healthy adult mixed-breed dogs	Increased superoxide dismutase activity and reduced glutathione concentration.
Roman-Blas et al. (2009) (Spain)	Synoviocytes and chondrocytes collected from knee of patients	Diminished prostaglandin E2 release and the expression of both cyclooxygenase-2 and MMP-13 protein only in synoviocytes but failed to modify their production in chondrocytes. Inhibited NF- κ B activation in synoviocytes. Did not inhibit nitric oxide synthesis in chondrocytes. No change in MMP-1 protein expression in synoviocytes or chondrocytes.
Jallali et al. (2005) United Kingdom	Chondrocytes obtained from the articular cartilage of the femoral head of 3 month (young), 6 month (mature) and 15-18 month (old) male rats	Reduced ROS levels only in old chondrocytes.
Ayache et al. (2002) (France)	Human chondrocytes derived from the knee of osteoarthritic patients	Stimulated nitric oxide production. Elevated inducible nitric oxide synthase. Counteracted TGF- β 2 expression. Increased TGF- β RI expression but reduced TGF- β RII.

NAC: N-acetylcysteine; ROS: reactive oxygen species; MMP-13: matrix metalloproteinase-13; MMP-1: matrix metalloprotein-1; NF- κ B: factor nuclear kappa B; TGF- β 2: transforming growth factor- β 2; TGF- β RI and TGF- β RII: receptors I and II of the TGF- β .

No serious adverse events were reported in NAC-treated patients (16). In addition, oral NAC administration did not induce significant changes in body weight or water and food intake in osteoarthritis rats (21).

Discussion

This scoping review aimed to present a synthesis of the literature on the effect of NAC in LBP management. LBP is the leading cause of disability worldwide (1–7) and NAC has been investigated in the treatment of chronic pain (15). A scoping review was chosen because it is considered an ideal tool to give a clear indication of the volume of literature and studies available on a given topic (31). A meta-analysis was not performed because of the high heterogeneity in experimental designs and outcomes of the studies.

We found 16 articles, indicating that the number of studies concerning NAC effects in LBP is still relatively low. This apparent paucity is also evident from the country of origin of first authors - only nine countries. The small number of studies did not allow definitive conclusions to be drawn.

Clinical and preclinical studies showed promising results. Despite the fact that the NAC dose and/or route of administration differed across clinical trials and experimental animal LBP model studies, NAC consistently

relieved LBP. Therefore, these findings highlight NAC's potential in LBP treatment. In this context, NAC has been considered an oral medication to prevent osteoarthritis development (21) and a cheaper alternative for LBP treatment, since it appears effective in slowing progressive cartilage destruction and improving clinical and functional status (23). Since the studies included in our review did not report adverse effects or side effects of NAC treatment in humans and experimental animal models, NAC appears to be safe for treating LBP, as reported in recent reviews (8,16,32). However, a previous study found an association between long-term use of oral NAC and a higher risk of knee osteoarthritis, although a dose-response relationship could not be determined (7).

The effects of NAC administration appear to depend on treatment concentration and duration. It was suggested that a high NAC dose is necessary to produce anti-inflammatory effects, while a low NAC dose might aggravate the lesion, especially in infection-associated disease (8,9). However, a previous study using A549 human epithelial cells reported that chronic administration of a low NAC dose had sustained antioxidant and anti-inflammatory effects, and acute treatment with a high NAC dose exerted a strong antioxidant and anti-inflammatory response (33). These results suggest that chronic administration of low NAC doses might be a good strategy for managing LBP. Better NAC effects occurred at a higher

dose, but more adverse events were found at a higher dose (34). In this context, low-dose NAC has been shown to be effective as adjunctive therapy in improving neuropathic pain associated with diabetic neuropathy (35).

Nevertheless, NAC had no effect on deep pain sensation in dogs with intervertebral disc disease (28). According to these authors, the limitation of their study was the sample size, as only 54 of the 70 dogs completed the study. In healthy cats, the pharmacokinetics of oral and intravenous administration of NAC paralleled that reported in humans, but there was a shorter elimination half-life due to the faster clearance in cats (36). A similar condition may occur in dogs.

