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Preliminary communication

1,8-Naphthyridines IV. 9-Substituted *N*,*N*-dialkyl-5-(alkylamino or cycloalkylamino) [1,2,4]triazolo[4,3-*a*][1,8]naphthyridine-6-carboxamides, new compounds with anti-aggressive and potent anti-inflammatory activities

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Abstract – The title compounds (**8**) were synthesized through the cyclocondensation of the corresponding *N*-substituted 4-amino-2-chloro-1,8-naphthyridine-3-carboxamides (**4**) with the proper hydrazides, in order to evaluate their anti-inflammatory and anti-aggressive properties. Several compounds **8** exhibited high anti-inflammatory activity (carrageenin-induced paw edema assay in the rat) along with appreciable anti-aggressive properties (isolation-induced aggressiveness test in mice). With respect to anti-inflammatory activity, the most active compounds (**8n** and **8c**) produced a 61% edema inhibition at the 25 mg/kg dose, and 50 or 35% inhibition, respectively, at the 12.5 mg/kg dose. The structure–activity relationships are discussed. © 2000 Éditions scientifiques et médicales Elsevier SAS

[1,2,4]triazolo[4,3-*a*][1,8]naphthyridine derivatives / anti-inflammatory activity / anti-aggressive activity / structure–activity relationships

1. Introduction

As we previously described [1], the reaction of 2-aminonicotinic acid with the Vilsmeier type reagent ethyl *N*,*N*-dialkylmalonamate/phosphorus oxychloride afforded the 1,8-naphthyridine derivatives **1**, from which *N*,*N*-dialkyl-2,4-dichloro-1,8-naphthyridine-3-carboxamides **2** were easily obtained. In turn, compounds **2** were treated with primary amines to give the isomeric amino derivatives **3** and **4** (*figure 1*).

In a wide preliminary pharmacological screening of some of the compounds **1**–**4**, compounds **2** and **3** exhibited significant anti-inflammatory, anti-aggressive, or anti-hypertensive properties, depending on the structure [1].

Subsequently, a number of new compounds **2** and **3** were synthesized by us, their anti-inflammatory and anti-aggressive activities evaluated, and some information derived about structure–activity relationships (SAR). Some compounds showed one or both of these activities to a satisfactory extent, along with quite low acute toxicity [2].

Besides, following a literature suggestion [3], we had previously obtained the 4*H*-[1,2,4]triazolo[4,3-*a*]-[1,5]benzodiazepin-5-amines **6**, endowed with anti-inflammatory and/or analgesic activities, by fusing the inactive 1,5-benzodiazepines **5** with the 1,2,4-triazole ring [4, 5] (*figure 2*).

Considering the above results, we more recently [6] prepared the 5-chloro[1,2,4]triazolo[4,3-*a*][1,8]-naphthyridine-6-carboxamides **7**, in order to obtain more active anti-inflammatory agents and/or to separate anti-inflammatory from anti-aggressive activity. Actually, none of the 15 compounds tested showed anti-aggressive activity, but only four of them produced a statistically significant anti-inflammatory activity in the carrageenin-induced edema assay in the rat (the highest degree of protection was 40% at the 200 mg/kg dose) [6].

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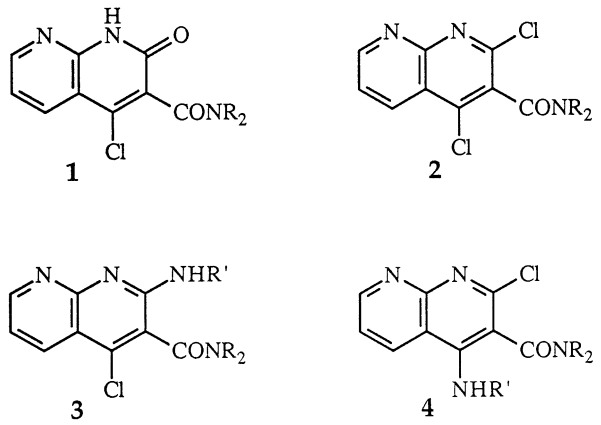


Figure 1. Structures of 1,8-naphthyridine-3-carboxamide derivatives **1–4**.

Taking into account the structure–activity relationships suggested by the pharmacological data of compounds **2**, **3**, **7** [1, 2, 6], we have now synthesized a number of the novel 5-amino[1,2,4]triazolo[4,3-*a*]-[1,8]naphthyridine-6-carboxamides **8**, designed with the aim of obtaining more interesting anti-inflammatory agents (*figure 2*).

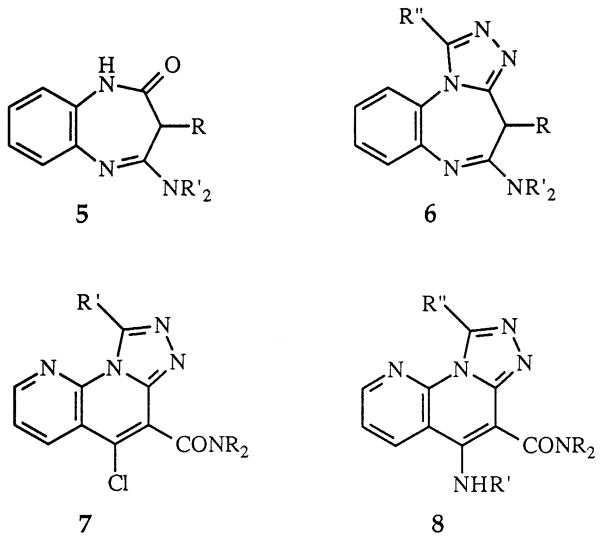


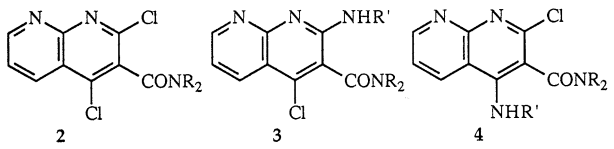
Figure 2. Structures of 1,5-benzodiazepine derivatives **5**, **6** and [1,2,4]triazolo[4,3-*a*][1,8]naphthyridine derivatives **7**, **8**.

2. Chemistry

The reaction of *N*,*N*-dialkyl-2,4-dichloro-1,8-naphthyridine-3-carboxamides **2a–d** [1, 2] with excess primary amines (anhydrous ethanol, room temperature, 1–24 h) afforded a mixture of the *N*,*N*-dialkyl-2-(alkylamino or cycloalkylamino)-4-chloro-1,8-naphthyridine-3-carboxamides **3a,j,k,m,n** and the isomeric 4-amino-2-chloro derivatives **4a,j,k,m,n** (*figure 3*, *table I*).

The desired 9-substituted *N*,*N*-dialkyl-5-(alkylamino or cycloalkylamino) [1,2,4]triazolo[4,3-*a*][1,8]-naphthyridine-6-carboxamides **8a–x** (*table II*) were then obtained through the cyclocondensation of the corresponding compounds **4a–n** (*table I*) with the proper hydrazides (Dowtherm A, 130–200 °C, 20–75 min) (*figure 3*). The starting compounds **4b–i,l** were previously described by us [1, 2].

Table I. Structures of 1,8-naphthyridine-3-carboxamide derivatives **2a–d**, **3a,j,k,m,n**, and **4a–n**.



Compound	NR_2	R'	Ref.
2a	$N(CH_3)_2$	-	[1]
2b	$N(C_3H_7)_2$	-	[2]
2c	$N(i\text{-}C_3H_7)_2$	-	[1]
2d		-	[1]
3a, 4a	$N(CH_3)_2$	$i\text{-}C_4H_9$	a
4b	$N(C_2H_5)_2$	C_2H_5	[1]
4c	$N(C_2H_5)_2$	C_3H_7	[2]
4d	$N(C_2H_5)_2$	$i\text{-}C_3H_7$	[1]
4e	$N(C_2H_5)_2$	$CH_2CH{=}CH_2$	[2]
4f	$N(C_2H_5)_2$		[1]
4g	$N(C_2H_5)_2$	C_4H_9	[2]
4h	$N(C_2H_5)_2$	$i\text{-}C_4H_9$	[2]
4i	$N(C_2H_5)_2$		[1]
3j, 4j	$N(C_3H_7)_2$		a
3k, 4k	$N(C_3H_7)_2$	$i\text{-}C_4H_9$	a
4l	$N(C_3H_7)_2$		[2]
3m, 4m	$N(i\text{-}C_3H_7)_2$		a
3n, 4n		$i\text{-}C_4H_9$	a

[a]Described in the present paper.

