Design, Synthesis, and Biological Evaluation of New Inhibitors of the Endocannabinoid Uptake: Comparison with Effects on Fatty Acid Amidohydrolase

María L. López-Rodríguez,*,† Alma Viso,† Silvia Ortega-Gutiérrez,† Christopher J. Fowler,‡ Gunnar Tiger,‡ Eva de Lago,§ Javier Fernández-Ruiz,§ and José A. Ramos§

Departamento de Química Orgánica I, Facultad de Ciencias Químicas, Universidad Complutense, E-28040 Madrid, Spain, Department of Pharmacology and Clinical Neuroscience, Umeå University, Umeå, Sweden, and Departamento de Bioquímica y Biología Molecular III, Facultad de Medicina, Universidad Complutense, E-28040 Madrid, Spain

Received October 28, 2002

A new series of arachidonic acid derivatives were synthesized and evaluated as inhibitors of the endocannabinoid uptake. Most of them are able to inhibit anandamide uptake with IC_{50} values in the low micromolar range ($IC_{50} = 0.8-24\ \mu M$). In general, the compounds had only weak effects upon CB_1, CB_2, and VR_1 receptors ($K_i > 1000-10000$ nM). In addition, there was no obvious relationship between the abilities of the compounds to affect anandamide uptake and to inhibit anandamide metabolism by fatty acid amidohydrolase (FAAH; $IC_{50} = 30-113\ \mu M$). This indicates that the compounds do not exert their effects secondarily to FAAH inhibition. It is hoped that these compounds, particularly the most potent in this series (compound **5**, UCM707, with IC_{50} values for anandamide uptake and FAAH of 0.8 and 30 μM, respectively), will provide useful tools for the elucidation of the role of the anandamide transporter system in vivo.

Introduction

The structural elucidation of Δ^9-tetrahydrocannabinol, achieved in the mid-1960s,[1] marked a milestone for the understanding and rationalization of effects attributed to *Cannabis sativa*. However, it has not been until the past decade when cannabinoids have become a topic of extensive research culminating with the characterization of the endogenous cannabinoid system (ECS).[2] The ECS is primarily responsible for all the effects mediated by cannabinoids, and it is constituted of two G protein coupled receptors named CB_1[3] and CB_2,[4] its endogenous ligands such as anandamide (AEA),[5] 2-arachidonoylglycerol,[6] 2-arachidonyl glyceryl ether,[7] and the recently reported virodhamine,[8] and an inactivation system that degrades these endocannabinoids. This termination system consists of the uptake of anandamide followed by its intracellular metabolism by fatty acid amidohydrolase (FAAH).[9] The ECS is involved in the regulation of a wide variety of physiological functions[10−17] such as antinociception, brain development, memory, retrograde neuronal communication, control of movement, cardiovascular and immune regulation, and cellular proliferation. In consequence, the compounds affecting ECS function are potential therapeutic agents for the treatment of diverse pathologies[18−21] including neurodegenerative disorders, nociceptive alterations, and malignant tumors. In particular, compounds preventing the cellular removal of anandamide are promising pharmacological objectives in the search of new agents able to regulate the ECS but have no undesirable side effects.[22] In this respect, preliminary results obtained for the potential use of synthetic inhibitors for the treatment of pain[23] and neurodegenerative diseases such as multiple sclerosis[24] and Huntington's chorea[25] are very promising. However, the relative paucity of structure−affinity relationship (SAFIR) studies reported so far, together with the lack of molecular characterization of the anandamide uptake process, has hampered the development of potent and selective inhibitors of the endocannabinoid uptake and only a few have been described to date.[26−31]

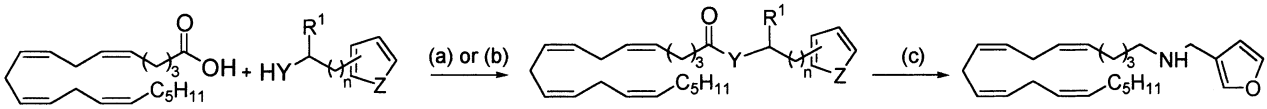
The nature of the transport system for AEA is rather controversial. It is generally described as a system of facilitated transport,[26,32] termed the anandamide transporter system (ANT), that can be inhibited by arachidonic acid derivatives such as *N*-(4-hydroxyphenyl)-arachidonamide (AM404).[26] However, it is clear that FAAH, by removing the intracellularly accumulated anandamide, contributes to the anandamide gradient across the plasma membrane and hence its uptake.[33,34] Indeed, it has been argued that at least some of the available ANT inhibitors may in fact be acting primarily on FAAH,[33,35] although some cannabidiol derivatives, with partial selectivity for the ANT vs FAAH and cannabinoid receptors have also been reported.[36] In addition, compounds such as AM404 can affect other cellular systems, such as the vanilloid receptor (VR_1).[37] There is thus a major need for (a) the identification of potent and selective ANT inhibitors and (b) further SAFIR data comparing effects of the compounds on ANT and FAAH.

