



Aalborg Universitet

AALBORG UNIVERSITY  
DENMARK

## Bridging the translational gap

*adenosine as a modulator of neuropathic pain in preclinical models and humans*

Arendt-Nielsen, Lars; Klitgaard, Henrik; Hansen, Stine N.

*Published in:*  
Scandinavian Journal of Pain

*DOI (link to publication from Publisher):*  
[10.1515/sjpain-2023-0048](https://doi.org/10.1515/sjpain-2023-0048)

*Creative Commons License*  
CC BY 4.0

*Publication date:*  
2024

*Document Version*  
Publisher's PDF, also known as Version of record

[Link to publication from Aalborg University](#)

*Citation for published version (APA):*  
Arendt-Nielsen, L., Klitgaard, H., & Hansen, S. N. (2024). Bridging the translational gap: adenosine as a modulator of neuropathic pain in preclinical models and humans. *Scandinavian Journal of Pain*, 24(1), Article 20230048. Advance online publication. <https://doi.org/10.1515/sjpain-2023-0048>

### General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- You may not further distribute the material or use it for any profit-making activity or commercial gain
- You may freely distribute the URL identifying the publication in the public portal -

### Take down policy

If you believe that this document breaches copyright please contact us at [vbn@aub.aau.dk](mailto:vbn@aub.aau.dk) providing details, and we will remove access to the work immediately and investigate your claim.

## Topical Review

Lars Arendt-Nielsen\*, Henrik Klitgaard and Stine N. Hansen

# Bridging the translational gap: adenosine as a modulator of neuropathic pain in preclinical models and humans

<https://doi.org/10.1515/sjpain-2023-0048>

Received April 14, 2023; accepted November 22, 2023;

published online December 11, 2023

### Abstract

**Objectives:** This review aims to analyse the published data on preclinical and human experimental and clinical adenosine modulation for pain management. We summarise the translatability of the adenosine pathway for further drug development and aim to reveal subgroups of pain patients that could benefit from targeting the pathway.

**Content:** Chronic pain patients suffer from inadequate treatment options and drug development is generally impaired by the low translatability of preclinical pain models. Therefore, validating the predictability of drug targets is of high importance. Modulation of the endogenous neurotransmitter adenosine gained significant traction in the early 2000s but the drug development efforts were later abandoned. With the emergence of new drug modalities, there is a renewed interest in adenosine modulation in pain management. In both preclinical, human experimental and clinical research, enhancing adenosine signalling through the adenosine receptors, has shown therapeutic promise. A special focus has been on the A<sub>1</sub> and A<sub>3</sub> receptors both of which have shown great promise and predictive validity in neuropathic pain conditions.

**Summary:** Adenosine modulation shows predictive validity across preclinical, human experimental and clinical investigations. The most compelling evidence is in the field

of neuropathic pain, where adenosine has been found to alleviate hyperexcitability and has the potential to be disease-modifying.

**Outlook:** Adenosine modulation show therapeutic potential in neuropathic pain if selective and safe drugs can be developed. New drug modalities such as RNA therapeutics and cell therapies may provide new options.

**Keywords:** adenosine; neuropathic pain; translation; animal models; human studies

## Introduction

Pain conditions are complex, multifactorial disorders associated with several severe comorbidities such as depression, anxiety, sensitisation, and sleep disorders significantly impacting patient life-quality and increasing healthcare costs for societies [1]. Development of new drugs for pain has been impaired by the low translatability of animal pain models to humans and the patients are often left with insufficient or addictive treatment options. Therefore, there is a large unmet need for new drug candidates with minimal adverse effects for treatment of specific sub-categories of pain preferable with a personalised medicine approach considering the individual patient needs. This is evident after the opioid crisis and the concerns with alternative approaches such as the long-term benefits of cannabis and cannabidiol [2]. Translation from bench to bedside is associated with a high attrition rate and only one in 10 pain drug candidates entering phase I clinical studies ends up becoming an approved drug [3].

Adenosine is a ubiquitous, endogenous neurotransmitter that exerts its function through four distinct G-protein coupled receptors (AR), A<sub>1</sub>R, A<sub>2A</sub>R, A<sub>2B</sub>R and A<sub>3</sub>R. The receptors are widely distributed through the body including in central nervous system (CNS) areas known to be involved in spinal and supraspinal pain modulation [4]. This distribution equates adenosine modulation with the antinociceptive properties but also several other pharmacological effects through presence of AR in e.g. the cardiovascular system.

