Experimental Brain Research (2019) 237:2645–2651
https://doi.org/10.1007/s00221-019-05615-x

RESEARCH ARTICLE

Protective effect of ibuprofen in a rat model of chronic oxaliplatin-induced peripheral neuropathy

Thomas Krøigård[1,4] · Athanasios Metaxas[2] · Martin Wirenfeldt[3,4] · Bente Finsen[2]

Received: 23 December 2018 / Accepted: 26 July 2019 / Published online: 6 August 2019

Abstract

Despite extensive preclinical and clinical investigations, a clinically relevant neuroprotective agent against oxaliplatin-induced peripheral neuropathy, which affects the quality of life following chemotherapy, has not been identified. Epidemiological data suggest that ibuprofen may reduce the risk of neuropathy. Male rats were treated with oxaliplatin ($n=6$), oxaliplatin and ibuprofen ($n=5$) or vehicle ($n=5$) every second day for 15 days. Neuropathy was evaluated using mechanical detection thresholds (MDT) at the hind paw and sensory nerve conduction velocity (SNCV) in the tail nerve at baseline, right after and 3 weeks after the end of treatment. Intraepidermal nerve fibre density (IENFD) was evaluated in the hind paw and inflammation in the dorsal root ganglia 3 weeks after treatment. Inflammation in the dorsal root ganglia was assessed using quantitative real-time RT-PCR (qPCR) of the mRNA levels for the pro-inflammatory cytokines, TNF-α and IL-1β, and by immunohistochemical staining for Iba1$^+$ macrophages. SNCV was reduced in rats treated with oxaliplatin and with oxaliplatin and ibuprofen compared to control rats 3 weeks after treatment. No differences were found for MDT 3 weeks after treatment. IENFD was reduced in rats treated with oxaliplatin. There was a trend towards up-regulation of TNF-α mRNA levels in rats treated with oxaliplatin and with oxaliplatin and ibuprofen. Morphological changes of Iba1$^+$ macrophages suggested activation, but no differences were found in area fraction or size of macrophage cell bodies. The results did not support a neuroprotective effect of ibuprofen but indicated that inflammation may play a role in oxaliplatin-induced peripheral neuropathy.

Keywords Ibuprofen · Neuroprotection · Neuroinflammation · Oxaliplatin · Rat

Background

Oxaliplatin is a widely used chemotherapeutic agent in the treatment of colorectal cancer and other solid tumours. However, therapy is often complicated by chronic, sensory polyneuropathy, which affects the quality of life. Almost all of the patients also have symptoms of an acute polyneuropathy following each injection of oxaliplatin. The chronic polyneuropathy is primarily related to the cumulative dose of oxaliplatin (Gamelin et al. 2002), and is probably caused by accumulation of platinum in the dorsal root ganglia as demonstrated in rats exposed to oxaliplatin (Ta et al. 2006).

Xiao et al. (2012) found that oxaliplatin evokes mechanic allodynia, hyperalgesia, and cold allodynia, but has no effect on heat sensitivity. They found a significant loss of intraepidermal nerve fibres in skin biopsies from the hind paw 4 weeks after treatment. Sensory fibres, but not motor fibres were affected in nerve conduction studies. Signs of neuropathy peaked at 2.5–4 weeks after treatment, and had disappeared after 15 weeks. Loss of sensory neurons has not been demonstrated.

Peripheral nerve lesion is known to lead to an increase in the number of microglia in the dorsal horn (Beggs and Salter 2007), and in at rat model of paclitaxel-induced neuropathy, the number of CD68$^+$ activated macrophages was increased in the dorsal root ganglia and the sciatic nerve (Peters et al. 2007). Following a low dose of oxaliplatin, no evidence of microglial activation in the lumbar spinal cord was found,

✉ Thomas Krøigård
thomas.kroigard@rsyd.dk

1 Department of Neurology, Odense University Hospital, Odense, Denmark

2 Department of Neurobiology Research, Institute of Molecular Medicine, University of Southern Denmark, Odense, Denmark

3 Department of Pathology, Odense University Hospital, Odense, Denmark

4 Institute of Clinical Research, University of Southern Denmark, J. B. Winsløws Vej 4, 5000 Odense C, Denmark

but the dorsal root ganglia were not examined (Zheng et al. 2011).

As oxaliplatin-induced neuropathy represents a substantial clinical problem, a large number of interventions for preventing the condition have been tested in the clinical setting including amifostine, vitamin E, calcium and magnesium, glutathione, acetylcysteine, oxcarbazepine and others. A Cochrane review from 2011 concluded that the data are still insufficient (Albers et al. 2011).

