4-Amino-5-substituted-3(2*H*)-pyridazinones as Orally Active Antinociceptive Agents: Synthesis and Studies on the Mechanism of Action

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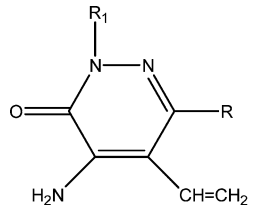
A number of 4-amino-5-vinylpyridazinones and 4-amino-5-heterocyclic-pyridazinones were synthesized and tested for their analgesic activity. Many of these compounds, tested at doses of 3−20 mg kg^{-1} po, showed good antinociceptive activity, reducing by more than 50% the number of writhes with respect to controls. Compounds **16c**, **19a**, **20a**, and **28** were the most potent of the series because they were able to induce a potent antinociceptive effect at a dose of 3 mg kg^{-1} po. None of the active compounds at the analgesic dose provoked any visible change in normal behavior, as demonstrated in the rotarod test. Studies on the mechanism of action showed that the analgesia induced by the active compounds was completely prevented by pretreatment with the α_2-antagonist yohimbine, suggesting an involvement of α_2-adrenoceptors. Further investigation demonstrated an indirect activation of the noradrenergic system through an amplification of noradrenaline release.

Introduction

Pain is widely accepted to be one of the most important determinants of quality of life. A study reported by the World Health Organization demonstrated that individuals who live with persistent pain suffer 4-fold more from depression or anxiety compared to healthy subjects.[1] Moreover, the costs associated with pain are very high, not only in medical terms but also with regard to reduced work productivity.[2] Advances in pharmaceutical research have greatly increased the options for analgesic therapy, which fall under three chief categories: nonsteroidal anti-inflammatory drugs (NSAIDs), opioids, and "analgesic adjuvants".[3] As is well-known, this last category has primary application for conditions other than pain but is also used for specific forms of pain such as neuropathic, musculoskeletal, and cancer pain.[4] In spite of the large number of analgesic drugs available on the market, the research in this field is very lively with more potent molecules devoid of the secondary effects typical of NSAIDs and opioids[5,6] being sought. Since pain is a very complex process in which many neuromodulators are involved[7,8] (i.e., adrenaline, acetylcholine, adenosine, etc.), researchers have also focused their efforts on molecules that work on these systems.[9]

Our research in the field of antinociceptive agents has led us to obtain pyridazine derivatives with very potent analgesic activity.[10−16]

In our previous papers[11,13,16] we reported a number of 4-amino-5-vinyl-3(2*H*)-pyridazinones that exhibited interesting antinociceptive activity, with the most active compounds having an ED_{50} of <20 mg/kg sc in an abdominal constriction test (Figure 1). Structure−activity relationships revealed that modifications at positions 2 and 6 of the pyridazine ring are well-tolerated, while at position 4, the presence of an amino or low alkylamino group is necessary for the activity. Furthermore, replacement of the vinyl group at position 5 with a variety of different functionalities produced inactive compounds. The only modifications allowed are the homologation of the vinyl group (propenyl) and the introduction of a bromine in position β of the vinyl.



A: R= Ph, R_1= Me

B: R= Ph, R_1= nBut

Figure 1. 4-Amino-5-vinyl-3(2*H*)-pyridazinones with antinociceptive activity.

Considering these data, we decided to examine in more depth the structure−activity relationships of the above compounds, performing modifications at position 6 while maintaining at the other positions the groups that award the maximum activity. At the same time, we replaced the vinyl group at position 5 with nitrogen heterocyclic systems. In fact, the vinyl group is potentially toxic,[17] since it could generate the reactive epoxidic function in vivo. Finally, we investigated the mechanism of action of all synthesized compounds.

Design of the new molecules was carried out following a modification of Lipinski's rule of five,[18] which is the most widely accepted technique for identifying druglike compounds. According to this method,[19] our molecules fulfill the following requirements: (a) hydrogen bond donors (OH and NH), ≤5; (b) hydrogen bond acceptors (O and N), ≤10; (c) molecular weight, ≤500; (d) log $P \leq 5$. Evaluation of lipophilicity was performed by measuring log P with ACD/LogP Free LogP calculator. The value met the requirement, since our compounds showed log P in the range 0.22−2.24.

Chemistry

All new compounds were synthesized as reported in Schemes 1−6.

Schemes 1 and 2 depict the synthetic procedures that afford final products **5a**−**i** (Table 2) and **12**, which were modified at

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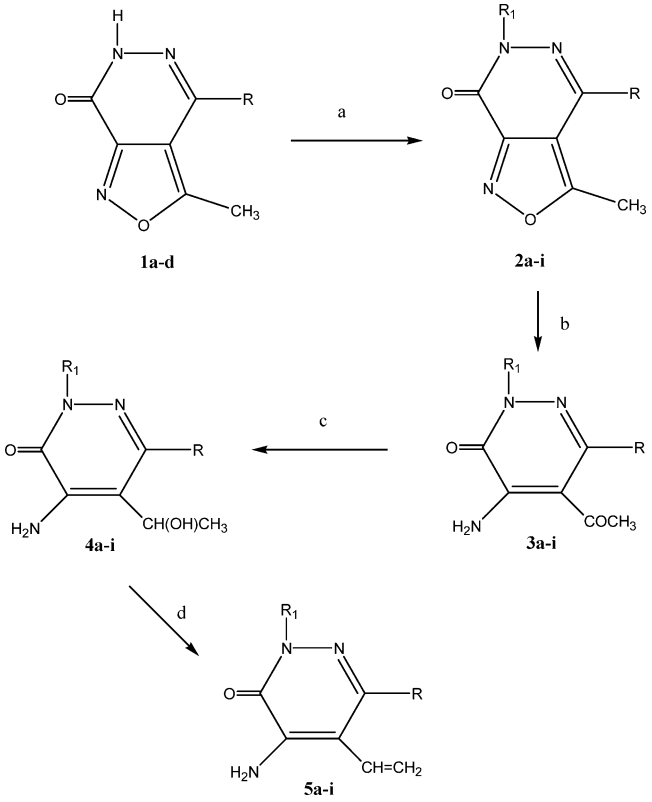
Table 1. Details for Compounds **1a**−**d**

compd	R
1a	4-pyridyl
1b	2-thienyl
1c	4-F−Ph
1d	cyclopentyl

Table 2. Details for Compounds **2**−**5(a**−**i)**

compds **2**−**5**	R	R_1
a	4-pyridyl	nC_4H_9
b	2-thienyl	nC_4H_9
c	4-F−Ph	nC_4H_9
d	4-F−Ph	CH_3
e	cyclopentyl	CH_3
f	4-pyridyl	CH_3
g	2-thienyl	CH_3
h	4-Br−Ph	CH_3
i	CH_3	nC_4H_9

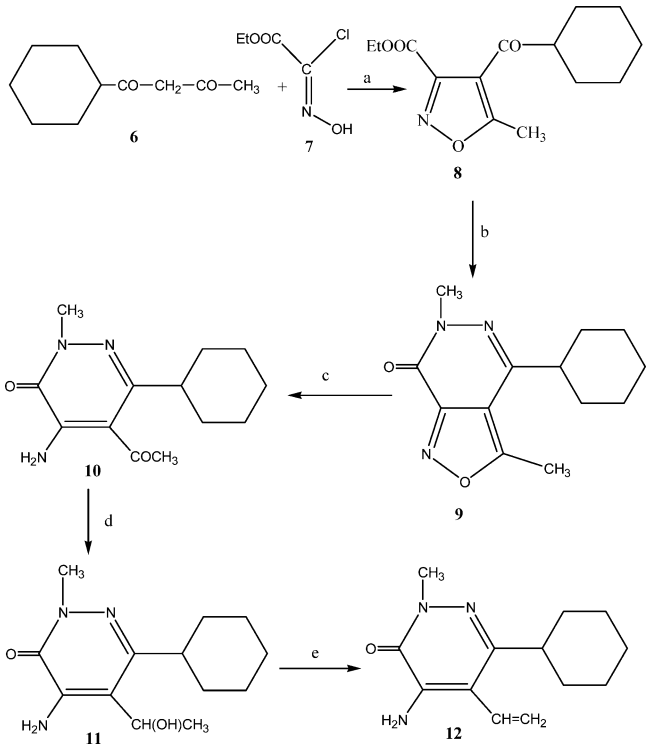
Scheme 1. Synthesis of Final Compounds **5a**−**i**[a]



[a] (a) Appropriate alkyl halide, anhydrous acetone, K_2CO_3, reflux, 2−3 h; (b) 10% Pd/C, ammonium formate, EtOH, reflux, 1 h; (c) $NaBH_4$, MeOH, 40-60 min, room temp; (d) PPA, 60 °C, 2−6 h.

