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ALTERNATIVE TREATMENTS FOR PAIN MEDICINE (M JONES, SECTION EDITOR)

Flavonoids in the Treatment of Neuropathic Pain

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Abstract

Purpose of Review Chronic pain continues to present a large burden to the US healthcare system. Neuropathic pain, a common class of chronic pain, remains particularly difficult to treat despite extensive research efforts. Current pharmacologic regimens exert limited efficacy and wide, potentially dangerous side effect profiles. This review provides a comprehensive, preclinical evaluation of the literature regarding the role of flavonoids in the treatment of neuropathic pain.

Recent Findings Flavonoids are naturally occurring compounds, found in plants and various dietary sources, which may have potential benefit in neuropathic pain. Numerous animal-model studies have demonstrated this benefit, including reversal of hyperalgesia and allodynia. Flavonoids have also exhibited an anti-inflammatory effect relevant to neuropathic pain, as evidenced by the reduction in multiple pro-inflammatory mediators, such as TNF-α, NF-κB, IL-1β, and IL-6.

Summary Flavonoids represent a potentially new treatment modality for neuropathic pain in preclinical models, though human clinical evidence is yet to be explored at this time.

Keywords Neuropathic pain · Flavonoids · Neuroinflammation · Chronic pain · Treatment

Introduction

Chronic pain management continues to challenge health systems worldwide as a leading cause of disability and disease burden [1]. Over 100 million US adults suffer from chronic pain, implicating it as one of the most prevalent and disabling health conditions [1, 2]. Furthermore, accounting for healthcare costs and lost productivity, the annual cost of chronic pain ranges from approximately $560 to $635 billion annually, exceeding the combined costs of cancer, heart disease, and diabetes [2, 3]. Despite its large impact, an overall increasing trend persists in the prevalence of chronic pain among the US population [1].

Neuropathic pain, a common class of chronic pain, has been described as "pain caused by a lesion or disease of the somatosensory nervous system" [4]. The etiology of neuropathic pain is often multifactorial and the underlying pathophysiology is still not fully understood. When treating neuropathic pain, the importance of a multidisciplinary approach has shown to decrease pain, and improve mood and function [5]. Commonly used analgesics, such as non-steroidal anti-inflammatory drugs and opioids which are typically used for nociceptive pain, are less effective in neuropathic pain [6]. The first-line pharmacological therapies in neuropathic pain include tricyclic antidepressants (TCAs), serotonin-norepinephrine reuptake inhibitors (SNRIs), gabapentinoids, and topical substances [7]. These medications can be efficacious, but carry the potential risk of multiple side effects, which are especially crucial to consider in patients with renal or hepatic dysfunction, cardiac arrhythmias, psychiatric

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comorbidities requiring medication, and elderly patients [7, 8]. Many patients are prescribed multiple agents for combination therapy to maximize benefit while minimizing the risk of adverse effects [6, 7, 9]. Opioids also tend to be lower-line pharmacologic options for neuropathic pain; per multiple systematic reviews, there is minimal to low evidence supporting the use of opioids for long-term use in chronic neuropathic pain with the possible benefit for only short-term use [6, 10–12].

Despite the multiple agents and modalities available for the treatment of neuropathic pain, current treatment options provide an average reduction in neuropathic pain by 30–50% with 20–40% of patients achieving less than 30% improvement in pain [6]. The difficulty in optimally treating neuropathic pain has prompted significant research into newer analgesic agents with fewer side effects. Natural products, such as flavonoids, have demonstrated potential due to their role in inflammatory diseases [13•]. Given the emerging evidence surrounding the role of neuroinflammation in neuropathic pain [14–16], we conducted a review of the literature to examine the role of flavonoids in the treatment of neuropathic pain. A comprehensive literature search was conducted, including PubMed, EMBASE, MEDLINE, and Google Scholar databases. Articles published in peer-reviewed journals were reviewed systematically, including references cited in relevant articles. Search terms used included "neuropathic pain" AND "flavonoids" OR "inflammation".

Anti-inflammatory Effects of Flavonoids

Flavonoids have been studied extensively for various indications, including their role as anti-inflammatory, antioxidant, and antitumor agents [13, 17]. They are naturally occurring polyphenolic compounds that can be found in plants and various dietary sources, including fruits, vegetables, and legumes. Flavonoids, comprised of greater than 4000 individual compounds, can be divided into separate classes depending on specific modifications to the base carbon skeleton. Certain structural modifications have been thought to lead to greater anti-inflammatory activity, including an unsaturated C ring, presence of carbonyl group on C-4, and lack of glycosylation.