\*Corresponding author: Prof., Dr. Med. Lars Arendt-Nielsen, PhD, Department of Health Science and Technology, Center for Neuroplasticity and Pain, CNAP, School of Medicine, Aalborg University, Selma Lagerlöfs Vej 249, 12.02.024, 9260 Gistrup, Denmark; Department of Gastroenterology & Hepatology, Mech-Sense, Clinical Institute, Aalborg University Hospital, DK-9000 Aalborg, Denmark; and Steno Diabetes Center North Denmark, Clinical Institute, Aalborg University Hospital, DK-9000 Aalborg, Denmark, Phone: +4520940764, E-mail: LAN@hst.aau.dk

Henrik Klitgaard and Stine N. Hansen, NEUmiRNA Therapeutics, Copenhagen, Denmark, E-mail: hk@neumirna.com (H. Klitgaard), snh@neumirna.com (S.N. Hansen)

The early findings of antinociceptive effects of adenosine modulation triggered the design of adenosine analogues, positive allosteric modulators, and agonists – particularly for the inhibitory A<sub>1</sub>R and A<sub>3</sub>R receptors. However, no adenosine drugs for pain indications have reached the market. Increased levels of adenosine shows rapid and potent effects on the cardiovascular system where A<sub>1</sub>R activation of the heart leads to bradycardia and suppression of the sinus node, atrioventricular node and His bundle potentially leading to complete atrioventricular block [5]. Concurrently, the effects on A<sub>2</sub>R in the blood vessels are responsible for vasodilation leading to hypotension [5]. Due to these cardiovascular effects, adenosine is currently only approved for myocardial perfusion scintigraphy and treating supraventricular tachycardia [6, 7].

Another promising therapeutic approach has been to activate adenosine receptors through design of specific inhibitors for the enzyme responsible for adenosine breakdown; adenosine kinase (ADK) [8]. The two isoforms of this enzyme serve distinct physiological functions with the short isoform ADK-S being responsible for regulation of extracellular adenosine levels and the long isoform ADK-L being a nuclear epigenetic regulator [8]. Modulation of the adenosine system has seen a revival of interest as a novel pain target as new drug development options (e.g. RNA therapeutics and cell therapies) have appeared in recent years and may pave the way for interaction with the adenosine pathways in new ways [9].

This review aims to analyse published data on pre-clinical and human (both experimental and clinical) evaluations of adenosine approaches for pain management with the perspective to investigate translatability of adenosine regulation and to reveal subgroups of patients that could benefit the most from treatment. A brief bibliometric analysis on papers focusing on adenosine in selected pain relevant journals (Pain, European Journal of Pain, Clinical Journal of Pain, Anesthesia and Analgesia, Scandinavian Journal of Pain, Acta Anaesthesiologica Scandinavica) show a peak interest in the beginning of this millennium (Figure 1) and a decline thereafter.

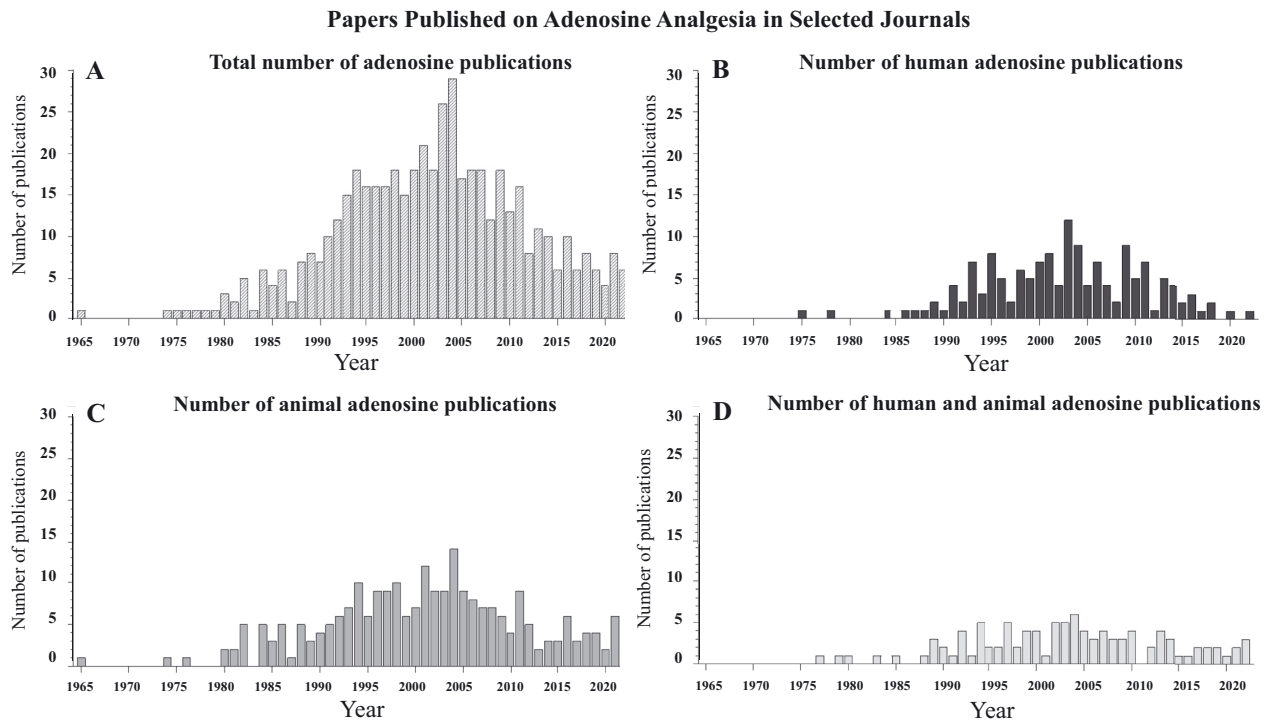
## The mechanism-based analgesic effect of adenosine in preclinical pain studies

