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# Synthesis and biological evaluation of novel dimiracetam derivatives useful for the treatment of neuropathic pain

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**Abstract**—Chemical modifications of dimiracetam, a bicyclic analogue of the nootropic drug piracetam, afforded a small set of novel derivatives that were investigated in in vivo models of neuropathic pain. Compounds **5**, **7** and **8** displayed a very promising antihyperalgesic profile in rat models of neuropathic pain induced by both chronic constriction injury of the sciatic nerve and streptozotocin. The compounds completely reverted the reduction of pain threshold evaluated by the paw pressure test. Importantly these derivatives did not induce any behavioural impairment as evaluated by the rotarod test. These results suggest that compounds **5**, **7** and **8** might represent novel and well-tolerated therapeutic agents for the relief of neuropathic pain.


## 1. Introduction

Neuropathic pain (NeP) is defined as chronic, persistent pain mostly caused by peripheral or central neural injury.[1] Unlike physiological pain it serves no useful purpose and is usually sustained and chronic. The characteristic symptoms of neuropathic pain are usually expressed as allodynia, hyperalgesia and spontaneous pain. The majority of the currently available treatments for NeP are not adequate: tricyclic antidepressants, anticonvulsants and opioids are only partially effective and/or are associated with significant side effects.[2] Even gabapentin, the gold standard for the treatment of NeP, reduces pain by 30–50% at best in less than 50% of patients.[3] Therefore, safer and more effective treatments for neuropathic pain need to be developed and significant efforts must be applied to the discovery of novel drug molecules able to alleviate this intractable pain.

Nootropic drugs comprise a series of derivatives of γ-aminobutyric acid (GABA) whose prototype, piracetam, is used in diseases characterized by learning and memory deficits (Fig. 1).[4] Recent studies have shown

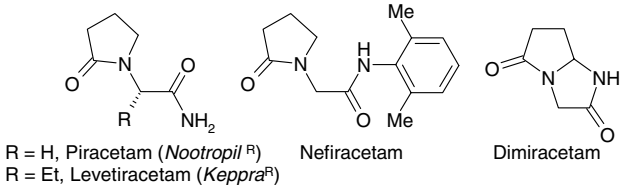


**Figure 1.** Nootropic drugs.

that some nootropic drugs such as nefiracetam or levetiracetam may be useful in treating neuropathic pain both in animals and in patients.

The validation of nootropic pyrrolidinones in neuropathic pain is noteworthy. For example, nefiracetam, a Piracetam derivative, featuring a lipophilic anilide moiety, dose dependently reversed the thermal or mechanical hyperalgesia induced by partial sciatic nerve ligation or streptozotocin treatment in mice.[5] Levetiracetam, an anticonvulsant drug targeting the synaptic vesicle protein SV2A, was recently shown to be effective in patients with chronic neuropathic pain, particularly when treatment with other anticonvulsant agents failed.[6]

The analgesic action induced by this class of derivatives was non-opioid in nature, as it was not reversed by the

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opioid antagonist naloxone. Together, these findings suggest that nefiracetam, and other cognition enhancers of the same pyrrolidinone nootropic class, could be good therapeutic tools against neuropathic pain, having no liabilities due to possible involvement of the opioid system, and an exceptional safety profile.[7]

Thus, taking nefiracetam as the model drug, we continued the research in this field with the aim of finding compounds endowed with increased potency, efficacy and with a good safety profile. Dimiracetam,[8] a bicyclic pyrrolidinone analogue of piracetam, was developed as a novel cognition enhancer until Phase I, before its development was discontinued. This compound, which maintains the backbone of piracetam with the acetamide side chain restricted in a folded conformation, was 10–100 times more potent than piracetam and, as for the other related derivatives, practically devoid of toxicity.

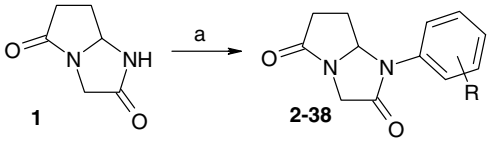
In the present study, a series of variously substituted dihydro-1*H*-pyrrolo[1,2-*a*]imidazole-2,5(3*H*,6*H*)-diones **2**–**38**, derivatives of **1**, were prepared and their antihyperalgesic effect was investigated in several models of neuropathic pain in rats.

## 2. Chemistry

Substituted aromatic rings were inserted into the dimiracetam structure through a microwave-enhanced Goldberg reaction, allowing the synthesis of a small set of 37 compounds (Scheme 1).[9]

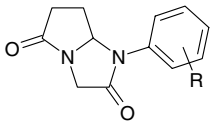
Microwave irradiation greatly accelerates this copper-catalysed arylation of the secondary amide. Compounds **2**–**38** (Table 1) were quickly obtained by irradiation of a mixture of dimiracetam and the corresponding aryl iodides or bromides, using NMP as solvent, in the presence of CuI as catalyst, in a microwave reactor at 150 °C for about 20 min. Notwithstanding the moderate yields obtained (ranging from 20% to 60%), the described procedure is time and energy-saving affording substantial amounts of the desired derivatives in a short timeframe. Moreover, after reaction work-up, the final compounds were obtained without any additional purification and with purities sufficient for in vivo biological screening.[10]

As during the continuation of our study, larger amounts of selected compounds were required for biological and toxicological studies, scaling-up of the Goldberg reaction was achieved by using DMF as a solvent and warming the reactions by conventional heating. The yields obtained using oil-bath heating were comparable, while




**Scheme 1.** (a) ArI or ArBr, CuI, $K_2CO_3$, NMP, MW, 150 °C, 20 min.

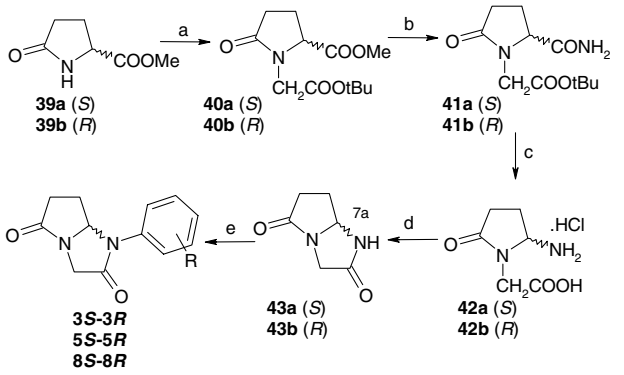
**Table 1.** Chemical and physical characteristics of compounds **2**–**38**