The above facts prompted us to address a study on the structural requirements for the recognition ANT-substrates. Consequently, within a program aimed at discovering potent and selective inhibitors of endocan-

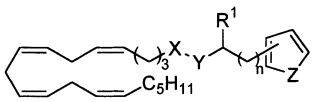
* To whom correspondence should be addressed. Phone: 34-91-3944239. Fax: 34-91-3944103. E-mail: mluzlr@quim.ucm.es.
† Departamento de Química Orgánica I, Universidad Complutense.
‡ Umeå University.
§ Departamento de Bioquímica y Biología Molecular III, Universidad Complutense.

10.1021/jm0210818 CCC: $25.00
Published on Web 03/15/2003

Scheme 1. Synthesis of Compounds of General Structure **I**[a]



[a] Reagents: (a) oxalyl chloride, DMF, CH_2Cl_2, rt; (b) DCC, DMAP, CH_2Cl_2, rt; (c) **5**, $LiAlH_4$, THF, Δ, Ar.



I (1-28)

X = CO, CH_2; Y = NH, NR^2, O, S; Z = N-CH_3, O, S; R^1 = H, CH_3; n = 0,1

Figure 1. Compounds of general structure **I**.

nabinoid uptake, we have designed and synthesized a new series of arachidonic acid derivatives of general structure **I** (Figure 1), in which we have replaced the ethanolamine moiety of anandamide with a fragment containing a heterocyclic ring, that have been characterized as highly potent and selective inhibitors of the anandamide reuptake.[38,39]

In the present study, we have considered a series of compounds of general structure **I** to gain insights on the requirements of the heterocyclic moiety (HM) and its position, the −X−Y− fragment, and the increase in the distance and branching between the arachidonic acid chain and the heterocyclic ring for affinity and selectivity for the ANT. In addition, we have compared the effects of the compounds on the ANT and the FAAH to determine whether the effects on these two target systems can be pharmacologically separated. The interaction of the compounds with CB_1 and CB_2 cannabinoid receptors and with VR_1 vanilloid receptors has also been determined. Our results have allowed us to carry out a structure−affinity relationship study focused on the molecular requirements involved in the uptake process, thus providing a basis for the design of more potent and selective ANT inhibitors with potential applications to the treatment of a wide variety of diseases.

Chemistry

The synthesis of the amides and esters of general structure **I** (**1**−**6**, **8**−**28**) listed in Table 1 is detailed in Scheme 1. These compounds were prepared from arachidonic acid by treatment of its acyl chloride with the appropriate amine or alcohol (method A) or by direct condensation between the arachidonic acid and the corresponding amine, alcohol, or thiol in the presence of dicyclohexylcarbodiimide (DCC) and catalytic amounts of *N*,*N*-dimethyl-4-aminopyridine (DMAP) (method B). Additionally, treatment of amide **5** with $LiAlH_4$ provided secondary amine **7**.

The synthesis of the noncommercial amines was carried out by direct reductive amination of the corresponding heterocyclic aldehydes or by reduction of the corresponding nitrovinyl derivatives using methods previously described in the literature.[39−44]

Biochemical in Vitro Assays

All new compounds were assessed for their ability to inhibit [^{3}H]anandamide uptake in human lymphoma U937 cells, for their affinity for CB_1 and CB_2 cannabinoid receptors in radioligand binding assays using [^{3}H]-WIN552122 in rat cerebellum membranes and [^{3}H]-CP55940 in HEK293EBNA human CB_2 receptor transfected cells, respectively, and for their affinity for the vanilloid VR_1 receptor in radioligand binding assays using [^{3}H]resiniferatoxin ([^{3}H]RTX) in rat spinal cord membranes (Table 1). Also, synthesized compounds **I** were evaluated for their capacity to inhibit the FAAH-catalyzed metabolism of anandamide by rat brain homogenates. For comparative purposes, we have also included results obtained for AM404.

Results and Discussion

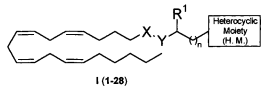
Most of the compounds with structure **I** display an excellent ability to inhibit anandamide reuptake with IC_{50} values in the low micromolar range as well as selectivity for the transporter, as deduced from their affinity constants for CB_1, CB_2, and VR_1 receptors (K_i > 1000−10000 nM), and for the enzyme FAAH, with IC_{50} values 1 order of magnitude greater than those for the ANT (Table 1). This contrasts with the data for previously reported inhibitors that inhibit FAAH and the ANT with similar potencies[28] such as, for example, AM404 (Table 1).