There was a large time gap between NAC administration and the effect in some of the included studies. Oral NAC relieved the pain in patients with acute lumbar radiculopathy associated with disc herniation, but a significant effect was found from week 4 to week 8 (15). NAC also had an analgesic effect on postoperative opioid consumption, and potentially improved pain scores later in the postoperative period (16). An analgesic effect of NAC also occurred with intra-articular injections, and the effect was measured 6 weeks after injection (23). In an experimental animal model, NAC also relieved the pain (18) and prevented osteoarthritis development and progression after oral administration for weeks (21). These results suggest that NAC's analgesic effect depends on a long-term course of treatment, whatever the administration route. NAC's low bioavailability may explain the need for longer treatment. In humans, the absolute bioavailability of oral NAC was low and ranged from 6–10%, which was suggested to be related to its rapid metabolism in the gut (37). However, the route of administration and doses used in the studies included in our review varied greatly, which limit a direct comparison of the results (32).

Among the experimental animal model studies included in this review, the majority used male rats. No study using female rats was found. A previous study showed that male and female rats have distinct relationships between intervertebral disc degeneration and pain, and the authors highlighted the need to treat females and males as different cohorts (38). Sex differences in behavioral responses to painful and non-painful stimuli were also observed when male and female rats were submitted to an LBP-inducing protocol (39). Thus, future preclinical research on the effects of NAC on LBP using female rats is needed. Although studies using female mice, rabbits, and dogs were found, a previous study highlighted the need to use more heterogeneous animal models in studies of pain to assess the generalization of pain in that experimental animal model (40). Since only one study used dogs as the experimental animal model, studies using large animal models are needed. A recent scoping review recommended preclinical studies using large animal models that assess sex, skeletal age, and

gonadal hormones as biological variables as fundamental to translational osteoarthritis research (41).

Many methods are used to quantify pain-like behaviors or nociception in animals. These methodologies can be categorized into spinal reflexive and non-reflexive pain tests, depending on whether or not an external stimulus is applied to elicit a withdrawal response, respectively (40,42). Studies included in our review assessed pain behavior using von Frey and acetone tests (18) and neurologic outcome including deep pain sensation (28). Von Frey and acetone tests are spinal reflexive tests (40). Spinal non-reflexive pain tests include spontaneous-related behaviors and movement-evoked hypersensitivity (42). None of the included studies assessed pain behaviors using spinal non-reflexive pain tests. However, the use of tests such as grimace scales, burrowing, weight bearing, and gait analysis have gained prominence in studies of pain using experimental animal models (40). A previous study suggested that non-reflexive pain tests should be tried in future studies on LBP (42). In experimental animal neuropathic pain models, it was suggested that measuring non-evoked pain could improve analgesic drug development and clinical translation (43). Thus, stimulus-evoked or non-stimulus-evoked nociception needs to be included in future studies using experimental animal models. We believe that the increase in the diversity of pain behavior measurements could increase clinical translation of studies on NAC's effects in LBP.

The evidence presented in this review regarding NAC potential mechanisms of action in LBP included its effects on ROS levels, inflammatory cytokines, and MAPK signaling pathways. All these mechanisms are known to participate and be altered in LBP (44–48). Oxidative stress parameters were the most evaluated. Our review found that 50% of the included studies used NAC as an antioxidant and a scavenger of ROS. NAC is a precursor to the antioxidant glutathione, the most abundant intracellular free thiol whose depletion triggers cell death pathways (8,10). A previous review highlighted that glutathione and its precursor NAC appear to have a protective effect in chronic inflammatory musculoskeletal disorders such as osteoarthritis (12). However, the sulfhydryl group in the NAC molecule directly scavenges ROS (10,32). NAC is also known to be converted into hydrogen sulfide and sulfane species inside cells (49). Studies evaluating these other NAC mechanisms of action in LBP are lacking. Nevertheless, NAC had no effect on urinary $15F_{2t}$ isoprostane excretion in dogs with intervertebral disease (28). A possible explanation to this result is the number of animals used, as previously discussed. Oxidative stress is considered the main pathogenic factor in intervertebral disc degeneration (50) and NAC has demonstrated a therapeutic role in disorders characterized by increased oxidative stress (8).