| Compound | R | Mp (°C) | Elemental analysis |
|---|---|---|---|
| **2** | H | 185–188 | $C_{12}H_{12}N_2O_2$ |
| **3** | 2-Me | 138–139 | $C_{13}H_{14}N_2O_2$ |
| **4** | 3-Me | 92–93 | $C_{13}H_{14}N_2O_2$ |
| **5** | 4-Me | 117–118 | $C_{13}H_{14}N_2O_2$ |
| **6** | 2-CN | 94–95 | $C_{13}H_{11}N_3O_2$ |
| **7** | 3-CN | 128–130 | $C_{13}H_{11}N_3O_2$ |
| **8** | 4-CN | 175–176 | $C_{13}H_{11}N_3O_2$ |
| **9** | 2-$CF_3$ | 95–97 | $C_{13}H_{11}F_3N_2O_2$ |
| **10** | 3-$CF_3$ | 124–125 | $C_{13}H_{11}F_3N_2O_2$ |
| **11** | 4-$CF_3$ | 124–125 | $C_{13}H_{11}F_3N_2O_2$ |
| **12** | 3-F | 148–149 | $C_{12}H_{11}FN_2O_2$ |
| **13** | 4-F | 158–159 | $C_{12}H_{11}FN_2O_2$ |
| **14** | 3-Cl | 123–125 | $C_{12}H_{11}ClN_2O_2$ |
| **15** | 4-Cl | 159–160 | $C_{12}H_{11}ClN_2O_2$ |
| **16** | 3-*i*-Pr | 81–82 | $C_{15}H_{18}N_2O_2$ |
| **17** | 4-*i*-Pr | 124–125 | $C_{15}H_{18}N_2O_2$ |
| **18** | 3-OMe | 94–95 | $C_{13}H_{14}N_2O_3$ |
| **19** | 4-OMe | 210–211 | $C_{13}H_{14}N_2O_3$ |
| **20** | 3-OH | 210–211 | $C_{12}H_{12}N_2O_3$ |
| **21** | 4-$SO_2$Me | 143–145 | $C_{13}H_{14}N_2O_4S$ |
| **22** | 4-Et | 98–99 | $C_{14}H_{16}N_2O_2$ |
| **23** | 4-$CH_2$OH | 159–161 | $C_{13}H_{14}N_2O_3$ |
| **24** | 4-COOEt | 158–159 | $C_{15}H_{16}N_2O_4$ |
| **25** | 4-COOH | 252–253 | $C_{13}H_{12}N_2O_4$ |
| **26** | 3,5-diF | 200 | $C_{12}H_{10}F_2N_2O_2$ |
| **27** | 3,4-Me | 142–143 | $C_{14}H_{16}N_2O_2$ |
| **28** | 3,5-Me | 103–104 | $C_{14}H_{16}N_2O_2$ |
| **29** | 2-F-5-Me | 96–97 | $C_{13}H_{13}FN_2O_2$ |
| **30** | 3-F-4-Me | 135–137 | $C_{13}H_{13}FN_2O_2$ |
| **31** | 2-Me-3-F | 135–137 | $C_{13}H_{13}FN_2O_2$ |
| **32** | 2-Me-3-Cl | 142–144 | $C_{13}H_{13}ClN_2O_2$ |
| **33** | 2-Me-4-F | 105–107 | $C_{13}H_{13}FN_2O_2$ |
| **34** | 2-Me-4-Cl | 138–139 | $C_{13}H_{13}ClN_2O_2$ |
| **35** | 2-Me-5-F | 147–148 | $C_{13}H_{13}FN_2O_2$ |
| **36** | 3-Me-4-F | 107–109 | $C_{13}H_{13}FN_2O_2$ |
| **37** | 3-Me-4-Cl | 138–139 | $C_{13}H_{13}ClN_2O_2$ |
| **38** | 3-Me-5-F | 111–113 | $C_{13}H_{13}FN_2O_2$ |

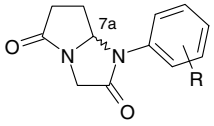
the reaction times were much longer (i.e., 5 h was necessary to get complete conversion into compound **5** vs 20 min of the MW irradiation).

Both enantiomers of the three selected compounds were synthesized in order to investigate the potential presence of a more active eutomer (Scheme 2). (*S*) and (*R*) methyl 5-oxo-pyrrolidin-2-carboxylate were used as enantiopure precursors and were alkylated with *tert*-butyl-bromoacetate to give, respectively, compounds **40a** and **40b**. Transamidation with ammonia afforded the primary amides that were readily converted into the corresponding amines by Hofmann-type rearrangement with the hypervalent iodine compound iodosobenzene–bistrifluoroacetate.[11] Acidic hydrolysis of *tert*-butyl esters and final cyclizations gave the (+)-(7*aS*) and (−)-(7*aR*) dimiracetam (**43a**, $[\alpha]_D$ + 39.9 ($c$ = 0.4, MeOH), and **43b** $[\alpha]_D$ = −39.2 ($c$ = 0.4, MeOH), respectively).




**Scheme 2.** (a) NaH, $BrCH_2COOtBu$; (b) 32% $NH_4OH$; (c) PIFA then TFA, HCl; (d) $Ac_2O$, AcONa and then $H^+$; (e) ArI or ArBr, CuI, $K_2CO_3$, NMP.

**Table 2.** Chemical and physical characteristics of compounds **3**–**5** (*R* and *S* pure enantiomers)




| Compound | 7a | R | Mp (°C) | $[\alpha]_D$ in MeOH | Elemental analysis |
|---|---|---|---|---|---|
| **3S** | S | 2-Me | 138–139 | −51.3 (*c* = 0.4) | $C_{13}H_{14}N_2O_2$ |
| **3R** | R | | 138–139 | +52.2 (*c* = 0.4) | $C_{13}H_{14}N_2O_2$ |
| **5S** | S | 4-Me | 146–148 | −134.3 (*c* = 0.4) | $C_{13}H_{14}N_2O_2$ |
| **5R** | R | | 144–146 | +134.0 (*c* = 0.4) | $C_{13}H_{14}N_2O_2$ |
| **8S** | S | 4-CN | 208–210 | −133.2 (*c* = 0.3) | $C_{13}H_{11}N_3O_2$ |
| **8R** | R | | 184–186 | +133.7 (*c* = 0.3) | $C_{13}H_{11}N_3O_2$ |

Finally, compounds **3**–**5** (*R* and *S* pure enantiomers), whose chemical and physical characteristics are reported in Table 2, were synthesized under the well-established experimental condition described before.

## 3. Pharmacology

The lack of a well-known mechanism of action for racetam derivatives prevents primary in vitro screening and the selection of active compounds generally relies on in vivo testing. All the compounds prepared were tested on the Bennett and Xie[12] model of neuropathic pain after i.c.v. injection. This route of administration was preferred to obtain a 'pure' measurement of efficacy, avoiding the influence of other parameters that could prevent an active compound from being detected (e.g., poor absorption and/or poor blood brain barrier penetration). The neuropathy was induced by right sciatic nerve ligation. Fourteen days after the operation a significant reduction of the pain threshold was established in the injured paw. By contrast, in the contralateral paw, the pain perception remained unchanged. Paw withdrawal threshold was measured using a Randall and Selitto apparatus exerting a force that increases at constant rate (32 g/s). Stimulus at which rats withdrew the paw was recorded before treatment and after drug administration (100 μg/kg, i.c.v.) at different times (30, 45 and 60 min). Based on their initial efficacy, the most promising compounds were tested further at lower concentrations and the results obtained for the minimal dose giving a statistical significant effect ($p < 0.01$) are reported in Table 3. The results represent means ± SEM of the mechanical threshold expressed as grams. Compounds were tested in different experiments. In vivo testing facilitates the selection of the compounds with the most promising clinical properties but does not allow establishing robust structure–activity relationships because the resulting activity is a consequence of the overall compound properties that can be significantly modulated even by small structural modifications. Seven derivatives showed good antihyperalgesic activity. Compounds **4**, **5**, **8** (10 μg/kg, i.c.v.) and **2**, **3**, **7** and **16** (30 μg/kg, i.c.v.) exhibited a dose dependent antihyperalgesic effect when compared to the animal group treated with saline. The pain threshold in contralateral non-operated paw was not modified in all the experiments. Importantly, at the dose of 10 μg/kg (30–45 min post-dosing), compounds **4**, **5** and **8** did increase the pressure threshold of the lesioned paw to the same level of the contralateral paw and were about 3-fold more effective than gabapentin or nefiracetam used as reference standards.

Since all the tested derivatives were racemic, both enantiomers of **3**, **5** and **8** were synthesized. All of these compounds displayed similar efficacy to the racemate in the animal models of neuropathic pain (data not shown), suggesting that the enantioselection is not important for the mechanism of action. We then focused our attention on compounds **5**, **7** and **8** which were tested on nerve injury-induced hyperalgesia in the rat paw pressure test after po administration (Fig. 2). Compound **5** increased the pressure threshold of the lesioned paw to the same level of the contralateral paw at a dose of 30 mg/kg po for 30–60 min post-dosing but not at a dose of 10 mg/kg. Compounds **7** and **8** produced a statistically significant antihyperalgesic effect at doses of 30 and 50 mg/kg po, respectively. At the highest dose tested (100 mg/kg po), both compounds completely reverted hyperalgesia (30–60 min post-treatment). All the compounds tested did not induce any significant effects on the mechanical threshold of the non-lesioned paw at all the doses and times tested.