The mechanism by which flavonoids exhibit their anti-inflammatory effect is thought to be multifactorial, including inhibition of protein kinases and transcription factors, antioxidant activity, and downregulation of immune cell activity, many of which are implicated in the neuroinflammation cascade [13, 17–19]. It has been well demonstrated that flavonoids inhibit the mitogen-activated protein kinase (MAPK) pro-inflammatory signaling cascade, particularly associated with downregulated levels of IL-1β, TNF-α, and IL-6 [17, 19]. Furthermore, there is also emerging evidence that flavonoids may modulate many metabolic pathways causing neuronal dysfunction, though more so studied in other disease states linked to neuroinflammation, such as Alzheimer's disease, Parkinson's disease, and multiple sclerosis [17, 20]. Increasing animal model data has importantly demonstrated that flavonoids may have a therapeutic role in the treatment of neuropathic pain.

Preclinical Evidence for Flavonoids in Neuropathic Pain

Naringenin

Naringenin is part of the flavonoid subgroup called flavanones, which are typically found in citrus fruits, herbal teas, potatoes, bergamot, and tomatoes [21]. The compound is derived from its precursor naringin via hydrolysis [22]. Several studies using animal-based models support its analgesic effect through its anti-inflammatory capabilities via inhibition of voltage-gated sodium channels leading to reduced calcium influx in dorsal root ganglion neurons [23]. Thus far, studies have demonstrated analgesic effects in the treatment of somatic pain and visceral pain, in addition to neuropathic pain [24]. A summation of studies on naringenin to date is presented in Table 1.

Multiple studies on naringenin have focused on its potential role in treating diabetic neuropathy. In these studies, diabetes was induced in these patients via an intraperitoneal injection of streptozotocin, which destroys pancreatic β-cells. In a study by Al-Rejaie et al., rats were injected with either 25 or 50 mg/kg/day of intraperitoneal naringenin, which was shown to reverse both clinical and biochemical measures of diabetic neuropathy [25]. The rats treated with naringenin had reduced serum glucose levels, increased insulin levels, decreased hyperalgesia based on increasing tail and paw withdrawal latency, and dose-dependent decreases in IL-1β, IL-6, and TNF-α, which are thought to provoke neural cell death leading to neuropathy [25]. A similar study by Singh et al. in 2020 corroborated these findings, in which they observed a dose-dependent reduction in serum glucose levels and hemoglobin A1C, multiple inflammatory cytokines (TNF-α, TGF-β, and MMP-9), response to painful stimuli suggesting improved hyperalgesia, and improved nerve conduction studies [26].

Hu and Zhao studied the benefits of naringenin using a peripheral neuropathy model using lumbar spinal nerve ligation in 2014 [27]. Similar to the prior two studies, rats were treated with various doses of intrathecal naringenin. Allodynia and hyperalgesia were both attenuated, and various inflammatory markers (IL-1B, TNF-α, and MCP-1) were reduced in rats administered naringenin [27].

Lastly, two other studies that have demonstrated the analgesic potential of naringin, the precursor of naringenin. The first study examined the role of naringin in reversing the

Table 1 Naringenin

Reference	Model	Dosing	Findings
Al-Rejaie et al. [25]	Diabetic neuropathy	Intraperitoneal 25 or 50 mg/kg/d	-Decreased serum glucose levels and increase insulin levels -Dose-dependent reduction in IL-1B, IL-6, and TNF-α -Decreased paw-withdrawal and tail-flick latency
Singh et al. [26]	Diabetic neuropathy	Intraperitoneal 25, 50, or 100 mg/kg/d	-Dose dependent reduction in serum glucose and HA1C levels -Reduced levels of TNF-α, TGF-β, and MMP-9 -Reduced reaction time to hot-plate and tail-immersion tests -Improved results on nerve conduction studies
Hu and Zhao [27]	Spinal nerve ligation	Intrathecal 50, 100, or 200 mg/kg/d	-Attenuation of mechanical allodynia and thermal hyperalgesia -Reduction in TNF-α, IL1B, and MCP-1 levels -Reduction in glial cell activation demonstrated by reduction in expression of GFAP and Mac-1

effects of cisplatin-based neuropathy. Their results showed that naringin had a dose-dependent reversal of behavioral, biochemical, and molecular alterations caused by cisplatin hypothesized to be due to a reduction in reactive oxidative species and nitric oxide levels [28]. Another study by Kandhare et al. in 2012 examined naringin using the streptozotocin-induced diabetic neuropathy model. Their findings showed that naringin reversed both the diabetic state and neuropathic pain similar to the findings in the studies shared above on naringenin [29].

