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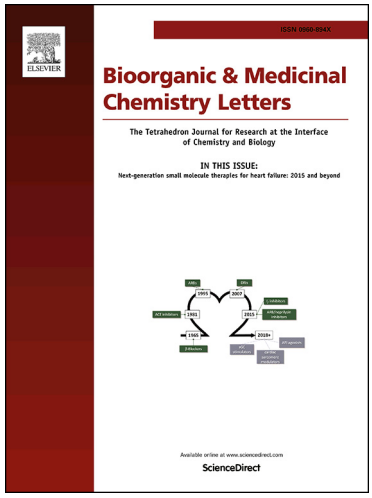

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Synthesis and Pharmacological Evaluation of Novel N-Mannich Bases Derived from 5,5-Diphenyl and 5,5-Di(propan-2-yl)imidazolidine-2,4-dione Core

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Running head: Imidazolidine-2,4-diones - anticonvulsant and antinociceptive activity

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Abstract

The aim of this study was to design and synthesize two series of N-Mannich bases with imidazolidine-2,4-dione core as a potential anticonvulsant with reduced toxicity and broad antiseizure activity. Preliminary screening revealed that the majority of synthesized compounds were effective in the maximal electroshock seizure (MES) and/or subcutaneous pentylenetetrazole (scPTZ) test. The most active *in vivo* compound, **18** (3-((4-methylpiperazin-1-yl)methyl)-5,5-diphenylimidazolidine-2,4-dione), exhibited an ED_{50} value comparable to that of phenytoin in the MES test (38.5 mg/kg vs 28.1 mg/kg), and more importantly, it showed four times higher potency than phenytoin in the 6 Hz test (12.2 mg/kg vs >60 mg/kg). Additionally, **18** exhibited antiallodynic properties in the von Frey test in neuropathic (oxaliplatin-treated) mice. Compound **18** also demonstrated a broader spectrum of anticonvulsant activity than phenytoin and showed statistically significant antinociceptive properties in selected models of chronic pain.

Keywords*:* anticonvulsants, antinociceptive, imidazolidine-2,4-dione, hydantoin

According to epidemiological studies, epilepsy is one of the most common disorders of the central nervous system (CNS) affecting more than 60 million people worldwide.[1] A characteristic feature of epilepsy is the occurrence of recurrent spontaneous seizures with or without the loss of consciousness, which are accompanied by motor, sensory, or autonomic disturbances.[2] Despite an enhanced understanding of the pathogenesis of the disease and the introduction of novel therapeutics whose mechanisms of action differ from commonly used antiepileptic drugs (AEDs), drug resistance is a common phenomenon in one-third of patients.[3]

The activity of most anticonvulsant drugs used clinically is related to neither one specific mechanism of action nor any particular site of action.[4] AEDs exhibit their activity *via* many different mechanisms of action. Insufficient understanding of the pathogenesis of human epilepsy and its complexity make it difficult to develop rational methodologies for the discovery of candidates for new AEDs. Therefore, at present, researchers mainly focus on investigations for new anticonvulsant agents through conventional ligand-based screening approaches, which are based on structure of existing drugs or different pharmacophore models. To obtain more efficacious compounds that will decrease seizures and will reduce adverse effects compared to those of the maternal compound, a variety of structural modifications are introduced.[5] Many lines of well-documented evidence emphasize that anticonvulsant activity is mainly attributed to the presence of the nitrogen heterocyclic system, usually a cyclic imide, with at least one carbonyl group and hydrophobic aryl/alkyl group attached to the heterocyclic system. These are essential requirements for substances to possess a potential antiepileptic activity. This common pharmacophore is present in the structures of well-known anticonvulsants, for example, ethosuximide and phenytoin (Figure 1).[6]