Drug discovery for pain indications has generally been marred by the low translatability of preclinical pain models into the clinic and novel approaches emphasizing the need for novel approaches [10, 11]. In the field of adenosine

modulation in pain, preclinical research has elucidated the contributions of agonism and antagonism of the four ARs in animal models of nociceptive and neuropathic pain [4, 12]. Activation of the adenosine pathway has been found in few studies to alleviate inflammatory and postoperative pain primarily through the A<sub>1</sub>R in animal models [13–15] but the most compelling evidence for adenosine's anti-hyperexcitability properties is in neuropathic pain [16–29]. Experimental research has primarily focused on activation of A<sub>1</sub>R and A<sub>3</sub>R as underlying the analgesic properties of adenosine in preclinical neuropathic pain but the A<sub>2A</sub>R has also been implicated [25, 26]. The A<sub>1</sub>R is expressed pre- and post-synaptically and is found in brain areas associated with supraspinal pain perception, on neuronal cell bodies in the dorsal spinal horn, and on the endings of peripheral sensory nerves [4]. A<sub>1</sub>R knockout mice exhibit increased neuropathic pain-like behaviours after partial nerve injury [30] and A<sub>1</sub>R modulation has been found to decrease allodynia and hyperalgesia in models of nerve injury [18, 24, 30, 31] indicating the potential role of adenosine in neuropathic pain conditions.

In relation to certain chemotherapies, neuropathic-like conditions associated with hyperalgesia and allodynia is often a side effect as the medications have become increasingly neurotoxic. In a rat model of chemotherapy-induced peripheral neuropathy, intrathecal administration of an A<sub>1</sub>R agonist alleviated the neuropathic symptoms [22], a finding which has been reproduced by others [21]. Another study showed a chemotherapy-induced increase in spinal ADK implying that imbalances in the adenosine pathway in itself can be an underlying cause of neuropathic pain conditions in the animal models [29]. Additionally, A<sub>1</sub>R activation has been found to alleviate mechanical allodynia in models of diabetic neuropathy [16, 23]. Thus, increased A<sub>1</sub>R activation has been found to counteract hyperalgesia and allodynia in preclinical neuropathic pain models of different aetiologies. Other studies have shown that the A<sub>3</sub>R may also be involved in neuropathic pain as activation of this receptor can reduce the allodynic and hyperalgesic reactions [4]. The A<sub>3</sub>R is expressed on immune and glial cells and activation has been found to counteract the pro-inflammatory and –algia factors released by these cells in pathological conditions [32]. A<sub>3</sub>R activation has likewise been found to alleviate chemotherapy-induced peripheral neuropathy by decreasing astrocyte activation and cytokine release [19, 20] and to decrease neuronal hyperexcitability [33, 34].

In addition to the evidence of adenosine modulation of hyperexcitability in neuropathic pain conditions. In a model of diabetic neuropathy adenosine administration reversed diabetic hyperalgesia in thermal and chemical stimuli tests



**Figure 1:** The total number of papers published in selected pain/analgesia journals (Pain, European Journal of Pain, Clinical Journal of Pain, Anesthesia and Analgesia, Scandinavian Journal of Pain, Acta Anaesthesiologica Scandinavica) (A). The separation of these publications into human studies; both experimental and clinical (B), preclinical (C) and combined preclinical and human (experimental and clinical studies) and the combined preclinical and human studies (D).

[16]. Interestingly, increased levels of adenosine have been found to be disease-modifying in epilepsy where mice with forebrain specific ADK reduction show reduced epileptogenesis and ADK-deficient ES cell – derived brain implants suppressed spontaneous seizures in mice even when implanted after the induction of epileptogenesis [35]. The nuclear long isoform of ADK responsible for modulating intranuclear adenosine levels has also been found to have epigenetics modulatory effects implying that the disease-modifying effects of the adenosine pathway may be AR-independent [8].

In summary, adenosine modulation shows therapeutic properties in neuropathic animal models with manifestations of allodynia and hyperalgesia.

## Mechanism-based analgesic effect of adenosine in human experimental pain studies

Between 1990 and 2004, there was a significant interest in proof-of-concept studies evaluating adenosine in human experimental pain. Most of those studies used quantitative,

mechanistic pain biomarkers in healthy volunteers to study the analgesic action of adenosine administered via various routes. In particular three groups have been involved in this work: the Karolinska Hospital, Stockholm, Sweden Group (Sollevi and Segerdahl), the Wake Forest School of Medicine, Winston-Salem, NC, USA Group, US (Eisenach) and the Aalborg University Group, Aalborg, Denmark (Arendt-Nielsen).