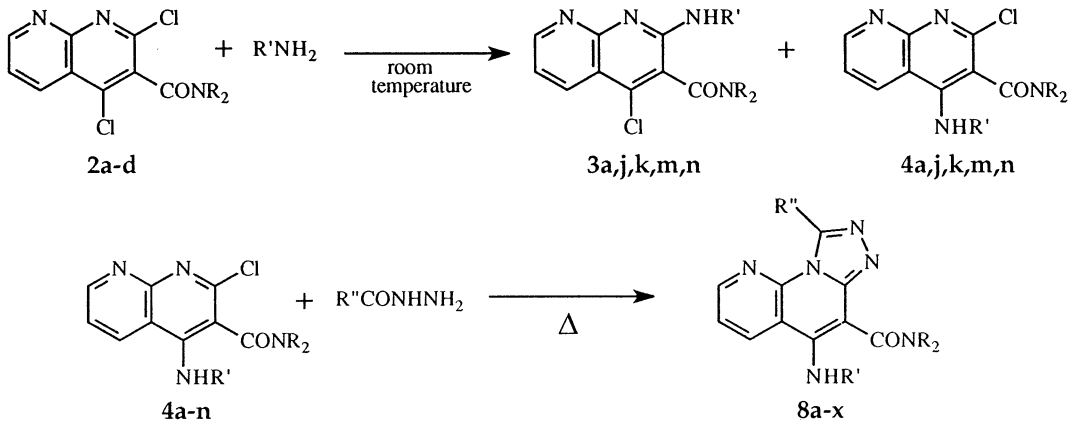


Figure 3. Synthetic route to substituted 5-amino[1,2,4]triazolo[4,3-*a*][1.8]naphthyridine-6-carboxamides **8a–x.**

The structures attributed to the compounds described in this paper are supported by the results of elemental analyses and IR and ^{1}H-NMR spectral data (see Section 5 and *table IV*).

The IR and ^{1}H-NMR spectra of compounds **3** and **4** now synthesized are in accordance with those of compounds **3** and **4** previously described by us [1, 2], respectively.

Concerning the ^{1}H-NMR spectra ($CDCl_3$) of tricyclic 5-(alkylamino) derivatives **8** (see *table IV*), they closely agree with those of the corresponding 5-chloro derivatives **7** [6]. Actually in both cases, as respects the alkyl groups, the CH_2 (particularly N–CH_2) signals and the CH_3 signals of –$CH(CH_3)_2$ moieties suggest the chirality of these molecules, most likely deriving from restricted rotation of 6-carboxamide substituent around the C–6–CO bond. For instance, in the case of compound **8k** the protons of two of the three N–CH_2 groups appear chemical shift not equivalent and clearly coupled with each other: two multiplets at δ 3.23–3.50 and 4.07 for one N–CH_2 group of the $CON(C_2H_5)_2$ substituent and two multiplets at δ 2.66 and 3.23–3.50 for the N–CH_2 group of the amino substituent. On the other hand, also the CH_3 groups of the isobutylamino substituent are not chemical shift equivalent (two doublets at δ 1.02 and 1.05). A similar behaviour was also shown by the *N*-alkyl substituents of the bicyclic compounds **3** and **4** (see Section 5).

Finally, with regard to the IR spectra of compounds **3**, **4** ($CHCl_3$) and **8** (KBr), the low frequencies of the tertiary amide ν CO bands (1600–1631 cm^{-1}) can be reasonably attributed both to the conjugation of the carbonyl group with the β-(alkylamino) substituent and the intermolecular H-bonding between these two groups.

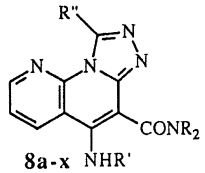
3. Pharmacological results and discussion

Compounds **8a–x** were tested in vivo for their anti-inflammatory and anti-aggressive activities. All the compounds were administered orally and assayed at the initial dose of 200 mg/kg. Compounds that exhibited a statistically significant activity at this dose were further tested at doses decreasing by a factor of two. The results of the pharmacological evaluation are listed in *table II*.

Fourteen of the 24 tested compounds exhibited a statistically significant but variably pronounced anti-inflammatory activity in the carrageeenin-induced paw edema assay in the rat. At the 200 mg/kg dose the degree of edema inhibition ranged from 32 to 74% and compounds **8n**, **8b**, **8c** showed the greatest activities (74, 73, and 65% protection, respectively). On the whole, the most active compounds were **8c** and **8n** that displayed a statistically significant inhibition of the paw edema down to the 6.25 mg/kg dose (33 and 27%, respectively). Compound **8m** exhibited a statistically significant activity down to 25 mg/kg (33% inhibition of the paw edema).

At the 200 mg/kg dose 15 compounds showed an interesting anti-aggressive activity, evaluated in the isolation-induced aggressiveness test in mice. At this dose the highest protection was observed with compounds **8b**, **8d**, **8e**, **8m** and **8n**, which inhibited the

Table II. Structures and pharmacological data of compounds **8a–x**.



8a-x

Compd.	NR_2	R'	R''	Dose (mg/kg p.o.)	Anti-inflammatory activity in rats[a] Edema (μL) (mean ± S.D.)	Inhibition (%)	Antiaggressive activity in mice[b]
8a	$N(CH_3)_2$	i-C_4H_9	C_2H_5	200	243±198	16	3/4
				100	-	-	0/4
8b	$N(C_2H_5)_2$	C_2H_5	C_2H_5	200	78±18	73**	4/4
				100	116±48	60**	2/4
				50	173±22	40**	0/4
				25	228±43	21	-
8c	$N(C_2H_5)_2$	C_2H_5	i-C_3H_7	200	101±51	65**	3/4
				100	107±74	63**	2/4
				50	107±75	63**	1/4
				25	113±49	61**	-
				12.5	188±45	35**	-
				6.25	194±62	33*	-
				3.12	251±20	13	-
8d	$N(C_2H_5)_2$	C_3H_7	CH_3	200	150±36	48**	4/4
				100	215±45	25	1/4
				50	-	-	0/4
8e	$N(C_2H_5)_2$	C_3H_7	C_2H_5	200	147±31	49**	4/4
				100	191±37	34*	1/4
				50	205±25	29*	0/4
				25	277±43	4	-
8f	$N(C_2H_5)_2$	i-C_3H_7	C_2H_5	200	116±22	60**	3/4
				100	176±37	39**	0/4
				50	208±23	28*	-
				25	275±47	5	-
8g	$N(C_2H_5)_2$	$CH_2CH{=}CH_2$	$CH_2C_6H_5$	200	228±71	21	0/4
8h	$N(C_2H_5)_2$		C_2H_5	200	173±28	40**	1/4
				100	246±25	15	0/4
8i	$N(C_2H_5)_2$	C_4H_9	$CH_2C_6H_5$	200	407±49	0	0/4
8j	$N(C_2H_5)_2$	i-C_4H_9	H	200	225±52	22	3/4
				100	-	-	2/4
				50	-	-	1/4
				25	-	-	0/4
8k	$N(C_2H_5)_2$	i-C_4H_9	CH_3	200	138±51	52**	2/4
				100	211±40	27*	0/4
				50	302±69	0	-
8l	$N(C_2H_5)_2$	i-C_4H_9	C_2H_5	200	142±44	51**	3/4
				100	145±46	50**	2/4
				50	150±28	48**	2/4
				25	216±50	25	1/4
				12.5	-	-	0/4

Table II. (*Continued*)