In rodents the administration of the pancreatic toxin streptozotocin causes both mechanical and thermal hyperalgesia, possibly by mimicking diabetic neuropathy. Rats were injected ip with 50 mg/kg streptozotocin and tested for mechanical hyperalgesia in the paw pressure test 21 days after toxin treatment. In this model, compound **5** produced a statistically significant increase of the paw mechanical threshold at 50 and 100 mg/kg after oral administration (Fig. 3. At 100 mg/kg its efficacy was comparable to gabapentin given at the same dose). In the same model, orally administered compounds **7** and **8** (30–100 mg/kg), increased in a dose dependent manner the paw pressure threshold reaching statistical significance at 100 mg/kg with slightly reduced efficacy compared to that of gabapentin (data not shown). The compounds did not induce any significant effect in control animals. Furthermore, in the range of

**Table 3.** Antihyperalgesic effect of **1**–**5**,**7**, **8** and **16** in a rat model of neuropathic pain evaluated in paw pressure test

| Compound | R | Minimal effective dose μg/kg (i.c.v.) | Paw | Paw pressure in rats | | | |
|---|---|---|---|---|---|---|---|
| | | | | Before treatment | After analgesic treatment | | |
| | | | | | 30 min | 45 min | 60 min |
| Vehicle | — | | Contra | 59.1 ± 3.7 | 62.6 ± 4.8 | 57.3 ± 3.3 | 59.4 ± 4.4 |
| | | | Les | 29.3 ± 3.5 | 27.1 ± 4.2 | 25.9 ± 5.1 | 28.8 ± 3.9 |
| Gabapentin | — | 30 | Les | 24.5 ± 4.7 | 46.3 ± 4.2* | 43.5 ± 6.1* | 46.1 ± 5.5* |
| Levetiracetam | — | 500 | Les | 22.4 + 3.5 | 51.8 ± 6.8* | 52.1 ± 4.9* | 49.3 ± 4.2* |
| Vehicle | | | Contra | 53.2 + 3.2 | 53.4 + 4.7 | 51.9 + 5.5 | 56.4 + 5.1 |
| | | | Les | 22.1 ± 3.9 | 23.7 ± 4.4* | 27.6 ± 4.5* | 28.5 ± 4.5* |
| Nefiracetam | | 30 | Les | 23.6 ± 4.1 | 46.7 ± 4.5* | 49.5 ± 5.2* | 47.6 ± 5.0* |
| **1** (dimiracetam) | | 100 | Les | 25.1 ± 2.4 | 41.0 ± 4.5* | 42.8 ± 2.5* | 39.2 ± 2.5* |
| **2** | H | 30 | Les | 27.6 ± 3.5 | 51.7 ± 5.2* | 52.7 ± 4.6* | 53.3 ± 3.2* |
| **3** | 2-Me | 30 | Les | 24.2 ± 2.8 | 46.7 ± 5.1* | 55.3 ± 6.3* | 54.8 ± 3.6* |
| Vehicle | — | | Contra | 60.1 ± 3.8 | 57.4 ± 4.7 | 62.2 ± 4.7 | 58.4 ± 4.4 |
| | | | Les | 28.6 ± 3.1 | 25.2 ± 2.8 | 28.3 ± 3.5 | 33.2 ± 4.2 |
| **4** | 3-Me | 10 | Les | 28.5 ± 3.5 | 59.3 ± 5.3* | 63.4 ± 6.0* | 51.2 ± 5.3* |
| **5** | 4-Me | 10 | Les | 32.0 ± 3.0 | 54.0 ± 5.3* | 57.4 ± 4.3* | 45.4 ± 3.6* |
| Vehicle | — | | Contra | 60.7 ± 4.7 | 64.5 ± 6.3 | 62.9 ± 5.6 | 60.4 ± 4.8 |
| | | | Les | 29.3 ± 3.2 | 28.1 ± 3.5 | 31.7 ± 3.9 | 26.2 ± 4.2 |
| **7** | 3-CN | 30 | Les | 28.5 ± 2.9 | 49.7 ± 5.2* | 55.4 ± 4.7* | 46.8 ± 5.3* |
| Vehicle | — | | Contra | 59.6 ± 3.7 | 62.9 ± 5.0 | 60.3 ± 4.4 | 57.8 ± 3.5 |
| | | | Les | 30.6 ± 2.9 | 33.5 ± 3.6 | 32.2 ± 3.5 | 35.6 ± 4.1 |
| **8** | 4-CN | 10 | Les | 31.7 ± 2.7 | 59.4 ± 5.8* | 55.3 ± 5.3* | 58.2 ± 6.2* |
| Vehicle | — | | Contra | 60.7 ± 4.7 | 64.5 ± 6.3 | 62.9 ± 5.6 | 60.4 ± 4.8 |
| | | | Les | 29.3 ± 3..2 | 28.1 ± 3.5 | 31.7 ± 3.9 | 26.2 ± 4.0 |
| **16** | 3-*i*-Pr | 30 | Les | 25.4 ± 2.7 | 48.5 ± 4.6* | 43.7 ± 5.0 | 42.5 ± 4.6 |

There were 8–14 rats per group. Les, lesioned; Contra, contralateral.
* $p < 0.01$ versus the saline lesioned dx paw.

antihyperalgesic doses (100 mg/kg po) compounds **5** and **7** did not produce any behavioural impairment as evaluated by the rat rotarod test (data not shown).[14]

## 4. Conclusion

Microwave assisted Goldberg reaction allowed rapid insertion of substituted phenyl rings onto a dimiracetam scaffold. Compounds **5**, **7** and **8** dose dependently reverted mechanical hyperalgesia in sciatic nerve-ligated rats and streptozotocin-treated animals after i.c.v. and oral administration without inducing any motor impairment. Interestingly the action of these compounds is neuropathy-specific since no significant activity is detected on the unlesioned contralateral paw in mononeuropathic and in control rats.

All the data suggest that these compounds represent a novel class of well-tolerated therapeutic agents for the relief of neuropathic pain. Additional work is in progress to gain more information on their mechanism of action and activity profile in other models of neuropathic pain.

## 5. Experimental

### 5.1. Chemistry

Reagents obtained from commercial sources were used without further purification. In order to monitor the progress of the reaction, thin layer chromatography (TLC) was performed using Merck silica gel 60 F254 precoated plates. Flash chromatography was performed using Merck silica gel 60, 230–400 mesh. The reported yields are unoptimized. Melting points were determined on a Buchi melting point B545 apparatus and are uncorrected. $^1$H NMR spectra were recorded on a Bruker AC 300. Mass spectra were recorded TSQ 700 instrument (Thermo-Finnigan), in EI mode with an external probe (DIS). Elemental analyses were within 0.4% of the theoretical value for the indicated elements.

The microwave-enhanced Goldberg reactions were performed on a CEM discover microwave apparatus, equipped with an internal probe that monitors reaction temperature and maintains the desired temperature by computer control.

#### 5.1.1. General procedure for dimiracetam arylation with aryl halides.

**5.1.1.1. Method A (MW heating).** To a solution of dimiracetam (**1**, 0.5 g, 3.5 mmol),[8a] in *N*-methylpyrrolidone (NMP 1 ml), CuI (0.19 g, 1 mmol), $K_2CO_3$ (0.5 g, 3.5 mmol) and aryl halide (7 mmol) were added under stirring. The suspension was heated in a microwave apparatus at 150 °C for 20 min. Ethyl acetate (50 ml) and water (5 ml) were added to the suspension and the mixture was stirred for 30 min the presence of Celite. The reaction mixture was filtered and the organic phase was washed with brine, dried over $Na_2SO_4$, filtered and concentrated to dryness. The residue was triturated with $Et_2O$ to give the desired final compounds. *CAUTION:* The microwave-enhanced Goldberg reaction should be performed in an open reaction system in order to enable