Quercetin

Quercetin is part of the flavonol subgroup, which is characterized by the hydroxyl group on the third position in the carbon ring and is frequently found in apples, berries, grapes, kale, leek, lettuce, onions, tomatoes, tea, and red wine [21]. In vitro studies have demonstrated that quercetin has properties as a mast cell stabilizer by inhibiting histamine release [30•].

A summation of studies on quercetin to date is presented in Table 2.

Much of the research on quercetin focuses on chemotherapy-induced neuropathy, which is often a side effect of platinum-based agents, such as cisplatin, paclitaxel, and oxaliplatin. Similar to other neuropathies, chemotherapy-induced neuropathy is also complicated by allodynia and hyperalgesia [30•]. Two initial studies both used an oxaliplatin-based rat model with conflicting results. The first study by Azevedo et al. in 2013 used mice treated with intraperitoneal quercetin (25–100 mg/kg) and intravenous oxaliplatin (1 mg/kg) and demonstrated that quercetin reversed oxaliplatin-induced allodynia [31]. Additionally, biochemical analysis of spinal cord tissue samples showed a reduction in oxidative stress with reduced levels of nitric oxide synthase and nitrotyrosine [31]. The following year, Schwingel et al. [32] conducted another study that did not replicate the results found by Azevedo et al. [31]. Schwingel et al. first pretreated rats with 10 mg/kg/week of oxaliplatin for 6 weeks, at the end of which they predictably demonstrated signs of mechanical allodynia [32]. The rats were then treated with either 20 mg/kg/day of either quercetin or quercetin nanoemulsion, and neither of these two groups demonstrated any improvement in allodynia after treatment with the two quercetin formulations in behavioral testing [32]. These results suggested that quercetin may need to be administered as a pretreatment to the chemotherapeutic agent to have its analgesic effect [32].

The analgesic efficacy of pretreatment with quercetin has since been supported by multiple studies, one of which also examined chemotherapy-induced neuropathy. Gao et al. in 2016 used a paclitaxel-induced neuropathy model, in which rats and mice were treated with 20 or 60 mg/kg/day of intraperitoneal quercetin, as well as paclitaxel 2 mg/kg every other day for the first 7 days of the experiment [30•]. Their findings demonstrated a reduction in paclitaxel-induced hyperalgesia, and tissue samples with reduced expression of PKC-epsilon and TRPV1, which are associated with paclitaxel-induced neuropathy [30•]. A more recent study in 2018 specifically compared outcomes between rats treated with quercetin before and after initiation of a spared nerve injury model of the sciatic nerve, in which the authors showed that there was attenuation of allodynia in pre-injury quercetin-treated rats, but not in those treated post-injury[33].

Ji et al. elucidated further insight regarding the temporal nature of quercetins' analgesic actions using a spinal nerve ligation model [34]. In this study, rats were treated with quercetin for 14 days either before or after surgical ligation. Both groups demonstrated attenuation of hyperalgesia compared to