Compd.	NR_2	R'	R"	Dose (mg/kg p.o.)	Anti-inflammatory activity in rats[a] Edema (μL) (mean ± S.D.)	Inhibition (%)	Antiaggressive activity in mice[b]
8m	$N(C_2H_5)_2$	i-C_4H_9	C_3H_7	200	124±56	57**	4/4
				100	127±62	56**	2/4
				50	133±50	54**	2/4
				25	194±39	33*	0/4
				12.5	246±42	15	-
8n	$N(C_2H_5)_2$	i-C_4H_9	i-C_3H_7	200	75±31	74**	4/4
				100	87±21	70**	2/4
				50	98±19	66**	0/4
				25	113±48	61**	-
				12.5	145±46	50**	-
				6.25	211±40	27*	-
				3.12	246±33	15	-
8o	$N(C_2H_5)_2$	i-C_4H_9	i-C_4H_9	200	357±36	0	2/4
				100	-	-	0/4
8p	$N(C_2H_5)_2$	i-C_4H_9	*tert*-C_4H_9	200	191±107	34*	1/4
				100	254±51	12	-
8q	$N(C_2H_5)_2$	i-C_4H_9	$CH_2C_6H_5$	200	188±49	35**	2/4
				100	254±60	12	1/4
				50	-	-	0/4
8r	$N(C_2H_5)_2$	i-C_4H_9	C_6H_5	200	179±36	38**	2/4
				100	245±59	15	1/4
				50	-	-	0/4
8s	$N(C_2H_5)_2$		$CH_2C_6H_5$	200	272±22	6	0/4
8t	$N(C_3H_7)_2$		C_2H_5	200	196±78	32*	2/4
				100	257±43	11	1/4
				50	-	-	0/4
8u	$N(C_3H_7)_2$	i-C_4H_9	C_2H_5	200	277±130	4	0/4
8v	$N(C_3H_7)_2$		$CH_2C_6H_5$	200	222±49	23	0/4
8w	$N(i\text{-}C_3H_7)_2$		C_2H_5	200	319±38	0	0/4
8x		i-C_4H_9	C_2H_5	200	327±95	0	0/4
Indomethacin				6	101±50	65**	-
Diazepam				10	-	-	3/4

[a]Carrageenin paw edema test (control value: 289±82 μL; n = 51); ** P<0.01, * P<0.05 (Student's *t*-test versus controls). [b]Number of animals showing less than 3 fighting episodes in 3 minutes in the isolation-induced aggressiveness test.

aggressive behaviour in all the four mice treated. However, the most active compounds were **8l** and **8m** that still afforded a 50% protection at the 50 mg/kg dose.

The three compounds that exhibited statistically significant anti-inflammatory activity at the lowest doses (**8c**, **8m** and **8n**) were further tested for antinociceptive activity (writhing test in mice) at the intermedi-

Table III. Analgesic activity of compounds **8c,m,n**.

Compound	Dose (mg/kg p.o.)	Analgesic activity[a] % protection
8c	50	31
8m	50	0
8n	50	13
Dipyrone	100	58*

[a] Writhing test in mice; statistical significance versus control group (54 ± 18.7 writhing movements) was evaluated by the Mann–Whitney test (* $P < 0.05$).

ate dose of 50 mg/kg p.o. Although compounds **8c** and **8n** displayed a 31 and 13% protection, respectively, none of the tested compounds showed a statistically significant activity (*table III*).

At the highest dose administered (200 mg/kg), none of the compounds **8a–x** produced lethal toxic effects in the animals treated. However, it is worth noting that in all rats treated with compound **8c** diarrhoea was observed.

The anti-inflammatory and anti-aggressive activity data obtained in the successive steps of the present study allowed us to draw the following structure–activity relationships.

(a) Taking into account the possible sterical interaction between $CONR_2$ and NHR' groups, first of all we synthesized and tested compound **8b** and its analogues **8e,f,h,l**, differing from it only in the R′ substituent. Compound **8b** was chosen as a lead in this study due to the intermediate sizes of its substituents and considering the pharmacological results previously afforded by compounds **1–4, 7** [1, 2, 6]. On the whole, pharmacological data of **8b** (R′=C_2H_5) and **8l** (R′=i-C_4H_9) appeared to be the most interesting in this series, but R′ = i-C_4H_9 was selected for the subsequent investigation step due to a better behaviour of **8l** at the 50 mg/kg dose in both tests.

(b) Compounds **8j,k,m–r** (differing from each other only in R″ structure) were then assayed, **8l** being the reference compound.

At the 200 mg/kg dose the anti-inflammatory activity increased regularly from **8j** (R″=H) up to **8n** (R″=i-C_3H_7; 74% edema inhibition) according to the size of the R″ alkyl substituent, then suddenly dropped when R″ was *tert*-C_4H_9 and, particularly, when it was i-C_4H_9 (**8o**: 0% edema inhibition). Both in the case of R″ (see **8n** versus **8m**) and of R′ (see **8f** versus **8e**) the branched substituent was more effective than its linear isomer.

A similar behaviour at the 200 mg/kg dose was observed in this series for anti-aggressive activity: it increased from **8k** (R″=CH_3, 50% protection) up to **8m** and **8n** (R″=C_3H_7 and i–C_3H_7, respectively; 100% protection), then declined to 50 and 25% protection for compounds **8o** (R″ = i–C_4H_9) and **8p** (R″ = *tert*-C_4H_9), respectively.

Relatively low activities were displayed in both tests by compounds **8q** and **8r** (R″ = $CH_2C_6H_5$ or C_6H_5, respectively).

(c) If we consider the series of compounds **8a**, **8l**, **8u** and **8x** (differing from each other only in NR_2 substituent), we can again observe that the lengthening of R alkyl chain initially produced an increase in both the activities, but further lengthening resulted in a sudden decrease in pharmacological effects. In both tests $N(C_2H_5)_2$ proved to be the most effective NR_2 group.

Finally, we can point out that, for some compounds, an increase in the dose administered increased anti-inflammatory activity until a maximum was reached, further dose increase (up to 200 mg/kg) being practically ineffective. This behaviour was particularly evident in the case of compounds **8c**, **8l** and **8m**, and might be due to their low water solubility and consequent uncompleted absorption at high doses.

4. Conclusions

In the light of pharmacological properties shown by the 1,8-naphthtyridine derivatives **2**, **3**, **7** previously described by us [1, 2, 6], we have designed and synthesized the 5-amino[1,2,4]triazolo[4,3-*a*][1,8]naphthyridine-6-carboxamides **8**, a new class of 1,8-naphthyridine derivatives, which have been tested for their anti-inflammatory and anti-aggressive activities.

Several compounds **8** exhibited high anti-inflammatory activity along with significant anti-aggressive properties: this interesting behaviour prompts us to plan for the near future the study of their mechanism of action. As expected on the basis of SAR suggestions, **8n** proved to be the most potent anti-inflammatory agent among compounds tested (74% and 50% inhibition of carrageenin-induced paw edema in the rat, at 200 and 12.5 mg/kg doses, respectively).

5. Experimental protocols

5.1. Chemistry

Melting points were determined using a Fisher–Johns

apparatus and are uncorrected. IR spectra were recorded on a Perkin-Elmer 398 spectrophotometer. ^{1}H-NMR spectra were recorded on a Varian Gemini 200 (200 MHz) spectrometer, using $(CH_3)_4Si$ as an internal reference ($\delta = 0$), and chemical shifts (δ) are reported in ppm. Analyses of all new compounds, indicated by the symbols of the elements, were within $\pm 0.4\%$ of the theoretical values and were performed by the Laboratorio di Microanalisi, Dipartimento di Scienze Farmaceutiche, Università di Genova.

Thin layer chromatograms were run on Merck silica gel 60 F_{254} precoated plastic sheets (layer thickness 0.2 mm). Column chromatography was performed using Carlo Erba silica gel (0.05–0.20 mm) or Carlo Erba neutral aluminium oxide (Brockmann activity I).

*5.1.1. General procedure for compounds **3a,j,k,m,n** and **4a,j,k,m,n***

A mixture of 4.0 mmol of the suitable compound **2** [1, 2], 40.0 mmol of the proper amine and 40 mL of anhydrous ethanol was stirred at room temperature for the time reported below for each case.

The reaction mixture was then poured into water (100 mL) and the resulting solution was exhaustively extracted with dichloromethane. The combined extracts were dried (anhydrous Na_2SO_4), then evaporated to dryness under reduced pressure to give a viscous oil which was subjected to column chromatography (silica gel), eluting first with petroleum ether–dichloromethane–triethylamine (6:3:1). The eluate obtained was evaporated affording a whitish thick oil from which, after treatment with a little ethyl ether–petroleum ether, the pure compound **3** separated out as white or whitish crystalline solid which was then recrystallized from the suitable solvent.

The corresponding isomer **4** was then recovered by eluting with methanol. The thick oil obtained after removing the solvent from this eluate was partitioned between 0.5 N aqueous NaOH and chloroform, then the aqueous phase was thoroughly extracted with chloroform. The combined chloroform phases were dried (anhydrous Na_2SO_4) and the solvent was removed to give a whitish thick oil which, after treatment with some ethyl acetate and standing, afforded the pure compound **4** as white crystalline solid which was then recrystallized from the proper solvent.