Initial research focused on the hypothesis that adenosine functioned as a mediator of pain sensation. In 1989, Jonzon et al. studied adenosine in ischaemic pain sensation by using the  $A_2R$  and  $A_2R$  blocker theophylline [36]. In healthy volunteers, muscle ischaemia was induced, and the pain sensation was continuously reported. A small, significant inhibitory effect on the ischaemic pain sensation was detected and found to be compatible with a hyperalgesic effect of endogenous adenosine. The background for this study was the finding that adenosine caused ischemic angina-like pain due to the AR in the heart muscles [37] and the role of adenosine as a stimulator of angina-like pain.

Fifteen years later, the theophylline study was followed up by the same group later using a sub-maximum effort forearm tourniquet ischemic test [38]. It was shown that the peak Visual Analog Scale (VAS) pain score was lowered by the theophylline treatment. Theophylline therefor seemed to

both have the potential to both inhibit and facilitate pain caused by ischaemia.

The view on adenosine as a possible algescic substance was later replaced by the view that adenosine has analgesic properties. The analgesic effects were supported by Rae et al. who concluded that continuous intravenous (IV) adenosine infusion inhibited ischaemic pain, though efficacy was not sustained after discontinuation of the infusion [39]. A series of human experimental pain studies expanded on the ischemic muscle pain model and found that IV adenosine alleviated experimentally-induced ischemic muscle pain in healthy volunteers [40]. The study suggested an additive effect on pain reduction when adenosine was combined with morphine or ketamine suggesting that adenosine may function as an opioid sparing drug.

The next steps involved other human experimental pain tests such as the heat pain stimulus. IV adenosine revealed a significantly increase in cutaneous heat pain threshold without effecting ketamine or morphine (as known from other studies). This highlighted the interesting aspect that adenosine in combination with morphine and ketamine may provide synergistic effects on different pain modalities [41].

In 1998, the first clinical trial of experimental pain investigating the effect of intrathecal adenosine was conducted and explored the effects on cold-pain rating of the foot (submersion in ice water for 1 min), the forearm ischemic pain rating during a 30 min tourniquet test, and the thermal and tactile pain thresholds on healthy and inflamed skin after 4 min application of mustard oil to the calf [42]. Adenosine treatment reduced the areas of secondary allodynia after mustard oil-induced skin inflammation and reduced the pain intensity scores from pain induced by ischaemia. This was the first experimental study to show the effects of adenosine on the central sensitization phenomena as a manifestation in neuropathic pain.

The findings were followed up by additional studies investigating the effect of adenosine on other central pain manifestations such as hyperexcitability (hyperalgesia and allodynia).

The first study in a series investigated the effects of IV adenosine on a superficial cutaneous burn injury by 4 min topical application of mustard oil or by heat injury (47 °C for 7 min). Adenosine significantly reduced the area of secondary hyperalgesia in both models with no other differences in sensory functions [43]. These findings underlined the selective effect on the peripheral and central sensitization phenomena supporting the previous finding by Rane et al. [42]. However, conflicting evidence exist. Dirks et al.

found no effects on acute nociceptive pain induced by heat stimulation or on secondary hyperalgesia induced by heat/capsaicin sensitization in healthy volunteers [44]. This suggests that adenosine modulation may show different efficacy based on the pain aetiology.

Central sensitization was further studied in two studies by Eisenach et al. investigating the effects of intrathecal adenosine on intradermal capsaicin injection in humans. No decreases in acute pain evoked by thermal or chemical stimulation was found but a reduction in mechanical hyperalgesia and allodynia after intradermal capsaicin injection was shown, further substantiating the possible central actions of adenosine on sensitisation [45]. The follow-up dose-response study in the same experimental models showed no difference in adenosine efficacy against experimental hypersensitivity between the largest experimental dose of intrathecal adenosine and a 75 % lower dose, but side effects are more common with the larger dose [46]. The controversies between studies can be related to different methods used, different dosing regimens or sample size differences resulting in possible false positive or false negative studies.

This role of adenosine as particularly a modulator of sensitization was further consolidated in an IV adenosine study by Chizh et al. [47] where electrically evoked hyperalgesia was tested. The area of pinprick hyperalgesia was reduced during the adenosine infusion compared with placebo but there was no significant effect on tactile allodynia or evoked pain ratings. In addition, adenosine did not have long-term analgesic effects as efficacy were only seen during the infusion. In a study by Morélot-Panzini et al. [48], healthy volunteers received an IV administration of adenosine or placebo and the nociceptive withdrawal RIII reflex was assessed. Adenosine induced dyspnea, tachycardia, and significant RIII reflex inhibition suggesting a role of adenosine on the central descending pain modulatory pathways [48]. Another study investigated trigeminal nociception using the nociceptive blink reflex as read out. The experimental A<sub>1</sub>R agonist GR79236 inhibited trigeminal nociception in humans which could hint a possible therapeutic role for A<sub>1</sub>R agonists in headache disorders [49] suggesting direct acute, anti-nociceptive effect.

In summary, different routes of exogenous adenosine are consistently showing efficacy on pain sensitisation phenomena such as experimentally induced hyperalgesia, allodynia, and descending pain modulation which can be via a modulation of the afferent drive or a direct effect on the central mechanisms. The next important question is if this translates into a beneficial effect on neuropathic pain.