SAFIR of Inhibitors of Anandamide Uptake. As an exploration of the SAFIR between compounds of general structure **I** and ANT, we have examined the role of the heterocyclic ring and its position, the −X−Y− fragment, and the increase in the distance and branching between the arachidonic acid chain and the heterocyclic moiety. The most important points can be expressed in the following terms:

(i) To evaluate the influence of the heterocyclic moiety, we have considered compounds with five-membered rings with one heteroatom (furan, thiophene, and pyrrole). Regarding furan derivatives, amides exhibit a higher potency as ANT inhibitors compared to their corresponding isosteric esters, as shown from the IC_{50} values for analogues **2** vs **1** (5 vs 24 μM) and **5** vs **4** (0.8 vs 14 μM). Also, the presence of the arachidonic acid chain at position 3 of the furan ring enhances the affinity for the transporter (**4** vs **1**, with IC_{50} values of 14 vs 24 μM, respectively, and **5** vs **2**, with IC_{50} values of 0.8 vs 5 μM, respectively). However, within thienyl derivatives, these structural factors are not so relevant, since the IC_{50} values for compounds **16**, **18**, and **19** are comparable (between 3 and 5 μM). The lack of activity of compound **15** seems to indicate, however, an exception. Subsequently, we have compared the pyrrole ring with its corresponding furan and thienyl analogues, observing equipotent IC_{50} values for compounds **20**, **2**, and **16**, respectively.

With respect to the aromaticity of the heterocyclic moiety, when the arachidonic chain is attached to position C-2, the equipotent IC_{50} values observed for

Table 1. Inhibition of ANT and FAAH and Binding Data of Compounds with Structure **I** (**1**−**28**)[l]



I (1-28)

					ANT[a]		FAAH[b]		Receptor Affinity		
Compound	H M	X-Y	R^1	n	Max. Inh. (%)	IC_{50} (μM)	Max. Inh. (%)	IC_{50} (μM)	$K_i CB_1$ (nM)[c]	$K_i CB_2$ (nM)[d]	$K_i VR_1$ (nM)[e]
1		CO-O	H	0	>85	24±14	100	53	>5000	>5000	>5000
2		CO-NH	H	0	>85	5±2	100	34	>5000	>5000	*f*
3		CO-S	H	0	>85	9±2	36±7 at 100 μM[g]		>5000	>5000	>5000
4		CO-O	H	0	>85	14±2	100	103	>5000	>5000	*f*
5		CO-NH	H	0	>85	0.8±0.4	100	30	4700±80	67±6	>5000
6		CO-N(Me)	H	0	>85	111±51	24±4 at 100 μM[g]		>1000	>1000	*h*
7		CH_2-NH	H	0	>85	*i*	57±11	29	>1000	>1000	>5000
8		(±) CO-O	H	0	>85	18±7	23±1 at 100 μM[g]		>5000	>5000	>5000
9		(±) CO-NH	H	0	>85	8±2	47±14	44	>5000	>5000	>5000
10		(±) CO-O	H	0	>85	*j*	100	113	>5000	>10000	>5000
11		(±) CO-O	CH_3	0	70±2	*i*	45±11 at 100 μM[g]		>1000	>1000	>5000
12		(*R*) (+) CO-O	CH_3	0	37±2	*i*	19±5 at 100 μM[g]		>1000	>5000	>5000
13		(*S*) (-) CO-O	CH_3	0	55±4	*i*	24±0.6 at 100 μM[g]		>1000	>5000	>5000
14		(±) CO-NH	CH_3	0	>85	21±12	75±11	36	>5000	>5000	>5000
15		CO-O	H	0	Inactive		80±6	12	>5000	>5000	>5000
16		CO-NH	H	0	>85	5.7±0.6	100	112	>1000	>10000	>5000
17			H	0	>85	16±4	*h*	*h*	>1000	>1000	>5000
18		CO-O	H	0	>85	3±2	100	84	>5000	>5000	>5000
19		CO-NH	H	0	>85	5±2	76±6	12	>1000	>1000	>5000
20		CO-NH	H	0	>85	5.0±0.7	*h*	*h*	124±1	70±5	>5000
21		CO-NH	H	1	>85	12.8±0.4	38±1 at 100 μM[g]		>5000	>1000	>5000
22		CO-NH	H	1	>85	3.3±0.9	*h*	*h*	>1000	400±100	>5000
23		CO-O	H	1	>85	12±1	68±13	24	>1000	>5000	>5000
24		CO-NH	H	1	>85	19±3	100	102	3000±1000	>1000	>5000
25		(±) CO-NH	CH_3	1	66±2	22±1	36±6	15	>1000	>1000	*f*
26		CO-O	H	1	75±8	26±9	100	28	>1000	>5000	>5000
27		CO-NH	H	1	60±11	1.3±0.5	*h*	*h*	>1000	>1000	*f*
28		(±) CO-NH	CH_3	1	78±3	21±6	37±3	*g*	>1000	>1000	*f*
AM404					>85	4±2	100[k]	3.7[k]	>1000	>1000	>5000