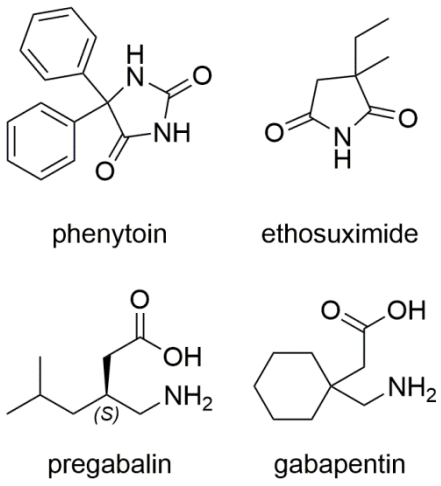


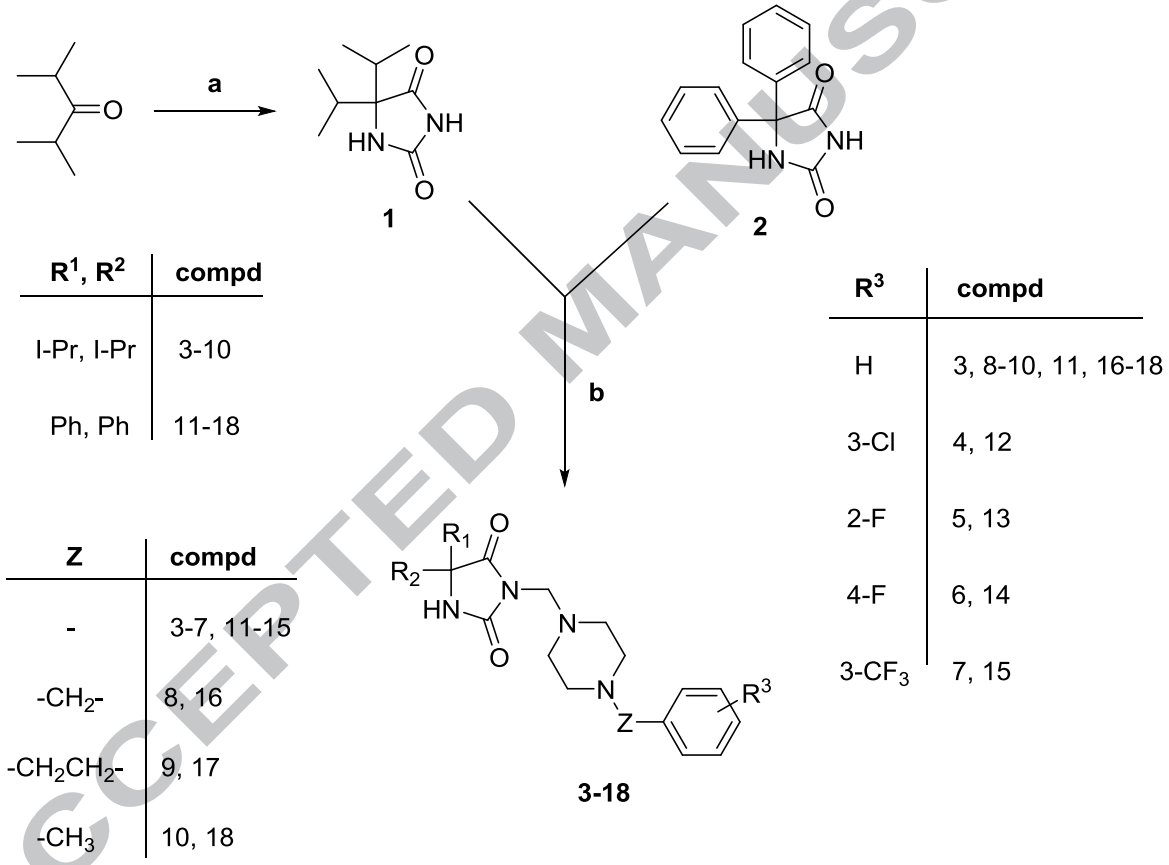
Figure 1. Structures of known antiepileptic drugs.

In addition to antiepileptic activity, AEDs exhibit other promising therapeutic activities. They are used in the treatment of many CNS diseases such as migraine, anxiety, and bipolar disorder, which appear to be linked to varying hyperexcitability within specific structures of the CNS.[7]