*5.1.1.1. 2-(Isobutylamino)-4-chloro-N,N-dimethyl-1,8-naphthyridine-3-carboxamide **3a** and 4-(isobutylamino)-2-chloro-N,N-dimethyl-1,8-naphthyridine-3-carboxamide **4a***

The reaction (2 h) of **2a** [1] (1.08 g) with isobutylamine (2.93 g) afforded **3a** (0.53 g, 43%) and **4a** (0.46 g, 37%).

Compound **3a**. M.p. 95–96 °C (from isopropyl ether); IR ($CHCl_3$), cm^{-1}: 3430 (NH), 1631 (CO), 1608, 1553 weak, 1527; ^{1}H-NMR ($CDCl_3$), δ: 0.96 [d, 6H, $NCH_2CH(CH_3)_2$], 2.00 [m, 1H, $NCH_2CH(CH_3)_2$], 2.94 and 3.20 [2 s, 6H, $N(CH_3)_2$], 3.47 [m, 2H, $NCH_2CH(CH_3)_2$], 5.32 (near t, 1H, NH; disappeared with D_2O), 7.22 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.30 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.85 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{15}H_{19}ClN_4O$ (C, H, N, Cl).

Compound **4a**. M.p. 187–188 °C (from ethyl acetate); IR ($CHCl_3$), cm^{-1}: 3345 and 3320 broad (NH), 1626 (CO), 1600 weak, 1578, 1561, 1520 broad; ^{1}H-NMR ($CDCl_3$), δ: 1.01 [d, 6H, $NCH_2CH(CH_3)_2$], 1.93 [m, 1H, $NCH_2CH(CH_3)_2$], 2.99 and 3.17 [2 s, 6H, $N(CH_3)_2$], 3.05 and 3.36 [2 m, 2H, $NCH_2CH(CH_3)_2$], 5.46 (near t, 1H, NH; disappeared with D_2O), 7.38 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.25 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.98 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{15}H_{19}ClN_4O$ (C, H, N, Cl).

*5.1.1.2. 4-Chloro-2-(cyclopropylamino)-N,N-dipropyl-1,8-naphthyridine-3-carboxamide **3j** and 2-chloro-4-(cyclopropylamino)-N,N-dipropyl-1,8-naphthyridine-3-carboxamide **4j***

The reaction (24 h) of **2b** [2] (1.30 g) with cyclopropylamine (2.28 g) yielded **3j** (0.45 g, 32%) and **4j** (0.76 g, 55%).

Compound **3j**. M.p. 104–105 °C (from isopropyl ether); IR ($CHCl_3$), cm^{-1}: 3415 (NH), 1628 (CO), 1604, 1551, 1512; ^{1}H-NMR ($CDCl_3$), δ: 0.53 (m, 2H, cyclopropyl CH_2), 0.70 and 1.02 [2 t, 6H, $N(CH_2CH_2CH_3)_2$], 0.80–1.08 (m, 2H, cyclopropyl CH_2), 1.32–1.58 and 1.64–1.86 [2 m, 4H, $N(CH_2CH_2CH_3)_2$], 2.95–3.32 [m, 4H, 3H of $N(CH_2CH_2CH_3)_2$ + cyclopropyl CH], 3.82 [m, 1H, 1H of $N(CH_2CH_2CH_3)_2$], 5.43 (s, 1H, NH; disappeared with D_2O), 7.29 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.34 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.90 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{18}H_{23}ClN_4O$ (C, H, N, Cl).

Compound **4j**. M.p. 124–125 °C (from ethyl acetate); IR ($CHCl_3$), cm^{-1}: 3445 weak and 3380 broad (NH), 1617 (CO), 1598, 1583, 1555, 1503; ^{1}H-NMR ($CDCl_3$), δ: 0.62–0.80 (m, 2H, cyclopropyl CH_2), 0.73 and 1.01 [2 t, 6H, $N(CH_2CH_2CH_3)_2$], 0.90–1.10 (m, 2H, cyclopropyl CH_2), 1.32–1.89 [m, 4H, $N(CH_2CH_2CH_3)_2$], 3.01–3.27 [m, 4H, 3H of $N(CH_2CH_2CH_3)_2$ + cyclopropyl CH], 3.82 [m, 1H, 1H of $N(CH_2CH_2CH_3)_2$], 5.81 (s, 1H, NH; disappeared with D_2O), 7.36 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.93 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H,

H-5), 9.00 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{18}H_{23}ClN_4O$ (C, H, N, Cl).

5.1.1.3. 2-(Isobutylamino)-4-chloro-N,N-dipropyl-1,8-naphthyridine-3-carboxamide ***3k*** *and 4-(isobutylamino)-2-chloro-N,N-dipropyl-1,8-naphthyridine-3-carboxamide* ***4k***

The reaction (2 h) of **2b** [2] (1.30 g) with isobutylamine (2.93 g) afforded **3k** (0.49 g, 34%) and **4k** (0.57 g, 39%).

Compound **3k**. M.p. 102–103 °C (from isopropyl ether); IR ($CHCl_3$), cm^{-1}: 3435 (NH), 1628 (CO), 1613, 1559, 1530; ^{1}H-NMR ($CDCl_3$), δ: 0.73 and 1.02 [2 t, 6H, $N(CH_2CH_2CH_3)_2$], 0.97 [d, 6H, $NCH_2CH(CH_3)_2$], 1.52 and 1.76 [2 m, 4H, $N(CH_2CH_2CH_3)_2$], 1.96 [m, 1H, $NCH_2CH(CH_3)_2$], 3.14 [t, 2H, 2H of $N(CH_2CH_2CH_3)_2$], 3.21–3.61 [m, 3H, 1H of $N(CH_2CH_2CH_3)_2$ + $NCH_2CH(CH_3)_2$], 3.78 [m, 1H, 1H of $N(CH_2CH_2CH_3)_2$], 5.26 (near t, 1H, NH; disappeared with D_2O), 7.26 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.33 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.86 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{19}H_{27}ClN_4O$ (C, H, N, Cl).

Compound **4k**. M.p. 150–151.5 °C (from ethyl acetate/petroleum ether); IR ($CHCl_3$), cm^{-1}: 3450 and 3350 broad (NH), 1619 (CO), 1601, 1582, 1560, 1516 broad; ^{1}H-NMR ($CDCl_3$), δ: 0.74 and 1.00 [2 t, 6H, $N(CH_2CH_2CH_3)_2$], 1.05 [d, 6H, $NCH_2CH(CH_3)_2$], 1.39–1.84 [m, 4H, $N(CH_2CH_2CH_3)_2$], 1.94 [m, 1H, $NCH_2CH(CH_3)_2$], 3.16 [t + m, 3H, 2H of $N(CH_2CH_2CH_3)_2$ + 1H of $NCH_2CH(CH_3)_2$], 3.25–3.52 [m, 2H, 1H of $N(CH_2CH_2CH_3)_2$ + 1H of $NCH_2CH(CH_3)_2$], 3.62 [m, 1H, 1H of $N(CH_2CH_2CH_3)_2$], 5.29 (near t, 1H, NH; disappeared with D_2O), 7.38 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.28 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.99 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{19}H_{27}ClN_4O$ (C, H, N, Cl).

5.1.1.4. 4-Chloro-2-(cyclopropylamino)-N,N-diisopropyl-1,8-naphthyridine-3-carboxamide ***3m*** *and 2-chloro-4-(cyclopropylamino)-N,N-diisopropyl-1,8-naphthyridine-3-carboxamide* ***4m***

The reaction (24 h) of **2c** [1] (1.30 g) with cyclopropylamine (2.28 g) afforded **3m** (0.49 g, 35%) and **4m** (0.72 g, 52%).

Compound **3m**. M.p. 145–145.5 °C (from isopropyl ether); IR ($CHCl_3$), cm^{-1}: 3419 (NH), 1629 (CO), 1608, 1553, 1511; ^{1}H-NMR ($CDCl_3$), δ: 0.53 (m, 2H, cyclopropyl CH_2), 0.80–1.04 (m, 2H, cyclopropyl CH_2), 1.09 and 1.21 [2 d, 6H, $NCH(CH_3)_2$], 1.59 and 1.61 [2 d, 6H, $NCH(CH_3)_2$], 3.09 (m, 1H, cyclopropyl CH), 3.60 {m, 2H, $N[CH(CH_3)_2]_2$}, 5.23 (near s, 1H, NH; disappeared with D_2O), 7.29 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.34 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.90 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{18}H_{23}ClN_4O$ (C, H, N, Cl).