[a] Inhibition of anandamide transport was determined using human lymphoma U937 cells and [^{3}H]anandamide. Maximum inhibition values are expressed as the percentage of inhibition produced by the highest dose of the compounds tested (50 μM). [b] Inhibition of FAAH enzymatic activity was determined using rat brain homogenates as the enzyme source and [^{3}H]anandamide. Maximum inhibition values were calculated by GraphPad Prism analyses[53] of the inhibition curves over eight concentrations ranging from 0.5 to 100 μM. [c] Affinity of compounds for the CB_1 receptor was evaluated using rat cerebellum membranes and [^{3}H]WIN552122. [d] Affinity of compounds for the CB_2 receptor was assayed using [^{3}H]CP55940 in HEK293EBNA human CB_2 receptor transfected cells. [e] Affinity of compounds for the VR_1 receptor was evaluated using rat spinal cord membranes and [^{3}H]RTX. Maximum inhibition, IC_{50}, and K_i values were obtained from two to four independent experiments carried out in triplicate and are expressed as the mean ± standard error. [f] These compounds exhibited a partial capacity to inhibit [^{3}H]RTX binding (K_i < 5000 nM). [g] When the calculated IC_{50} value was well over the highest inhibitor concentration tested (i.e., >120 μM) or alternatively no curve could be fitted for statistical reasons, the observed inhibition at 100 μM is given. [h] Not determined. [i] These compounds presented hyperbolic inhibitory profiles instead of the sigmoid dose−response curves. [j] This compound presented a strange profile that did not fit the sigmoid dose−response characteristic of the rest of the compounds. [k] Data from ref 53. [l] IC_{50} (ANT) data and receptor affinity for compounds **1**, **2**, **4**, **5**, **8**−**10**, **15**, **16**, **18**, and **20** were previously reported.[39]

compounds **1** vs **8** (24 vs 18 μM) and **2** vs **9** (5 vs 8 μM) indicate that the presence of an aromatic fragment is not essential for affinity. However, when the arachidonic chain is attached to position C-3 (compound **4** vs **10**), aromaticity becomes an important factor in the inhibitory trend observed, which does not fit the sigmoid dose−response typical of the rest of compounds. This unusual behavior of compound **10** may be caused by the existence of different mechanisms of interaction or recognition between the transporter and its substrates.

(ii) With respect to the nature of the −X−Y− fragment, in general, secondary amides, esters, and thioesters are all able to compete with anandamide for transport, although the presence of the NH group seems to enhance the affinity for the carrier. Some representative examples include the higher potency of amide **2** vs its corresponding isosters **1** and **3** (IC_{50} values of 5 vs 24 and 9 μM, respectively), the increase in affinity of nearly 20-fold between compounds **4** and **5** (14 and 0.8 μM), and especially the recovery of inhibitory ability observed between compounds **15** (inactive) and **16** (5.7 μM). Tertiary amides critically diminish the affinity for the ANT. This effect can be appreciated in the 3-fold loss in affinity displayed by compound **17** vs **16** (16 vs 5.7 μM) as well as in the dramatic decrease in the inhibitory ability of compound **6** vs **5** (IC_{50} values of 111 and 0.8 μM). However, these results do not seem to indicate the involvement of the NH group in a hydrogen bond, since simple tertiary amides with IC_{50} values ranging from 10 to 30 μM have been reported.[27]

Isosteric replacement of a carbonyl group for a methylenic group (compound **5** vs **7**) implies a change in the inhibitory trend observed for derivative **7**. Although amine **7** is able to interact efficaciously with the ANT, as deduced from its high maximum inhibition value (90%), it does not display a sigmoid profile. These results suggest a different mode of interaction with the ANT for both substrates, probably due to the presence of the carbonyl group, which could fix the bioactive conformation of compound **5**.