AEDs also have applications in the treatment of pain; for example, gabapentin and pregabalin (Figure 1), second-generation AEDs, are currently recommended as first-line pharmacological treatment for neuropathic pain,[8] and phenytoin (5,5-diphenylimidazolidine-2,4-dione), a first-generation AED, shows analgesic properties.[9] Other differently substituted imidazolidine-2,4-dione (hydantoin) derivatives also exhibit antinociceptive properties; for instance, a recently published series of amide derivatives with a hydantoin moiety exhibited analgesic properties in an acute model of pain.[10] Other research studies involving imidazolidin-2,4-dione fragments have shown their antidepressant, antipsychotic,[11,12] and antiarrhythmic properties.[13] All these findings highlight that modifications in the heterocyclic core of imidazolidine-2,4-dione create a variety of opportunities for designing and obtaining new promising therapeutic agents. Considering the abovementioned information, our research focused on systematic structural modifications of different heterocyclic systems to elucidate their pharmacological properties.[10,14–17] Therefore, as a continuation of chemical and pharmacological studies in the designing of new anticonvulsant agents, in the present study, a new series of N-Mannich bases with imidazolidine-2,4-dione core was obtained. The imidazolidine-2,4-dione ring, present in phenytoin structure, was modified to obtain less toxic molecules than the maternal compound. Furthermore, to evaluate the role of aromatic rings in the neurotoxic and anticonvulsant activity, two phenyl groups present in the phenytoin structure were replaced by alkyl groups.

Structure–activity relationship studies have demonstrated[12,14–16] that anticonvulsant activity may be enhanced by the presence of the arylpiperazine fragment in the compound structure, and therefore, favorable, selected amines, for example, phenylpiperazines (unsubstituted or substituted with electron-withdrawing atoms), benzylpiperazines, phenylethylpiperazines, and methylpiperazines, have been introduced. To determine whether the introduced modifications affect their pharmacological properties, the designed molecules were evaluated for their anticonvulsant and neurotoxic properties in the Antiepileptic Drug Development (ADD) Program in Epilepsy Branch, National Institutes of Health, National Institute of Neurological Disorders and Stroke (NIH/NINDS), Rockville, MD. Further, the most active compound from the MES test was chosen for the assessment of its antinociceptive activity by using the formalin test and the oxaliplatin-induced neuropathic pain model.

Compounds **3**-**18** were obtained according to Scheme 1. The starting materials, namely 5,5-di(propan-2-yl) (**1**) and 5,5-diphenyl-imidazolidine-2,4-diones (**2**), were either purchased from Merck or prepared in line with Bucherer-Bergs reaction.[18] The final compounds **3**–**18** were obtained via aminoalkylation reaction from appropriately substituted imidazolidine-2,4-diones (**1**,**2**), formaldehyde, and corresponding arylpiperazines. The reaction was carried out in absolute ethanol at reflux for 24 h. The final products after recrystallization from absolute ethanol were obtained as free bases in yields ranging from 60% to 95%. The purity of compound and their structures were confirmed by both elemental and spectral (^{1}H-NMR) analyses (see Supplementary Materials).



R^1, R^2	compd
I-Pr, I-Pr	3-10
Ph, Ph	11-18

R^3	compd
H	3, 8-10, 11, 16-18
3-Cl	4, 12
2-F	5, 13
4-F	6, 14
3-CF_3	7, 15

Z	compd
-	3-7, 11-15
-CH_2-	8, 16
-CH_2CH_2-	9, 17
-CH_3	10, 18

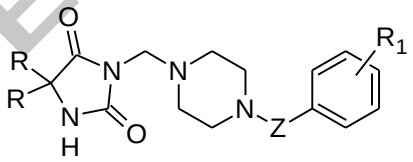
Scheme 1. Reagents and reaction conditions: (a) KCN, $(NH_4)_2CO_3$, 50% ethyl alcohol; (b) 4-substituted piperazines, 37% formaldehyde, 96% ethyl alcohol.

The designed compounds were evaluated for their anticonvulsant activity in the Antiepileptic Drug Development Program (ADD) developed by the NIH (Rockville). Initially, the anticonvulsant activity was established by the maximal electroshock (MES)[19] and subcutaneous pentylenetetrazole (*sc*PTZ) tests,[20] while the neurotoxic properties were measured by the minimal motor impairment rotarod screening test (NT). The preliminary results are presented in Table 1. To identify entities with broad-spectrum anticonvulsant

activity, the most active compounds in the MES and *sc*PTZ tests were chosen for the psychomotor seizure (6 Hz) test. The experimental procedure used for pharmacological studies are described in Supplementary Materials.