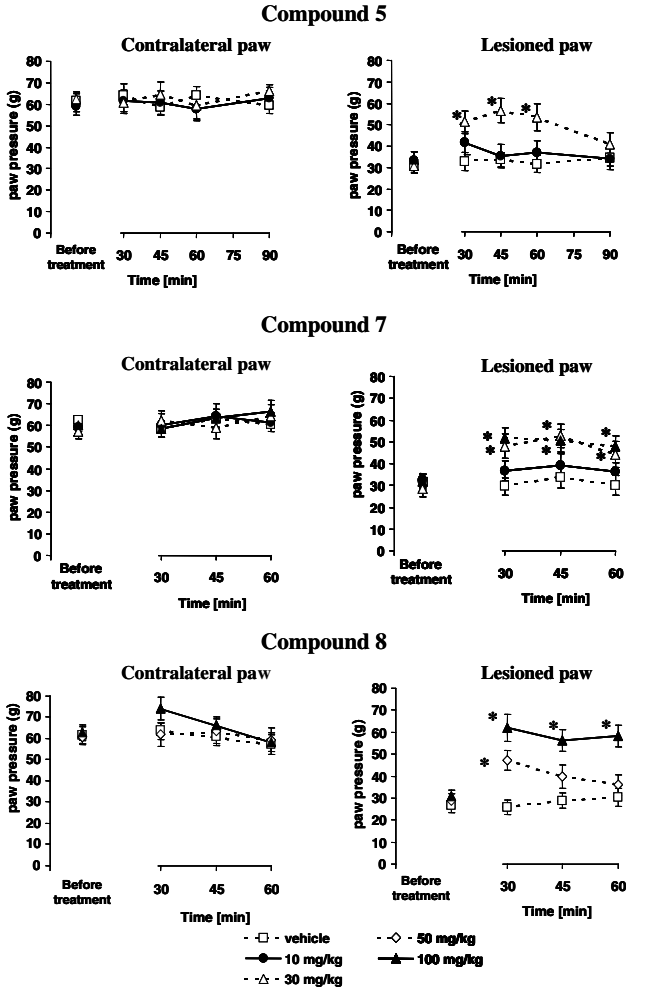



**Figure 2.** Compounds **5**, **7** and **8** after po administration in nerve injury-induced hyperalgesia in the rat paw pressure test. Pain threshold is reported as the force inducing the first struggling from the rat (g). Data are means ± SEM of 6–11 rats. $^{*}p < 0.01$, statistically significant from vehicle administration. Test was performed 14 days after sciatic nerve ligation.

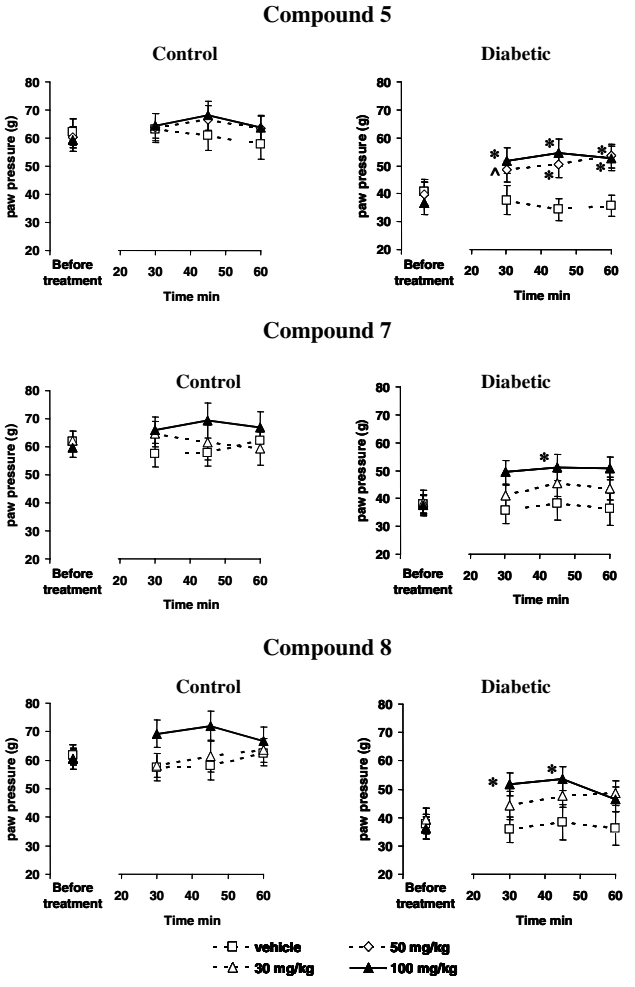



**Figure 3.** Compounds **5**, **7** and **8** after po administration in streptozotocin-induced hyperalgesia in the rat paw pressure test. Pain threshold is reported as the force inducing the first struggling from the rat (g). Data are means ± SEM of 9–10 rats.$^{\cdot}p < 0.05$ and $^{*}p < 0.01$, statistically significant from vehicle administration.

gaseous products to escape, thus avoiding the risk of explosion.

**5.1.1.2. Method B (conventional heating).** To a solution of dimiracetam (**1**, 0.5 g, 3.5 mmol),[8a] in DMF (5 ml), CuI (0.19 g, 1 mmol), $K_2CO_3$ (0.5 g, 3.5 mmol) and aryl halide (7 mmol) were added under stirring. The suspension was refluxed for 5 h. The solvent was removed under vacuum and the crude was portioned between ethyl acetate and water. The organic layer was filtered over Celite and the filtrate was washed with 10% HCl, then with a saturated solution of $NaHCO_3$ and finally with brine. The organic layer was dried over $Na_2SO_4$, filtered and concentrated to dryness. The residue was triturated with $Et_2O$ to give the desired final compounds.

**5.1.2. Compound 2.** Yield: 20%. $^1$H NMR ($CDCl_3$) $\delta$: 7.35–7.50 (m, 4H), 7.18–7.32 (m, 1H), 5.84 (t, $J$ = 5.8 Hz, 1H), 4.48 (d, $J$ = 16.7 Hz, 1H), 3.74 (d, $J$ = 16.7 Hz, 1H), 2.57–2.82 (m, 2H), 2.36–2.54 (m, 1H), 1.92–2.16 (m, 1H). MS: 216 $(M)^+$, 160, 97.

**5.1.3. Compound 3.** Yield: 45%. $^1$H NMR ($CDCl_3$) $\delta$: 7.20–7.39 (m, 3H), 7.03–7.14 (m, 1H), 5.42–5.75 (m, 1H), 4.44 (d, $J$ = 16.7 Hz, 1H), 3.75 (d, $J$ = 16.4 Hz, 1H), 2.67 (dt, $J$ = 17.0, 9.8 Hz, 1H), 2.46 (dd d, $J$ = 17.3, 10.1, 3.1 Hz, 1H), 2.33 (dd dd, $J$ = 16.2, 6.5, 6.4, 3.0 Hz, 1H), 2.24 (s, 3H), 1.79–2.09 (m, 1H). MS: 230 $(M)^+$, 143, 118, 97.

**5.1.4. Compound 4.** Yield: 40%. $^1$H NMR ($CDCl_3$) $\delta$: 7.30 (dd, $J$ = 7.9 Hz, 1H), 7.24 (br s, 1H), 7.13 (dd, $J$ = 8.2, 1.3 Hz, 1H), 7.06 (d, $J$ = 7.6 Hz, 1H), 5.76–5.85 (m, 1H), 4.47 (d, $J$ = 16.4 Hz, 1H), 3.73 (d, $J$ = 16.4 Hz, 1H), 2.54–2.80 (m, 2H), 2.40–2.53 (m, 1H), 2.38 (s, 3H), 1.91–2.13 (m, 1H). MS: 230 $(M)^+$, 174, 118.

**5.1.5. Compound 5.** Yield: 62%. $^1$H NMR ($CDCl_3$) $\delta$: 77.30 (m, 2H), 7.24 (m, 2H), 5.76–5.87 (m, 1H), 4.48

(d, $J$ = 16.4 Hz, 1H), 3.75 (d, $J$ = 16.4 Hz, 1H), 2.56–2.83 (m, 2H), 2.41–2.55 (m, 1H), 2.37 (s, 3H), 1.94–2.12 (m, 1H). $^{13}$C NMR ($CDCl_3$) $\delta$: 177.3 (CO); 169.1 (CO); 136.6 (Ar); 133.0 (Ar); 130.4 (Ar); 122.1 (Ar), 75.9 (C7a); 47.8 (C3); 30.7 (C6); 28.2 (C7); 21.4 ($CH_3$). MS: 230 $(M)^+$, 174, 118.