Compound **4m**. M.p. 219.5–220 °C (from ethyl acetate); IR ($CHCl_3$), cm^{-1}: 3450 weak and 3390 (NH), 1619 (CO), 1599, 1582, 1554, 1500; ^{1}H-NMR ($CDCl_3$), δ: 0.70 and 1.00 (2 m, 4H, cyclopropyl CH_2's), 1.07 and 1.25 [2 d, 6H, $NCH(CH_3)_2$], 1.58 and 1.60 [2 d, 6H, $NCH(CH_3)_2$], 3.11 (m, 1H, cyclopropyl CH), 3.57 and 3.72 {2 m, 2H, $N[CH(CH_3)_2]_2$}, 5.48 (near s, 1H, NH; disappeared with D_2O), 7.36 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.96–9.06 (m, 2H, H-5,7). Anal. $C_{18}H_{23}ClN_4O$ (C, H, N, Cl).

5.1.1.5. 2-(Isobutylamino)-4-chloro-N,N-pentamethylene-1,8-naphthyridine-3-carboxamide ***3n*** *and 4-(isobutylamino)-2-chloro-N,N-pentamethylene-1,8-naphthyridine-3-carboxamide* ***4n***

The reaction (1 h) of **2d** [1] (1.24 g) with isobutylamine (2.93 g) afforded **3n** (0.47 g, 34%) and **4n** (0.58 g, 42%).

Compound **3n**. M.p. 180–181 °C (from ethyl acetate); IR ($CHCl_3$), cm^{-1}: 3430 (NH), 1624 (CO), 1610, 1554, 1527; ^{1}H-NMR ($CDCl_3$), δ: 0.98 [d, 6H, $NCH_2CH(CH_3)_2$], 1.34–1.85 (m, 6H, piperidine $\beta + \gamma$-CH_2's), 1.98 [m, 1H, $NCH_2CH(CH_3)_2$], 3.21–3.45 (m, 3H, 3H of piperidine α-CH_2's), 3.52–3.70 [m, 2H, $NCH_2CH(CH_3)_2$], 3.94–4.09 (m, 1H, 1H of piperidine α-CH_2), 5.25 (t, 1H, NH; disappeared with D_2O), 7.24 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.30 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.86 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{18}H_{23}ClN_4O$ (C, H, N, Cl).

Compound **4n**. M.p. 197–198 °C (from ethyl acetate); IR ($CHCl_3$), cm^{-1}: 3455 and 3310 broad (NH), 1619 (CO), 1602, 1581, 1563, 1525; ^{1}H-NMR ($CDCl_3$), δ: 1.02 and 1.04 [2d, 6H, $NCH_2CH(CH_3)_2$], 1.40–1.90 (m, 6H, piperidine $\beta + \gamma$–CH_2's), 1.96 [m, 1H, $NCH_2CH(CH_3)_2$], 3.00–3.16 [m, 1H, 1H of $NCH_2CH(CH_3)_2$], 3.20–3.48 [m, 3H, 1H of $NCH_2CH(CH_3)_2$ + 2H of piperidine α-CH_2's], 3.78 (m, 2H, 2H of piperidine α-CH_2's), 5.40 (near s, 1H, NH; disappeared with D_2O), 7.36 (dd, $J_{6,5} = 8.5$ Hz, $J_{6,7} = 4.2$ Hz, 1H, H-6), 8.23 (dd, $J_{5,6} = 8.5$ Hz, $J_{5,7} = 2$ Hz, 1H, H-5), 8.97 (dd, $J_{7,6} = 4.2$ Hz, $J_{7,5} = 2$ Hz, 1H, H-7). Anal. $C_{18}H_{23}ClN_4O$ (C, H, N, Cl).

5.1.2. General procedure for N,N-dialkyl-9-alkyl or phenyl-5-(alkylamino or cycloalkylamino)[1,2,4]-triazolo[4,3-a][1,8]naphthyridine-6-carboxamides **8a–x**

A mixture of 4.0 mmol of the proper compound **4** (1.23 g of **4a** or **4b** [1], 1.28 g of **4c** [2] or **4d** [1], 1.27 g

of **4e** [2] or **4f** [1], 1.34 g of **4g** [2] or **4h** [2], 1.44 g of **4i** [1], 1.39 g of **4j**, **4m** or **4n**, 1.45 g of **4k**, 1.55 g of **4l** [2], 8.0 mmol of the suitable hydrazide (0.54 g of formylhydrazine, 0.59 g of acetylhydrazine, 0.70 g of propionylhydrazine, 0.80 g of butyrylhydrazine or isobutyrylhydrazine, 0.90 g of isovalerylhydrazine or pivaloylhydrazine, 1.18 g of phenylacetylhydrazine, or 1.09 g of benzoylhydrazine) and 10 mL of Dowtherm A was stirred at 160 °C for 20 min (compounds **8c,n,o**) or 1 h (compounds **8a,b,d–g,i–m, q–s,u,v,x**) or at 150 °C for 15 min then at 200 °C for 1 h (compound **8p**). For the preparation of compounds **8h,t,w** only 4.0 mmol (0.35 g) of propionylhydrazine were used and the reaction was carried out at 130 °C for 1 h.

After cooling, compound **8** was recovered through one of the three following procedures.

In most cases (compounds **8c–e,g,i–t,v,w**) the reaction mixture was partitioned between chloroform and 10% aqueous Na_2CO_3. The organic layer was collected and the aqueous phase was extracted several more times with chloroform. The combined extracts were dried (anhydrous Na_2SO_4), then evaporated to dryness under reduced pressure to give an oily residue which was chromatographed on a silica gel (compounds **8d,e,g,i–m,p–t,v,w**) or neutral aluminium oxide column (compounds **8c,n,o**) eluting with dichloromethane [petroleum ether–dichloromethane (9:1) in the case of **8n**] until Dowtherm A was completely removed. The nearly pure compound **8** was then recovered by eluting with the suitable solvent [dichloromethane for compound **8n**; ethyl acetate for compounds **8c,o**; acetone for **8g,i,p–s**; chloroform–methanol (9:1) for **8d,e,j–m,t**; acetone–chloroform–methanol (50:45:5) for **8v**; benzene-triethylamine (9:1) for **8w**]. The solid or thick oily residue obtained from the eluate was then crystallized from the proper solvent.

In the case of compounds **8a,b,u**, the reaction mixture was nearly devoid of the starting compound **4** (TLC) and was partitioned between ethyl ether and 2N aqueous HCl. The aqueous layer was collected and the organic phase was alternately extracted several times with 2N aqueous HCl and water. The combined aqueous phases were then cooled in an ice-bath, then carefully made alkaline with 6N aqueous NaOH. The resulting turbid solution was exhaustively extracted with chloroform and the combined extracts (dried over anhydrous Na_2SO_4) were evaporated in vacuo to give a solid residue which was then crystallized from the appropriate solvent.

In the case of compounds **8f,h,x,** the crystalline solid present in the final reaction mixture (hydrochloride of compound **8**) was recovered by filtration, washed with ethyl ether and dried. This solid was then suspended and stirred some minutes at room temperature in 10% aqueous Na_2CO_3, then the mixture was exhaustively extracted with chloroform. The combined extracts were dried (anhydrous Na_2SO_4), then evaporated to dryness in vacuo to give the nearly pure compound **8** as a solid which was crystallized from the suitable solvent.

Compounds **8a–x** are yellow or pale yellow crystalline solids. Their data are reported in *table IV*.

5.2. Pharmacology

Male albino Swiss mice (22–30 g) and male Sprague–Dawley rats (100–120 g) were used. All the test compounds were administered by oral gavage in a 0.5% carboxymethylcellulose suspension.