In the first step of anticonvulsant screening, the activity of titled compounds was determined by the MES and *sc*PTZ tests. In the MES test, the majority of compounds were effective at doses of 100 or 300 mg/kg at 4 h. Three compounds (**13**, **17,** and **18**) exhibited anticonvulsant activity in this test at the initial dose of 30 mg/kg, but only for compound **18,** the initial dose was gradually reduced to 10 mg/kg (Table 1). Furthermore, eight compounds (**4**, **5**, **10**, **11**, and **15**–**18**) were also active in the MES test at doses of 100 and 300 mg/kg at 0.5 h. Based on the aforementioned results in the MES test, the anticonvulsant activity of 13 compounds can be classified as either long-lasting (**4, 5, 11, and 15–18**) or delayed (**3, 6, 7, and 12–14**) (Table 1), while compound **10** exhibited a rapid-onset and short-lived activity in the same test. It is worth noting that the most potent molecule in this test (**18**), representing methylpiperazine derivative, showed a long-lasting activity up to 4 h at the fixed dose of 10 mg/kg. A slightly weaker effect was observed for its 2-fluorophenyl (**13**) and phenethyl (**17**) analogues, which showed protective effect at 4 h at a dose of 30 mg/kg.

Table 1. The results of screening anticonvulsant activity after *ip* administration into mice (**3**-**18**).



				Intraperitoneal administration in mice[a]						**ASP class**
Compd	**R/R**	**Z**	**R_1**	**MES** [b]		**scPTZ** [c]		**NT** [d]		
				0.5 h	4 h	0.5 h	4 h	0.5 h	4 h	
3	iPr/iPr	-	H	-	300	-	-	-	-	2
4	iPr/iPr	-	3-Cl	100	100	-	-	-	-	1
5	iPr/iPr	-	2-F	100	100	-	100	300	300	1
6	iPr/iPr	-	4-F	-	100	-	-	300	300	1
7	iPr/iPr	-	3-CF_3	-	100	-	-	-	-	1
8	iPr/iPr	-CH_2-	H	-	-	-	-	300	-	3
9	iPr/iPr	-CH_2CH_2-	H	-	-	-	100	300[1]	-	1
10	iPr/iPr	-CH_3	-	300	-	-	-	300	-	4

11	Ph/Ph	-	H	300	100	-	-	-	-	1
12	Ph/Ph	-	3-Cl	-	100	-	-	-	-	1
13	Ph/Ph	-	2-F	-	30	-	-	-	-	1
14	Ph/Ph	-	4-F	-	100	-	-	-	-	1
15	Ph/Ph	-	3-CF_3	300	100	-	-	-	-	1
16	Ph/Ph	-CH_2-	H	300	100	-	-	-	-	1
17	Ph/Ph	-CH_2CH_2-	H	100	30	-	-	-	-	1
18	Ph/Ph	-CH_3	-	10	10	-	-	100	100	1
PHT[e]		-	-	30	30	-	-	100	100	1

[a] Doses of 30, 100, and 300 mg/kg were administered. The data indicate the minimum dose whereby bioactivity was demonstrated. The animals were examined at 0.5 and 4.0 h. A dash indicates the absence of anticonvulsant activity and neurotoxicity at the maximum dose administered (300 mg/kg). iPr – isopropyl; Ph – phenyl; Response comments: [1] death.
[b] Maximal electroshock test
[c] Subcutaneous pentylenetetrazole test
[d] Neurotoxicity screening using rotarod test
[e] Reference drugs, data for phenytoin (PHT) from Ref.[33]

In the *sc*PTZ test, anticonvulsant activity was observed only for compounds **5** and **9** at the dose of 100 mg/kg (Table 1). Thus, both compounds exhibited delayed anticonvulsant activity at 4 h in the scPTZ test. It should be emphasized that only compound **5** was active in both seizure models (MES and scPTZ), whereas the other molecules showed anticonvulsant activity exclusively in one test. In the whole series, only one compound (**8**), representing benzylpiperazine derivative, was completely devoid of any anticonvulsant activity.