**5.1.6. Compound 6.** Yield: 20%. $^1$H NMR ($CDCl_3$) $\delta$: 7.79 (dd, $J$ = 7.9, 1.5 Hz, 1H), 7.72 (td, $J$ = 7.9, 1.5 Hz, 1H), 7.45–7.54 (m, $J$ = 7.6, 7.6, 1.2 Hz, 1H), 7.40 (dd, $J$ = 8.2, 0.9 Hz, 1H), 5.89–6.03 (m, 1H), 4.52 (d, $J$ = 16.7 Hz, 1H), 3.83 (dt, $J$ = 16.7, 1.0 Hz, 1H), 2.63–2.82 (m, 1H), 2.40–2.60 (m, 2H), 1.92–2.13 (m, 1H). MS: 242.22 $(MH)^+$.

**5.1.7. Compound 7.** Yield: 31%. $^1$H NMR ($CDCl_3$) $\delta$: 7.67–7.80 (m, 2H), 7.47–7.61 (m, 2H), 5.86 (t, $J$ = 5.7 Hz, 1H), 4.53 (d, $J$ = 17.0 Hz, 1H), 3.77 (d, $J$ = 17.0 Hz, 1H), 2.67–2.86 (m, 2H), 2.39–2.57 (m, 1H), 1.92–2.14 (m, 1H). MS: 241 $(M)^+$, 185, 129.

**5.1.8. Compound 8.** Yield: 55%. $^1$H NMR ($CDCl_3$) $\delta$: 7.73 (m, 2H), 7.60 (m, 2H), 5.75–5.98 (m, 1H), 4.54 (d, $J$ = 16.7 Hz, 1H), 3.77 (d, $J$ = 17.0 Hz, 1H), 2.65–2.91 (m, 2H), 2.41–2.62 (m, 1H), 1.94–2.17 (m, 1H). MS: 242.2 $(MH)^+$.

**5.1.9. Compound 9.** Yield: 20%. $^1$H NMR ($CDCl_3$) $\delta$: 7.81 (dd, $J$ = 7.9, 1.6 Hz, 1H), 7.67 (t, $J$ = 7.2 Hz, 1H), 7.47–7.62 (m, 1H), 7.26–7.34 (m, 1H), 5.65 (t, $J$ = 5.7 Hz, 1H), 4.27–4.56 (m, 1H), 3.64–3.88 (m, 1H), 2.57–2.78 (m, 1H), 2.40–2.57 (m, 1H), 2.19–2.39 (m, 1H), 1.87–2.19 (m, 1H). MS: 284 $(M)^+$, 228; 192, 172, 145.

**5.1.10. Compound 10.** Yield: 35%. $^1$H NMR ($CDCl_3$) $\delta$: 7.44–7.75 (m, 4H), 5.88 (t, $J$ = 5.5 Hz, 1H), 4.51 (d, $J$ = 16.7 Hz, 1H), 3.76 (d, $J$ = 16.7 Hz, 1H), 2.61–2.91 (m, 2H), 2.35–2.57 (m, 1H), 1.88–2.16 (m, 1H). MS: 284 $(M)^+$, 228, 172, 145.

**5.1.11. Compound 11.** Yield: 51%. $^1$H NMR ($CDCl_3$) $\delta$: 7.68 (m, 2H), 7.57 (m, 2H), 5.88 (dd, $J$ = 6.0, 5.3 Hz, 1H), 4.51 (d, $J$ = 16.7 Hz, 1H), 3.75 (d, $J$ = 16.7 Hz, 1H), 2.63–2.84 (m, 2H), 2.35–2.59 (m, 1H), 1.92–2.15 (m, 1H). MS: 284 $(M)^+$, 228; 172; 145.

**5.1.12. Compound 12.** Yield: 15%. $^1$H NMR ($CDCl_3$) $\delta$: 7.38 (td, $J$ = 8.2, 6.3 Hz, 1H), 7.29 (dt, $J$ = 10.4, 2.2 Hz, 1H), 7.15 (dd d, $J$ = 8.2, 2.0, 0.8 Hz, 1H), 6.88–7.02 (m, $J$ = 8.2, 8.2, 2.4, 0.9 Hz, 1H), 5.81 (t, $J$ = 5.5 Hz, 1H), 4.49 (d, $J$ = 16.7 Hz, 1H), 3.74 (d, $J$ = 16.7 Hz, 1H), 2.59–2.87 (m, 2H), 2.33–2.57 (m, 1 H), 1.91–2.16 (m, 1H). MS: 235.1 $(MH)^+$.

**5.1.13. Compound 13.** Yield: 10%. $^1$H NMR ($CDCl_3$) $\delta$: ($CDCl_3$) δ: 7.37 (m, 2H), 7.12 (m, 2H), 5.78 (t, $J$ = 5.7 Hz, 1H), 4.47 (d, $J$ = 16.7 Hz, 1H), 3.74 (d, $J$ = 16.7 Hz, 1H), 2.55–2.86 (m, 2H), 2.37–2.54 (m, 1H), 1.88–2.10 (m, 1H). MS: 235.1 $(MH)^+$.

**5.1.14. Compound 14.** Yield: 65%. $^1$H NMR ($CDCl_3$) $\delta$: 7.44–7.49 (m, 1H), 7.28–7.39 (m, 2H), 7.16–7.25 (m, 1H), 5.81 (t, $J$ = 5.5 Hz, 1H), 4.49 (d, $J$ = 16.7 Hz, 1H), 3.74 (d, $J$ = 16.7 Hz, 1H), 2.60–2.79 (m, 2H), 2.36–2.56 (m, 1H), 1.84–2.14 (m, 1H). MS: 250 $(M)^+$, 194, 138, 111.

**5.1.15. Compound 15.** Yield: 44%. $^1$H NMR ($CDCl_3$) $\delta$: 7.37 (m, 4H), 5.80 (t, $J$ = 6.0 Hz, 1H), 4.47 (d, $J$ = 16.7 Hz, 1H), 3.73 (d, $J$ = 16.7 Hz, 1H), 2.57–2.83 (m, 2H), 2.36–2.54 (m, 1H), 1.89–2.11 (m, 1H). MS: 250 $(M)^+$, 194.

**5.1.16. Compound 16.** Yield: 23%. $^1$H NMR ($CDCl_3$) $\delta$: 7.04–7.43 (m, 4H), 5.83 (t, $J$ = 5.8 Hz, 1H), 4.47 (d, $J$ = 16.7 Hz, 1H), 3.74 (d, $J$ = 16.7 Hz, 1H), 2.93 (spt, $J$ = 7.2 Hz, 1H), 2.56–2.80 (m, 2 H), 2.36–2.56 (m, 1H), 1.93–2.18 (m, 1H), 1.26 (d, $J$ = 6.9 Hz, 6H). MS: 259 $(MH)^+$.

**5.1.17. Compound 17.** Yield: 33%. 1H NMR (CDCl3) δ: 7.20–7.36 (m, 4H), 5.80 (t, $J$ = 6.0 Hz, 1H), 4.47 (d, $J$ = 16.4 Hz, 1H), 3.73 (d, $J$ = 16.4 Hz, 1H), 2.91 (spt, $J$ = 6.9 Hz, 1H), 2.55–2.81 (m, 2H), 2.37–2.53 (m, 1H), 1.90–2.18 (m, 1H), 1.25 (d, $J$ = 6.9 Hz, 6H). MS: 259.1 $(MH)^+$.

**5.1.18. Compound 18.** Yield: 63%. $^1$H NMR ($CDCl_3$) $\delta$: 7.44–7.75 (m, 4H), 5.88 (t, $J$ = 5.5 Hz, 1H), 4.51 (d, $J$ = 16.7 Hz, 1H), 3.82 (s, 3H), 3.76 (d, $J$ = 16.7 Hz, 1H), 2.61–2.91 (m, 2H), 2.35–2.57 (m, 1H), 1.88–2.16 (m, 1H). MS: 246 $(M)^+$, 190.

**5.1.19. Compound 19.** Yield: 25%. $^1$H NMR ($CDCl_3$) $\delta$: 7.16–7.41 (m, 2H), 6.95 (m, 2H), 5.74 (t, $J$ = 6.0 Hz, 1H), 4.45 (d, $J$ = 16.4 Hz, 1H), 3.81 (s, 3H), 3.73 (d, $J$ = 16.7 Hz, 1H), 2.35–2.84 (m, 3H), 1.86–2.14 (m, 1H). MS: 246 $(M)^+$, 190, 134.