Table 2 Quercetin

Reference	Model	Dosing	Findings
Gao et al. [30•]	Paclitaxel-induced neuropathy	Intraperitoneal 20 or 60 mg/kg/d	-Dose-dependent reduction in hyperalgesia and allodynia based on mechanical nociceptive thresholds and tail withdrawal latency -Tissue samples demonstrated that quercetin inhibited expression of PKC-epsilon and TRPV1
Azevado et al. [31]	Oxaliplatin-induced neuropathy	Intraperitoneal 25–100 mcg/kg twice a week	-Reversal of oxaliplatin-induced allodynia -Reduced levels of nitric oxide synthase and nitrotyrosine
Schwingel et al. [32]	Oxaliplatin-induced neuropathy	Intraperitoneal quercetin 20 mg/kg/d or quercetin nanoemulsion 20 mg/kg/d	-No change in mechanical allodynia caused by oxaliplatin -Reduction in c-Fos levels in DRG tissue samples
Muto et al. [33]	Spared nerve injury of sciatic nerve	Rodent diet with 1% quercetin supplementation	-Attenuation of allodynia with pre-injury treatment of quercetin but not post-injury -Quercetin inhibited GFAP in satellite glial cells in DRG
Ji et al. [34]	Spinal nerve ligation	10–100 mg/kg/d for 14 d either before or after surgical ligation	-Both groups demonstrated attenuation of hyperalgesic response -Magnitude of analgesic effect of pre-injury group decreased over 14 days after injury while the magnitude increased in post-treatment group -Quercetin reduced phosphorylation of TAK1, IKK, and JNK2 in astrocytes
Civi et al. [35]	Chronic constriction injury of sciatic nerve	100 mg/kg	-Pre-injury treatment showed long-term attenuation of mechanical and thermal hypersensitivity
Yang et al. [36]	Diabetic neuropathy model	Intraperitoneal 50 mg/kg/d for 14 d	-Increased threshold and latency in diabetic rats treated with quercetin -Increased levels of P2X4 mRNA in diabetic rats that was reversed in rats who received quercetin

the control group not treated with quercetin, but the rats that received pre-injury quercetin had the greatest attenuation of hyperalgesia assessed on the last day of quercetin administration [34]. Following treatment, the degree of attenuation resembled that of the control arm [34]. The group that received post-injury quercetin initially had hyperalgesia comparable to the control arm, though with progressive attenuation of hyperalgesia over the 14-day course of treatment with quercetin, suggesting that the analgesic effects of quercetin increase with regular dosing and wane shortly after treatment is stopped [34].

Additionally, quercetin has been studied using two other models for neuropathic pain in mice and rats. First, a study by Civi et al. showed that pre-injury quercetin administration led to long-term reduction of hypersensitivity in a chronic constriction nerve injury (CCI) model, and the effect was superior to that of gabapentin and morphine [35]. Second, a recent study by Yang et al. in 2019 demonstrated that quercetin reduced mechanical allodynia and hyperalgesia in a diabetic neuropathy model via decreased upregulation of the p38MAPK signaling cascade, which is implicated in the upregulation of pro-inflammatory mediators in neuropathic pain [36].

Hesperidin

Hesperidin, hesperetin-7-rhamnoglucoside, is a primarily citrus flavanone that has been studied for its anti-inflammatory, analgesic, and anti-diabetic properties [21, 37]. Though the exact molecular mechanism of action is yet to be elucidated, hesperidin is overall thought to mediate its anti-inflammatory properties via downregulation of multiple pro-inflammatory

cytokines, some of which are implicated in neuropathic pain models [38, 39]. A summation of studies on hesperidin to date is presented in Table 3.

Visnagri et al. induced diabetes and diabetic neuropathy in rat models with streptozotocin [39]. Four weeks later, after confirming the induction of diabetes and baseline neuropathic pain, they administered hesperidin in dosages ranging from 25 to 100mg/kg, with or without coadministration of insulin, and subsequently analyzed behavioral tests for assessing hyperalgesia and allodynia, nerve conduction velocity, plasma