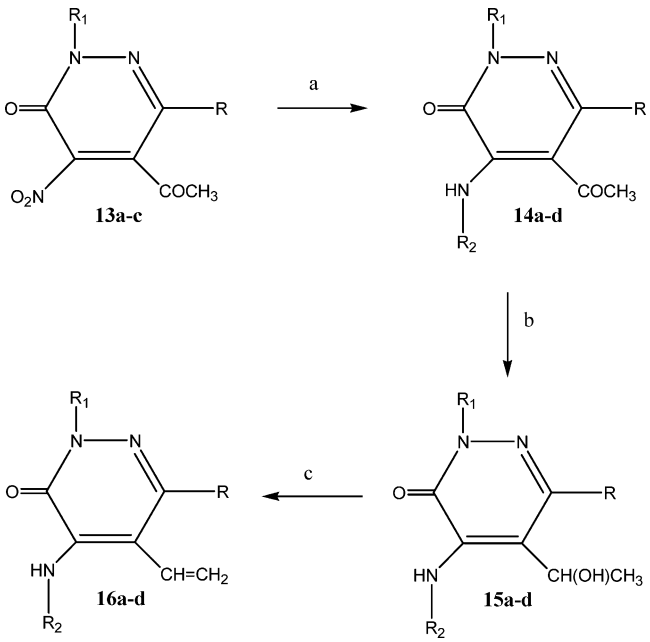
position 6 of the pyridazine ring. The isoxazolo[3,4-*d*]pyridazinones **2a**−**i** represent the starting material (Scheme 1). Compounds **2a**−**e** were prepared by alkylation under standard conditions of the precursors **1a**−**d** (**1a**,[10] **1b**,[20] **1c**,[21] **1d**;[22] Table 1), whereas compounds **2f**−**i** were previously described (**2f**,[10] **2g**,[23] **2h**,[24] **2i**[22]). Reductive cleavage with ammonium formate and Pd/C gave the 4-amino-5-acetyl derivatives **3a**−**i** in good yield. Reduction of these intermediates with sodium borohydride in methanol at room temperature afforded the corresponding secondary alcohols, which were transformed into the final 4-amino-5-vinyl derivatives **5a**−**i** by treatment with polyphosphoric acid (PPA). The same procedure was followed to obtain compound **12** (Scheme 2), but in this case it was necessary to synthesize the 4-cyclohexanecarbonyl-5-methylisoxazole **8** starting from 1-cyclohexylbutane-1,3-dione **6** and commercially available chloro(hydroximino)acetate **7**.

Scheme 2. Synthesis of Compound **12**[a]



[a] (a) NaOEt, anhydrous EtOH, 0 °C; (b) $MeNHNH_2$, EtOH, room temp, 2 h; (c) 10% Pd/C, ammonium formate, EtOH, reflux, 1 h; (d) $NaBH_4$, MeOH, 1 h, room temp; (e) PPA, 60 °C, 2 h.

Scheme 3. Synthesis of Final Compounds **16a**−**d**[a]



[a] (a) Alkylamine, EtOH, room temp, 30−120 min; (b) $NaBH_4$, MeOH, 40−60 min, room temp; (c) PPA, 60 °C, 2−6 h.

Modifications at position 4 of the pyridazine ring (Scheme 3) were performed by displacement of the nitro group in compounds **13a**−**c** (Table 3) with methyl or ethylamine at room temperature (compounds **14a**−**d**). These intermediates were converted into the secondary alcohols **15a**−**d**, which in turn were transformed into the vinyl derivatives **16a**−**d** (Table 4).

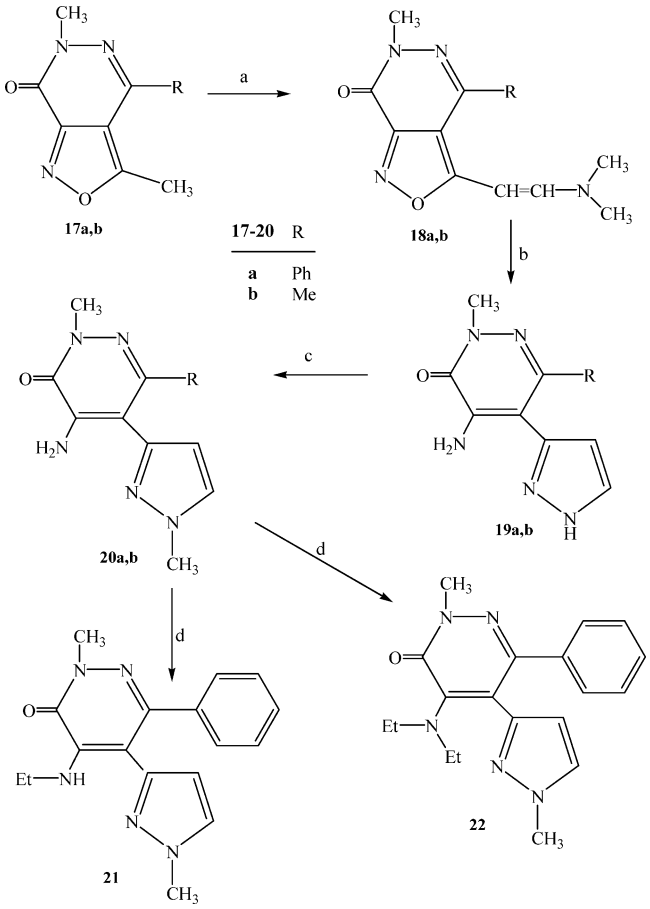
Table 3. Details for Compounds **13a**−**c**

compd	R	R_1
13a	C_4H_9	Ph
13b	CH_3	CH_3
13c	CH_3	Ph

Table 4. Details for Compounds **14**−**16**(**a**−**d**)

compds **14**−**16**	R	R_1	R_2
a	Ph	C_4H_9	CH_3
b	CH_3	CH_3	CH_3
c	Ph	CH_3	C_2H_5
d	CH_3	CH_3	C_2H_5

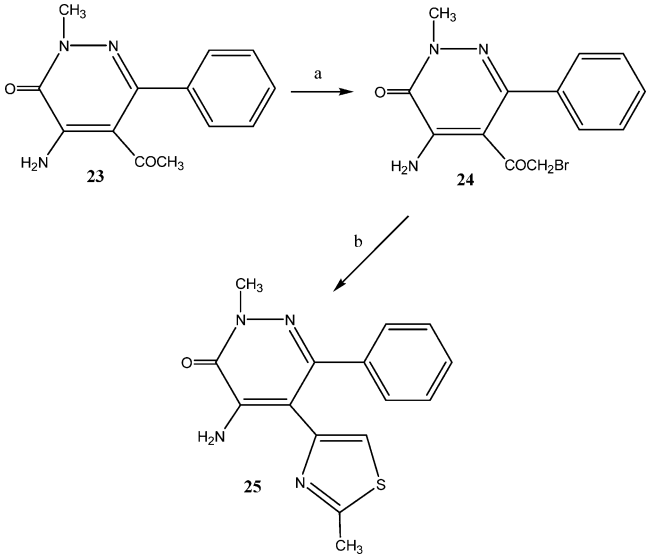
Scheme 4. Synthesis of Final Compounds **19**−**22**[a]



[a] (a) *N*,*N*-Dimethylformammide dimethyl acetal, 100−110 °C, 3−4 h; (b) hydrazine hydrate, EtOH, 80−90 °C, 1−2 h; (c) CH_3I, anhydrous acetone, K_2CO_3, reflux, 2−3 h; (d) for compound **20a**, EtBr, NaH, anhydrous DMSO, 80 °C, 90 min.

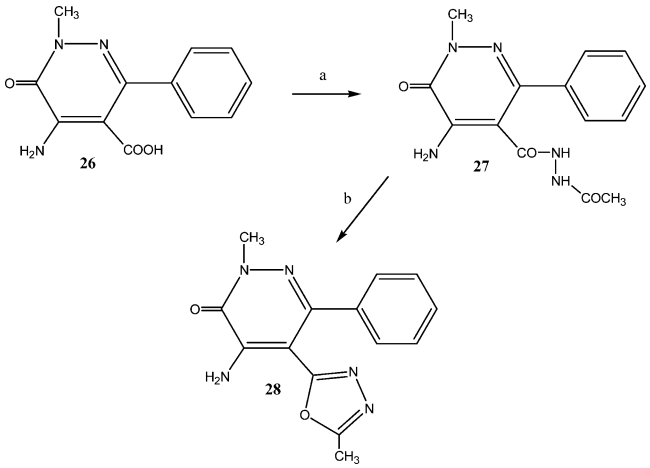
Modification of the vinyl group was performed as in Schemes 4−6 by inserting different heterocycles at position 5. Final compounds **19**−**22** (Scheme 4) were obtained starting from precursors **17a**,**b** which when treated with *N*,*N*-dimethylformamide dimethylacetal, afforded the corresponding 3-dimethylaminovinyl derivatives **18a**,**b**. Treatment with the hydrazine hydrate of **18a**,**b** gave rise to 5-(1*H*)-pyrazolyl derivatives **19a**,**b**, which when alkylated under standard conditions, afforded products **20a**,**b**. The regioselectivity of alkylation was confirmed by a positive NOE effect, detected between the methyl group on the pyrazole ring and the proton at position 5′. Alkylation performed on final compound **20a** under drastic conditions (EtBr, NaH, DMSO at 90 °C) permitted us to obtain compounds **21** and **22**, depending on the amount of ethyl bromide.

Scheme 5. Synthesis of Compound **25**[a]



[a] (a) Br_2, AcOH, HBr, 50 °C, 4 h; (b) thioacetamide, EtOH, 80 °C, 1 h.

Scheme 6. Synthesis of Compound **28**[a]



[a] (a) $SOCl_2$, 60 °C, 1 h, acetohydrazide, anhydrous dioxane, room temp, 3 h; (b) $POCl_3$, 60 °C, 2 h.

The synthesis of 5-methylthiazolyl derivative **25** (Scheme 5) was performed starting from the previously described compound **23**. Treatment of **23** with bromine in acetic acid afforded product **24**, which with thioacetamide in ethanol gave rise to final product **25**.

Finally, the 5-oxadiazolyl derivative **28** (Scheme 6) was synthesized by treatment of precursor **26** with $SOCl_2$. The crude chloride was reacted with acetohydrazide affording compound **27**, which in turn was cyclized with $POCl_3$ to give compound **28**.