In a retrospective study of 52 patients, treatment with non-steroidal anti-inflammatory drugs (NSAIDs) for more than 2 weeks prior to examination reduced the risk of neuropathy by 60% (Kanbayashi et al. 2010). It was not possible to determine whether the effect of the NSAIDs was neuroprotective or symptomatic due to lack of information regarding the exact temporal relationship with oxaliplatin treatment. Furthermore, neuropathy assessment was based on neuropathy symptoms only and not on objective measures of neuropathy.

Non-steroidal anti-inflammatory drugs may have a neuroprotective effect by inhibition of cyclooxygenase-2, which plays an important role in neuropathic pain by up-regulation of ion channels and cytokines (Ma and Quirion 2008). Up-regulation of cyclooxygenase-2 has been demonstrated in the rat dorsal horn at day 3 after nerve lesion in a model for neuropathic pain (O'Rielly and Loomis 2006).

The aims of the present study were to test the hypotheses that chronic oxaliplatin-induced neuropathy in the rat is related to inflammation in the dorsal root ganglia and that treatment with the NSAID ibuprofen can ameliorate chronic oxaliplatin-induced neuropathy, likely due to its anti-inflammatory action.

Materials and methods

Animals

Eighteen male Sprague–Dawley rats weighing approximately 450 g at the beginning of the experiment were housed in a self-contained, temperature-controlled unit, and set to 12 h dark–light cycles with access to food and water ad libitum. Approval to conduct the experiments was obtained from The Danish Animal Experiments Inspectorate (Case no. 2014-15-2934-01032) and they were performed in accordance with Directive 2010/63/EU on the protection of animals used for scientific purposes. Six rats (group A) were treated with oxaliplatin, six rats (group B) were treated with oxaliplatin and ibuprofen, and six rats (group C) served as controls. Rats were counterbalanced prior to treatment so that there were no between group differences according to weight. One rat in group B and one rat in group C died during treatment and were not included in the analysis.

Pharmacological treatments

Rats in groups A and B were treated with oxaliplatin (Hospira, Lake Forest, IL, USA) administered intraperitoneally (i.p.) at a dose of 4.5 mg/kg in a solution of 5% dextrose every second day for 15 days (a total of eight injections) to obtain a cumulative dose which is comparable with the dose known to induce chronic polyneuropathy in patients (Di Cesare Mannelli et al. 2015). Rats randomised to group B were also treated with ibuprofen (Merck KGaA, Darmstadt, Germany) administered subcutaneously at a dose of 60 mg/kg in a solution of 50% phosphate-buffered saline and 50% alcohol (70%) once a day for 19 days (from 2 days prior to initiation of oxaliplatin treatment to 2 days after treatment). A similar ibuprofen dosing regimen has previously attenuated post-hypoxic brain injury (Wixey et al. 2012). Control animals received equivalent volumes of vehicles. Rats were weight monitored once weekly for adjustment of drug doses and for monitoring of their general health status (Krohn et al. 2001).

Behavioural tests and nerve conduction

Animals were examined before treatment, 2 days after end of treatment, and 3 weeks after treatment. Testing for mechanical detection threshold was carried out while rats were sitting on an elevated metal rod flooring and after approximately 15 min of habituation. Von Frey hairs were applied to the hind paw with increasing pressure until the rat withdrew its paw. This was repeated twice after reduction of the paw pressure by two steps. The mechanical detection threshold was calculated as the geometric mean of the three measurements. The orthodromic sensory nerve conduction velocity in the tail nerve was assessed by stimulating distally and recording 5 and 10 cm proximal to stimulation using disposable sensory needle electrodes (Alpine Biomed ApS, Skovlunde, Denmark) for both stimulation and recording. A Dantec KeyPoint (Natus Medical Incorporated, San Carlos, CA, USA) was used for stimulation and recording. The latencies of the potentials recorded at the two sites (5 and 10 cm) were determined, and nerve conduction velocity calculated based on the difference between latencies as previously described (Leandri et al. 2006). During nerve conduction studies, rats were anaesthetized using fentanyl (0.315 mg/ml)/fluanisone (10 mg/ml) at a dose of 0.2–0.3 ml administered i.p. and cooling of the rat tail was prevented using a heating pad.