(iii) Increase in the distance between the arachidonic chain and the heterocyclic subunit (compounds with $n = 1$) produces in all cases important decreases in the inhibitory potency of the compounds. For instance, a 3-fold loss of affinity is observed for furan derivatives **21** vs **2** (12.8 vs 5 μM) and **22** vs **5** (3.3 vs 0.8 μM). A similar effect was obtained for thienyl derivatives with marked decreases in affinity (**24** vs **16**, with IC_{50} values of 19 and 5.7 μM, respectively) or in maximum inhibition capacity (**27** vs **19**, with 60% and >85%) or both (**26** vs **18**, with 75% and 26 μM vs >85% and 3 μM, respectively). Compound **23** follows the general trend observed for derivatives with $n = 1$.

Branching (R^1) in the fragment between the arachidonic chain and the heterocyclic ring implies, in all analyzed cases ($n = 0, 1$), a critical decrease in the inhibitory potency, as reflected by the IC_{50} values of compounds **14** and **2** (21 vs 5 μM) and **28** vs **27** (21 vs 1.3 μM) or by the maximum inhibitory capacity of compounds **11** (70%), **12** (37%), and **13** (55%) vs **1** (>85%) and **25** (66%) vs **24** (>85%).

In general, the fact that these compounds show maximum inhibition values clearly under the 85% indicates an important decay in the efficacy of their interaction with the transporter.

Additionally, compounds **12** and **13** allow us to analyze the effect of stereochemistry in the capacity to interact with the transporter. From their maximum inhibition values, it is deduced that the capacity of the *S* enantiomer to inhibit anandamide uptake is superior to that shown for the *R* enantiomer (55% vs 37%, respectively). This result is in accordance with previously reported data in the literature for other anandamide analogues, which also describes the enantiomeric preference of the ANT for the *S* isomer.[27,45]

Transporter Selectivity. (i) With respect to cannabinoid receptors, most of the compounds analyzed here turned out to have little affinity for both CB_1 and CB_2 (K_i > 1000−10000 nM). However, it should be pointed out that the presence of the *N*-methylpyrrole fragment dramatically increases the affinity for both cannabinoid receptors as deduced from the K_i values obtained for **20** ($K_i(CB_1) = 124$ nM; $K_i(CB_2) = 70$ nM). Moreover, the presence of the fur-3-ylmethyl moiety selectively enhances the affinity for CB_2, as shown by compounds **5** and **22** [$K_i(CB_2)$ values of 67 and 400 nM and $K_i(CB_1)$ values of 4700 and >1000 nM, respectively].

(ii) Regarding the interaction of compounds with vanilloid VR_1 receptors, the data obtained here show that most of the compounds synthesized are essentially unable to bind to VR_1 (K_i > 5000 nM), especially when these data are compared with the K_i values of high-affinity VR_1 ligands such as RTX ($K_d = 25$ pM,[46] which is in accordance with the value determined in our experiment; see methods section).

(iii) With respect to the capacity of compounds of general structure **I** to inhibit the catalytic activity of FAAH, the majority of them have been characterized as weak FAAH inhibitors. In contrast to the equipotent effects of AM404 on ANT and FAAH, many of the compounds in the present series were better inhibitors of ANT than of FAAH. The most pronounced difference was for compound **5**, with a ~40-fold selectivity between effects on ANT and FAAH. With respect to effects on FAAH, the compounds either showed a low potency or alternatively were not able to produce a complete inhibition of anandamide metabolism. Examples of the former are compounds **5**, **16**, and **18**, with IC_{50} values for the FAAH of 30, 112, and 84 μM, respectively. Examples of the latter are compounds **3**, **9**, and **21**, with maximum inhibition values in the FAAH ranging from 36% to 47%.

Conclusions

From all the above considerations, it is clear that small structural variations in compounds of structure **I** have allowed us to obtain different selectivities and affinities in the four systems that constitute the ECS. Perhaps the most important finding from the present study is that there is no obvious correlation between the effects of the compounds on FAAH and on the ANT. Thus, for example, compound **15** is more potent than compound **5** as an inhibitor of FAAH; indeed, compound **15** is inactive in the ANT. Similarly, compounds **2**, **5**, **14**, and **26** have essentially identical potencies for inhibition of anandamide metabolism but vary considerably in their abilities to inhibit the uptake of ananda-

mide (IC_{50} values range from 0.8 to 26 μM). This would suggest that the ANT and FAAH can be pharmacologically separated and that the potencies of the present compounds on the ANT system investigated do not simply reflect their abilities to prevent the metabolism of the intracellularly accumulated anandamide. Of the compounds tested, compound **5** (UCM707) remains as the most potent ANT inhibitor so far described in the literature. The excellent IC_{50} value of 0.8 μM exhibited by this compound and its selectivity for the ANT vs CB_1 receptors (K_i = 4700 nM), VR_1 (K_i > 5000 nM), and FAAH (IC_{50} = 30 μM) make it a valuable candidate for future and deeper pharmacological studies. In addition, the fact that this compound, the most potent derivative in the transporter reported to date, displays a moderate affinity only to CB_2 (K_i = 67 nM) raises the possibility of selectively affecting the activity of this receptor and simultaneously inhibiting the metabolism of endocannabinoids, with all the interesting and attractive therapeutic applications that this implies.