Results and Discussion

In the present study, the antinociceptive activity of the investigated compounds was evaluated in the experimental model of the abdominal constriction test in mice in which a painful chemical stimulus was applied. All compounds were administered po,[a] and their activity was compared with that of two representative terms of the previous series (compounds **A** and **B**, Figure 1), which were tested again to make the data uniform. Compounds **5f**, **12**, **16a**−**d**, **19a**, **20a**,**b**, **21**, **25**, and **28**

[a] Abbreviations: po, per os; SGU, signal generation unit; LTQ, linear trap quadruple.

Table 5. Antinociceptive Effect of Final Compounds in the Writhing Test and Effect of Compounds **5a**,**h**,**i**, **12**, **16d**, **19a**, **20a**,**b**, **21**, and **25** on Noradrenaline Extracellular Levels from Rat Cerebral Cortex

treatment[a]	dose (mg kg^{-1})	no. of writhes in the absence of yohimbine	no. of writhes in the presence of yohimbine	% of NA release
CMC		33.2 ± 2.1		100
yohimbine	3		31.7 ± 3.1	
5a	20	35.3 ± 4.4		97.5 ± 8.1
5b	20	25.2 ± 4.2		
5c	20	33.6 ± 4.1		
5d	20	27.2 ± 3.3		
5e	10	27.7 ± 3.9		
5f	20	18.7 ± 3.1[b]	34.1 ± 4.4[c]	
5g	20	30.2 ± 5.1		
5h	20	35.7 ± 3.8		107.0 ± 9.4
5i	20	36.7 ± 4.3		95.9 ± 8.8
12	10	14.4 ± 4.2[b]	32.5 ± 3.4[c]	182.1 ± 9.2[b]
16a	20	21.6 ± 4.5[b]	31.9 ± 2.8[c]	
16b	10	17.6 ± 2.8[b]	32.0 ± 3.3[c]	
16c	3	17.4 ± 3.3[b]	28.1 ± 3.5[c]	
16d	10	16.8 ± 4.9[b]	29.0 ± 4.3[c]	176.2 ± 8.5[b]
19a	3	15.9 ± 3.1[b]	26.5 ± 3.7[c]	168.4 ± 7.7[b]
20a	3	13.6 ± 3.0[b]	31.1 ± 3.5[c]	190.1 ± 10.6[b]
20b	30	15.9 ± 4.3[b]	31.7 ± 3.2[c]	188.5 ± 9.5[b]
21	20	14.1 ± 3.7[b]	29.6 ± 3.2[c]	173.7 ± 6.8[b]
22	20	27.1 ± 4.2		
25	10	14.7 ± 2.7[b]	33.6 ± 3.0[c]	181.3 ± 9.4[b]
28	3	19.5 ± 2.8[b]	34.2 ± 4.1[c]	
A	10	19.7 ± 3.6[b]	29.3 ± 3.1[c]	
B	10	20.4 ± 3.9[b]	33.1 ± 2.6[c]	

[a] All drugs were administered per os 30 min before test. [b] $P < 0.01$ versus CMC treated mice. Each value represents the mean of at least two experiments. [c] $P < 0.05$ in comparison with yohimbine-treated mice. Each value represents the mean of at least two experiments.

were able to reduce the number of abdominal constrictions and showed a potency comparable to that of reference compounds **A** and **B**. In particular **12**, **20a**, **21**, and **25**, tested respectively at 10, 3, 20, and 10 mg kg^{-1} po, were able to exhibit a good antinociceptive activity reducing by more than 50% the number of writhes with respect to controls (Table 5). Antinociceptive effect induced by the above-mentioned compounds was therefore exhibited at an interesting level since these compounds showed potency and efficacy comparable to that shown by some clinically employed analgesic drugs such as diphenhydramine, baclofen, amitriptiline etc..[25] Compounds **16c**, **19a**, **20a** and **28** were the most potent of the series because they induce a statistically significant antinociceptive effect at a dose of 3 mg kg^{-1} po. Compounds **5f**, **16a**,**b**, **16d**, and **20b** reduced by less than 50% the number of abdominal constrictions, and finally compounds **5a**−**e**,**g**−**i** and **22** were devoid of any effect in the abdominal constriction test.

Previously we demonstrated that the analgesia induced by structurally related compounds[15] was completely prevented by pretreatment with the α_2-antagonist yohimbine. In order to clarify whether the α_2-adrenoceptor was involved in the mechanism of action of this series, we pretreated animals with yohimbine as antagonist.

The dose of yohimbine employed to prevent the analgesia induced by the above-reported compounds is the minimal dose able to antagonize antinociception induced by activation of α_2-adrenoceptor as demonstrated by the block exerted on amitriptyline and imipramine antinociception.[26] A complete reversal of the antinociceptive effect of compounds **5f**, **12**, **16a**−**d**, **19a**, **20a**,**b**, **21**, **25**, and **28** by the α_2-antagonist yohimbine (3 mg kg^{-1} ip) was evidenced in the mouse abdominal constriction test, suggesting the involvement of α_2-adrenoceptors in the mechanism of analgesic action of the above-mentioned compounds (Table 5).

Table 6. Effect of **5f**, **12**, **16a**−**d**, **19a**, **20a**,**b**, **21**, **25**, and **28** in the Mouse Rotarod Test[a]

		no. of falls in 30 s			
			after treatment		
treatment	dose po (mg kg^{-1})	before treatment	15 min	30 min	45 min
CMC		4.5 ± 0.3	3.6 ± 0.4	3.0 ± 0.3	2.2 ± 0.4
5f	20	4.8 ± 0.4	3.5 ± 0.3	2.8 ± 0.6	2.1 ± 0.2
12	10	5.0 ± 0.5	4.1 ± 0.5	3.4 ± 0.5	2.9 ± 0.4
16a	20	4.7 ± 0.6	4.0 ± 0.5	3.3 ± 0.3	2.5 ± 0.3
16b	10	4.9 ± 0.7	3.7 ± 0.3	2.5 ± 0.3	2.0 ± 0.2
16c	3	4.4 ± 0.3	3.4 ± 0.5	2.3 ± 0.4	1.9 ± 0.3
16d	10	5.1 ± 0.5	3.9 ± 0.5	3.2 ± 0.4	2.5 ± 0.4
19a	3	4.9 ± 0.5	3.7 ± 0.4	2.8 ± 0.6	2.3 ± 0.5
20a	3	4.8 ± 0.4	4.2 ± 0.6	3.1 ± 0.5	2.3 ± 0.4
20b	30	5.0 ± 0.5	3.9 ± 0.3	2.6 ± 0.4	1.8 ± 0.5
21	20	4.5 ± 0.3	2.6 ± 0.4	1.8 ± 0.4	2.1 ± 0.3
25	10	4.9 ± 0.5	3.8 ± 0.5	2.7 ± 0.3	2.0 ± 0.4
28	3	4.3 ± 0.5	2.7 ± 0.4	2.3 ± 0.5	1.5 ± 0.4

[a] Each value represents the mean at least of five mice.

In our experimental conditions, the α_2-antagonist yohimbine did not modify the pain threshold of mice in comparison with control animals. The lack of effect of this antagonist agrees with results of studies in which this compound did not modify the nociceptive threshold against either thermal (hot-plate) or chemical (writhing) noxious stimuli.[27] We can therefore exclude the possibility that the prevention of the antinociception induced by assayed compounds was due to a hyperalgesic effect of the α_2-adrenoceptor antagonist used.

None of the antinociceptive doses of the compounds provoked any visible change in the normal behavior of the mice as demonstrated in rotarod experiments in which no impairment of mouse rotarod performance was observed (Table 6), as revealed by the decrease of the number of falls in each session.

The α_2-mediated analgesia can be induced by a direct α_2-adrenoceptor activation or by an indirect α_2-adrenoceptor system activation, such as an increase of noradrenaline release. To elucidate whether the antinociceptive mechanism of action was due to an indirect noradrenergic mechanism, noradrenaline extracellular level from rat cerebral cortex treated with most active compounds (**12**, **16d**, **19a**, **20a**,**b**, **21**, **and 25**) was evaluated. Noradrenaline release was determined after administration of the above compounds at the doses able to induce a reduction of writhes in the abdominal constriction test (Table 5). The increase of noradrenaline release exerted by **12** (+182%), **16d** (+176%), **19a** (+168%), **20a** (+190%), **20b** (+188%), **21** (+173%), and **25** (+181%) indicates the existence of a pharmacological correlation between the reduction of writhes and the enhancement of noradrenaline levels. This observation suggests that the antinociceptive action of the above compounds could be due to an indirect activation of the noradrenergic system through an amplification of noradrenergic release. By contrast, **5a**, **5h**, and **5i**, which were not endowed with antinociceptive effect, were also unable to increase noradrenaline release in the same experimental conditions.

Taking into account the fact that the tested compounds were administered by oral route, structure−activity relationships can be only partly determined. In fact the observed activity is a consequence of both pharmacokinetic and pharmacodynamic properties that could be affected differently by structural modifications. Nevertheless, some SARs can be inferred. First of all, we examined the role played by the substituent in position 6 in the subseries of 5-vinyl derivatives in comparison with the lead **A**, keeping the methyl group at position 2 and amino group at position 4. Thus, we observed that replacement of phenyl with 4-fluorophenyl (**5d**), 4-bromophenyl (**5h**), 2-thienyl (**5g**),

and cyclopentyl (**5e**) at the same or at higher doses with respect to the lead **A** was associated with complete loss of activity, whereas the presence at position 6 of 4-pyridyl (**5f**) or cyclohexyl (**12**) determined the appearance of good antinociceptive effects (44% and 57% inhibition of the writhes respectively).

In the same subseries, replacement of the amino group with NHEt (**16c**) in the structure of **A** led to a still active compound (48% inhibition at 3 mg/kg).