Cytokine mRNA evaluation

After the final nerve conduction recording, rats received a lethal injection of pentobarbital and they were perfused

transcardially with phosphate-buffered saline. Dorsal root ganglia (L5–L6) were dissected immediately and frozen. Messenger RNA levels of the pro-inflammatory cytokines TNF-α and IL-1β, which are elevated in the dorsal root ganglia 21 days after sciatic nerve lesion (Chamaa et al. 2016) were determined using quantitative real-time RT-PCR (qPCR) of pooled dorsal root ganglia (L5–L6). Primers were designed using the web-based NCBI Gene Bank. All primers were designed to span exon–exon junctions and sequence specificity was checked using NCBI Primer-BLAST. Primer sequences were: TNF-α: GGTGTGTGACGTTCCATT AGA (forward) and GTAGATAAGGTACAGCCCATCTG (reverse) and IL-1β: GGTGTGTGACGTTCCATTAGA (forward) and CATGGAGAATATCACTTGTTGGTTGA (reverse). Primers were purchased from TAG-Copenhagen A/S (Copenhagen, DK). RNA was extracted using TRIzol® (Invitrogen, Grand Island, NY, USA) and cDNA was synthesized using a High-Capacity cDNA Reverse Transcription Kit (Applied Biosystems, CA, USA) according to manufacturer's instructions. The qPCR was controlled by an Applied BioSystem iCycler and was performed using diluted cDNA, 400 nM primers and Maxima™ SYBR green Master (Life Technologies, Nærum, DK). The conditions for the PCR were as follows: 10 min at 95 °C, 40 cycles of 15 s (sec) at 95 °C and 1 min at the annealing temperature followed by 15 s at 95 °C, 1 min at 60 °C and finally 15 s at 95 °C for generation of melting curves. Samples were run against a standard curve generated from serially diluted cDNA from the control group (group C). After normalization to HPRT mRNA (forward: GTTAAGCAGTACAGCCCCAAAATG, reverse: AAATCCAACAAAGTCTGGCCTGTA), the relative qPCR data for each cytokine mRNA were expressed as fold changes relative to the mean of the control group which was set to one.

Immunohistochemistry

To evaluate the morphology of macrophages, dorsal root ganglia L5–L6 were dissected as described above from the contralateral side and frozen in an embedding medium (Killik, Bio-Optica, Milan, Italy) immediately after dissection. They were sectioned at a thickness of 16 µm and immersion fixed in 4% paraformaldehyde for 24 h at 4 °C and for 24 h at 30 °C. Endogenous peroxidase activity was blocked using H_2O_2 and sections were incubated with 0.25% freshly prepared sodium borohydride (NaBH4) to reduce unreactive aldehydes and Schiff bases. They were incubated with a polyclonal rabbit-anti-Iba-1 antibody (Fujifilm WakoPure Chemical Corporation, Osaka, Japan, Cat no 019-19741) at a concentration of 0.1 µg/mL for 24 h at room temperature. The primary antibody was visualized using EnVision (Agilent Technologies, Inc., Santa Clara, CA, USA) and diaminobenzidine following incubation with imidazole and 0.5% nickel ammonium hexahydrate. Rabbit immunoglobulin (IgG) (Agilent Technologies Inc, Santa Clara, CA, USA, Cat no X0903) was used as a substitution control in the same concentration as used for the primary antibody.

Macrophage Iba1 immunoreactivity was quantified in a blinded manner using the particle analysis plugin in ImageJ (v1.51s; National Institutes of Health, MD, USA). Photomicrographs were acquired with an Olympus DP71 digital camera, mounted on an Olympus BX51 microscope. The % area of Iba1 immunoreactivity was measured in at least six sections/animal. The shortest diameter of five randomly selected Iba-1^+ cells/section was also measured.

For the estimation of intraepidermal nerve fibre density (IENFD), 3 mm skin biopsies from the hind paw were fixed overnight in a 10% formalin buffer, sectioned at a thickness of 50 µm and stained as free floating sections using a PGP9.5 primary antibody (Bio-Rad Laboratories, product code MCA4750GA). Intraepidermal nerve fibre density was defined as the number of nerve fibres crossing the dermoepidermal junction divided by the total length of the surface of the biopsies.

Statistics

Data are presented graphically as mean ± SE or in dot diagrams with indications of means. Statistical analysis is performed with one- or two-way analysis of variance (ANOVA) followed by a Tukey post hoc analysis, using Prism 4.0 software (GraphPad Software). One data point identified as an outlier in dataset for IL-1β mRNA by ROUT analysis was excluded from the analysis. Changes were considered statistically significant if $p < 0.05$ and indicated as follows: $^*p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$; $^{****/\#\#\#\#}p < 0.0001$.