## The mechanism-based analgesic effects of adenosine in neuropathic pain

The effect of exogenous adenosine on clinical pain conditions have a long track record expanding almost 40 years (Figure 1).

The Swedish group headed by Alf Sollevi pioneered the evaluation of adenosine in clinical pain conditions. In 1995, the group published a study where two patients with peripheral neuropathic pain received IV adenosine which showed that particularly hyperalgesia and allodynia was attenuated by treatment [50]. This critical study paved the way for the experimental pain studies investigation the effect of adenosine on neuropathic pain.

The study was followed by a larger IV adenosine study in neuropathic pain patients where perception thresholds for touch, touch-evoked pain, cold, warmth, painful heat, and cold were assessed. In the neuropathic area, VAS ratings to pin prick stimulation and perception threshold for touch-evoked pain using von Frey filaments were assessed [51]. The study demonstrated that adenosine infusion alleviated spontaneous neuropathic pain, tactile allodynia, and pinprick hyperalgesia in patients with peripheral neuropathic pain [47].

In a case story of a patient with severe erythromelalgia due to a  $Na_v1.7$  mutation and presenting with ongoing burning dysesthesia and pain in the legs, sustained thermal hyperalgesia and allodynia, and intolerable pressure pain on standing and walking, symptoms were modulated by both IV and intrathecal adenosine administration [52]. In complex regional pain syndrome, another neuropathic condition, intrathecal adenosine reduced areas of hyperalgesia and allodynia and also inhibited temporal summation; another important biomarker strongly facilitated in neuropathic pain [53]. However, spontaneous pain was not dramatically reduced [53]. Lynch et al. likewise showed differentiated effects on central sensitisation mechanisms in neuropathic pain patients as IV adenosine caused significant reduction in pinprick hyperalgesia, but not in allodynia and only a few patients experienced long-term pain alleviation following IV adenosine [54]. The study further supports that IV adenosine may have long-term effects on neuropathic pain and hyperalgesia suggesting a potential disease-modifying effects of adenosine modulation.

The effect of intrathecal adenosine was further substantiated by Belfrage et al. in neuropathic pain patients with tactile hyperalgesia and/or allodynia primarily of traumatic origin finding that spontaneous and evoked pain intensity decreased in most patients with an effect lasting for a median of 24 h suggesting a possible difference in duration

of action depending on IV or intrathecal administration [51]. Another neuropathic pain study by Eisenach and colleagues further investigated this and found that intrathecal adenosine reduced allodynic area and pain intensity, whereas the same dose of adenosine IV was ineffective [55] supporting that high levels of adenosine is required in the central component to inhibit the central manifestations in neuropathic pain patients. Importantly, intrathecal adenosine has in animal models been found to be safe and not elicit any histopathological or behavioural adverse effects [56, 57].

Another important question was whether adenosine could potentiate anaesthesia. It was shown that a low-dose perioperative infusion reduced the perioperative anaesthetic requirements and demand for post-operative analgesics [58] as well as the requirements for postoperative opioids [59]. However, later studies showed that intrathecal adenosine did not influence the requirement of anaesthetic drug or postoperative analgesics [60, 61]. Another study showed no effect of intrathecal adenosine on postoperative pain relief as well as no preventive effect [62]. Additional controlled studies are needed to investigate the role of adenosine in postoperative management regimes.

In summary, in neuropathic pain patients different routes of exogenous administered adenosine show analgesic effects on central pain sensitisation mechanisms such as hyperalgesia and allodynia. In contrast, the evidence the efficacy against spontaneous pain and the role of administration route is less complete, although intrathecal administration seems favourable. Adenosine has not shown consistent results in managing post-operative pain but may have a sparing effect on other analgesics/anaesthetics. The possible synergistic action with opioids needs further investigations.

## Conclusions

Adenosine effects in animal models have to a large extent replicated the anti-hyperexcitability (anti-hyperalgesia and anti-allodynia) properties in human experimental and clinical pain studies. In addition direct anti-nociceptive effects have been shown. This correlation between human and animal findings is unique and rarely seen for drugs developed in the pain space. This emphasises that drugs targeting the adenosine system screened in pre-clinical neuropathic pain models may be predictive for human neuropathic pain conditions. Early in the new millennium, the fundamental interest in adenosine and preclinical models of pain and human pain trials started to fade (Figure 1) as especially adenosine receptor activation studies in humans showed various unwanted side effects [5] and hence drug development in the field was abandoned. As the four different,

identified adenosine receptors have different physiological roles on various tissues the selectivity associated with pain processing is essential to minimise unwanted side effect and is a critical issue to pursue in the development of new adenosine-based pain therapeutics. However, the current short review indicates that adenosine may be a drug targets where the anti-hyperexcitability efficacy found in preclinical models translate into clinic and hence provide new options in management of neuropathic pain if the side effects can be minimised. New drug development options (e.g. RNA therapeutics and cell therapies) have appeared in recent years and may pave the way for interaction with selective, restricted adenosine pathways in new ways a thereby minimise unwanted side effects.