The pathophysiological mechanism of how ROS regulate LBP is still unclear. In rats with LBP, it was

demonstrated that ROS stimulate nucleus pulposus cells to secrete substance P, a peptide thought to be involved in synaptic transmission of pain. ROS-mediated stimulation of nucleus pulposus cells to secrete substance P appears to promote LBP (18). According to these authors, LBP was relieved after administration of an antagonist of substance P receptor. Unfortunately, the study did not assess the effect of NAC on substance P or its receptors. In cardiac mast cells, NAC had no effect on substance P-induced histamine release, but it attenuated histamine release in response to hemokinin-1, another tachykinin (51). Preclinical findings can serve as the foundation upon which scientists can construct potential breakthroughs that directly impact human health (52).

NAC also reduced prostaglandin E2 release, the expression of both cyclooxygenase-2 and metalloproteinase-13 (MMP-13) (27), and inhibited tumor necrosis factor alpha (TNF α) and MMP-13 expressions (21). These mechanisms are indicators of inflammation, which are increased in LBP (21,53,54). Thus, anti-inflammatory properties are also involved in NAC's effect in LBP. Anti-inflammatory properties can be a direct action of NAC or attributed to its antioxidant activity (32).

NAC rescued cell viability and prevented changes in MAPKs and apoptosis (19–22,26). MAPKs are a family of protein kinases that have been implicated in the regulation of virtually all cellular processes (55). In intervertebral disc degeneration, the MAPK/ERK signaling pathway appears to be closely related to progressive destruction of extracellular matrix, cell aging, apoptosis, inflammation, and impairment of tissue biomechanical function (46). A previous study suggested that the p38 MAPK signaling pathway is a curative target for intervertebral disc degeneration (44).

NO (nitric oxide) is an important signaling molecule that participates in various physiological functions. It has been demonstrated that NO increased in the periradicular region in patients with LBP (56). An association between variants in genes encoding enzymes involved in NO synthesis and LBP was observed (57). Finally, NAC has other mechanisms of action that should be explored in LBP, such as its ability to assist the activity of deficient or otherwise misfolded proteins and its role in the regulation of

glutamate homeostasis by cysteine-glutamate exchangers (32).

This scoping review had several strengths. A robust search strategy was carried out across databases to retrieve published literature, without limitations of language or year of publication. The PRISMA-ScR checklist was used to ensure the integrity of the review. However, our review had limitations. The included studies had a small sample size, which limited the conclusions. There was a large heterogeneity of study designs, which made it difficult to draw definitive conclusions on NAC's effects in LBP and was a limitation in conducting a meta-analysis. Further clinical studies are necessary to adequately validate preclinical findings included in the present review. Preclinical studies play a vital role in understanding mechanisms behind chronic pain and in developing new pain-relief drugs (52). NAC modulates N-methyl-D-aspartate receptor (NMDAR), a key regulator of pain processing (8–10), and NMDAR showed different functional properties in rats and humans (58), which is a species difference that should be considered in future studies searching new therapeutic candidates.

Conclusion

The objective of this scoping review was to synthesize the existing literature and provide a robust foundation for clinicians and researchers on the state of the art regarding NAC for LBP treatment. Although NAC has proven to be beneficial for the treatment of LBP, data are derived from few clinical trials and preclinical studies. The results appear promising, but the dosing regimens and route of administration of NAC need to be further investigated. Thus, further clinical studies need to be performed before NAC can be clinically recommended for the treatment of LBP.

Funding

This study was supported by grants from the Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (FAPERGS) and the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq).

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