Table 3 Hesperidin

Reference	Model	Dosing	Findings
Carballo-Villalobos et al. [38]	Chronic constriction injury of sciatic nerve	Intraperitoneal hesperidin 100mg/kg	- Reduced hyperalgesia - Decreased protein levels of TNF-α, IL-6, and IL-1β in both sciatic nerve and throughout spinal cord
Visnagri et al. [39]	Diabetic neuropathy model	Oral hesperidin 50mg/kg and 100mg/kg	- Significantly reduced hyperglycemia - Dose-dependent reduction in mechano-tactile allodynia and thermal hyperalgesia, with synergistic effect when combined with insulin - Ameliorated decrease in nerve conduction velocity - Dose-dependent downregulation of TNF-α and IL-1β mRNA, with synergistic effect when combined with insulin
Carballo-Villalobos et al. [40]	Chronic constriction injury of sciatic nerve	Intraperitoneal hesperidin 10mg/kg, 100mg/kg, 316.2mg/kg, 562.3mg/kg, 1000mg/kg; intraperitoneal diosmin 10mg/kg, 100mg/kg	- Significantly decreased mechanical and thermal hyperalgesia - Hesperidin effect synergistically improved with coadministration of diosmin - Naloxone, bicuculline, and haloperidol variably inhibited antihyperalgesic effects of hesperidin/diosmin
Aswar et al. [41]	Partial sciatic nerve ligation	Oral hesperetin 20mg/kg and 50mg/kg	- Attenuated development of mechanical hyperalgesia and mechano-tactile allodynia - Improved nerve conduction velocity - Significant downregulation of TNF-α, IL-6, and IL-1β mRNA
Tao et al. [42]	Chronic constriction injury of sciatic nerve	Intraperitoneal hesperidin 50mg/kg	- Decreased mechanical and thermal hyperalgesia - Decreased expression of P2X3 mRNA and protein in dorsal root ganglia - Hesperidin both directly inhibits P2X3, and suppresses upregulation of P2X3 receptors in DRG neurons

glucose levels, and pro-inflammatory cytokine mRNA expression at 8 weeks [39]. The authors demonstrated that both hesperidin and insulin were independently capable of reducing plasma glucose levels, but the effect was synergistic when both hesperidin and insulin were administered together [39]. In addition to this anti-hyperglycemic effect, the same synergistic effect between hesperidin and insulin was observed when assessing mechano-tactile allodynia, thermal hyperalgesia, and nerve conduction velocity [39]. Furthermore, hesperidin and insulin coadministration again led to a supra-additive effect resulting in downregulation of TNF-α and IL-1β mRNA in the sciatic nerve [39]. Thus, it follows that although hesperidin may have an independent effect in ameliorating diabetes and downregulating the associated pro-inflammatory cytokines implicated in neuropathic pain, adequate glycemic control is also an important factor in reducing the effects of diabetic neuropathy.

Multiple studies of hesperidin in chronic constriction injury neuropathic pain models revealed similar benefits in reducing neuropathic pain. In 2016, Caraballo-Villalobos et al. induced neuropathic pain via CCI of the sciatic nerve in rats, and assessed paw withdrawal threshold and latency following administration of hesperidin and/or diosmin (another flavonoid compound), and gabapentin [40]. Both paw withdrawal threshold and latency, reflecting mechanical and thermal hyperalgesia respectively, were improved with hesperidin [40]. In addition, a supra-additive effect was observed with coadministration of hesperidin with diosmin in both mechanical and thermal hyperalgesia, but this antihyperalgesic effect was diminished in the presence of naloxone, bicuculline (a selective $GABA_A$ antagonist), and haloperidol, potentially implicating opioids, $GABA_A$, and D2 receptors as mediators in the antihyperalgesic effect of flavonoids [40]. A subsequent study in 2017 also utilizing CCI models of rat sciatic nerves again demonstrated the antihyperalgesic effect of hesperidin, while also reporting decreased protein levels of TNF-α, IL-6, and IL-1β in the sciatic nerve, corroborating evidence from an earlier study utilizing a partial sciatic nerve ligation model [38, 41]. An interesting aspect of the CCI rat model study by Caraballo-Villalobos et al. in 2017 was the downregulation of protein levels of pro-inflammatory cytokines in not only the sciatic nerve but also in the spinal cord following hesperidin treatment [38]. Furthermore, the authors observed an increase in TNF-α in the spinal cord samples of neuropathic rats prior to any treatment, without significant changes in IL-1β or IL-6, which was attenuated by hesperidin [38]. Thus, in addition to peripheral antihyperalgesic effects, hesperidin may also have a secondary role in decreasing central sensitization in neuropathic pain, which may be mediated by TNF-α.

Tao et al. again demonstrated the antihyperalgesic effects of hesperidin in CCI rats in 2019, though focused on the P2X3 receptor, which amplifies nociceptive signaling secondary to release of ATP following peripheral nerve injury [42]. The authors showed that there was decreased expression of P2X3 mRNA and protein in the dorsal root ganglia (DRG) neurons following hesperidin treatment in CCI rats and that hesperidin both directly inhibited P2X3 and suppressed upregulation of P2X3 receptors in DRG neurons [42]. This study further highlights the multifactorial development of neuropathic pain, highlighting various signaling cascades and mediators that may also be affected by flavonoid compounds.