Replacement of phenyl with a methyl group was well-tolerated, as demonstrated by the activity of **16b**,**d** in this series and of **20b** in the 5-heterocyclic series.

In this last subseries, good results were obtained with a variety of five-membered systems at position 5. In fact, all the synthesized compounds showed interesting levels of activity (52−59% inhibition at doses ranging from 3 to 20 mg/kg) with the exception of **22**, which proved to be inactive. In this case, the presence of the tertiary amino group NEt_2 in position 4 is probably responsible for the loss of activity. Taken together, the obtained results in this subseries clearly demonstrate that replacement of the metabolically suspect vinyl group with several five-membered heterocycles did not affect the antinociceptive activity. Moreover, these heterocyclic substituted pyridazinones proved to have the same mechanism of action with respect to the 5-vinyl series, as confirmed by results related to antinociception evaluation after pretreatment with yohimbine and percent increase of noradrenaline (Table 5).

Finally, for position 2, methyl groups proved to be better than *n*-butyl substituents at least in the 5-vinyl series, as demonstrated by the inactivity of **5a** and **5i** in comparison with **5f** and **5g**, respectively.

In conclusion, with the present work we found that the best groups around the pyridazinone core are a methyl group at position 2, a primary or secondary amino group at position 4, and a five-membered heterocycle, in particular a methylpyrazole, at position 5. At position 6 a variety of groups like methyl, cyclohexyl, phenyl, and 4-pyridyl are tolerated. Taking into account the fact that all the new compounds were administered by oral route, the obtained results may warrant further development of some representative agents to investigate in more depth the pharmacological profile of the present series.

Experimental Section

Chemistry. All melting points were determined on a Büchi apparatus and are uncorrected. IR spectra were measured as Nujol mulls with a Perkin-Elmer spectrometer (FT-IR, Spectrum 1000). ^{1}H NMR spectra were recorded with Avance 400 instruments (Bruker Biospin, version 002 with SGU). Chemical shifts are reported in ppm, using the solvent as internal standard. Mass spectra (*m*/*z*) were recorded on a LTQ mass spectrometer (ThermoFisher, San Jose, CA). Extracts were dried over Na_2SO_4, and the solvents were removed under reduced pressure. Merck F-254 commercial plates were used for analytical TLC to follow the course of reaction. Silica gel 60 (Merck 70−230 mesh) was used for column chromatography. Reagents and starting material **7** were commercially available.

General Procedure for 2a−e. A mixture of isoxazolopyridazinones **1a**−**d**[10,20−22] (**1a**,[10] **1b**,[20] **1c**,[21] **1d**[22]) (1.2 mmol), K_2CO_3 (2.4−3.6 mmol), and the appropriate alkyl halide (2.0−5.5 mmol) in anhydrous acetone (5 mL) was refluxed under stirring for 2−3 h. Then the mixture was concentrated in vacuo and diluted with cold water, and the precipitate was recovered by suction.

6-*n*-Butyl-3-methyl-4-pyridin-4-ylisoxazolo[3,4-*d*]pyridazin-7(6*H*)-one 2a. Yield = 69%; mp = 120−121 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H,$(CH_2)_3CH_3$), 1.40−1.50 (m, 2H, $(CH_2)_2CH_2CH_2CH_3$), 1.85 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.70 (s, 3H, 3-CH_3), 4.25 (t, 2H, $CH_2CH_2CH_2CH_3$), 7.65 (d, 2H, Ar), 8.85 (d, 2H, Ar).

6-*n*-Butyl-3-methyl-4-thiophen-2-ylisoxazolo[3,4-*d*]pyridazin-7(6*H*)-one 2b. Yield = 73%; mp = 92−95 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H,$(CH_2)_3CH_3$), 1.40−1.50 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.80−1.90 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.75 (s, 3H, 3-CH_3), 4.20 (t, 2H, $CH_2CH_2CH_2CH_3$), 7.20 (m, 1H, Ar), 7.35 (m, 1H, Ar), 7.50 (m, 1H, Ar).

6-*n*-Butyl-4-(4-fluorophenyl)-3-methylisoxazolo[3,4-*d*]pyridazin-7(6*H*)-one 2c. Yield = 93%; mp = 97−100 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H, $(CH_2)_3CH_3$), 1.40 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.80−1.90 (m, 5H (3H, 3-CH_3; 2H, $CH_2CH_2CH_2CH_3$)), 4.20 (t, 2H, $CH_2CH_2CH_2CH_3$), 7.15 (m, 2H, Ar), 7.45 (m, 2H, Ar).

4-(4-Fluorophenyl)-3,6-dimethylisoxazolo[3,4-*d*]pyridazin-7(6*H*)-one 2d. Yield = 71%; mp = 223−225 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 2.55 (s, 3H, 3-CH_3), 3.85 (s, 3H, N−CH_3), 7.25 (m, 2H, Ar), 7.55 (m, 2H, Ar).

4-Cyclopentyl-3,6-dimethylisoxazolo[3,4-*d*]pyridazin-7(6*H*)-one 2e. Yield = 20%; mp = 103−104 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.45−2.05 (m, 8H, *c*C_5H_9), 2.90 (s, 3H, 3-CH_3), 3.25−3.40 (m, 1H, *c*C_5H_9), 3.75 (s, 3H, N−CH_3).

General Procedure for 3a−i. A mixture of appropriate 2-substituted isoxazolo[3,4-*d*]pyridazinones **2a**−**i** (**2f**,[10] **2g**,[23] **2h**,[24] **2i**[22]) (0.6 mmol), 10% Pd/C (30−40 mg), and ammonium formate (2.0 mmol) in EtOH (5−6 mL) was refluxed for 1 h. After the mixture was cooled, CH_2Cl_2 (6 mL) was added, the catalyst was filtered off, and the solvent was evaporated in vacuo to afford **3a**−**i**.

5-Acetyl-4-amino-2-*n*-butyl-6-pyridin-4-ylpyridazin-3(2*H*)-one 3a. Yield = 80%; mp = 103 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H,$(CH_2)_3CH_3$), 1.45 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.80−1.90 (m, 5H (3H, $COCH_3$; 2H, $CH_2CH_2CH_2CH_3$)), 4.20 (t, 2H, $CH_2CH_2CH_2CH_3$), 6.90 (exch br s, 2H, NH_2), 7.50 (d, 2H, Ar), 8.75 (d, 2H, Ar).

5-Acetyl-4-amino-2-*n*-butyl-6-thiophen-2-ylpyridazin-3(2*H*)-one 3b. Yield = 82%; mp = 109−112 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H,$(CH_2)_3CH_3$), 1.45 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.85 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.00 (s, 3H, $COCH_3$), 4.20 (t, 2H, $CH_2CH_2CH_2CH_3$), 7.10 (m, 2H, Ar), 7.45 (m, 1H, Ar), 7.55 (exch br s, 2H, NH_2).

5-Acetyl-4-amino-2-butyl-6-(4-fluorophenyl)pyridazin-3(2*H*)-one 3c. Yield = 95%; mp = 143−145 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.00 (m, 3H,$(CH_2)_3CH_3$), 1.40 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.75−1.85 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.55 (s, 3H, $COCH_3$), 4.20 (t, 2H, $CH_2CH_2CH_2CH_3$), 6.80 (exch br s, 2H, NH_2), 7.25 (m, 2H, Ar), 7.55 (m, 2H, Ar).

5-Acetyl-4-amino-6-(4-fluorophenyl)-2-methylpyridazin-3(2*H*)-one 3d. Yield = 79%; mp = 215−216 °C, dec (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.80 (s, 3H, $COCH_3$), 3.85 (s, 3H, N−CH_3), 7.15 (m, 2H, Ar), 7.45 (m, 2H, Ar), 7.65 (exch br s, 2H, NH_2).

5-Acetyl-4-amino-6-cyclopentyl-2-methylpyridazin-3(2*H*)-one 3e. Yield = 80%; mp = 106−107 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.50−2.00 (m, 8H, *c*C_5H_9), 2.60 (s, 3H, $COCH_3$), 3.20−3.35 (m, 1H, *c*C_5H_9), 3.75 (s, 3H, N−CH_3), 6.95 (exch br s, 2H, NH_2).

5-Acetyl-4-amino-2-methyl-6-pyridin-4-ylpyridazin-3(2*H*)-one 3f. Yield = 84%; mp = 195−198 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.90 (s, 3H, $COCH_3$), 3.85 (s, 3H, N−CH_3), 6.90 (exch br s, 2H, NH_2), 7.55 (d, 2H, Ar), 8.75 (d, 2H, Ar).

5-Acetyl-4-amino-2-methyl-6-thiophen-2-ylpyridazin-3(2*H*)-one 3g. Yield = 82%; mp = 163−166 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 2.00 (s, 3H, $COCH_3$), 3.85 (s, 3H, N−CH_3), 5.85 (exch br s, 2H, NH_2), 7.10 (m, 2H, Ar), 7.50 (m, 1H, Ar).

5-Acetyl-4-amino-6-(4-bromophenyl)-2-methylpyridazin-3(2*H*)-one 3h. Yield = 87%; mp = 203−206 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.85 (s, 3H, $COCH_3$), 3.85 (s, 3H, N−CH_3), 5.90 (exch br s, 2H, NH_2), 7.30−7.50 (m, 4H, Ar).

5-Acetyl-4-amino-2-*n*-butyl-6-methylpyridazin-3(2*H*)-one 3i. Yield = 79%; mp = 90−93 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.40 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.55 (s, 3H, $COCH_3$), 2.60 (s, 3H, 6-CH_3), 4.10 (t, 2H, $CH_2CH_2CH_2CH_3$), 7.75 (exch br s, 2H, NH_2).