**5.1.20. Compound 20.** Yield: 35%. $^1$H NMR ($CDCl_3$) $\delta$: 77.16 (t, $J$ = 8.2 Hz, 1H), 6.96 (t, $J$ = 2.2 Hz, 1H), 6.75 (dd, $J$ = 7.9, 1.9 Hz, 1H), 6.69 (dd, 1H), 5.74 (t, $J$ = 5.7 Hz, 1H), 4.38 (d, $J$ = 16.4 Hz, 1H), 3.67 (d, $J$ = 16.4 Hz, 1H), 2.49–2.76 (m, 2H), 2.28–2.49 (m, 1H), 1.83–2.11 (m, 1H). MS: 232 $(M)^+$, 176.

**5.1.21. Compound 21.** Yield: 25%. $^1$H NMR ($CDCl_3$) $\delta$: 7.98 (d, 2H), 7.65 (d, $J$ = 8.8 Hz, 2H), 5.90 (t, $J$ = 5.2 Hz, 1H), 4.52 (d, $J$ = 17.0 Hz, 1H), 3.77 (d, $J$ = 16.7 Hz, 1H), 3.05 (s, 3H), 2.61–2.91 (m, 2H), 2.31–2.60 (m, 1H), 1.83–2.16 (m, 1H). MS: 295.1 $(MH)^+$.

**5.1.22. Compound 22.** Yield: 46%. $^1$H NMR ($CDCl_3$) $\delta$: 7.27 (m, 4H), 5.80 (t, $J$ = 6.0 Hz, 1H), 4.46 (d, $J$ = 16.4 Hz, 1H), 3.74 (d, $J$ = 16.4 Hz, 1H), 2.65 (q, $J$ = 7.6 Hz, 2H), 2.55–2.80 (m, 2H), 2.39–2.52 (m, 1H), 1.90–2.14 (m, 1H), 1.11–1.32 (m, 3H). MS: 245.1 $(MH)^+$.

**5.1.23. Compound 23.** Yield: 48%. $^1$H NMR ($CDCl_3$) $\delta$: 7.37–7.50 (m, 4H), 5.81–5.91 (m, 1H), 4.72 (s, 2H), 4.49 (d, $J$ = 16.7 Hz, 1H), 3.76 (d, $J$ = 16.7 Hz, 1H), 2.58–2.83 (m, 2H), 2.39–2.55 (m, 1H), 1.94–2.13 (m, 1H), 1.56 (br s, 1H). MS: 247.2 $(MH)^+$.

**5.1.24. Compound 24.** Yield: 62%. $^1$H NMR ($CDCl_3$) $\delta$: 7.96–8.22 (m, 2H), 7.40–7.65 (m, 2H), 5.89 (t, $J$ = 5.7 Hz, 1H), 4.51 (d, $J$ = 16.7 Hz, 1H), 4.38 (q, $J$ = 7.2 Hz, 2H), 3.75 (d, $J$ = 16.7 Hz, 1H), 2.63–2.90 (m, 2H), 2.35–2.58 (m, 1H), 1.87–2.16 (m, 1H), 1.39 (t, $J$ = 7.2 Hz, 3H). MS: 289.4 $(MH)^+$, 218.4.

**5.1.25. Compound 25.** Yield: 24%. $^1$H NMR (DMSO-$d_6$) $\delta$: 12.87 (br s, 1H), 7.98 (m, $J$ = 8.8 Hz, 2H), 7.65 (m, $J$ = 8.8 Hz, 2H), 5.89–6.06 (m, 1H), 4.20 (d, $J$ = 16.1 Hz, 1H), 3.79 (d, $J$ = 16.1 Hz, 1H), 2.57–2.81 (m, 2H), 2.18–2.36 (m, 1H), 1.79–1.99 (m, 1H). MS: 261.2 $(MH)^+$.

**5.1.26. Compound 26.** Yield: 15%. $^1$H NMR ($CDCl_3$) $\delta$: 6.98–7.16 (m, 2H), 6.70 (tt, $J$ = 8.8, 2.2 Hz, 1H), 5.67–5.84 (m, 1H), 4.51 (d, $J$ = 16.7 Hz, 1H), 3.76 (d, $J$ = 16.7 Hz, 1H), 2.65–2.86 (m, 2H), 2.40–2.60 (m, 1H), 1.93–2.16 (m, 1H). MS: 253.2 $(MH)^+$.

**5.1.27. Compound 27.** Yield: 57%. $^1$H NMR ($CDCl_3$) $\delta$: 7.18 (d, $J$ = 2.2 Hz, 1H), 7.16 (d, $J$ = 7.9 Hz, 1H), 7.05 (dd, $J$ = 8.2, 2.2 Hz, 1H), 5.77 (t, $J$ = 6.0 Hz, 1H), 4.44 (d, $J$ = 16.4 Hz, 1H), 3.72 (d, $J$ = 16.4 Hz, 1H), 2.63–2.79 (m, 1H), 2.53–2.63 (m, 1H), 2.37–2.51 (m, 1H), 2.28 (s, 3H), 2.25 (s, 3H), 1.91–2.12 (m, 1H). MS: 245 $(MH)^+$.

**5.1.28. Compound 28.** Yield: 54%. $^1$H NMR ($CDCl_3$) $\delta$: 6.98 (s, 2H), 6.89 (s, 1H), 5.79 (t, $J$ = 5.8 Hz, 1H), 4.45 (d, $J$ = 16.4 Hz, 1H), 3.72 (d, $J$ = 16.7 Hz, 1H), 2.71 (dt, $J$ = 16.0, 9.8 Hz, 1H), 2.53–2.65 (m, 1H), 2.37–2.51 (m, 1H), 2.33 (s, 6H), 1.89–2.13 (m, 1H). MS: 244 $(M)^+$, 188, 97.

**5.1.29. Compound 29.** Yield: 10%. $^1$H NMR ($CDCl_3$) $\delta$: 7.24–7.31 (m, 1H), 7.10–7.17 (m, 1H), 7.07 (dd, $J$ = 8.8 Hz, 1H), 5.72–5.84 (m, 1H), 4.48 (d, $J$ = 16.7 Hz, 1H), 3.76 (dt, $J$ = 16.7, 1.0 Hz, 1H), 2.57–2.81 (m, 2H), 2.43–2.54 (m, 1H), 2.32 (d, $J$ = 2.1 Hz, 3H), 1.93–2.13 (m, 1H). MS: 249.3 $(MH)^+$.

**5.1.30. Compound 30.** Yield: 46%. $^1$H NMR ($CDCl_3$) $\delta$: 7.14–7.25 (m, 2H), 7.03 (dd, $J$ = 8.2, 2.2 Hz, 1H), 5.77 (t, $J$ = 5.8 Hz, 1H), 4.46 (d, $J$ = 16.4 Hz, 1H), 3.72 (d, $J$ = 16.4 Hz, 1H), 2.59–2.81 (m, 2H), 2.37–2.55 (m, 1H), 2.26 (d, $J$ = 1.6 Hz, 3H), 1.94–2.12 (m, 1H). MS: 249 $(MH)^+$, 192, 97.

**5.1.31. Compound 31.** Yield: 35%. $^1$H NMR ($CDCl_3$) $\delta$: 7.18–7.28 (m, 1H), 7.08 (t, $J$ = 8.3 Hz, 1H), 6.92 (d, $J$ = 7.9 Hz, 1H), 5.73 (br s, 1H), 4.46 (d, $J$ = 16.7 Hz, 1H), 3.78 (d, $J$ = 16.4 Hz, 1H), 2.70 (dt, $J$ = 17.3, 9.4 Hz, 1H), 2.48 (dd d, $J$ = 17.0, 10.1, 2.8 Hz, 1H), 2.29–2.43 (m, 1H), 2.14 (s, 3H), 1.75–2.01 (m, 1H). MS: 248 $(M)^+$, 136, 109.