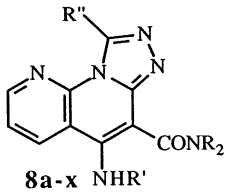
5.2.1. Anti-inflammatory activity

The carrageenin-induced paw edema test [7] was used on groups of five rats. Sixty minutes after the administration of the test compound, 0.1 mL of a 1% carrageenin solution in saline was injected into the plantar surface of the right hind paw of each rat. Paw volume, as determined by measuring the amount of water displaced after immersing the paw to the level of the lateral malleolus, was recorded immediately after the carrageenin injection, and again 3 h later. The difference between these two values was taken as edema volume. The percent inhibition of the edema of treated rats with respect to controls was calculated. Indomethacin (6 mg/kg p.o.) was used as reference standard.

5.2.2. Anti-aggressive activity

The isolation-induced aggressiveness test [8] was used on groups of four mice. Mice at least 4-weeks-old were isolated in a cage for several weeks. Sixty minutes before test compound administration aggressive isolated mice were selected by placing an intruder in the cage of the isolated mouse. Mice showing more than three fighting episodes in 3 min were chosen. The same procedure was repeated 60 min after the administration of the test compound: mice showing less than three fighting episodes in 3 min were considered protected. Diazepam (10 mg/kg p.o.) was used as reference standard.

Table IV. Physical and chemical data of compounds **8a–x**.



8a-x

Compound	Yield (%)	M.p. (° C) (solvent)[a]	Molecular formula[b]	IR[c] (cm^{-1})	^{1}H-NMR[d] (δ, ppm)
8a	63	209–210 (A)	$C_{18}H_{24}N_6O$	3245 (NH), 1607 s, br (CO), 1550, 1520 w	1.01 [d,6H,$NCH_2CH(CH_3)_2$], 1.48 (t,3H,9-CH_2CH_3), 1.99 [m,1H,$NCH_2CH(CH_3)_2$], 2.70 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.05 and 3.23 [2s,6H,$N(CH_3)_2$], 3.15–3.30 [m, 1H, 1H of $NCH_2CH(CH_3)_2$], 3.46 (q,2H, 9-CH_2CH_3), 6.22[e] (m, 1H,NH), 7.00 (dd,1H,H-3), 8.18 (dd,1H,H-4), 8.34 (dd,1H,H-2)
8b	70	225–225.5 (B)	$C_{18}H_{24}N_6O$	3290 (NH), 1610 s, br (CO), 1546, 1520	1.18 and 1.33 [2t,6H,$N(CH_2CH_3)_2$], 1.40 (t,3H,NCH_2CH_3), 1.50 (t,3H,9-CH_2CH_3), 3.04 (m,1H,1H of NCH_2CH_3), 3.28–3.63 [m,6H,3H of $N(CH_2CH_3)_2$+1H of NCH_2CH_3+9-CH_2CH_3], 3.86 [m,1H,1H of $N(CH_2CH_3)_2$], 6.04[e] (m,1H,NH), 6.97 (dd,1H,H-3), 8.14 (dd,1H,H-4), 8.35 (dd,1H,H-2)
8c	77	189–191 (A)	$C_{19}H_{26}N_6O$	3250 br (NH), 1608 s, br (CO), 1546, 1518 w	1.18 and 1.33 [2t,6H,$N(CH_2CH_3)_2$], 1.38 [t+d,6H,NCH_2CH_3+3H of 9-$CH(CH_3)_2$], 1.62 [d,3H,3H of 9-$CH(CH_3)_2$], 3.04 (m,1H,1H of NCH_2CH_3), 3.32–3.87 [m, 5H,$N(CH_2CH_3)_2$+1H of NCH_2CH_3], 4.13 [m,1H,9-$CH(CH_3)_2$], 6.12[e] (d,1H,NH), 6.93 (dd,1H,H-3), 8.14 (dd,1H,H-4), 8.32 (dd,1H,H-2)
8d	76	217–218 (C)	$C_{18}H_{24}N_6O$	3270 (NH), 1616 sh, 1601 s (CO), 1550, 1521 w	1.08 (t,3H,$NCH_2CH_2CH_3$),1.18 and 1.28 [2t,6H,$N(CH_2CH_3)_2$], 1.71–1.93 (m,2H,$NCH_2CH_2CH_3$), 2.90 (m,1H,1H of $NCH_2CH_2CH_3$), 2.98 (s,3H,9-CH_3), 3.25–3.57 [m, 4H, 3H of $N(CH_2CH_3)_2$+1H of $NCH_2CH_2CH_3$], 4.05 [m,1H,1H of $N(CH_2CH_3)_2$], 6.40[e] (m,1H,NH), 6.90 (dd,1H,H-3), 8.14 (dd,1H,H-4), 8.30 (dd,1H,H-2)

Table IV. (Continued)

Compound	Yield (%)	M.p. (° C) (solvent)[a]	Molecular formula[b]	IR[c] (cm^{-1})	^{1}H-NMR[d] (δ, ppm)
8e	52	145–146 (D)	$C_{19}H_{26}N_6O$	3250 (NH), 1600 s, br (CO), 1543, 1520 sh	1.04 (t,3H,$NCH_2CH_2\mathit{CH_3}$), 1.19 and 1.33 [2t,6H,$N(CH_2\mathit{CH_3})_2$], 1.49 (t,3H,9-$CH_2\mathit{CH_3}$), 1.67–1.93 (m,2H,$NCH_2\mathit{CH_2}CH_3$), 2.95 (m,1H,1H of $N\mathit{CH_2}CH_2CH_3$), 3.28–3.57 [m, 6H,3H of $N(\mathit{CH_2}CH_3)_2$ + 1H of $N\mathit{CH_2}CH_2CH_3$ + 9-$\mathit{CH_2}CH_3$], 3.93 [m,1H,1H of $N(\mathit{CH_2}CH_3)_2$], 6.19[e] (near d,1H,NH), 6.97 (dd,1H,H-3), 8.16 (dd,1H,H-4), 8.32 (dd,1H,H-2)
8f	79	159–160 (A)	$C_{19}H_{26}N_6O$	3370 (NH), 1631 (CO), 1601, 1536, 1515 sh	1.20 and 1.34 [2t,6H,$N(CH_2\mathit{CH_3})_2$], 1.26 and 1.29 [2d,6H,$NCH(\mathit{CH_3})_2$], 1.50 (t,3H,9-$CH_2\mathit{CH_3}$), 3.21–3.99 [m,7H,$N(\mathit{CH_2}CH_3)_2$ + $N\mathit{CH}(CH_3)_2$ + 9-$\mathit{CH_2}CH_3$], 4.33[e] (d,1H,NH), 7.47 (dd,1H,H-3), 8.30 (dd,1H,H-4), 8.68 (dd,1H,H-2)
8g	65	181–182 (A)	$C_{24}H_{26}N_6O$	3295 (NH), 1608 s (CO), 1596 s, 1560 sh, 1533	1.18 and 1.34 [2t,6H,$N(CH_2\mathit{CH_3})_2$], 3.26–4.11 [m,6H,$N(\mathit{CH_2}CH_3)_2$ + $N\mathit{CH_2}$-CH = CH_2], 4.93 (s,2H,$\mathit{CH_2}C_6H_5$), 5.27 (m,2H,NCH_2-CH = $\mathit{CH_2}$), 5.51[e] (m,1H,NH), 5.90–6.13 (m,1H,NCH_2-CH = CH_2), 7.02–7.34 (m,4H,3H of C_6H_5 + H-3), 7.47 (m,2H,2H of C_6H_5), 8.14 (dd,1H,H-4), 8.34 (dd,1H,H-2)
8h	59	198–199 (A)	$C_{19}H_{24}N_6O$	3240 (NH),1612 s, br (CO), 1538, 1513 sh	0.30–0.63 and 0.76–0.96 (2 m,4H,cyclopropyl CH_2's), 1.23 and 1.33 [2t,6H,$N(CH_2\mathit{CH_3})_2$], 1.50 (t,3H, 9-$CH_2\mathit{CH_3}$), 3.05 (m, 1H, cyclopropyl CH), 3.21–3.91 [m, 6H,$N(\mathit{CH_2}CH_3)_2$ + 9-$\mathit{CH_2}CH_3$], 6.81 (dd,1H,H-3), 7.27[e] (s,1H,NH), 8.07 (dd,1H,H-4), 8.21 (dd,1H,H-2)
8i	78	156–157 (E)	$C_{25}H_{30}N_6O$	3260 (NH),1603 s, br (CO), 1536, 1518 sh	0.96 (t,3H,$NCH_2CH_2CH_2\mathit{CH_3}$), 1.18 and 1.33 [2t,6H,$N(CH_2\mathit{CH_3})_2$], 1.30–1.53 (m,2H,$NCH_2CH_2\mathit{CH_2}CH_3$), 1.65–1.90 (m,2H,$NCH_2\mathit{CH_2}CH_2CH_3$), 2.95 (m,1H,1H of $N\mathit{CH_2}CH_2CH_2CH_3$), 3.28–3.58 [m,4H,3H of $N(\mathit{CH_2}CH_3)_2$ + 1H of $N\mathit{CH_2}CH_2CH_2CH_3$], 3.91 [m,1H,1H of $N(\mathit{CH_2}CH_3)_2$], 4.82 and 4.88 (AB system, J = 16 Hz,2H,$\mathit{CH_2}C_6H_5$), 6.28[e] (near d,1H,NH), 6.80 (dd,1H,H-3), 7.13–7.38 (m, 3H, 3H of C_6H_5), 7.50 (m,2H,2H of C_6H_5), 7.96–8.12 (m, 2H, H-2,4)
8j	64	230–231 (C)	$C_{18}H_{24}N_6O$	3248 (NH), 1600 s, br (CO), 1542, 1516 sh	1.03 and 1.06 [2d,6H,$NCH_2CH(\mathit{CH_3})_2$], 1.21 and 1.33 [2t,6H,$N(CH_2\mathit{CH_3})_2$], 2.11 [m,1H,$NCH_2\mathit{CH}(CH_3)_2$], 2.65 [m,1H,1H of $N\mathit{CH_2}CH(CH_3)_2$], 3.24–3.55 [m,4H,3H of $N(\mathit{CH_2}CH_3)_2$ + 1H of $N\mathit{CH_2}CH(CH_3)_2$], 4.08 [m,1H,1H of $N(\mathit{CH_2}CH_3)_2$], 6.68[e] (d,1H,NH), 6.87 (dd,1H,H-3), 8.15 (dd,1H,H-4), 8.37 (dd,1H,H-2), 8.94 (s,1H,H-9)