Results

Body weight is impacted by time and treatment

Body weight monitoring was performed to assess the general health status of animals. Two-way ANOVA revealed a significant effect both of time ($[F_{(2,39)} = 12.80, p < 0.001]$ and treatment $[F_{(2,39)} = 10.92, p < 0.001]$, and an interaction between time and treatment $[F_{(4,39)} = 4.47, p < 0.01]$, on body weight during the 5-week study period (Fig. 1a). As expected, there was a significant increase in body weight during this period in control animals (Fig. 1a, $p < 0.0001$, Tukey's test). No increase was seen in groups of rats treated with oxaliplatin and with oxaliplatin and ibuprofen which both had significantly lower weight compared to control rats 3 weeks after treatment (Fig. 1a, $p < 0.001$ and $p < 0.01$), with no difference between rats treated with oxaliplatin and with oxaliplatin and ibuprofen (Fig. 1a, $p = 0.99$).

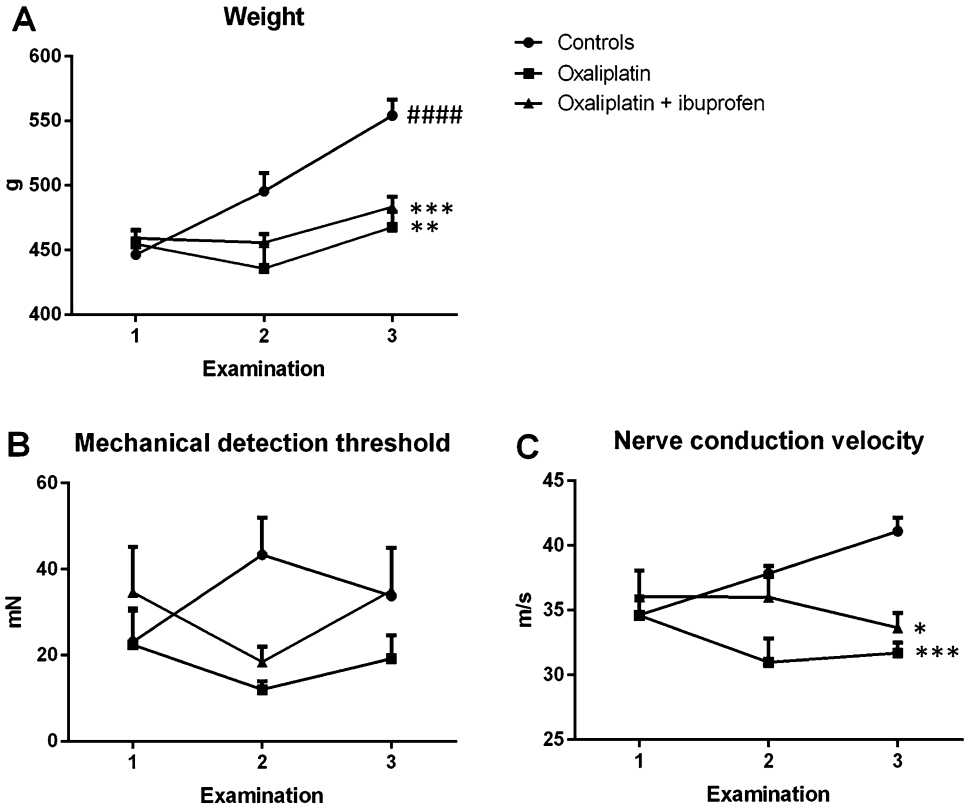

Fig. 1 Weight (**a**), mechanical detection threshold (**b**), and sensory nerve conduction velocity (**c**) in the tail nerve before treatment start (examination 1), 2 days after end of treatment (examination 2), and 3 weeks after end of treatment (examination 3). Mean + SEM. Data were analysed by two-way ANOVA followed by Tukey post hoc test $^{\#\#\#\#}p < 0.0001$ (compared to examination 1) $^{*}p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$ (comparison of experimental group(s) to control group at examination 3)

Effect of treatment on mechanical detection threshold and nerve conduction velocity