General Procedure for 4a−i. To a stirred solution of the appropriate 4-amino-5-acetyl derivative **3a−i** (0.35 mmol) in MeOH (3−6 mL), sodium borohydride (4.0−4.5 mmol) was added portionwise. The reaction mixture was stirred for 40−60 min at room temperature, concentrated in vacuo, and diluted with cold water. The final products were recovered by suction.

4-Amino-2-*n*-butyl-5-(1-hydroxyethyl)-6-pyridin-4-ylpyridazin-3(2*H*)-one 4a. Yield = 87%; mp = 150−153 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.30−1.40 (m, 5H (3H, CH(OH)CH_3; 2H, $(CH_2)_2CH_2CH_3$)), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.80 (exch br s, 1H, OH), 4.20 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.75 (m, 1H, $CH(OH)CH_3$), 6.00 (exch br s, 2H, NH_2), 7.40 (m, 2H, Ar), 8.80 (m, 2H, Ar).

4-Amino-2-*n*-butyl-5-(1-hydroxyethyl)-6-thiophen-2-ylpyridazin-3(2*H*)-one 4b. Yield = 89%; mp = 84−86 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.40 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.55 (d, 3H, CH(OH)CH_3), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.20 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.30 (exch br s, 1H, OH), 5.10 (q, 1H, $CH(OH)CH_3$), 7.10 (m, 2H, Ar), 7.40 (m, 1H, Ar), 7.65 (exch br s, 2H, NH_2).

4-Amino-2-*n*-butyl-6-(4-fluorophenyl)-5-(1-hydroxyethyl)pyridazin-3(2*H*)-one 4c. Yield = 86%; mp = 119−120 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.40−1.55 (m, 5H (3H, CH_3CH(OH); 2H, $(CH_2)_2CH_2CH_3$)), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.20 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.30 (exch br s, 1H, OH), 4.80 (q, 1H, $CH(OH)CH_3$), 5.95 (exch br s, 2H, NH_2), 7.20 (m, 2H, Ar), 7.30 (m, 2H, Ar).

4-Amino-6-(4-fluorophenyl)-5-(1-hydroxyethyl)-2-methylpyridazin-3(2*H*)-one 4d. Yield = 66%; mp = 208−210 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.45 (d, 3H, CH(OH)CH_3), 2.00 (exch br s, 1H, OH), 3.80 (s, 3H, N−CH_3), 4.80 (q, 1H, $CH(OH)CH_3$), 5.95 (exch br s, 2H, NH_2), 7.15 (m, 2H, Ar), 7.30 (m, 2H, Ar).

4-Amino-6-cyclopentyl-5-(1-hydroxyethyl)-2-methylpyridazin-3(2*H*)-one 4e. Yield = 70%; mp = 171−173 °C (cyclohexane); ^{1}H NMR ($CDCl_3$) δ 1.55 (d, 3H, CH(OH)CH_3), 1.70−2.00 (m, 8H, cC_5H_9), 2.90−3.00 (m, 1H, cC_5H_9), 3.75 (s, 3H, N−CH_3), 4.50 (exch br s, 1H, OH), 5.20 (q, 1H, $CH(OH)CH_3$), 5.75 (exch br s, 2H, NH_2).

4-Amino-5-(1-hydroxyethyl)-2-methyl-6-pyridin-4-ylpyridazin-3(2*H*)-one 4f. Yield = 60%; mp = 217−220 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.50 (d, 3H, CH(OH)CH_3), 3.80 (s, 3H, N−CH_3), 4.10 (exch br s, 1H, OH), 4.75 (q, 1H, $CH(OH)CH_3$), 6.00 (exch br s, 2H, NH_2), 7.30 (d, 2H, Ar), 8.70 (d, 2H, Ar).

4-Amino-5-(1-hydroxyethyl)-2-methyl-6-thiophen-2-ylpyridazin-3(2*H*)-one 4g. Yield = 69%; mp = 220−221 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.55 (d, 3H, CH(OH)CH_3), 3.85 (s, 3H, N−CH_3), 4.85 (exch br s, 1H, OH), 5.10 (m, 1H, $CH(OH)CH_3$), 6.20 (exch br s, 2H, NH_2), 7.10 (m, 2H, Ar), 7.40 (m, 1H, Ar).

4-Amino-6-(4-bromophenyl)-5-(1-hydroxyethyl)-2-methylpyridazin-3(2*H*)-one 4h. Yield = 60%; mp = 223−226 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.45 (d, 3H, CH(OH)CH_3), 3.80 (s, 3H, N−CH_3), 4.50 (exch br s, 1H, OH), 4.80 (q, 1H, $CH(OH)CH_3$), 5.95 (exch br s, 2H, NH_2), 7.35 (d, 2H, Ar), 7.45 (d, 2H, Ar).

4-Amino-2-*n*-butyl-5-(1-hydroxyethyl)-6-methylpyridazin-3(2*H*)-one 4i. Yield = 53%; mp = 75−77 °C (cyclohexane); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.35−1.50 (m, 5H (3H, CH_3CH(OH); 2H, $(CH_2)_2CH_2CH_3$)), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.20 (s, 3H, 6-CH_3), 4.10 (t, 2H, $CH_2CH_2CH_2CH_3$), 4.30 (exch br s, 1H, OH), 5.05 (q, 1H, $CH(OH)CH_3$), 6.80 (exch br s, 2H, NH_2).

General Procedure for 5a−i. The appropriate derivative **4a−i** (0.35 mmol) was treated with PPA (35 mmol) at 60 °C for 2−6 h. After treatment with ice−water (20 mL), the mixture was neutralized with 3 N NaOH and compounds **5a−i** were recovered by suction.

4-Amino-2-*n*-butyl-6-pyridin-4-yl-5-vinylpyridazin-3(2*H*)-one 5a. Yield = 64%; mp = 76−78 °C (EtOH); IR (cm^{-1}) 3390 and 3420 (NH_2), 1640 (CO); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H, $(CH_2)_3CH_3$), 1.40 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.20 (t, 2H, $CH_2CH_2CH_2CH_3$), 5.55 (exch br s, 2H, NH_2), 5.60 (d, 1H, CH=CH_2), 5.75 (d, 1H, CH=CH_2), 6.30 (dd, 1H, CH=CH_2), 7.50 (d, 2H, Ar), 8.80 (d, 2H, Ar); MS (ESI) *m*/*z* 271.15 ($[M + H]^+$).

4-Amino-2-*n*-butyl-6-thiophen-2-yl-5-vinylpyridazin-3(2*H*)-one 5b. Yield = 80%; mp = 80−83 °C (EtOH); IR (cm^{-1}) 3375 and 3400 (NH_2), 1635 (CO); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H, $(CH_2)_3CH_3$), 1.40−1.50 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.80−1.90 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.20 (m, 2H, $CH_2CH_2CH_2CH_3$), 5.40 (exch br s, 1H, NH_2), 5.65 (d, 1H, CH=CH_2), 5.80 (d, 1H, CH=CH_2), 6.60 (dd, 1H, CH=CH_2), 7.05 (m, 1H, Ar), 7.35 (m, 2H, Ar); MS (ESI) *m*/*z* 276.37 ($[M + H]^+$).

4-Amino-2-*n*-butyl-6-(4-fluorophenyl)-5-vinylpyridazin-3(2*H*)-one 5c. Yield = 89%; mp = 83−85 °C (EtOH); IR (cm^{-1}) 3380 and 3435 (NH_2), 1645 (CO); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H, $(CH_2)_3CH_3$), 1.40 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.85 (m, 2H, $CH_2CH_2CH_2CH_3$), 4.25 (t, 2H, $CH_2CH_2CH_2CH_3$), 5.45 (exch br s, 2H, NH_2), 5.50 (d, 1H, CH=CH_2), 5.70 (d, 1H, CH=CH_2), 6.25 (dd, 1H, CH=CH_2), 7.15 (m, 2H, Ar), 7.45 (m, 2H, Ar); MS (ESI) *m*/*z* 288.33 ($[M + H]^+$).

4-Amino-6-(4-fluorophenyl)-2-methyl-5-vinylpyridazin-3(2*H*)-one 5d. Yield = 72%; mp = 74−76 °C (EtOH); IR (cm^{-1}) 3385 and 3420 (NH_2), 1640 (CO); ^{1}H NMR ($CDCl_3$) δ 3.85 (s, 3H, N−CH_3), 5.45 (exch br s, 2H, NH_2), 5.55 (d, 1H, CH=CH_2), 5.65 (d, 1H, CH=CH_2), 6.25 (dd, 1H, CH=CH_2), 7.05−7.15 (m, 2H, Ar), 7.40−7.50 (m, 2H, Ar); MS (ESI) *m*/*z* 246.25 ($[M + H]^+$).

4-Amino-6-cyclopentyl-2-methyl-5-vinylpyridazin-3(2*H*)-one 5e. Yield = 49%; mp = 74−75 °C (EtOH); IR (cm^{-1}) 3375 and 3450 (NH_2), 1620 (CO); ^{1}H NMR ($CDCl_3$) δ 1.70−1.95 (m, 8H, cC_5H_9), 2.95−3.10 (m, 1H, cC_5H_9), 3.75 (s, 3H, N−CH_3), 5.15 (exch br s, 2H, NH_2), 5.55 (d, 1H, CH=CH_2), 5.75 (d, 1H, CH=CH_2), 6.50−6.65 (m, 1H, CH=CH_2); MS (ESI) *m*/*z* 220.18 ($[M + H]^+$).