Table IV. (Continued)

Compound	Yield (%)	M.p. (° C) (solvent)[a]	Molecular formula[b]	IR[c] (cm^{-1})	^{1}H-NMR[d] (δ, ppm)
8k	72	252–253 (C)	$C_{19}H_{26}N_6O$	3260 (NH), 1603 s, br (CO), 1550, 1520 w	1.02 and 1.05 [2d,6H,$NCH_2CH(CH_3)_2$], 1.18 and 1.31 [2t,6H,$N(CH_2CH_3)_2$], 2.09 [m,1H,$NCH_2CH(CH_3)_2$], 2.66 [m,1H,1H of $NCH_2CH(CH_3)_2$], 2.98 (s,3H,9-CH_3), 3.23–3.50 [m,4H,3H of $N(CH_2CH_3)_2$+1H of $NCH_2CH(CH_3)_2$], 4.07 [m,1H,1H of $N(CH_2CH_3)_2$], 6.44[e] (d,1H,NH), 6.91 (dd,1H,H-3), 8.15 (dd,1H,H-4), 8.28 (dd,1H,H-2)
8l	64	223–224 (A)	$C_{20}H_{28}N_6O$	3260 (NH), 1601 s, br (CO), 1548, 1518 w	1.02 and 1.05 [2d,6H,$NCH_2CH(CH_3)_2$], 1.18 and 1.31 [2t,6H,$N(CH_2CH_3)_2$], 1.48 (t,3H,9-CH_2CH_3), 2.09 [m, 1H,$NCH_2CH(CH_3)_2$], 2.67 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.20–3.56 [m,6H,3H of $N(CH_2CH_3)_2$+1H of $NCH_2CH(CH_3)_2$+9-CH_2CH_3], 3.97 [m,1H,1H of $N(CH_2CH_3)_2$], 6.43[e] (d,1H,NH), 6.89 (dd,1H,H-3), 8.14 (dd,1H,H-4), 8.27 (dd,1H,H-2)
8m	53	176–177 (D)	$C_{21}H_{30}N_6O$	3263 (NH), 1603 s, br (CO), 1550, 1520 w	0.96–1.22 [m,9H,$NCH_2CH(CH_3)_2$+9-$CH_2CH_2CH_3$], 1.18 and 1.33 [2t,6H,$N(CH_2CH_3)_2$], 1.77–2.18 [m, 3H,$NCH_2CH(CH_3)_2$+9-$CH_2CH_2CH_3$], 2.72 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.22–3.52 [m, 6H, 3H of $N(CH_2CH_3)_2$+1H of $NCH_2CH(CH_3)_2$+9-$CH_2CH_2CH_3$], 3.97 [m,1H,1H of $N(CH_2CH_3)_2$], 6.13[e] (near d, 1H,NH), 6.99 (dd,1H,H-3), 8.17 (dd,1H,H-4), 8.33 (dd,1H,H-2)
8n	64	189.5–190.5 (D)	$C_{21}H_{30}N_6O$	3265 (NH), 1602 s, br (CO), 1548, 1520 w	1.01 and 1.04 [2d,6H,$NCH_2CH(CH_3)_2$], 1.19 and 1.33 [2t,6H,$N(CH_2CH_3)_2$], 1.38 and 1.62 [2d,6H, 9-$CH(CH_3)_2$], 2.07 [m,1H,$NCH_2CH(CH_3)_2$], 2.74 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.25–3.63 [m,4H,3H of $N(CH_2CH_3)_2$+1H of $NCH_2CH(CH_3)_2$], 3.86 [m,1H,1H of $N(CH_2CH_3)_2$], 4.16 [m,1H,9-$CH(CH_3)_2$], 6.05[e] (near d,1H,NH), 6.99 (dd,1H,H-3), 8.17 (dd, 1H,H-4), 8.35 (dd,1H,H-2)
8o	84	204–205 (F)	$C_{22}H_{32}N_6O$	3250 (NH), 1616 sh, 1606 s (CO), 1547, 1522 w	1.03, 1.04, 1.05 and 1.09 [4d,12H,$NCH_2CH(CH_3)_2$+9-$CH_2CH(CH_3)_2$], 1.18 and 1.33 [2t,6H, $N(CH_2CH_3)_2$], 2.05 [m,1H,$NCH_2CH(CH_3)_2$], 2.32 [m,1H,9-$CH_2CH(CH_3)_2$], 2.77 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.19–3.55 [m,6H,3H of $N(CH_2CH_3)_2$+9-$CH_2CH(CH_3)_2$+1H of $NCH_2CH(CH_3)_2$], 3.96 [m,1H,1H of $N(CH_2CH_3)_2$], 5.93[e] (m,1H,NH), 7.06 (dd,1H,H-3), 8.17 (dd,1H, H-4), 8.38 (dd, 1H, H-2)

Table IV. (Continued)