Indeed, the compound is active in vivo and is able to enhance the hypokinetic and/or antinociceptive actions of a subeffective dose of anandamide,[47] so more detailed in vivo studies are in progress. Further syntheses aimed at developing new furan derivatives bearing different substituents are currently under investigation in our laboratory and will be reported in due course.

Experimental Section

Chemistry. Infrared (IR) spectra were determined on a Perkin-Elmer 781 or Shimadzu-8300 infrared spectrophotometer. Optical rotation [α] was measured using a Perkin-Elmer 781 polarimeter. ^{1}H and ^{13}C NMR spectra were recorded on a Varian VXR-300S, Bruker Avance 300-AM or Bruker 200-AC instrument at room temperature (rt) unless stated otherwise. Chemical shifts (δ) are expressed in parts per million relative to internal tetramethylsilane; coupling constants (J) are in hertz. Satisfactory elemental analyses were obtained for all the newly synthesized analogues and are within ±0.4% of the theoretical values. Thin-layer chromatography (TLC) was run on Merck silica gel 60 F-254 plates. For normal pressure chromatography, Merck silica gel type 60 (size 70−230) was used. Unless stated otherwise, starting materials used were high-grade commercial products from Aldrich, Acros, Fluka, Merck, or Panreac except arachidonic acid (90% pure), which was purchased from Sigma. Anhydrous dimethylformamide (DMF) was obtained by stirring over CaH_2, filtering, and vacuum distillation. Methylene chloride was used freshly distilled over CaH_2. Anhydrous tetrahydrofuran (THF) was used freshly distilled under argon over sodium and in the presence of benzophenone as indicator.

Synthesis of Noncommercial Amines. The synthesis of noncommercial amines (n = 0) was carried out by catalytic hydrogenation (Raney nickel) of the corresponding aldehydes or oximes in the presence of NH_4OH or CH_3NH_2 using methods previously reported.[39−41,44]

The synthesis of ethylamino derivatives (n = 1) was performed by reduction ($LiAlH_4$ or BH_3) of the corresponding nitrovinyl compounds obtained from the corresponding aldehydes and nitromethane or nitroethane using routes described in the literature.[42,43] Representative spectroscopic data of these amines are gathered below.

(Fur-3-ylmethyl)amine. R_f = 0.35 (diethyl ether/methanol/ammonium hydroxide, 1:1:0.02). IR ($CHCl_3$, cm^{-1}): 3377, 3134, 3113, 2866, 1500, 1387, 1161, 1020, 874. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.72 (br s, 2H, NH_2), 3.68 (s, 2H, CH_2), 6.34 (m, 1H, H-4), 7.28−7.30 (m, 2H, H-2, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 36.8, 109.6, 126.8, 138.7, 143.0.

(1-Methyl-1*H*-pyrrol-2-ylmethyl)amine. R_f = 0.18 (chloroform/methanol, 95:5). IR ($CHCl_3$, cm^{-1}): 3018, 2854, 1159, 930. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.66 (br s, 2H, NH_2), 3.63 (s, 3H, CH_3), 3.80 (s, 2H, CH_2), 6.01−6.10 (m, 2H, H-3, H-4), 6.59−6.64 (m, 1H, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 35.1, 36.9, 106.3, 112.8, 126.7, 128.2.

(±)-1-(Fur-2-yl)ethylamine. R_f = 0.17 (chloroform/methanol, 9:1). IR (CH_2Cl_2, cm^{-1}): 3369, 3298, 3051, 2972, 2930, 2874, 1678, 1601, 1572, 1470, 1150, 1009, 928, 883, 872, 806, 702. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.21 (d, 3H, J = 6.6 Hz, CH_3), 1.59 (br s, 2H, NH_2), 3.86 (q, 1H, J = 6.6 Hz, C*H*−NH_2), 5.90 (dt, 1H, J = 3.2, 0.7 Hz, H-3), 6.09 (dd, 1H, J = 3.2, 2.0 Hz, H-4), 7.13 (dd, 1H, J = 1.9, 0.7 Hz, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 21.9, 45.0, 103.1, 109.9, 141.0, 160.2.