Epigallocatechin-3-gallate

Epigallocatechin-3-gallate (EGCG) belongs to the flavan-3-ol subgroup of flavonoids and is the most abundant and active component of green tea. Many studies have explored its role in reducing inflammation, inhibiting oxidative stress, and possibly modulating tumorigenesis [19, 37, 43]. Here, we highlight several studies validating its preclinical role in the treatment of neuropathic pain in various rat models. A summation of studies on EGCG to date is presented in Table 4.

Both Xifro et al. and Bosch-Mola et al. demonstrated the efficacy of EGCG in reducing thermal hyperalgesia in their respective CCI rat models [44, 45]. Xifro et al. examined EGCG and two related synthetic derivatives, compound 23 and compound 30, and demonstrated that both EGCG and compound 30 were effective in reducing thermal hyperalgesia [44]. However, EGCG was more effective in the acute phase with a statistically significant reduction in thermal hyperalgesia until 21 days post-injury, whereas compound 30 was shown to be effective in the same regard from 14 to 56 days post-injury, which is postulated to be related to the increased stability of the synthetic compound 30 [44]. Additionally, Xifro et al. observed greater inhibition of fatty acid synthase (FASN) activity, which has also been implicated in neuropathic pain following nerve injury, in the dorsal horn of the spinal cord in EGCG and compound 30 treated CCI-mice, with again a more sustained effect observed in compound 30 treated CCI-mice [44]. Furthermore, as seen with other flavonoids, EGCG and compound 30 downregulated TNF-α, IL-6, IL-1β, and NF-κB mRNA and protein expression in the dorsal horn of the spinal cord, though this effect was only observed at 14 days post-injury and not sustained at 56 days post injury [44]. Thus, EGCG and compound 30 may modulate the earlier phases of development of neuropathic pain via downregulation of pro-inflammatory cytokines, but other molecular pathways likely also contribute to the persistence of neuropathic pain in the chronic phase. Additionally, the efficacy and sustained effect of compound 30 in reducing hyperalgesia and inflammation highlight an interesting avenue for further research in synthetic formulations of natural flavonoids.

Bosch-Mola et al. explored the effect of EGCG on chemokine fractalkine (CX3CL1), which is thought to be a mediator in the interaction between neurons and microglia in the

Table 4 Epigallocatechin-3-gallate (EGCG)

Reference	Model	Dosing	Findings
Xifro et al. [44]	Chronic constriction injury of sciatic nerve	Intraperitoneal 10mg/kg, 30mg/kg, 50mg/kg, 100mg/kg	- Reduced thermal hyperalgesia - Reduction in FASN activity in dorsal horn of spinal cord - Decreased expression of TNF-α, IL-6, IL-1β, and NF-κB mRNA and protein in dorsal horn of spinal cord
Bosch-Mola et al. [45]	Chronic constriction injury of sciatic nerve	Intraperitoneal 50mg/kg	- Reduced thermal hyperalgesia - Downregulated CX3CL1 protein expression in spinal cord
Renno et al. [46]	Spinal cord contusion injury	Intravenous 20mg/kg/h for 36h	- Improved recovery of motor and sensory function following injury, sustained at 1 year - Decreased thermal hyperalgesia and tactile allodynia - Preservation of neuronal morphology following injury - Reduced spinal cord gliosis
Renno et al. [47]	Sciatic nerve crush injury	Intraperitoneal 50mg/kg	- Improvement of motor recovery following crush injury - Reduced allodynia and mechanical/thermal hyperalgesia - Markedly improved axonal and myelin regeneration - Decreased expression of TNF-α, Nrf2, HO-1, and IL-1β
Alvarez-Perez et al. [48]	Spinal cord contusion injury	Intraperitoneal 30mg/kg	- Reduced thermal hyperalgesia, but no motor recovery - Downregulated RhoA, TNF-α, and FASN in spinal cord - Reduced gliosis following injury, but no effect of sprouting of afferent nociceptive fibers
Kuang et al. [49]	Chronic constriction injury of sciatic nerve	Intrathecal 1mg/kg	- Reduced mechanical and thermal hyperalgesia - Downregulated TNF-α and IL-1β protein expression in spinal cord, but increased IL-10 protein expression - Decreased mRNA and protein expression of TLR4, HMGB1, and NF-κB in spinal cord

development of neuropathic pain [45]. Previous studies have shown that exogenous CX3CL1 can induce hyperalgesia and that NF-κB is implicated in the upregulation of both CX3CL1 in neurons and its receptor in microglial cells [50, 51]. Bosch-Mola et al. demonstrated that EGCG reduced thermal hyperalgesia as discussed above, and downregulated protein expression, but not mRNA expression, of CX3CL1 [45]. This again further emphasizes the numerous molecular pathways involved in the development of neuropathic pain, though EGCG seems to modulate its effect via multiple mediators.