4-Amino-2-methyl-6-pyridin-4-yl-5-vinylpyridazin-3(2*H*)-one 5f. Yield = 86%; mp = 172−174 °C (EtOH); IR (cm^{-1}) 3370 and 3450 (NH_2), 1625 (CO); ^{1}H NMR ($CDCl_3$) δ 3.85 (s, 3H, N−CH_3), 5.60 (d, 1H, CH=CH_2), 5.70 (d, 1H, CH=CH_2), 5.80 (exch br s, 2H, NH_2), 6.30 (dd, 1H, CH=CH_2), 7.40 (d, 2H, Ar), 8.70 (d, 2H, Ar); MS (ESI) *m*/*z* 229.25 ($[M + H]^+$).

4-Amino-2-methyl-6-thiophen-2-yl-5-vinylpyridazin-3(2*H*)-one 5g. Yield = 65%; mp = 95−98 °C (EtOH); IR (cm^{-1}) 3390 and 3410 (NH_2), 1645 (CO); ^{1}H NMR ($CDCl_3$) δ 3.85 (s, 3H, N−CH_3), 5.40 (exch br s, 2H, NH_2), 5.65 (d, 1H, CH=CH_2), 5.80 (d, 1H, CH=CH_2), 6.60 (dd, 1H, CH=CH_2), 7.10 (m, 1H, Ar), 7.30−7.40 (m, 2H, Ar); MS (ESI) *m*/*z* 234.29 ($[M + H]^+$).

4-Amino-6-(4-bromophenyl)-2-methyl-5-vinylpyridazin-3(2*H*)-one 5h. Yield = 85%; mp = 98−101 °C (EtOH); IR (cm^{-1}) 3375 and 3470 (NH_2), 1635 (CO); ^{1}H NMR ($CDCl_3$) δ 3.85 (s, 3H, N−CH_3), 5.45 (exch br s, 2H, NH_2), 5.60 (m, 2H, CH=CH_2), 6.30 (dd, 1H, CH=CH_2), 7.30−7.60 (m, 4H, Ar); MS (ESI) *m*/*z* 307.16 ($[M + H]^+$).

4-Amino-2-*n*-butyl-6-methyl-5-vinylpyridazin-3(2*H*)-one 5i. Yield = 81%; mp = 40−43 °C (EtOH); IR (cm^{-1}) 3330 and 3430 (NH_2), 1630 (CO); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.35−1.45 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.25 (s, 3H, 6-CH_3), 4.10 (t, 2H, $CH_2CH_2CH_2CH_3$), 5.20 (exch br s, 2H, NH_2), 5.60 (d, 1H, CH=CH_2), 5.70 (d, 1H, CH=CH_2), 6.50 (dd, 1H, CH=CH_2); MS (ESI) *m*/*z* 208.14 ($[M + H]^+$).

4-Cyclohexanecarbonyl-5-methylisoxazole-3-carboxylic Acid Ethyl Ester 8. To a cooled (0 C°) and stirred solution of sodium ethoxide, obtained from sodium (9.15 mmol) and anhydrous ethanol (30 mL), a solution of 1-cyclohexylbutane-1,3-dione **6**[28] (9.3 mmoL) in the same solvent (13 mL) was slowly added. After the mixture was further cooled to −5 °C, a solution of ethyl chloro(hydroximino)acetate **7** (6.2 mmol) in anhydrous EtOH (10.5 mL) was added dropwise. The mixture was neutralized with 6 N HCl, and the solvent was evaporated in vacuo. The residue oil was washed with cold 0.5 N NaOH and cold water, and finally it was extracted with CH_2Cl_2 (3 × 20 mL). Evaporation of the solvent afforded compound **8**, which was purified by column chromatography using cyclohexane/ethyl acetate, 9:1, as eluent. Yield = 28%; oil; ^{1}H NMR ($CDCl_3$) δ 1.20−1.90 (m, 10H, cC_6H_{11}), 1.45 (t, 3H,

CH_2CH_3), 2.60 (s, 3H, 5-CH_3), 2.85−2.95 (m, 1H, cC_6H_{11}), 4.50 (q, 2H, CH_2CH_3).

4-Cyclohexyl-3,6-dimethylisoxazolo[3,4-*d*]pyridazin-7(6*H*)-one 9. To a solution of **8** (0.75 mmol) in EtOH (2 mL), methylhydrazine (4.2 mmol) was added. The mixture was stirred at room temperature for 2 h. After dilution with cold water (10−15 mL), the suspension was extracted with ethyl acetate (3 × 20 mL) and the solvent was evaporated to afford **9**, which was purified by column chromatography using cyclohexane/ethyl acetate, 2:1, as eluent. Yield = 24%; mp = 166−167 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.25−2.00 (m, 10H, cC_6H_{11}), 2.70−2.80 (m, 1H, cC_6H_{11}), 2.85 (s, 3H, 3-CH_3), 3.75 (s, 3H, N−CH_3).

5-Acetyl-4-amino-6-cyclohexyl-2-methylpyridazin-3(2*H*)-one 10. Compound **10** was obtained from compound **9** following the general procedure described for **3a**−**i**. Yield = 54%; mp = 133−134 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.25−1.90 (m, 10H, cC_6H_{11}), 2.55 (s, 3H, $COCH_3$), 2.75−2.85 (m, 1H, cC_6H_{11}), 3.75 (s, 3H, N−CH_3), 7.00 (exch br s, 2H, NH_2).

4-Amino-6-cyclohexyl-5-(1-hydroxyethyl)-2-methylpyridazin-3(2*H*)-one 11. Compound **11** was obtained starting from **10** following the general procedure described for **4a**−**i**. Yield = 34%; mp = 203−204 °C (cyclohexane); ^{1}H NMR ($CDCl_3$) δ 1.25−1.90 (m, 13H (10H, cC_6H_{11}; 3H, CH(OH)CH_3)), 2.40−2.50 (m, 1H, cC_6H_{11}), 3.75 (s, 3H, N−CH_3), 5.15 (q, 1H, CH(OH)CH_3), 5.80 (exch br s, 2H, NH_2).

4-Amino-6-cyclohexyl-2-methyl-5-vinylpyridazin-3(2*H*)-one 12. Compound **12** was obtained starting from **11** following the general procedure described for **5a**−**i**. Yield = 65%; mp = 118−119 °C (EtOH); IR (cm^{-1}) 3385 and 3430 (NH_2), 1640 (CO); ^{1}H NMR ($CDCl_3$) δ 1.20−1.90 (m, 10H, cC_6H_{11}), 2.45−2.60 (m, 1H, cC_6H_{11}), 3.75 (s, 3H, N−CH_3), 5.15 (exch br s, 2H, NH_2), 5.55 (d, 1H, CH=CH_2), 5.75 (d, 1H, CH=CH_2), 6.50 (dd, 1H, CH=CH_2); MS (ESI) *m*/*z* 234.18 ([M + H]$^+$).

General Procedure for Compounds 14a−d. A mixture of the required 5-acetyl-2-alkyl-4-nitropyridazinone **13a**−**c**[29,30] (0.3 mmol) and the appropriate amine (1.5−1.8 mmol) in EtOH (3−4 mL) was stirred at room temperature for 30−120 min. Then the reaction mixture was concentrated in vacuo and addition of cold water (10 mL) furnished compounds **14a**,**b**, which were recovered by suction. For compounds **14c**,**d** the suspension was extracted with CH_2Cl_2 (3 × 15 mL) and evaporation of the solvent afforded a crude precipitate for **14c** and an oil for **14d**. This last one was purified by column chromatography using cyclohexane/ethyl acetate/methanol, 1:2:0.1, as eluent.

5-Acetyl-2-*n*-butyl-4-methylamino-6-phenylpyridazin-3(2*H*)-one 14a. Yield = 45%; mp = 105 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.45 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.75−1.90 (m, 5H (3H, $COCH_3$; 2H, $CH_2CH_2CH_2CH_3$)), 2.95 (d, 3H, NH−CH_3), 4.15 (t, 2H, $CH_2CH_2CH_2CH_3$), 6.25 (exch br m, 1H, *NH*-CH_3), 7.50 (m, 5H, Ar).

5-Acetyl-2,6-dimethyl-4-methylaminopyridazin-3(2*H*)-one 14b. Yield = 57%; mp = 99−102 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 2.20 (s, 3H, $COCH_3$), 2.50 (s, 3H, 6-CH_3), 2.90 (d, 3H, NH-CH_3), 3.75 (s, 3H, N−CH_3), 5.50 (exch br m, 1H, *NH*-CH_3).

5-Acetyl-4-ethylamino-2-methyl-6-phenylpyridazin-3(2*H*)-one 14c. Yield = 71%; mp = 136−138 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 1.30 (t, 3H, CH_2CH_3), 1.90 (s, 3H, $COCH_3$), 3.35 (m, 2H, CH_2CH_3), 3.80 (s, 3H, N−CH_3), 4.85 (exch br m, 1H, *NH*-CH_2CH_3), 7.45 (s, 5H, Ar).

5-Acetyl-4-ethylamino-2,6-dimethylpyridazin-3(2*H*)-one 14d. Yield = 35%; mp = 85−86 °C (cyclohexane); ^{1}H NMR ($CDCl_3$) δ 1.30 (t, 3H, CH_2CH_3), 2.20 (s, 3H, $COCH_3$), 2.50 (s, 3H, 6-CH_3), 3.15 (m, 2H, CH_2CH_3), 3.75 (s, 3H, N−CH_3), 6.15 (exch br s, 1H, *NH*−CH_2CH_3).

General Procedure for 15a−d. Compounds **15a**−**d** were obtained starting from **14a**−**d** following the same general procedure described for **4a**−**i**. **15c** was obtained after extraction with ethyl acetate (3 × 20 mL) and evaporation in vacuo.