**5.1.32. Compound 32.** Yield: 25%. $^1$H NMR (DMSO-$d_6$) $\delta$: 7.42–7.52 (m, 1H), 7.28–7.34 (m, 2H), 5.75 (br. s., 1H), 4.17 (d, $J$ = 16.1 Hz, 1H), 3.79 (d, $J$ = 15.8 Hz, 1H), 2.54–2.69 (m, 1H), 2.22–2.41 (m, 2H), 2.21 (s, 3H), 1.89 (br. s., 1H). MS: 265.3 $(MH)^+$.

**5.1.33. Compound 33.** Yield: 20%. $^1$H NMR (DMSO-$d_6$) $\delta$: 7.32 (dd, $J$ = 8.8, 5.3 Hz, 1H), 7.16 (dd, $J$ = 9.4, 2.8 Hz, 1H), 7.07 (td, $J$ = 8.5, 2.8 Hz, 1H), 5.60–5.77 (m, 1H), 4.15 (d, $J$ = 16.0 Hz, 1H), 3.68–3.86 (m, $J$ = 16.0, 0.9, 0.9 Hz, 1H), 2.54–2.69 (m, 1H), 2.21–2.41 (m, 2H), 2.19 (s, 3H), 1.75–2.01 (m, 1H). MS: 249.1 $(MH)^+$.

**5.1.34. Compound 34.** Yield: 41%. $^1$H NMR ($CDCl_3$) $\delta$: 7.33 (d, $J$ = 2.2 Hz, 1H), 7.25 (dd, $J$ = 8.5, 2.2 Hz, 1H), 7.04 (d, $J$ = 8.5 Hz, 1H), 5.64 (br. s., 1H), 4.45 (d, $J$ = 16.7 Hz, 1H), 3.77 (d, $J$ = 16.7 Hz, 1H), 2.69 (dt, $J$ = 17.3, 9.8 Hz, 1H), 2.42–2.55 (m, $J$ = 17.3, 10.1, 2.8 Hz, 1H), 2.36 (dd dd, $J$ = 16.2, 6.4, 6.3, 2.8 Hz, 1H), 2.22 (s, 3H), 1.90 (br s, 1H). MS: 264 $(M)^+$, 97.

**5.1.35. Compound 35.** Yield: 24%. $^1$H NMR (DMSO-$d_6$) $\delta$: 7.31 (dd, $J$ = 8.2, 6.2 Hz, 1H), 7.04 (td, $J$ = 8.5, 2.7 Hz, 1H), 6.87 (dd, $J$ = 8.8, 2.7 Hz, 1H), 5.66 (br s, 1H), 4.48 (d, $J$ = 16.5 Hz, 1H), 3.72–3.86 (m, $J$ = 16.5, 1.2, 1.2 Hz, 1H), 2.63–2.82 (m, 1H), 2.32–2.58 (m, 2H), 2.22 (s, 3H), 1.81–2.12 (m, 1H). MS: 249.1 $(MH)^+$.

**5.1.36. Compound 36.** Yield: 25%. $^1$H NMR ($CDCl_3$) $\delta$: 7.01–7.22 (m, 3H), 5.70–5.85 (m, 1H), 4.46 (d, $J$ = 16.4 Hz, 1H), 3.77 (dt, $J$ = 16.4, 1.2 Hz, 1H), 2.61–2.79 (m, 1H), 2.37–2.54 (m, 2H), 2.36 (s, 3H), 1.89–2.10 (m, 1H). MS: 249.3 $(MH)^+$.

**5.1.37. Compound 37.** Yield: 35%. $^1$H NMR ($CDCl_3$) $\delta$: 7.37 (d, $J$ = 8.5 Hz, 1H), 7.33 (d, $J$ = 2.8 Hz, 1H), 7.13 (dd, $J$ = 8.3, 3.0 Hz, 1H), 5.79 (t, $J$ = 6.0 Hz, 1H), 4.46 (d, $J$ = 16.4 Hz, 1H), 3.73 (d, $J$ = 16.4 Hz, 1H), 2.56–2.81 (m, 2H), 2.41–2.52 (m, 1H), 2.37–2.41 (m, 3H), 1.91–2.11 (m, 1H). MS: 265 $(MH)^+$.

**5.1.38. Compound 38.** Yield: 43%. $^1$H NMR ($CDCl_3$) $\delta$: 6.94–7.07 (m, 2H), 6.77 (d, $J$ = 9.1 Hz, 1H), 5.78 (t, $J$ = 5.7 Hz, 1H), 4.47 (d, $J$ = 16.7 Hz, 1H), 3.73 (d, $J$ = 16.4 Hz, 1H), 2.61–2.81 (m, 2H), 2.39–2.53 (m, 1H), 2.37 (s, 3H), 1.93–2.11 (m, 1H). MS: 249 $(MH)^+$.

**5.1.39. Compound 40a.** To a solution of (*S*)-5-oxo-pyrrolidine-2-carboxylic acid methyl ester (30 g, 0.19 mol) in $CH_3CN$ (500 ml), 55% NaH (9.12 g, 0.21 mol) was added portionwise over 30 min at a temperature ranging between 0 °C and 5 °C. The mixture was stirred for additional 90 min and then a solution of bromoacetic acid *tert*-butyl ester (33.6 ml, 0.21 mol) in $CH_3CN$ (35 ml) was added dropwise over 15 min. The reaction mixture was warmed to room temperature and stirred for 3 h. The solvent was evaporated and the solid crude was dissolved in water and EtOAc (100/300 ml). The organic layers were separated and washed with a saturated solution of $NaHCO_3$, then with water and finally dried over $Na_2SO_4$, filtered and evaporated to get a gummy mass, which was triturated with hexane to afford the title compound as a white powder (39 g, 75%). Mp 70–71 °C, $[\alpha]_D$ = −41 ($c$ = 5, DCM). Compound **40b** was prepared following the same procedure of **40a**. Mp 68–69 °C, $[\alpha]_D$ = +39.9 ($c$ = 5, DCM).

**5.1.40. Compound 41a.** Compound **40a** (36.5 g, 0.13 mol) was added portionwise to a solution of 32% $NH_4OH$ (365 ml). The resulting suspension was stirred for 90 min at room temperature. The aqueous phases were extracted with EtOAc (6 × 200 ml). The combined organic layer was dried and evaporated to get a crude solid which on trituration with AcOEt afforded the desired compound as a white solid (21.2 g, 65%). Mp 128–129 °C, $[\alpha]_D = -31.3$ ($c = 5$, DCM). Compound **41b** was prepared following the same procedure as **41a**. Mp 132–135 °C, $[\alpha]_D = +31.4$ ($c = 5$, DCM).

**5.1.41. Compound 42a.** Compound **41a** (2 g, 8.2 mmol) was added to a solution of iodosobenzene–bistrifluoroacetate (3.5 g, 8.2 mmol) in $CH_3CN$ (27 ml) and water (14 ml). The reaction mixture was stirred for 4 h. The organic solvent was evaporated under vacuum and the aqueous phase was washed with diethylether, basified with solid $Na_2CO_3$ and extracted with DCM (5 × 80 ml). The combined organic layers were dried and evaporated to give (*S*)-(2-amino-5-oxo-pyrrolidin-1-yl)-acetic acid *tert*-butyl ester as a yellow oil (1.5 g, Yield: 86%). One gram of this compound (4.7 mmol) was dissolved in DCM (1 ml) and then TFA (5 ml) was added dropwise maintaining the temperature in the range of 15–20 °C. The solution was stirred for 3 h and then was evaporated under vacuum. The reaction crude was redissolved in 2 N HCl (4.7 ml, 9.4 mmol) and after few minutes the solution was evaporated to get a gummy solid that after trituration with diethylether afforded the desired compound as a white solid (0.65 g, 71%). Mp 175–177 °C, $[\alpha]_D = +1.9$ ($c = 1$, $H_2O$). Compound **42b** was prepared following the same procedure as **42a**. Mp 174–176 °C, $[\alpha]_D = -1.1$ ($c = 1$, $H_2O$).