Interestingly, treatment was found to have a significant effect on MDT ($[F_{(2,39)} = 3.35, p < 0.05]$, two-way ANOVA), whereas time had no effect $[F_{(2,39)} = 0.28, p = 0.76]$, and there was also no effect on the interaction between treatment and time on MDT $[F_{(4,39)} = 1.60, p = 0.19]$, and no statistically significant differences could be detected in the Tukey post hoc analysis (Fig. 1b). Treatment was also found to have a significant effect on SNCV $[F_{(2,37)} = 10.51, p < 0.001]$, whereas time had no effect $[F_{(2,37)} = 0.11, p = 0.90]$. Also in the case of SNCV, there was a significant effect on the interaction between treatment and time on the nerve conduction $[F_{(4,37)} = 3.97, p < 0.01]$. The post hoc analysis showed that rats treated with oxaliplatin or with oxaliplatin and ibuprofen both had significantly lower nerve conduction velocity compared to control rats at 3 weeks after treatment (Fig. 1c, $p < 0.001$ and $p < 0.05$, respectively), with no difference between rats treated with oxaliplatin and with oxaliplatin and ibuprofen (Fig. 1c, $p = 0.98$).

Intraepidermal nerve fibre density is impacted by treatment

Treatment had a statistically significant effect on the IENFD $([F_{(2, 13)} = 10.21, p < 0.01]$, one-way ANOVA). As expected, rats treated with oxaliplatin had a significantly lower IENFD than control animals (Fig. 2, $p < 0.01$, Tukey post hoc). There were no significant differences between rats treated with oxaliplatin and rats treated with oxaliplatin and ibuprofen or controls rats (Fig. 3, $p > 0.05$, both comparisons).

Effect of treatment on inflammation in the DRG

There was a trend towards up-regulation of TNF-α mRNA levels in the two treatment groups $([F_{(2,12)} = 3.04, p = 0.08]$, one-way ANOVA), but no statistically significant differences in IL1-β mRNA levels $[F_{(2,11)} = 0.43, p = 0.54]$ in dorsal root ganglia 3 weeks after treatment, and no significant differences between treatment groups (Fig. 3a, b, $p > 0.05$ for all comparisons, Tukey post hoc). $Iba1^+$ macrophages with enlarged cell bodies and processes encircling the neuronal cell bodies were observed in dorsal root ganglia of rats treated with oxaliplatin and with oxaliplatin and ibuprofen, which suggested macrophage activation (Fig. 3c–e). However, one-way ANOVA showed no effects of treatment on the area fraction for macrophage Iba1 immunoreactivity $[F_{(2,13)} = 0.56, p = 0.58]$ (Fig. 3f). Although differences in the diameter (smallest diameter) of $Iba1^+$ macrophage cell bodies in the oxaliplatin vs. control (Cohen's $d = 0.92$) and oxaliplatin vs. oxaliplatin + ibuprofen (Cohen's $d = 0.86$) groups were consistent with large effect sizes, one-way ANOVA

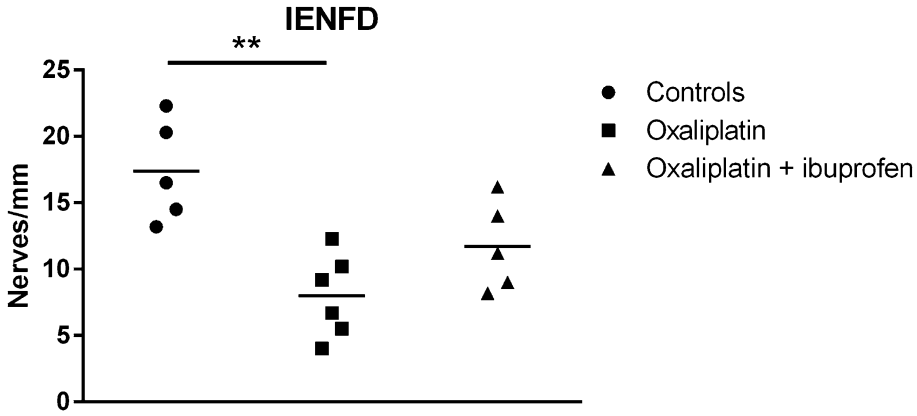

Fig. 2 Intraepidermal nerve fibre density (IENFD) in skin biopsies from the hind paw. Horizontal bars indicate means. Data were analysed by one-way ANOVA followed by Tukey post hoc test $^{**}p < 0.01$