Generally, compounds containing 5,5-di(propan-2-yl)imidazolidine-2,4-dione ring (**3**–**10**) were less active and more toxic than their 5,5-diphenyl analogues (**11**–**18**). Only six of the tested compounds (**5**, **6**, **8**–**10,** and **18**) showed neurotoxic activity in the NT test; compounds **5**, **6**, **8**–**10,** and **18** exhibited toxicity at doses of 300 or 100 mg/kg at 0.5 h, and compounds **5, 6,** and **18** were also neurotoxic at 4 h (Table 1). It is worth noting that the dose at which anticonvulsant activity was observed was 3- to 6-fold higher than that at which neurotoxicity appeared.

Oral bioavailability is one of the important properties of clinical candidates as it allows to inhibit convulsions after administration by the oral route. Therefore, the selected compounds that showed the highest activity in mice in the MES test (**3**, **4**, **7**, and **11**–**18**) were examined for their anticonvulsant activity after oral (*po*) administration in rats at a fixed dose of 30 mg/kg. All tested compounds, namely **3**, **4**, **7**, and **11**–**18**, exhibited satisfactory activity (at least in 50% of tested animals) in the MES test after oral administration (Table 2). Among them, two compounds were active at all time points (**11** and **18**), but only compound **18**

afforded protection against convulsions in up to 100% of the examined rats during this period. Both compounds **7** and **13** protected rats against convulsions starting from 0.5 h until 4 h, and compound **17** was active for a shorter time until 2 h. Unexpectedly, compound **3** showed delayed onset of anticonvulsant activity after 6 h, probably due to the action of its metabolites.

Table 2. Anticonvulsant activity and toxicity of selected compounds **3**, **4**, **7** and **11**-**18** administrated orally (30 mg/kg, *po*) into rats.

Compd	MES test [a]						NT [b]					
	0.25 h	0.5 h	1h	2h	4h	6h	0.25 h	0.5 h	1h	2h	4h	6h
3	0	0	0	0	0	2	0	0	0	0	0	0
4	0	0	0	1	2	-	0	0	0	0	0	-
7	0	1	1	2	3	-	0	0	0	0	0	-
11	2	2	1	1	3	-	0	0	0	0	0	-
12	1	0	2	1	1	-	0	0	0	0	0	-
13	0	2	3	3	1	-	0	0	0	0	0	-
14	0	2	0	0	2	-	0	0	0	0	0	-
15	0	1	4	4	4	-	0	0	0	0	0	-
16	1	2	0	0	1	-	0	0	0	0	0	-
17	0	4	1	3	0	-	0	0	0	0	0	-
18	1	3	4	3	1	-	0	0	0	0	0	-
PHT[c]	1	4	3	3	3	-	-	-	-	-	-	-

[a]The data indicate the number of rats of four that were protected at a dose of 30 mg/kg.
[b]Neurotoxicity screening using rotarod test. The data indicate the number of rats of four in which neurotoxicity was observed at a dose of 30 mg/kg.
[c]PHT – phenytoin reference drug, data from Ref.[34]

Subsequently, four compounds (**13, 15, 17,** and **18**) were chosen for the psychomotor seizure test (6 Hz, Table 3). All tested compounds showed satisfactory activity at a dose of 100 mg/kg for various time intervals (compounds **7** and **17** from 1 to 4 h, compounds **13** and **15** only at 2 h, and compound **18** from 0.25 to 4 h). The most promising anticonvulsant properties were detected for compound **18** containing 4-methylpiperazine moiety, which showed up to 100% protection against convulsions at lower doses of 25 and 50 mg/kg, starting from 0.25 to 1 h.

Table 3. Anticonvulsant activity 6 Hz test of selected compounds **7**, **13**, **15**, **17** and **18**, after administrated intraperitoneally into mice (current 32 mA).

Compd	Dose	6 Hz test[a]					NT[b]				
	(mg/kg)	0.25 h	0.5 h	1h	2h	4h	0.25 h	0.5 h	1h	2h	4h

7	100	0	1	2	3	3	0	0	0	0	0
13	100	1	1	1	4	1	0	0	0	0	0
15	100	0	0	1	4	3	0	0	0	0	0
17	50	1	1	3	3	4	0	0	0	0	0
18	25	3	3	4	0	0	0	0	0	0	0
18	50	4	4	4	2	2	0	0	0	0	0
18	100	3	4	4	4	4	3[c]	4[c]	4[c]	4[c]	4[c]

[a]The data indicate the number of mice of four that were protected at a respective dose (mg/kg).
[b]Neurotoxicity screening using rotarod test. The data indicate the number of mice of four in which neurotoxicity was observed at a respective dose (mg/kg).
[c]Number of animals of four which were unable to grasp rotarod or had muscle spasms.