***N*-(Fur-3-ylmethyl)-*N*-methylamine.** R_f = 0.13 (chloroform/methanol, 8:2). IR ($CDCl_3$, cm^{-1}): 3325, 3132, 2941, 1638, 1500, 1377, 1163, 874. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.98 (br s, 1H, NH), 2.36 (s, 3H, CH_3), 3.52 (s, 2H, CH_2), 6.28−6.31 (m, 1H, H-4), 7.28−7.31 (m, 2H, H-2, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 36.7, 46.3, 110.5, 123.8, 140.6, 143.1.

(Thien-3-ylmethyl)amine. R_f = 0.11 (chloroform). IR (CH_2Cl_2, cm^{-1}): 3366, 3296, 3049, 2924, 2854, 1638, 1558, 1456, 1244, 779. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.70 (br s, 2H, NH_2), 3.88 (s, 2H, CH_2), 7.03 (dd, 1H, J = 4.9, 1.0 Hz, H-4), 7.10 (m, 1H, H-2), 7.28 (dd, 1H, J = 4.9, 2.9 Hz, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 41.7, 120.3, 126.0, 126.9, 144.6.

***N*,*N*-Bis(thien-2-ylmethyl)amine.** R_f = 0.22 (chloroform:methanol, 95:5). ^{1}H NMR (200 MHz, $CDCl_3$, δ): 4.01 (s, 4H, $2CH_2$), 4.44 (br s, 1H, NH), 6.90−7.01 (m, 4H, 2H-3, 2H-4), 7.20−7.22 (m, 2H, 2H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 46.0 (2C), 125.5 (2C), 126.2 (2C), 126.7 (2C), 127.1 (2C).

2-(Fur-2-yl)ethylamine. R_f = 0.06 (chloroform/methanol, 9:1). IR ($CDCl_3$, cm^{-1}): 3310, 3155, 2926, 1597, 1560, 1466, 1429, 1148, 1011, 845. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.89 (br s, 2H, NH_2), 2.79 (t, 2H, J = 6.6 Hz, C*H*$_2$−C), 2.94 (t, 2H, J = 6.6 Hz, C*H*$_2$−NH_2), 6.06 (dd, 1H, J = 3.2, 0.7 Hz, H-3), 6.30 (dd, 1H, J = 3.2, 1.5 Hz, H-4), 7.33 (dd, 1H, J = 1.7, 0.7 Hz, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 31.8, 40.4, 105.9, 110.0, 141.1, 153.6.

2-(Fur-3-yl)ethylamine. R_f = 0.55 (chloroform/methanol/ammonium hydroxide, 7:3:0.1). IR (CH_2Cl_2, cm^{-1}): 3132, 3111, 2928, 1638, 1578, 1466, 1439, 1383, 1159, 1067, 874. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.56 (br s, 2H, NH_2), 2.56 (t, 2H, J = 6.8 Hz, C*H*$_2$−C), 2.89 (t, 2H, J = 6.6 Hz, C*H*$_2$−NH_2), 6.28−6.29 (m, 1H, H-4), 7.27 (dd, 1H, J = 1.5, 1.0 Hz, H-2), 7.38 (t, 1H, J = 1.7 Hz, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 29.0, 42.2, 110.9, 122.4, 139.6, 143.0.

2-(Thien-3-yl)ethylamine. R_f = 0.15 (chloroform/methanol, 9:1). Yield = 60%. IR (CH_2Cl_2, cm^{-1}): 3360, 3277, 3099, 2961, 2855, 1458, 1080, 704. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 2.06 (br s, 2H, NH_2), 2.79 (t, 2H, J = 6.7 Hz, C*H*$_2$−C), 2.96 (t, 2H, J = 6.8 Hz, C*H*$_2$−NH_2), 6.96 (dd, 1H, J = 4.9, 1.5 Hz, H-4), 7.00 (dd, 1H, J = 2.9, 1.2 Hz, H-2), 7.28 (dd, 1H, J = 4.9, 3.2 Hz, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 34.1, 42.6, 121.0, 125.6, 128.1, 139.9.

(±)-1-(Methyl-2-thien-2-yl)ethylamine. R_f = 0.11 (chloroform). Yield = 48%. IR ($CDCl_3$, cm^{-1}): 3368, 2962, 2928, 1583, 1456, 1437, 1381, 1148, 1059. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.12 (d, 3H, J = 6.1 Hz, CH_3), 1.91 (br s, 2H, NH_2), 2.70 (ddd, 1H, J = 14.3, 8.1, 0.5 Hz, 1H−CH_2), 2.89 (ddd, 1H, J = 14.4, 4.9, 0.7 Hz, 1H−CH_2), 3.06−3.22 (m, 1H, CH), 6.79−6.82 (m, 1H, H-3), 6.92 (dd, 1H, J = 5.1, 3.4 Hz, H-4), 7.13 (dd, 1H, J = 5.1, 1.2 Hz, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 23.1, 40.4, 48.6, 123.7, 125.6, 126.8, 141.8.