**5.1.42. Compound 43a.** A mixture of compound **42a** (4 g, 20.6 mmol) and anhydrous NaOAc (1.69 g, 20.6 mmol) in acetic anhydride (60 ml) was refluxed for 4 h under nitrogen. The solvent was then removed under vacuum and the crude was partitioned between EtOAc (200 ml) and water (50 ml), the organic layer was separated and washed with a saturated solution of $NaHCO_3$ (50 ml) and water (50 ml), dried over $Na_2SO_4$, filtered and evaporated to get 1-acetyl-tetrahydro-pyrrolo[1,2-*a*]imidazole-2,5-dione as a brown oil (2.4 g, 64%). The protected compound (2.2 g, 12 mmol) was dissolved in water (44 ml) and then Amberlite IR 120 (2.2 g, 16–45 mesh, $H^+$ form) was added. The reaction mixture was filtered and the water was evaporated under vacuum. The obtained solid was triturated with isopropanol to afford the desired compound (0.9 g, 54%). Mp 197–200 °C $[\alpha]_D = +39.9$ ($c = 0.4$, MeOH). Compound **43b** was prepared following the same procedure as **43a**. Mp 200–201 °C, $[\alpha]_D = -39.2$ ($c = 0.4$, MeOH).

**5.1.43. Compound 3S.** Yield: 48%. $^1$H NMR ($CDCl_3$) $\delta$: 7.20–7.39 (m, 3H), 7.03–7.14 (m, 1H), 5.42–5.75 (m, 1H), 4.44 (d, $J = 16.7$ Hz, 1H), 3.75 (d, $J = 16.4$ Hz, 1H), 2.67 (dt, $J = 17.0$, 9.8 Hz, 1H), 2.46 (dd d, $J = 17.3$, 10.1, 3.1 Hz, 1H), 2.33 (dd dd, $J = 16.2$, 6.5, 6.4, 3.0 Hz, 1H), 2.24 (s, 3H), 1.79–2.09 (m, 1H). MS: 230 $(M)^+$, 143, 118, 97.

**5.1.44. Compound 3R.** Yield: 44%. $^1$H NMR ($CDCl_3$) $\delta$: $^1$H NMR ($CDCl_3$) $\delta$: 7.20–7.39 (m, 3H), 7.03–7.14 (m, 1H), 5.42–5.75 (m, 1H), 4.44 (d, $J = 16.7$ Hz, 1H), 3.75 (d, $J = 16.4$ Hz, 1H), 2.67 (dt, $J = 17.0$, 9.8 Hz, 1H), 2.46 (ddd, $J = 17.3$, 10.1, 3.1 Hz, 1H), 2.33 (dd dd, $J = 16.2$, 6.5, 6.4, 3.0 Hz, 1H), 2.24 (s, 3H), 1.79–2.09 (m, 1H). MS: 230 $(M)^+$, 143, 118, 97.

**5.1.45. Compound 5S.** Yield: 60%. $^1$H NMR ($CDCl_3$) $\delta$: 77.30 (m, 2H), 7.24 (m, 2H), 5.76–5.87 (m, 1H), 4.48 (d, $J = 16.4$ Hz, 1H), 3.75 (d, $J = 16.4$ Hz, 1H), 2.56–2.83 (m, 2H), 2.41–2.55 (m, 1H), 2.37 (s, 3H), 1.94–2.12 (m, 1H). $^{13}$C NMR ($CDCl_3$) $\delta$: 177.3 (CO); 169.1 (CO); 136.6 (Ar); 133.0 (Ar); 130.4 (Ar); 122.1 (Ar), 75.9 (C7a); 47.8 (C3); 30.7 (C6); 28.2 (C7); 21.4 ($CH_3$). MS: 230 $(M)^+$, 174, 118.

**5.1.46. Compound 5R.** Yield: 54%. $^1$H NMR ($CDCl_3$) $\delta$: 77.30 (m, 2H), 7.24 (m, 2H), 5.76–5.87 (m, 1H), 4.48 (d, $J = 16.4$ Hz, 1H), 3.75 (d, $J = 16.4$ Hz, 1H), 2.56–2.83 (m, 2H), 2.41–2.55 (m, 1H), 2.37 (s, 3H), 1.94–2.12 (m, 1H). $^{13}$C NMR ($CDCl_3$) $\delta$: 177.3 (CO); 169.1 (CO); 136.6 (Ar); 133.0 (Ar); 130.4 (Ar); 122.1 (Ar), 75.9 (C7a); 47.8 (C3); 30.7 (C6); 28.2 (C7); 21.4 ($CH_3$). MS: 230 $(M)^+$, 174, 118.

**5.1.47. Compound 8S.** Yield: 58%. $^1$H NMR ($CDCl_3$) $\delta$: 7.73 (m, 2H), 7.60 (m, 2H), 5.75–5.98 (m, 1H), 4.54 (d, $J = 16.7$ Hz, 1H), 3.77 (d, $J = 17.0$ Hz, 1H), 2.65–2.91 (m, 2H), 2.41–2.62 (m, 1H), 1.94–2.17 (m, 1H). MS: 242.2 $(MH)^+$.

**5.1.48. Compound 8R.** Yield: 53%. $^1$H NMR ($CDCl_3$) $\delta$: 7.73 (m, 2H), 7.60 (m, 2H), 5.75–5.98 (m, 1H), 4.54 (d, $J = 16.7$ Hz, 1H), 3.77 (d, $J = 17.0$ Hz, 1H), 2.65–2.91 (m, 2H), 2.41–2.62 (m, 1H), 1.94–2.17 (m, 1H). MS: 242.2 $(MH)^+$.

### 5.2. Pharmacology

Male Sprague–Dawley rats weighting 150–200 g were used in all experiments (Harlan, Italy). Four rats were housed per cage. The cages were placed in the experimental room 24 h before the test for acclimatization. The animals were fed a standard laboratory diet and tap water ad libitum and kept at 23 ± 1 °C with a 12 h light/dark cycle, light at 7 a.m. All experiments were carried out in accordance with the NIH Guide for the Care and Use of Laboratory animals. All efforts were made to minimize animal suffering, and to reduce the number of animals used.

Results are given as means ± SEM; analysis of variance (ANOVA), followed by Fisher's PLSD procedure for post hoc comparison, was used to verify the significance between two means. *P* values less than 0.05 were considered significant. Data were analysed with the StatView for the Macintosh computer program

**5.2.1. Chronic constriction injury model.** A peripheral mononeuropathy was produced in adult rats by placing loosely constrictive ligatures around the common sciatic nerve according to the method described by Bennett and Xie.[12]

Rats were anaesthetized with chloral hydrate. The common sciatic nerve was exposed at the level of the middle of the thigh by blunt dissection through the biceps femoris. Proximal to sciatica's trifurcation, about 1 cm of the nerve was freed of adhering tissue and four ligatures (3/0 silk tread) were tied loosely around it with about 1 mm spacing. The length of the nerve thus affected was 1 cm long. Great care was taken to tie the ligatures such that the diameter of the nerve was seen to be just barely constricted when viewed with 40x magnification. The left paw was untouched. Animals were tested for neuropathic pain 14 days after nerve injury.

**5.2.2. Streptozotocin treatment.** Rats were injected intraperitoneally with streptozotocin (50 mg/kg). Animals were tested for mechanical hyperalgesia 21-day after toxin treatment in the paw pressure test.

**5.2.3. Paw pressure test.** The nociceptive threshold in the rat was determined with an analgesimeter (Ugo Basile, Varese, Italy), according to the method described by Leighton et al.[13] The stimulus at which rats withdrawn the paw was evaluated before and at different times after drug treatment. Rats scoring below 40 g or over 75 g during the test before drug administration (25%) were rejected. Results represent the mean of mechanical thresholds expressed as grams. To avoid any possible damage to the animal paw the maximum applied force was fixed at 240 g.

**5.2.4. Rat rotarod test.** Potential motor incoordination/ataxia caused by compounds' injection was evaluated using a rotarod apparatus in which animals were required to walk against the motion of rotating drum over 30 s.[14] The time taken to fall off the rotarod was recorded as the number of falls in 30 s. The rod (30 cm long) was divided into 5 equal sections by 6 discs. Thus, up to 5 rats were tested simultaneously on the apparatus, with a rod-rotation of 16 rpm.

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