2-*n*-Butyl-5-(1-hydroxyethyl)-4-methylamino-6-phenylpyridazin-3(2*H*)-one 15a. Yield = 79%; mp = 102 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 0.95 (t, 3H,$(CH_2)_3CH_3$), 1.30−1.50 (m, 5H (3H, CH_3CH-(OH)); 2H, $(CH_2)_2CH_2CH_3$), 1.80 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.50 (exch br s, 1H, OH),3.40 (d, 3H, NH−CH_3), 4.15 (t, 2H, $CH_2CH_2CH_2CH_3$), 4.80 (q, 1H, CH(OH)CH_3), 6.20 (exch br s, 1H, *NH*−CH_3), 7.30−7.45 (m, 5H, Ar).

5-(1-Hydroxyethyl)-2,6-dimethyl-4-methylaminopyridazin-3(2*H*)-one 15b. Yield = 84%; oil (purified by column chromatography using $CHCl_3$/CH_3OH, 9:1, as eluent); ^{1}H NMR ($CDCl_3$) δ 1.50 (d, 3H, CH(OH)CH_3), 2.25 (s, 3H, 6-CH_3), 3.25 (d, 3H, NH-CH_3), 3.65 (s, 3H, N−CH_3), 4.60 (exch br s, 1H, OH), 5.10 (q, 1H, CH(OH)CH_3), 6.10 (exch br s, 1H, *NH*−CH_3).

4-Ethylamino-5-(1-hydroxyethyl)-2-methyl-6-phenylpyridazin-3(2*H*)-one 15c. Yield = 77%; mp = 137−138 °C (cyclohexane); ^{1}H NMR ($CDCl_3$) δ 1.30 (t, 3H, CH_2CH_3), 1.45 (d, 3H, CH(OH)CH_3), 3.75 (s, 3H, N−CH_3), 3.75−3.85 (m, 2H, CH_2CH_3), 3.90 (exch br s, 1H, OH), 4.80 (q, 1H, CH(OH)CH_3), 5.70 (exch br s, 1H, *NH*−CH_2CH_3),7.30−7.50 (m, 5H, Ar).

4-Ethylamino-5-(1-hydroxyethyl)-2,6-dimethylpyridazin-3(2*H*)-one 15d. Yield = 53%; mp = 102−103 °C (cyclohexane); ^{1}H NMR ($CDCl_3$) δ 1.25 (t, 3H, CH_2CH_3), 1.55 (d, 3H, CH(OH)CH_3), 2.25 (s, 3H, 6-CH_3), 3.65−3.75 (m, 5H (3H, N−CH_3; 2H, CH_2CH_3)), 4.10 (exch br s, 1H, OH), 5.10 (q, 1H, CH(OH)CH_3,), 5.70 (exch br s, 1H, *NH*−CH_2CH_3).

General Procedure for 16a−d. Compounds **16a**−**d** were obtained starting from **15a**−**d** following the general procedure described for **5a**−**i**. For compounds **16b**−**d**, after dilution with water the mixture was neutralized with 3 N NaOH and extracted with CH_2Cl_2 (3 × 15 mL) and the solvent was evaporated in vacuo. Compounds **16c**,**d** were purified by column chromatography, using cycloexane/ethyl acetate, 2:1, as eluent for compound **16c** and using cycloexane/ethyl acetate, 1:1, for compound **16d**.

2-Butyl-4-methylamino-6-phenyl-5-vinylpyridazin-3(2*H*)-one 16a. Yield = 67%; mp = 65−67 °C (EtOH); IR (cm^{-1}) 3290 (NH), 1640 (CO); ^{1}H NMR ($CDCl_3$) δ 1.00 (t, 3H,$(CH_2)_3CH_3$), 1.40 (m, 2H, $(CH_2)_2CH_2CH_3$), 1.85 (m, 2H, $CH_2CH_2CH_2CH_3$), 2.95 (d, 3H, NH−CH_3), 4.20 (t, 2H, $CH_2CH_2CH_2CH_3$), 5.00 (d, 1H, CH=CH_2), 5.45 (d, 1H, CH=CH_2), 6.10 (exch br s, 1H, *NH*−CH_3), 6.50 (dd, 1H, CH=CH_2), 7.40 (s, 5H, Ar); MS (ESI) *m*/*z* 284.17 ([M + H]$^+$).

2,6-Dimethyl-4-methylamino-5-vinylpyridazin-3(2*H*)-one 16b. Yield = 88%; mp = 88−91 °C (EtOH); IR (cm^{-1}) 3310 (NH), 1620 (CO); ^{1}H NMR ($CDCl_3$) δ 2.20 (s, 3H, 6-CH_3), 2.95 (d, 3H, NH-CH_3), 3.75 (s, 3H, N−CH_3), 5.20 (d, 1H, CH=CH_2), 5.65 (d, 1H, CH=CH_2), 5.95 (exch br s, 1H, *NH*-CH_3), 6.65 (dd, 1H, CH=CH_2); MS (ESI) *m*/*z* 179.22 ([M + H]$^+$).

4-Ethylamino-2-methyl-6-phenyl-5-vinylpyridazin-3(2*H*)-one 16c. Yield = 11%; mp = 74−76 °C (EtOH); IR (cm^{-1}) 3300 (NH), 1630 (CO); ^{1}H NMR ($CDCl_3$) δ 1.25 (t, 3H,CH_2CH_3), 3.30 (m, 2H, CH_2CH_3), 3.85 (s, 3H, N−CH_3), 5.00 (d, 1H, CH=CH_2), 5.45 (d, 1H, CH=CH_2), 6.20 (exch br s, 1H, *NH*−CH_3), 6.50 (dd, 1H, CH=CH_2), 7.40 (s, 5H, Ar); MS (ESI) *m*/*z* 256.18 ([M + H]$^+$).

4-Ethylamino-2,6-dimethyl-5-vinylpyridazin-3(2*H*)-one 16d. Yield = 40%; mp = 65−66 °C (cyclohexane); IR (cm^{-1}) 3295 (NH), 1635 (CO); ^{1}H NMR ($CDCl_3$) δ 1.20 (t, 3H,CH_2CH_3), 2.20 (s, 3H, 6-CH_3), 3.30 (m, 2H, CH_2CH_3), 3.75 (s, 3H, N−CH_3), 5.25 (d, 1H, CH=CH_2), 5.62 (d, 1H, CH=CH_2), 5.80 (exch br s, 1H, *NH*-CH_3), 6.60 (dd, 1H, CH=CH_2); MS (ESI) *m*/*z* 194.12 ([M + H]$^+$).

General Procedure for 18a,b. A suspension of **17a**,**b**[29] (1.1 mmol) in *N*,*N*-dimethylformammide dimethyl acetal (3 mL, 22 mmol) was stirred at 100−110 °C for 3−4 h. After the mixture was cooled, the precipitate was recovered by suction.

3-(2-Dimethylaminovinyl)-6-methyl-4-phenyisoxazolo[3,4-*d*]-pyridazin-7(6*H*)-one 18a. Yield = 73%; mp = 203−204 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 2.80 (s, 6H, N(CH_3)$_2$, 3.80 (s, 3H, N−CH_3), 4.75 (d, 1H, CH=CH−N), 7.40−7.60 (m, 6H (5H, Ar; 1H, CH=CH−N)).

3-(2-Dimethylaminovinyl)-4,6-dimethylisoxazolo[3,4-*d*]pyridazin-7(6*H*)-one 18b. Yield = 85%; mp = 197−198 °C (EtOH); ^{1}H NMR ($CDCl_3$) δ 2.40 (s, 3H, CH_3), 3.05 (s, 6H, N(CH_3)$_2$), 3.65 (s, 3H, N−CH_3), 5.15 (d, 1H, CH=CH−N), 7.50 (d, 1H, CH=CH−N).

General Procedure for 19a,b. A suspension of compounds **18a**,**b** (0.9 mmol) in ethanol (2−3 mL) and hydrazine hydrate (15−20 mmol) was stirred at 80−90 °C for 1−2 h. Then the mixture was concentrated in vacuo and cooled for 3−4 h and the precipitate was recovered by suction.

4-Amino-2-methyl-6-phenyl-5-(1*H*-pyrazol-3-yl)pyridazin-3(2*H*)-one 19a. Yield = 54%; mp = 267−268 °C (cyclohexane); IR (cm^{-1}) 3385 and 3430 (NH_2), 3290 (NH), 1640 (CO); ^1H NMR ($CDCl_3$) δ 3.70 (s, 3H, N−CH_3), 5.15 (d, 1H, Ar), 6.40 (exch br s, 2H, NH_2), 7.10−7.40 (m, 5H, Ar), 7.60 (d, 1H, Ar), 13.05 (exch br s, 1H, NH); MS (ESI) *m*/*z* 268.18 ([M + H]$^+$).

4-Amino-2,6-dimethyl-5-(1*H*-pyrazol-3-yl)pyridazin-3(2*H*)-one 19b. Yield = 73%; mp = 250−253 °C, dec (EtOH); ^1H NMR ($CDCl_3$) δ 2.40 (s, 3H, 6-CH_3), 3.75 (s, 3H, N−CH_3), 6.55 (d, 1H, Ar), 7.35 (exch br s, 2H, NH_2), 7.85 (d, 1H, Ar), 11.05 (exch br s, 1H, NH).

General Procedure for 20a,b. A mixture of compounds **19a**,**b** (0.24 mmol), K_2CO_3 (0.48 mmol), and CH_3I (0.5−1 mmol) in anhydrous DMF (1.5−2.5 mL) was stirred at 90−100 °C for 3 h. After cooling, the suspension was diluted with cold water (10−15 mL) and extracted with ethyl acetate (3 × 15 mL) for compound **20a** and with CH_2Cl_2 (3 × 15 mL) for compound **20b.** Evaporation of the solvent afforded compounds **20a**,**b**, which were purified by column chromatography using $CHCl_3$/MeOH, 9:1, as eluent.