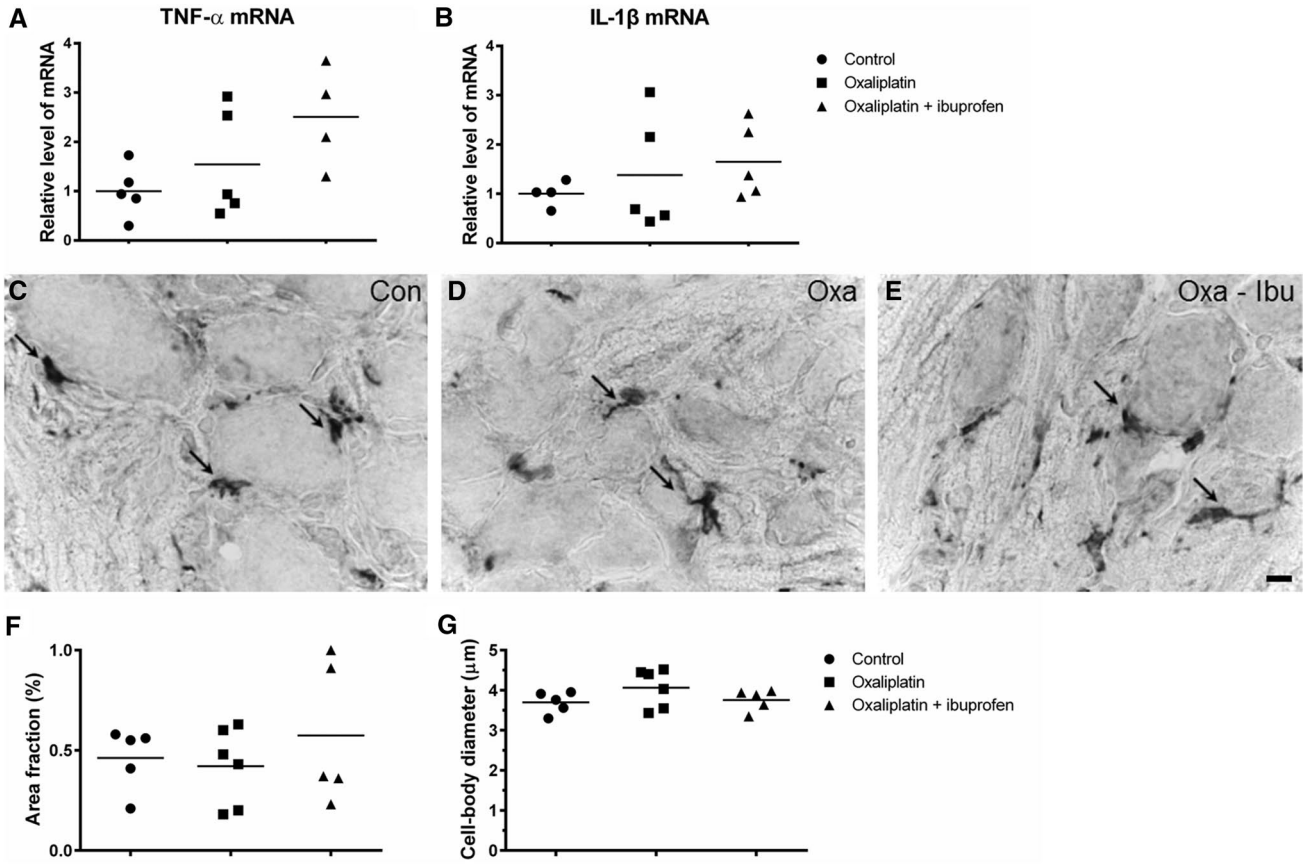

Fig. 3 Effect of treatment on inflammation. **a**, **b** Relative levels of TNF-α (**a**) and IL-1β (**b**) mRNA 3 weeks after end of treatment. Horizontal bars indicate means. Data were analysed by one-way ANOVA followed by Tukey post hoc test, which showed no differences between groups. **c**–**e** Photomicrographs of Iba1$^+$ macrophages (arrows) in dorsal root ganglia from control rats (**c**) and rats treated with oxaliplatin (**d**) and oxaliplatin and ibuprofen (**e**). In oxaliplatin-treated rats (**d**, **e**) Iba1$^+$ macrophages had enlarged cell bodies and processes encircling cell bodies of sensory neurons forming ring-like structures. Scale bar: 10 µm. **f**, **g** Quantification of macrophage activation given by the area fraction of Iba1 immunoreactivity (**f**) and the diameter of the Iba1$^+$ macrophage cell bodies (**g**). One-way ANOVA showed no differences between groups

showed no effect of treatment on the diameter of Iba-1$^+$ macrophage cell bodies [$F_{(2,13)} = 1.65$; $p = 0.23$] (Fig. 3g). Iba1$^+$ macrophage cell body diameters were 3.70 ± 0.12, 4.06 ± 0.20 and 3.76 ± 0.12 µm for control rats, and rats treated with oxaliplatin and oxaliplatin and ibuprofen, respectively.