**Acknowledgments:** Center for Neuroplasticity and Pain (CNAP) is supported by the Danish National Research Foundation (DNRF121). The team at Aalborg University Library are acknowledged for their help with the bibliometric analysis (J T Pedersen, S Dreier, A L Høj, L Thomsen).

**Research ethics:** Not applicable.

**Informed consent:** Not applicable.

**Author contributions:** Lars Arendt-Nielsen drafted the human part of the review, Stine N. Hansen drafted the preclinical part of the review, Henrik Klitgaard critically commented and corrected the review. All authors reviewed and corrected the review.

**Competing interests:** Stine N. Hansen is an employee and Henrik Klitgaard is a co-founder of NEUmiRNA Therapeutics; a biotech company developing RNA drugs against neurological disorders.

**Research funding:** This work was funded by Innovation Fund Denmark (2056-00008B) and Danish National Research Foundation (DNRF121).

**Data availability:** Not applicable.

## References

- Mills SEE, Nicolson KP, Smith BH. Chronic pain: a review of its epidemiology and associated factors in population-based studies. *Br J Anaesth* 2019;123:e273–83.
- Rice ASC, Belton J, Arendt-Nielsen L. Presenting the outputs of the IASP presidential task force on cannabis and cannabinoid analgesia. *Pain* 2021;162:S3–4.
- Hay M, Thomas DW, Craighead JL, Economides C, Rosenthal J. Clinical development success rates for investigational drugs. *Nat Biotechnol* 2014;32:40–51.
- Sawynok J. Adenosine receptor targets for pain. *Neuroscience* 2016; 338:1–18.
- Guiou R, Deharo J-C, Maille B, Crotti L, Torresani E, Brignole M, et al. Adenosine and the cardiovascular system: the good and the bad. *J Clin Med* 2020;9:1366.
- Drug Bank. Adenosine. <https://go.drugbank.com/drugs/DB00640> [cited 10 Aug 2023].
- Singh S, McKintosh R. Adenosine. *StatPearls* [Internet]. Treasure Island (FL): StatPearls Publishing; 2023.
- Murugan M, Fedele D, Millner D, Alharfoush E, Vegunta G, Boison D. Adenosine kinase: an epigenetic modulator in development and disease. *Neurochem Int* 2021;147:105054.
- Jarvis MF. Therapeutic potential of adenosine kinase inhibition – revisited. *Pharmacol Res Perspect* 2019;7:e00506.
- Abboud C, Duveau A, Bouali-Benazzouz R, Massé K, Mattar J, Brochoire L, et al. Animal models of pain: diversity and benefits. *J Neurosci Methods* 2021;348:108997.
- Obeng S, Hiranita T, León F, McMahon LR, McCurdy CR. Novel approaches, drug candidates, and targets in pain drug discovery. *J Med Chem* 2021;64:6523–48.
- Zhou M, Wu J, Chang H, Fang Y, Zhang D, Guo Y. Adenosine signaling mediate pain transmission in the central nervous system. *Purinergic Signal* 2023;19:245–54.
- Bai H-H, Liu J-P, Yang L, Zhao J-Y, Suo Z-W, Yang X, et al. Adenosine A1 receptor potentiated glycinergic transmission in spinal cord dorsal horn of rats after peripheral inflammation. *Neuropharmacology* 2017;126:158–67.
- Diao X-T, Yao L, Ma J-J, Zhang T-Y, Bai H-H, Suo Z-W, et al. Analgesic action of adenosine A1 receptor involves the dephosphorylation of glycine receptor  $\alpha 1$ ins subunit in spinal dorsal horn of mice. *Neuropharmacology* 2020;176:108219.
- Zahn PK, Straub H, Wenk M, Pogatzki-Zahn EM. Adenosine A1 but not A2a receptor agonist reduces hyperalgesia caused by a surgical incision in rats: a pertussis toxin-sensitive G protein – dependent process. *Anesthesiology* 2007;107:797–806.
- Balasubramanyan S, Sharma S. Protective effect of adenosine in diabetic neuropathic pain is mediated through adenosine A1 receptors. *Indian J Physiol Pharmacol* 2008;52:233–42.
- Chen Z, Janes K, Chen C, Doyle T, Bryant L, Tosh DK, et al. Controlling murine and rat chronic pain through A3 adenosine receptor activation. *FASEB J* 2012;26:1855–65.
- Draper-Joyce CJ, Bholra R, Wang J, Bhattarai A, Nguyen ATN, Cowie-Kent I, et al. Positive allosteric mechanisms of adenosine A1 receptor-mediated analgesia. *Nature* 2021;597:571–6.
- Janes K, Wahlman C, Little JW, Doyle T, Tosh DK, Jacobson KA, et al. Spinal neuroimmune activation is independent of T-cell infiltration and attenuated by A3 adenosine receptor agonists in a model of oxaliplatin-induced peripheral neuropathy. *Brain Behav Immun* 2015;44:91–9.
- Janes K, Esposito E, Doyle T, Cuzzocrea S, Tosh DK, Jacobson KA, et al. A3 adenosine receptor agonist prevents the development of paclitaxel-induced neuropathic pain by modulating spinal glial-restricted redox-dependent signaling pathways. *Pain* 2014;155:2560–7.
- Liu L, Xia L, Cui Y. Loss of astrocytic A1 adenosine receptor is involved in a chemotherapeutic agent-induced rodent model of neuropathic pain. *Muscle Nerve* 2023;67:417–26.
- Kim K, Jeong W, Jun IG, Park JY. Antiallodynic and anti-inflammatory effects of intrathecal R-PIA in a rat model of vincristine-induced peripheral neuropathy. *Korean J Anesth* 2020;73:434–44.
- Katz NK, Ryals JM, Wright DE. Central or peripheral delivery of an adenosine A1 receptor agonist improves mechanical allodynia in a mouse model of painful diabetic neuropathy. *Neuroscience* 2015;285:312–23.
- Li X, Bantel C, Conklin D, Childers SR, Eisenach JC. Repeated dosing with oral allosteric modulator of adenosine A1 receptor produces tolerance in rats with neuropathic pain. *Anesthesiology* 2004;100:956–61.
- Loram LC, Taylor FR, Strand KA, Harrison JA, Rzasalynn R, Sholar P, et al. Intrathecal injection of adenosine 2A receptor agonists reversed