4-Amino-2-methyl-5-(1-methyl-1*H*-pyrazol-3-yl)-6-phenylpyridazin-3(2*H*)-one 20a. Yield = 60%; mp = 151−153 °C (EtOH); IR (cm^{-1}) 3380 and 3410 (NH_2), 1630 (CO); ^1H NMR ($CDCl_3$) 3.85 (s, 3H, N−CH_3), 3.95 (s, 3H, 2′-N−CH_3), 5.20 (d, 1H, Ar), 7.10 (d, 1H, Ar), 7.35 (s, 5H, Ar), 7.55 (exch br s, 2H, NH_2); MS (ESI) *m*/*z* 282.18 ([M + H]$^+$).

4-Amino-2,6-dimethyl-5-(1-methyl-1*H*-pyrazol-3-yl)pyridazin-3(2*H*)-one 20b. Yield = 30%; mp = 167−169 °C (EtOH); IR (cm^{-1}) 3370 and 3435 (NH_2), 1645 (CO); ^1H NMR ($CDCl_3$) 2.35 (s, 3H, 6-CH_3), 3.75 (s, 3H, N−CH_3), 3.95 (s, 3H, 2′-N−CH_3), 6.40 (d, 1H, Ar), 7.45 (d, 1H, Ar), 7.80 (exch br s, 2H, NH_2); MS (ESI) *m*/*z* 220.11 ([M + H]$^+$).

4-Ethylamino-2-methyl-5-(1-methyl-1*H*-pyrazol-3-yl)-6-phenylpyridazin-3(2*H*)-one 21. A mixture of **20a** (0.35 mmol), bromoethane (0.7 mmol), and NaH (60% dispersion in mineral oil) (3.3 mmol) in anhydrous DMSO (1.5 mL) was stirred at 80 °C for 90 min. After dilution with cold water (10−15 mL), the suspension was extracted with CH_2Cl_2 (3 × 20 mL) and the solvent was evaporated in vacuo to afford **21**, which was purified by column chromatography using cyclohexane/ethyl acetate, 1:2, as eluent. Yield = 38%; oil; IR (cm^{-1}) 3300 (NH), 1640 (CO); ^1H NMR ($CDCl_3$) 1.00 (t, 3H, *C*H_3CH$_2$−N), 2.95 (q, 2H, CH$_3$*C*H_2−N), 3.90 (m, 6H (3H, N−CH_3; 3H, 2′-*N*−CH_3)), 5.85 (d, 1H, Ar), 7.10−7.35 (m, 5H, Ar), 7.40 (d, 1H, Ar); MS (ESI) *m*/*z* 310.36 ([M + H]$^+$).

4-Diethylamino-2-methyl-5-(1-methyl-1*H*-pyrazol-3-yl)-6-phenylpyridazin-3(2*H*)-one 22. Compound **22** was obtained from compound **20a** following the general procedure described for **21**. The reaction was carried out using a 1:10 molar ratio of substrate **20a** to bromoethane. Compound **22** was purified by column chromatography (cyclohexane/ethyl acetate, 1:3). Yield = 63%; mp = 79−81 °C (cyclohexane); IR (cm^{-1}) 1630 (CO); ^1H NMR ($CDCl_3$) 1.00 (m, 6H, (*C*H_3CH$_2$)$_2$−N), 3.10 (m, 4H, (CH$_3$*C*H_2)$_2$−N), 3.80 (s, 3H, N−CH_3), 3.90 (s, 3H, 2′-N−CH_3), 5.90 (d, 1H, Ar), 7.10−7.25 (m, 5H, Ar), 7.30 (d, 1H, Ar); MS (ESI) *m*/*z* 338.44 ([M + H]$^+$).

4-Amino-5-(2-bromoacetyl)-2-methyl-6-phenylpyridazin-3(2*H*)-one 24. To a stirred and heated (40 °C) suspension of **23**[31] (1.05 mmol) and a catalytic amount of HBr in AcOH (3.5 mL), a solution of Br_2 (1.05 mmol) in acetic acid (1.9 mL) was slowly added. The mixture was heated for 4 h at 50 °C. After dilution with cold water (20−30 mL) the precipitate was recovered by suction and purified by column chromatography using cyclohexane/ethyl acetate, 1:1, as eluent. Yield = 44%; mp = 191−193 °C (EtOH); ^1H NMR ($CDCl_3$) 3.50 (s, 2H, CH_2Br), 3.85 (s, 3H, N−CH_3), 6.80 (exch br s, 2H, NH_2), 7.50 (s, 5H, Ar).

4-Amino-2-methyl-5-(2-methylthiazol-4-yl)-6-phenylpyridazin-3(2*H*)-one 25. A suspension of **24** (0.34 mmol) and thioacetamide (0.34 mmol) in EtOH (3 mL) was heated at 80 °C for 1 h. After the mixture was cooled, water (10−15 mL) was added and the final product was recovered by suction. Yield = 21%; mp = 129−130 °C (cyclohexane); IR (cm^{-1}) 3385 and 3430 (NH_2), 1640 (CO); ^1H NMR ($CDCl_3$) 2.75 (s, 3H, 2′-C−CH_3), 3.85 (s, 3H, N−CH_3), 6.20 (s, 1H, Ar), 6.70 (exch br s, 2H, NH_2), 7.25−7.40 (m, 5H, Ar); MS (ESI) *m*/*z* 299.18 ([M + H]$^+$).

5-Amino-1-methyl-6-oxo-3-phenyl-1,6-dihydropyridazine-4-carboxylic Acid *N*′-Acetylhydrazide 27. A mixture of compound **26**[11] (0.4 mmol) in $SOCl_2$ (1.5 mL) was refluxed for 1 h. The excess of $SOCl_2$ was removed in vacuo, and then acetohydrazide (0.5 mmol) was added to the residue oil dissolved in cold anhydrous dioxane. The mixture was stirred at room temperature for 3 h and diluted with cold water, and the precipitate was recovered by suction. Yield = 40%; mp = 264−265 °C (EtOH); ^1H NMR ($CDCl_3$) 1.90 (s, 3H, $COCH_3$), 3.70 (s, 3H, N−CH_3), 7.15 (exch br s, 2H, NH_2), 7.35−7.50 (m, 5H, Ar), 10.25 (exch br s, 1H, NH), 10.35 (exch br s, 1H, NH).

4-Amino-2-methyl-5-(5-methyl-[1,3,4]oxadiazol-2-yl)-6-phenylpyridazin-3(2*H*)-one 28. A suspension of **27** (0.23 mol) in $POCl_3$ (2 mL) was stirred at 60 °C for 2 h. After cooling, the mixture was slowly poured into ice−water and then extracted with CH_2Cl_2 (3 × 20 mL). Evaporation of the solvent afforded **28**. Yield = 31%; mp = 183−184 °C (EtOH); IR (cm^{-1}) 3385 and 3430 (NH_2), 1640 (CO); ^1H NMR ($CDCl_3$) 2.25 (s, 3H, 5′-C−CH_3), 3.90 (s, 3H, N−CH_3), 7.20 (exch br s, 2H, NH_2), 7.35−7.50 (m, 5H, Ar); MS (ESI) *m*/*z* 284.29 ([M + H]$^+$).

Biological Assays. Animals. Male Swiss albino mice (23−30 g) from the Morini breeding farm (Italy) and male Sprague-Dawley (200−250) rats from Harlan Laboratories (Italy) were used. Fifteen mice and four rats were housed per cage. The cages were placed in the experimental room 24 h before the test for acclimatization. The animals were kept at 23 ± 1 °C with a 12 h light/dark cycle, light at 7 a.m., with food and water ad libitum. All experiments were carried out according to the guidelines of the European Community Council.

Abdominal Constriction Test. Mice were injected ip with a 0.6% solution of acetic acid (10 mL kg^{-1}), according to the procedure of Koster et al.[32] The number of stretching movements was counted for 10 min, starting 5 min after acetic acid injection.

Statistical Analysis. All experimental results are given as the mean ± SEM. Analysis of variance (ANOVA), followed by Fisher's protected least significant difference (PLSD) procedure for post hoc comparison, was used to verify the significance between two means. Data were analyzed with the StatView software for the Macintosh (1992). *P* values of less than 0.05 were considered significant.

Drugs. The following drug was used: yohimbine hydrochloride (Tocris). Other chemicals were of the highest quality commercially available. Yohimbine was dissolved in isotonic (NaCl, 0.9%) saline solution, and tested compounds were dispersed in sodium carboxymethylcellulose, 1%.

Noradrenaline Release from Rat Cerebral Cortex Evaluated by Microdialysis Technique. Experiments were performed following the methods described by Oropeza et al.[33]

Rats were anesthetized with 8% chloral hydrate (400 mg/kg) and placed in a stereotaxic apparatus with the skull flat. A small burr hole was made in the skull centered at 3.2 mm anterior, and ± 0.9 mM $MgCl_2$ and 4 mM KCl were continuously perfused through the probe by a microliter infusion pump. Approximately 18 h following surgery, dialysate samples were collected every 20 min. Dialisate samples were stored at −80 °C for subsequent analysis by HPLC-ED. Vertical concentric microdialysis probes were used.

The amount of NE in the dialysate samples was determined with HPLC-ED. Dialysate samples were injected into the HPLC system. The detection system consisted of an ESA Coulochem II electrochemical detector. The baseline value against which drug administration was compared to was derived from the average of three samples just prior to manipulation. The neurochemical data were expressed as the mean ± SEM. All statistics were performed using StatView software.

Supporting Information Available: Elemental analysis results for all target compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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