Based on the above preliminary results, in the next step, two of the most potent compounds (**9** and **18**) were chosen for further quantification studies (ED_{50} and TD_{50}) in the MES, *sc*PTZ, and 6 Hz tests, after intraperitoneal into mice and for the most active one after oral administration into rats. The aforementioned parameters were then compared to the data obtained in the same studies for well-known reference anticonvulsant such as phenytoin (Table 4). In the MES test, compound **18** showed slightly lower ED_{50} and TD_{50} values than reference drug phenytoin (e.g. ED_{50} *po* = 38.5 mg/kg vs 28.1 mg/kg, respectively), but its duration of anticonvulsant activity lasted longer than that of phenytoin (TPE: 2 h *po* or 4 h *ip* vs. 1 h *po/ip*). Additionally, compound **9** in *sc*PTZ test displayed 5.5-fold higher activity than phenytoin (ED_{50} values: 89.36 mg/kg vs > 500 mg/kg). It is worth mentioning that compound **18** in the 6 Hz test showed the median effective dose (ED_{50}) of 12.2 mg/kg and a toxic dose of 80.6 mg/kg at a TPE of 1 h. In quantitative studies, compound **18** showed 5-fold lower ED_{50} value (ED_{50} = 12.2 mg/kg) than that obtained for the reference drug, phenytoin (Table 4).

Table 4. Quantification studies of selected compounds **9** and **18** in the MES, *sc*PTZ, 6 Hz and neurotoxicity (NT) tests, after intraperitoneal and oral administration into mice (*ip*) or rats (*po*), respectively.

Compd	**TPE** (h)[a]	**MES ED_{50}**[b] (mg/kg)	***sc*PTZ ED_{50}**[b] (mg/kg)	**6 Hz ED_{50}**[b] (mg/kg)	**NT TD_{50}**[b] (mg/kg)	**PI**[c]
9 (*ip*)	4	-	89.36 (66.39 – 120.3)		> 200	>2.23(*sc*PTZ)
18 (*po*)	2	38.5 (30.6 – 51.7)	> 250		> 500	>12.99(MES)

18 (*ip*)	4 (MES) 1 (6 Hz, NT)	11.14 (9.4 -12.57)	> 80	12.2 (7.6 – 17.8)	75.55 (66.07 – 89.75)	6.78(MES) 6.19 (6 Hz)
PHT[d] (*po*)	1	28.1 (27.7 – 35.20)	> 500		> 1000	>35.58(MES)
PHT[d] (*ip*)	1	5.64 (4.74-6.45)	> 500	> 60[e]	41.00 (39.4 - 43)	7.27(MES)

[a] Time to peak effect.
[b] Results are represented as mean ± SEM at 95% confidence limit (MES-maximal electroshock test; *sc*PTZ-subcutaneous pentylenetetrazole test; 6 Hz test; NT-neurotoxicity).
[c] Protection index (TD_{50}/ED_{50}).
[d] PHT – Phenytoin, reference drug, MES test data from Ref.[34]
[e] 6 Hz test data for PHT from Ref.[32]

Apart from epilepsy, AEDs are widely used for many nonepileptic conditions, including neuropathic pain and other chronic pain types.[21] Hence, in the next stage of our experiments, following the ethical "3R rule,"[22] we selected the most promising compound, that is, compound **18**, to assess its ability to attenuate symptoms of chronic pain. This compound was chosen for further extended *in vivo* studies in mice considering its: (1) high anticonvulsant activity (equal or superior to the reference drugs) in the MES and 6 Hz tests, (2) broad therapeutic index, (3) anticonvulsant activity in both mice and rats, and (4) biological activity after oral administration at doses that were not neurotoxic to experimental animals. We used two mouse pain models reflecting distinct mechanisms of pain formation, namely the formalin test–a model of neurogenic and inflammatory pain,[23] and chemotherapy (oxaliplatin)-induced neuropathic pain model. Because the latter is thought to involve distinct mechanisms of pain development during the early phase (e.g., the overexpression of voltage-gated sodium and calcium channels, altered neuronal excitability, TRPA1 channel activation, and impaired calcium homeostasis) and the late phase (mitochondrial dysfunction, glial cell activation, oxidative stress, axon degeneration, and neuroinflammation),[24,25] the efficacy of analgesics used for pain relief in these two phases of chemotherapy-induced neuropathic pain might also be different.[26–28]