- neuropathic allodynia through protein kinase (PK)A/PKC signaling. *Brain Behav Immun* 2013;33:112–22.
26. Loram LC, Harrison JA, Sloane EM, Hutchinson MR, Sholar P, Taylor FR, et al. Enduring reversal of neuropathic pain by a single intrathecal injection of adenosine 2A receptor agonists: a novel therapy for neuropathic pain. *J Neurosci* 2009;29:14015–25.
  27. Ford A, Castonguay A, Cottet M, Little JW, Chen Z, Symons-Liguori AM, et al. Engagement of the GABA to KCC2 signaling pathway contributes to the analgesic effects of A3 AR agonists in neuropathic pain. *J Neurosci* 2015;35:6057–67.
  28. Kwilasz AJ, Ellis A, Wieseler J, Loram L, Favret J, McFadden A, et al. Sustained reversal of central neuropathic pain induced by a single intrathecal injection of adenosine A2A receptor agonists. *Brain Behav Immun* 2018;69:470–9.
  29. Wahlman C, Doyle TM, Little JW, Luongo L, Janes K, Chen Z, et al. Chemotherapy-induced pain is promoted by enhanced spinal adenosine kinase levels through astrocyte-dependent mechanisms. *Pain* 2018;159:1025–34.
  30. Wu W-P, Hao J-X, Halldnr L, Lövdahl C, DeLander GE, Wiesenfeld-Hallin Z, et al. Increased nociceptive response in mice lacking the adenosine A1 receptor. *Pain* 2005;113:395–404.
  31. Gong Q-J, Li Y-Y, Xin W-J, Wei X-H, Cui Y, Wang J, et al. Differential effects of adenosine A1 receptor on pain-related behavior in normal and nerve-injured rats. *Brain Res* 2010;1361:23–30.
  32. Borea PA, Varani K, Vincenzi F, Baraldi PG, Tabrizi MA, Merighi S, et al. The A3 adenosine receptor: history and perspectives. Sibley DR, editor. *Pharmacol Rev* 2015;67:74–102.
  33. Durante M, Squillace S, Lauro F, Giancotti LA, Coppi E, Cherchi F, et al. Adenosine A3 agonists reverse neuropathic pain via T cell – mediated production of IL-10. *J Clin Invest* 2021;131:e139299.
  34. Coppi E, Cherchi F, Fusco I, Failli P, Vona A, Dettori I, et al. Adenosine A3 receptor activation inhibits pronociceptive N-type Ca<sup>2+</sup> currents and cell excitability in dorsal root ganglion neurons. *Pain* 2019;160:1103–18.
  35. Li T, Ren G, Lusardi T, Wilz A, Lan JQ, Iwasato T, et al. Adenosine kinase is a target for the prediction and prevention of epileptogenesis in mice. *J Clin Invest* 2008;118:571–82.
  36. Jonzon B, Sylvén C, Kaijser L. Theophylline decreases pain in the ischaemic forearm test. *Cardiovasc Res* 1989;23:807–9.
  37. Sylvén C, Beermann B, Jonzon B, Brandt R. Angina pectoris-like pain provoked by intravenous adenosine in healthy volunteers. *Br Med J* 1986;293:227–30.
  38. Segerdahl M, Karelov A. Experimentally induced ischaemic pain in healthy humans is attenuated by the adenosine receptor antagonist theophylline. *Acta Physiol Scand* 2004;180:301–6.
  39. Rae CP, Mansfield MD, Dryden C, Kinsella J. Analgesic effect of adenosine on ischaemic pain in human volunteers. *Br J Anaesth* 1999;82:427–8.
  40. Segerdahl M, Ekblom A, Sollevi A. The influence of adenosine, ketamine, and morphine on experimentally induced ischemic pain in healthy volunteers. *Anesth Analg* 1994;79:787–91.
  41. Ekblom A, Segerdahl M, Sollevi A. Adenosine increases the cutaneous heat pain threshold in healthy volunteers. *Acta Anaesthesiol Scand* 1995;39:717–22.
  42. Rane K, Segerdahl M, Gojny M, Sollevi A. Intrathecal adenosine administration: a phase 1 clinical safety study in healthy volunteers, with additional evaluation of its influence on sensory thresholds and experimental pain. *Anesthesiology* 1998;89:1108–15.
  43. Sjolund K-F, Segerdahl M, Sollevi A. Adenosine reduces secondary hyperalgesia in two human models of cutaneous inflammatory pain. *Anesth Analg* 1999;88:605–10.