Discussion

There were some indications of an inflammatory response in rats treated with oxaliplatin regarding morphological changes in macrophages and a trend towards an increase in TNF mRNA but not IL-1β mRNA levels in the dorsal root ganglia, however, with no effect of ibuprofen. We were not able to demonstrate a neuroprotective effect of ibuprofen treatment in rats during oxaliplatin treatment, which was in line with the functional data on detection threshold and nerve conduction. The exact mechanisms responsible for oxaliplatin-induced peripheral neuropathy have not been established, and other possibilities such as mitochondrial dysfunction have been proposed (Canta et al. 2015).

Our results do not exclude that inflammation is the primary process responsible for oxaliplatin-induced peripheral neuropathy. Morphological changes similar to those observed in this study were found in the dorsal root ganglia as a part of an immunological response following peripheral nerve injury (Vega-Avilaira et al. 2009). It is possible that the failure to show clear up-regulation of TNF-α and IL1-β mRNA is related to the relatively long time period from cessation of treatment to cytokine evaluation. A recent study, examining glial activation found evidence of inflammation in the spinal cord and brain, but not in peripheral nerves or dorsal root ganglia following oxaliplatin treatment (Di Cesare Mannelli et al. 2013). Nevertheless, the trend towards an increase in TNF mRNA levels deserves attention, since increase in TNF mRNA levels is traditionally a good sensor of chronic pathology (Babcock et al. 2015). Also, celecoxib, another NSAID, reduced pain in a mouse model of oxaliplatin-induced neuropathy (Di Cesare Mannelli et al. 2017a).

There are several limitations in this study. First, our estimates of IENFD are approximately half of those previously reported in oxaliplatin-treated rats (Xiao et al. 2012), suggesting that we were not able to identify all intraepidermal nerve fibres. There may be several reasons, including suboptimal immunohistochemical detection due to, e.g., lack of penetration of the primary PGP9.5 antibody through the thick sections and inter-observer differences in visual evaluation of sections. However, IENFD similar to our findings in untreated rats have also been reported (Harte et al. 2017; Melli et al. 2006), which supports the validity of the present results and points to the possibility of a general lack of reproducibility for this measure. Second, we were not able to reproduce previously demonstrated changes in mechanical detection thresholds (Xiao et al. 2012). In a recent study (Di Cesare Mannelli et al. 2017b), however, only noxious stimuli, and not the non-noxious stimuli applied in the present study were impacted by the oxaliplatin treatment. Consequently, we were not able to assess the potential protective effect of ibuprofen regarding behavioural outcomes. It is also possible that subcutaneous administration of ibuprofen once daily was not sufficient to provide an acceptable level of ibuprofen during the study period. Continuous administration, e.g., via a mini-pump improving pharmacokinetics may have resulted in different outcomes. Finally, the relatively low number of animals included in the analysis limits the interpretation of results.

In conclusion, there is a need for further investigations into the pathophysiology of oxaliplatin-induced neuropathy and strategies for neuroprotection.

Funding The study was supported financially by Jacob og Olga Madsens fond.

Compliance with ethical standards

Conflict of interests The authors have no conflicts of interests to disclose.

References

Albers JW, Chaudhry V, Cavaletti G, Donehower RC (2011) Interventions for preventing neuropathy caused by cisplatin and related compounds. Cochrane Database Syst Rev 16:CD005228

Babcock AA, Ilkjær L, Clausen BH, Villadsen B, Dissing-Olesen L, Bendixen AT, Lyck L, Lambertsen KL, Finsen B (2015) Cytokine-producing microglia have an altered beta-amyloid load in aged APP/PS1 Tg mice. Brain Brain Behav Immun 48:86–101.