In the formalin test, in both the acute and inflammatory phases, compound **18** did not show a statistically significant activity. However, some nonsignificant effects may be observed in the late phase, 35 min after injection of formalin (Figure 2 and Supplementary Materials). This

discovery is likely to prove indirectly that the test compound **18** is devoid of anti-inflammatory properties and it does not affect nociceptors. This in turn excludes such an anti-inflammatory activity as a mechanism underlying this compound's effect observed *in vivo.*[23]

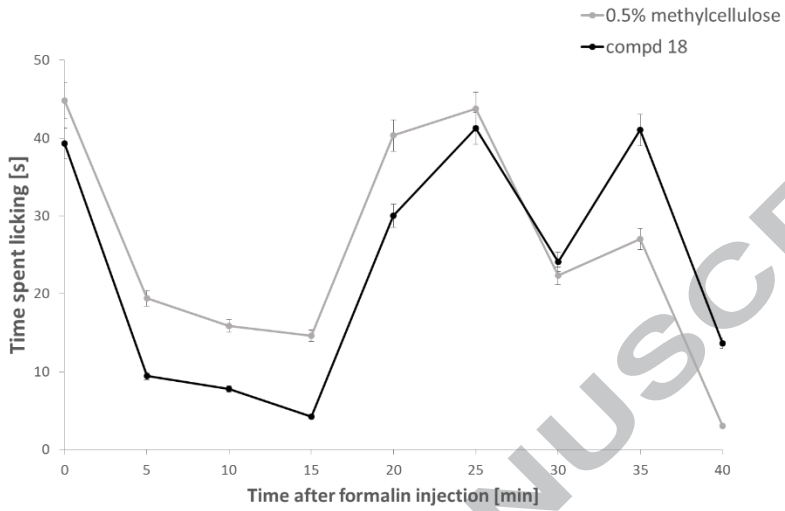


Figure 2. Antinociceptive activity of compound **18** in formalin test, after intraperitoneal administration into mice.

Subsequently, compound **18** was tested in the oxaliplatin-induced neuropathic pain model (Figure 3). In the von Frey test, an overall effect of treatment was observed ($F[4,36] = 21.49$; $p < 0.0001$). In animals not treated with oxaliplatin, the mean force that caused paw withdrawal in the von Frey test was 3.08 ± 0.3 g. Three hours after oxaliplatin injection, a significant reduction of pain sensitivity threshold was observed in mice ($p < 0.0001$ vs before oxaliplatin treatment). In this acute phase in neuropathic animals, the test compound **18** significantly elevated pain sensitivity threshold for mechanical stimulation by 53% ($p < 0.001$ vs pre-drug paw withdrawal force, Figure 3).

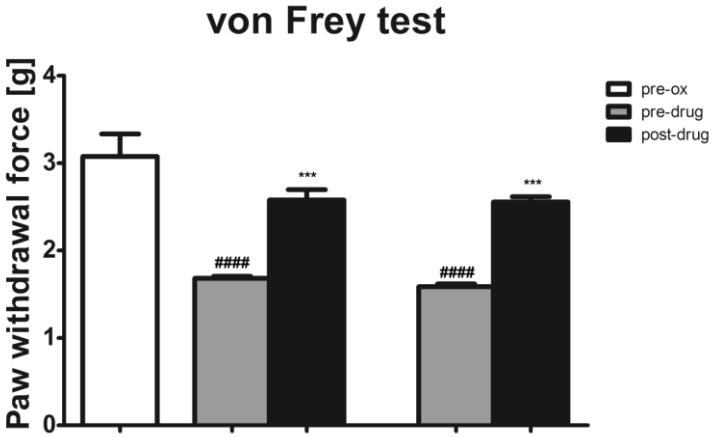


Figure 3. Antiallodynic effect of the compound **18** used at the dose of 11 mg/kg in the von Frey test in oxaliplatin-induced neuropathic pain model in mice. Results are shown as the mean force (±SEM) that caused paw withdrawal. Statistical analysis: repeated measures ANOVA followed by Bonferroni's multiple comparison. Significance *vs.* paw withdrawal force before oxaliplatin administration ('pre-ox'): #### $p < 0.0001$, and *vs.* pre-drug (baseline) paw withdrawal force: *** $p < 0.001$.