  44. Dirks J, Petersen K, Rowbotham M, Dahl J. Effect of systemic adenosine on pain and secondary hyperalgesia associated with the heat/capsaicin sensitization model in healthy volunteers. *Reg Anesth Pain Med* 2001;26:414–9.
  45. Eisenach JC, Hood DD, Curry R. Preliminary efficacy assessment of intrathecal injection of an American formulation of adenosine in humans. *Anesthesiology* 2002;96:29–34.
  46. Eisenach JC, Curry R, Hood DD. Dose response of intrathecal adenosine in experimental pain and allodynia. *Anesthesiology* 2002;97:938–42.
  47. Chizh BA, Dusch M, Puthawala M, Schmelz M, Cookson LM, Martina R, et al. The effect of intravenous infusion of adenosine on electrically evoked hyperalgesia in a healthy volunteer model of central sensitization. *Anesth Analg* 2004;99:816–22.
  48. Morelot-Panzini C, Corvol J-C, Demoule A, Raux M, Fiamma M-N, Willer J-C, et al. Intravenous adenosine activates diffuse nociceptive inhibitory controls in humans. *J Appl Physiol* 2013;115:697–703.
  49. Giffin NJ, Kowacs F, Libri V, Williams P, Goadsby PJ, Kaube H. Effect of the adenosine A1 receptor agonist Gr79236 on trigeminal nociception with blink reflex recordings in healthy human subjects. *Cephalalgia* 2003;23:287–92.
  50. Sollevi A, Belfrage M, Lundeberg T, Segerdahl M, Hansson P. Systemic adenosine infusion: a new treatment modality to alleviate neuropathic pain. *Pain* 1995;61:155–8.
  51. Belfrage M, Sollevi A, Segerdahl M, Sjolund K-F, Hansson P. Systemic adenosine infusion alleviates spontaneous and stimulus evoked pain in patients with peripheral neuropathic pain. *Anesth Analg* 1995;81:713–7.
  52. Lindblom U, Nordfors L-O, Sollevi A, Sydow O. Adenosine for pain relief in a patient with intractable secondary erythromelalgia. *Eur J Pain* 1997;1:299–302.
  53. Rauck RL, North J, Eisenach JC. Intrathecal clonidine and adenosine: effects on pain and sensory processing in patients with chronic regional pain syndrome. *Pain* 2015;156:88–95.
  54. Lynch ME, Clark AJ, Sawynok J. Intravenous adenosine alleviates neuropathic pain: a double blind placebo controlled crossover trial using an enriched enrolment design. *Pain* 2003;103:111–7.
  55. Eisenach JC, Rauck RL, Curry R. Intrathecal, but not intravenous adenosine reduces allodynia in patients with neuropathic pain. *Pain* 2003;105:65–70.
  56. Rane K, Karlsten R, Sollevi A, Gordh T Jr., Svensson BA. Spinal cord morphology after chronic intrathecal administration of adenosine in the rat. *Acta Anaesthesiol Scand* 1999;43:1035–40.
  57. Chiari A, Yaksh TL, Myers RR, Provencher J, Moore L, Lee C-S, et al. Preclinical toxicity screening of intrathecal adenosine in rats and dogs. *Anesthesiology* 1999;91:824–32.
  58. Segerdahl M, Ekblom A, Sandelin K, Wickman M, Sollevi A. Perioperative adenosine infusion reduces the requirements for isoflurane and postoperative analgesics. *Anesth Analg* 1995;80:1145–9.
  59. Segerdahl M, Irestedt L, Sollevi A. Antinociceptive effect of perioperative adenosine infusion in abdominal hysterectomy. *Acta Anaesthesiol Scand* 1997;41:473–9.
  60. Rane K, Sollevi A, Segerdahl M. Intrathecal adenosine administration in abdominal hysterectomy lacks analgesic effect. *Acta Anaesthesiol Scand* 2000;44:868–72.
  61. Rane K, Sollevi A, Segerdahl M. A randomised double-blind evaluation of adenosine as adjunct to sufentanil in spinal labour analgesia. *Acta Anaesthesiol Scand* 2003;47:601–3.
  62. Sharma M, Mohta M, Chawla R. Efficacy of intrathecal adenosine for postoperative pain relief. *Eur J Anaesthesiol* 2006;23:449–53.