In 2014, Renno et al. observed improved recovery of motor and sensory function following spinal cord contusion injury in rats treated with intravenous EGCG for 36 h beginning 4 h after spinal cord injury [46]. Locomotor recovery was sustained from 3 to 10 weeks post-injury, and even observed 1 year following spinal cord injury [46]. A different study by Renno et al. in 2017 corroborated the findings of improved motor recovery in EGCG-treated rats following sciatic nerve crush injury, but Alvarez-Perez et al. observed no motor recovery following spinal cord contusion injury in EGCG-treated rats [47, 48]. Of note, different methods of EGCG administration (intravenous vs. intraperitoneal) and doses were used in the Renno et al. study and the Alvarez-Perez study [46, 48]. Numerous studies have demonstrated the effect of EGCG on neuronal integrity and gliosis following injury. Both Renno et al. (2014) and Alvarez-Perez et al. demonstrated the preservation of neuronal morphology in EGCG-treated rats following injury, with reduced spinal cord gliosis [46, 48]. In 2017, Renno et al. revealed morphological evidence of a significantly greater degree of axonal and myelin regeneration in EGCG-treated rats in a sciatic nerve crush injury model [47]. In addition, numerous studies have again highlighted the role of EGCG in attenuating an increase in expression of pro-inflammatory mediators, such as TNF-α, Nrf2, HO-1, RhoA, FASN, and IL-1β [46–48].

Kuang et al. presented an early study of EGCG in a CCI rat model in 2012, in which they administered intrathecal EGCG and observed reduced mechanical and thermal hyperalgesia [49]. Interestingly, following EGCG treatment, TNF-α and IL-1β protein expression were markedly decreased in the spinal cord, whereas IL-10, an anti-inflammatory cytokine, was dramatically increased in the EGCG-treated group [49]. Intrathecal EGCG also decreased mRNA expression of TLR4 and HMGB1, which are important mediators in the inflammatory response [49]. TLR4 is an important pattern recognition receptor, for which HMGB1 is an endogenous ligand that can further activate TLR4 leading to the release of TNF-α, NF-κB, and IL-1β [49].

Future Directions

The literature supporting flavonoids for the treatment of neuropathic pain is limited to animal-based models at this point. We have summarized the literature for four well-studied, representative flavonoids above. Future studies will need to evaluate the effects of these flavonoids in the human population, starting with an evaluation of its side effect profile, including interactions with other commonly used neuropathic medications. Furthermore, it is potentially valuable to conduct additional animal-based model studies on other less studied flavonoids, such as luteolin, rutin, and morin, for more alternatives in neuropathic pain treatment.

Conclusion

Neuropathic pain remains a challenging condition for patients and providers, due to inadequate treatment options and its multifactorial nature. Numerous animal-model studies have demonstrated the potential benefit of flavonoids in the treatment of neuropathic pain, and the reversal of hyperalgesia and allodynia in numerous models, such as diabetic neuropathy, chemotherapy-induced peripheral neuropathy, and peripheral nerve injury. The anti-inflammatory effect of flavonoids is further demonstrated by the reduction in multiple pro-inflammatory mediators, such as TNF-α, NF-κB, IL-1β, and IL-6, which have been implicated in the pathophysiology of neuropathic pain. Overall, flavonoids present a potentially new treatment modality for neuropathic pain in preclinical models, though human clinical evidence is yet to be explored at this time.

Declarations

Conflict of Interest Richard D. Urman reports funding or fees from Medtronic, Merck, Acacia, Takeda, Pfizer, and AcelRx. Amitabh Gulati is a consultant for Medtronic, Flowonix, SPR Therapeutics, Nalu Medical, Bausch Health, and advisor for AIS. The other authors declare that no competing interests exist.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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