Beggs S, Salter MW (2007) Stereological and somatotopic analysis of the spinal microglial response to peripheral nerve injury. Brain Behav Immun 21:624–633

Canta A, Pozzi E, Carozzi VA (2015) Mitochondrial dysfunction in chemotherapy-induced peripheral neuropathy (CIPN). Toxics 3:198–223

Chamaa F, Chebaro M, Safieh-Garabedian B, Saadeh R, Jabbur SJ, Saadé NE (2016) Transcriptional expression of inflammatory mediators in various somatosensory relay centers in the brain of rat models of peripheral mononeuropathy and local inflammation. J Neuroimmunol 297:81–91

Di Cesare Mannelli L, Pacini A, Bonaccini L, Zanardelli M, Mello T, Ghelardini C (2013) Morphologic features and glial activation in rat oxaliplatin-dependent neuropathic pain. J Pain 14:1585–1600

Di Cesare Mannelli L, Pacini A, Corti F, Boccella S, Luongo L, Esposito E, Cuzzocrea S, Maione S, Calignano A, Ghelardini C (2015) Antineuropathic profile of *N*-palmitoylethanolamine in a rat model of oxaliplatin-induced neurotoxicity. PLoS One 10:e0128080

Di Cesare Mannelli L, Lucarini E, Micheli L, Mosca I, Ambrosino P, Soldovieri MV, Martelli A, Testai L, Taglialatela M, Calderone V, Ghelardini C (2017a) Effects of natural and synthetic isothiocyanate-based H_2S-releasers against chemotherapy-induced neuropathic pain: role of Kv7 potassium channels. Neuropharmacology 121:49–59

Di Cesare Mannelli L, Maresca M, Micheli L, Farina C, Scherz MW, Ghelardini C (2017b) A rat model of FOLFOX-induced neuropathy: effects of oral dimiracetam in comparison with duloxetine and pregabalin. Cancer Chemother Pharmacol 80:1091–1103

Gamelin E, Gamelin L, Bossi L, Quasthoff S (2002) Clinical aspects and molecular basis of oxaliplatin neurotoxicity: current management and development of preventive measures. Semin Oncol 29:21–33

Harte SE, Clauw DJ, Hayes JM, Feldman EL, St Charles IC, Watson CJ (2017) Reduced intraepidermal nerve fiber density after a sustained increase in insular glutamate: a proof-of-concept study examining the pathogenesis of small fiber pathology in fibromyalgia. Pain Rep 2:e590

Kanbayashi Y, Hosokawa T, Okamoto K et al (2010) Statistical identification of predictors for peripheral neuropathy associated with administration of bortezomib, taxanes, oxaliplatin or vincristine using ordered logistic regression analysis. Anticancer Drugs 21:877–881

Krohn TC, Hejgaard K, Hansen AK (2001) Methods for general assessment of the welfare of laboratory rats. Acta Agric Scand Sect A Anim Sci 30:118–123

Leandri M, Saturno M, Cilli M, Bisaglia M, Lunardi G (2006) Compound action potential of sensory tail nerves in the rat. Exp Neurol 203:145–157

Ma W, Quirion R (2008) Does COX2-dependent PGE2 play a role in neuropathic pain? Neurosci Lett 437:165–169

Melli G, Jack C, Lambrinus GL, Ringkamp M, Höke A (2006) Erythropoietin protects sensory axons against paclitaxel-induced distal degeneration. Neurobiol Dis 24:525–530

O'Rielly DD, Loomis CW (2006) Increased expression of cyclooxygenase and nitric oxide isoforms, and exaggerated sensitivity to prostaglandin E2, in the rat lumbar spinal cord 3 days after L5–L6 spinal nerve ligation. Anesthesiology 104:328–337

Peters CM, Jimenez-Andrade JM, Jonas BM et al (2007) Intravenous paclitaxel administration in the rat induces a peripheral sensory neuropathy characterized by macrophage infiltration and injury to sensory neurons and their supporting cells. Exp Neurol 203:42–54

Ta LE, Espeset L, Podratz J, Windebank AJ (2006) Neurotoxicity of oxaliplatin and cisplatin for dorsal root ganglion neurons correlates with platinum-DNA binding. Neurotoxicology 27:992–1002

Vega-Avilaira D, Géranton SM, Fitzgerald M (2009) Differential regulation and macrophage/neuron interactions in the dorsal root ganglion in young and adult rats following nerve injury. Molecular Pain 5:70

Wixey JA, Reinebrant HE, Buller KM (2012) Post-insult ibuprofen treatment attenuates damage to the serotonergic system after hypoxia-ischemia in the immature rat brain. J Neuropathol Exp Neurol 71:1137–1148

Xiao WH, Zheng H, Bennett GJ (2012) Characterization of oxaliplatin-induced chronic painful peripheral neuropathy in the rat and comparison with the neuropathy induced by paclitaxel. Neuroscience 203:194–206

Zheng FY, Xiao WH, Benett GJ (2011) The response of spinal microglia to chemotherapy-evoked painful peripheral neuropathies is distinct from that evoked by traumatic nerve injuries. Neuroscience 176:447–454