In the late phase, that is, 7 days after oxaliplatin injection, a statistically significant ($p < 0.0001$) reduction of pain sensitivity threshold was observed in oxaliplatin-treated mice as compared to that in nontreated animals (Figure 3). Treatment with compound **18** resulted in a statistically significant antiallodynic effect (61%, $p < 0.001$ vs pre-drug paw withdrawal force).

Summarizing, in this experiment, high antiallodynic activity of compound **18** was observed in the oxaliplatin-induced neuropathic pain model. Oxaliplatin is a third-generation platinum-based antitumor drug that induces the formation of DNA crosslinks, thus causing apoptotic death of dividing cells, but it has also affinity for the peripheral nervous system. In rodents, a single injection of oxaliplatin induces painful peripheral neuropathy accompanied by mechanical (tactile) allodynia.[29,30]

In neuropathic animals treated with oxaliplatin, the tested compound **18** elevated the nociceptive threshold in the acute and late phases of tactile allodynia. This *in vivo* study result, together with the results obtained in tests assessing anticonvulsant activity, might suggest potential mechanisms of action of compound **18**. It is thought that drugs that act by blockade of voltage-gated sodium and to a lesser degree calcium channels (with the exception of ethosuximide) are effective in the MES test, and they show no anticonvulsant activity in the PTZ model. In contrast, numerous GABAergic AEDs and the T-type calcium channel blocker ethosuximide are effective in the PTZ model. Mixed AEDs are active in both tests, which is probably due to their multitarget profile of the anticonvulsant activity.[20,31] Bearing that in mind, it can be concluded that the influence of the test compound **18** on ion channels as a mechanism underlying its anticonvulsant and antiallodynic activities should be considered. In terms of the oxaliplatin model of chronic pain, such ion channel-related mechanisms seem to have an impact on the activity of compound **18** mainly in the early-phase oxaliplatin-induced tactile allodynia. The mechanisms underlying the delayed antiallodynic effects of compound **18** remain unclear, but again–considering results from the formalin test, they do not seem to involve attenuation of inflammation. It should be therefore concluded that the

elucidation of the precise mechanism of action of compound **18** requires further extended studies (electrophysiological, biochemical, etc.), and the results obtained in the present research might be helpful for the establishment of directions for mechanistic studies.

The aim of this study was to synthesize a library of new 5,5-diphenyl- and 5,5-di(propan-2-yl)imidazolidine-2,4-dione derivatives and to evaluate their anticonvulsant properties in the "classical" animal models of seizures, namely MES and *sc*PTZ tests. Acute neurotoxicity of these compounds was determined by the rotarod test. After the initial screening, four most promising compounds were tested using the focal seizure model (6 Hz test). In the aforementioned screening evaluation, two compounds, namely **9** and **18**, were selected for further quantitative pharmacological studies. Among them, compound **9** showed significant ED_{50} values and protective index in the *sc*PTZ test, but compound **18** emerged as the most promising molecule with ED_{50} values similar to that of the reference drug phenytoin in the MES test and 4-fold lower ED_{50} dose than phenytoin in the 6 Hz test. Additionally, compound **18** was tested in a formalin model of tonic pain and in an oxaliplatin-induced neuropathic pain model. In the oxaliplatin-induced pain model, this molecule exhibited antiallodynic activity. Taken together, this research identified compound **18** (3-((4-methylpiperazin-1-yl)methyl)-5,5-diphenylimidazolidine-2,4-dione) to show very promising anticonvulsant activity, that is, a broad spectrum of protection against different type of seizures, namely generalized tonic–clonic type (MES) and focal (6 Hz) seizures and to have favorable antiallodynic properties. However, further extensive studies need to be carried out to explain the precise mechanism of action and toxicological effects of this molecule.

Conflict of interest

The authors declare no conflict of interest.

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