Compound	Yield (%)	M.p. (° C) (solvent)[a]	Molecular formula[b]	IR[c] (cm^{-1})	^{1}H-NMR[d] (δ, ppm)
8p	39	218–219 (A)	$C_{22}H_{32}N_6O$	3240 (NH), 1620 sh, 1606 s, br (CO), 1548, 1523 w	1.02 [d,6H,$NCH_2CH(CH_3)_2$], 1.19 and 1.34 [2t,6H,$N(CH_2CH_3)_2$], 1.69 [s,9H,9-$C(CH_3)_3$], 2.01 [m,1H,$NCH_2CH(CH_3)_2$], 2.90 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.26–3.87 [m, 5H,$N(CH_2CH_3)_2$+1H of $NCH_2CH(CH_3)_2$], 5.26[e] (m, 1H, NH), 7.25 (dd,1H,H-3), 8.25 (dd,1H,H-4), 8.55 (dd,1H,H-2)
8q	65	215–216 (G)	$C_{25}H_{30}N_6O$	3250 (NH), 1605 s, br (CO), 1546, 1520 sh	1.00 and 1.02 [2d,6H,$NCH_2CH(CH_3)_2$], 1.18 and 1.32 [2t,6H,$N(CH_2CH_3)_2$], 2.06 [m,1H,$NCH_2CH(CH_3)_2$], 2.70 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.22–3.58 [m, 4H, 3H of $N(CH_2CH_3)_2$+1H of $NCH_2CH(CH_3)_2$], 3.98 [m,1H,1H of $N(CH_2(CH_3)_2$], 4.86 and 4.92 (AB system, $J = 16$ Hz, 2H,$CH_2C_6H_5$), 6.28[e] (near d,1H,NH), 6.88 (dd,1H,H-3),7.13–7.38 (m,3H,3H of C_6H_5), 7.50 (m,2H,2H of C_6H_5), 8.06–8.18 (m,2H,H-2,4)
8r	48	231.5–233 (H)	$C_{24}H_{28}N_6O$	3260 (NH),1610 s, br (CO), 1550, 1520 sh	1.06 and 1.08 [2d,6H,$NCH_2CH(CH_3)_2$], 1.22 and 1.35 [2t,6H,$N(CH_2CH_3)_2$], 2.12 [m,1H,$NCH_2CH(CH_3)_2$], 2.73 [m,1H,1H of $NCH_2CH(CH_3)_2$], 3.28–3.56 [m,4H,3H of $N(CH_2CH_3)_2$+1H of $NCH_2CH(CH_3)_2$], 4.07 [m, 1H,1H of $N(CH_2CH_3)_2$], 6.59[e] (near d,1H,NH), 6.98 (dd,1H,H-3), 7.30–7.56 (m,3H, phenyl H-3′,4′,5′), 7.77 (m,2H,phenyl H-2′,6′), 8.08 (dd,1H,H-2), 8.28 (dd,1H,H-4)
8s	68	145–146 (D)	$C_{27}H_{32}N_6O$	3320 br (NH), 1610 s, br (CO), 1536 br	1.04–2.12 (m,10H,cyclohexyl CH_2's), 1.19 and 1.36 [2t,6H,$N(CH_2CH_3)_2$], 3.20–3.69 [m,4H,3H of $N(CH_2CH_3)_2$+cyclohexyl CH], 3.82 [m,1H,1H of $N(CH_2CH_3)_2$], 4.40[e] (d,1H,NH), 4.96 and 5.00 (AB system, $J = 16$ Hz, 2H, $CH_2C_6H_5$), 7.10–7.52 (m,6H, C_6H_5+H-3), 8.23 (dd,1H,H-4), 8.63 (dd,1H,H-2)
8t	47	167–168 (I)	$C_{21}H_{28}N_6O$	3230 br (NH), 1608 s, br (CO), 1537 br	0.36–0.92 (m,4H,cyclopropyl CH_2's), 0.70 and 1.07 [2t,6H,$N(CH_2CH_2CH_3)_2$], 1.51 (t, 3H,9-CH_2CH_3), 1.57–1.92 [m,4H,$N(CH_2CH_2CH_3)_2$], 3.04 (m, 1H,cyclopropyl CH), 3.15–3.73 [m, 6H,$N(CH_2CH_2CH_3)_2$+9-CH_2CH_3], 6.57[e] (near s,1H,NH), 7.02 (dd,1H,H-3), 8.21 (dd,1H,H-4), 8.38 (dd,1H,H-2)

Table IV. (Continued)

Compound	Yield (%)	M.p. (° C) (solvent)[a]	Molecular formula[b]	IR[c] (cm^{-1})	^{1}H-NMR[d] (δ, ppm)
8u	60	236–237 (A)	$C_{22}H_{32}N_6O$	3268 (NH), 1607 s (CO), 1546, 1516 w	0.72 [t,3H,3H of $N(CH_2CH_2\mathit{CH_3})_2$], 1.04 [d + t, 9H, $NCH_2CH(\mathit{CH_3})_2$ + 3H of $N(CH_2CH_2\mathit{CH_3})_2$], 1.50 (t,3H, 9-$CH_2\mathit{CH_3}$), 1.60–2.17 [m, 5H, $N(CH_2\mathit{CH_2}CH_3)_2$ + $NCH_2\mathit{CH}(CH_3)_2$], 2.79 [m,1H,1H of $N\mathit{CH_2}CH(CH_3)_2$], 3.29 [near t,3H,2H of $N(\mathit{CH_2}CH_2CH_3)_2$ + 1H of $N\mathit{CH_2}CH(CH_3)_2$], 3.40–3.71 (m,2H,2H of $N(\mathit{CH_2}CH_2CH_3)_2$], 3.46 (q,2H,9-$\mathit{CH_2}CH_3$), 5.93[e] (near d,1H,NH), 7.03 (dd,1H,H-3), 8.18 (dd,1H,H-4), 8.37 (dd,1H,H-2)
8v	77	87.5–89 (G)	$C_{29}H_{36}N_6O$	3360 br (NH), 1630 sh, 1605 s, br (CO), 1535 br	0.70 and 1.06 [2t,6H,$N(CH_2CH_2\mathit{CH_3})_2$], 1.05–2.10 [m,14H,cyclohexyl CH_2's + $N(CH_2\mathit{CH_2}CH_3)_2$], 3.04–3.46 [m,3H,2H of $N(\mathit{CH_2}CH_2CH_3)_2$ + cyclohexyl CH], 3.59 [t, 2H,2H of $N(\mathit{CH_2}CH_2CH_3)_2$], 4.32[e] (d,1H,NH), 4.97 and 5.02 (AB system, $J = 16$ Hz,2H,$\mathit{CH_2}C_6H_5$), 7.04–7.50 (m,6H,C_6H_5 + H-3), 8.24 (dd,1H,H-4), 8.66 (dd,1H,H-2)
8w	71	215–216 (E)	$C_{21}H_{28}N_6O$	3240 (NH), 1606 s, br (CO), 1550 sh, 1530, 1512 sh	0.40–1.00 (m,4H,cyclopropyl CH_2's), 1.14 and 1.38 [2d,6H,$NCH(\mathit{CH_3})_2$], 1.52 (t,3H,9-$CH_2\mathit{CH_3}$), 1.57 and 1.72 [2d,6H,$NCH(\mathit{CH_3})_2$], 3.17 (m,1H,cyclopropyl CH), 3.41 (q,2H, 9-$\mathit{CH_2}CH_3$), 3.60 and 3.97 {2m,2H,$N[\mathit{CH}(CH_3)_2]_2$}, 6.83[e] (near s,1H,NH), 6.93 (dd,1H,H-3), 8.17 (dd,1H,H-4), 8.28 (dd,1H,H-2)
8x	73	195–196 (A)	$C_{21}H_{28}N_6O$	3270 (NH), 1609 s, br (CO), 1540 br	1.01 and 1.05 [2d,6H,$NCH_2CH(\mathit{CH_3})_2$], 1.28–1.95 (m,6H,piperidine β + γ–CH_2's),1.48 (t,3H,9-$CH_2\mathit{CH_3}$), 2.06 [m,1H,$NCH_2\mathit{CH}(CH_3)_2$], 2.71 [m,1H,1H of $N\mathit{CH_2}CH(CH_3)_2$], 3.20–3.70 [m,4H,3H of piperidine α-CH_2's + 1H of $N\mathit{CH_2}CH(CH_3)_2$], 3.41 (q, 2H, 9-$\mathit{CH_2}CH_3$), 4.11 (m,1H,1H of piperidine α-CH_2), 6.50[e] (near d,1H,NH), 6.87 (dd,1H,H-3), 8.14 (dd,1H,H-4), 8.26 (dd,1H,H-2)

[a] Crystallization solvent: A = ethyl acetate, B = dichloromethane/ethyl acetate, C = dichloromethane/acetone, D = ethyl acetate/petroleum ether, E = ethyl acetate/isopropyl ether, F = dichloromethane/petroleum ether, G = acetone, H = isopropyl ether/acetone, I = ethyl acetate/ethyl ether.
[b] Anal. C,H,N.
[c] In KBr pellets. Abbreviations: s = strong, br = broad, w = weak, sh = shoulder.
[d] In $CDCl_3$ solutions. J values for H-2, H-3, H-4 signals (dd) of all compounds (when determinable): $J_{2,3} = J_{3,2} = 4.6$ Hz, $J_{2,4} = J_{4,2} = 1.5$ Hz, $J_{3,4} = J_{4,3} = 8.3$ Hz.
[e] Disappeared with D_2O.

5.2.3. Analgesic activity

The writhing test [9] was used on groups of six mice. One hour after the administration of the test compound, 0.01 mL/g of a 0.6% acetic acid solution was injected intraperitoneally in each mouse. The writhing movements of each animal were counted for 15 min (between the 5th and 20th min after the injection of the irritant). The antinociceptive effect was expressed as the percentage of protection compared with the control group. Dipyrone (100 mg/kg p.o.) was used as reference standard.

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