(±)-1-(Methyl-2-thien-3-yl)ethylamine. R_f = 0.10 (chloroform). Yield = 51%. IR (CH_2Cl_2, cm^{-1}): 3360, 3286, 3099, 3047, 2960, 1583, 1454, 1379, 1344, 1080, 833, 773. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.04 (d, 3H, J = 6.3 Hz, CH_3), 2.53 (dd, 1H, J = 13.7, 8.0 Hz, 1H−CH_2), 2.70 (ddd, 1H, J = 13.7, 5.1, 0.5 Hz, 1H−CH_2), 3.14 (sext, 1H, J = 6.3 Hz, CH), 4.72 (br s, 2H, NH_2), 6.92 (dd, 1H, J = 4.9, 1.2 Hz, H-4), 6.96 (ddd, 1H, J = 2.9, 1.0, 0.5 Hz, H-2), 7.24 (dd, 1H, J = 4.9, 2.9 Hz, H-5). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 23.8, 40.6, 47.6, 121.4, 125.3, 128.4, 139.8.

General Procedure for the Synthesis of Derivatives 1−6 and 8−28. Method A. Arachidonyl chloride was synthe-

sized by reaction of arachidonic acid (111.1 mg, 0.33 mmol) in methylene chloride (5 mL/mmol) with 2 equiv of oxalyl chloride in the presence of 1 equiv of DMF under argon atmosphere. After 1 h at room temperature, the solvent was removed under vacuum, arachidonyl chloride was dissolved in methylene chloride (5 mL/mmol), and the appropiate alcohol or amine (3.3 mmol approx) in a methylene chloride solution (1.5 mL/mmol) was added. The mixture was stirred (3−6 h) until no further evolution was observed by TLC (chloroform/methanol, 95:5) and then was washed with water and brine and dried over Na_2SO_4 or $MgSO_4$. Then, the solvent was evaporated under reduced pressure and the crude product was purified by column chromatography on silica gel using the appropriate eluent.

Method B. To a stirred solution of 1 equiv (0.33 mmol) of arachidonic acid in dry methylene chloride (1 mL/mmol) and the appropriate alcohol, thiol, or amine (1.5 equivalents) in dry methylene chloride (1 mL/mmol) at −20 °C in a salt/ice bath and under argon, a solution of DCC (1 equiv) and DMAP (0.068 equiv) in dry methylene chloride (3 mL/mmol DCC) was added dropwise. The mixture was stirred for 5 min at this temperature and then removed from the cooling bath and stirred at room temperature (3−6 h) until no further evolution was observed by TLC (chloroform/methanol, 95:5). The dicyclohexylurea was filtered off, the filtrate was evaporated under reduced pressure, and the obtained residue was taken up in methylene chloride (20 mL/mmol of fatty acid). This resulting organic phase was washed with a cooled 0.5 M hydrochloric acid solution and brine, and the organic extracts were dried over Na_2SO_4 or $MgSO_4$. Then the solvent was evaporated under reduced pressure and the product purified by column chromatography on silica gel using the appropriate eluent.

The synthesis of compounds **1**, **2**, **4**, **5**, **8**−**10**, **15**, **16**, **18**, and **20** was carried out as previously reported.[39]

Fur-2-ylmethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 1. Method A. R_f = 0.18 (hexane/methylene chloride, 2:1). Yield = 57%. IR ($CHCl_3$, cm^{-1}): 3013, 2957, 2928, 2856, 1736, 1456, 1377, 1229, 1153, 910, 734. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.78 (t, 3H, J = 6.8 Hz, 3H-20), 1.15−1.36 (m, 6H, 2H-17, 2H-18, 2H-19), 1.60 (quint, 2H, J = 7.5 Hz, 2H-3), 1.93−2.08 (m, 4H, 2H-4, 2H-16), 2.28 (t, 2H, J = 7.4 Hz, 2H-2), 2.68−2.79 (m, 6H, 2H-7, 2H-10, 2H-13), 4.99 (s, 2H, 2H-1″), 5.30−5.40 (m, 8H, vinylic-H), 6.35 (dd, 1H, J = 3.2, 1.8 Hz, H-4′), 6.39 (d, 1H, J = 3.1 Hz, H-3′), 7.41 (dd, 1H, J = 1.8, 0.8 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.1, 22.4, 24.6, 25.6 (3C), 26.6, 27.4, 29.5, 31.5, 33.6, 58.0, 110.7 (2C), 127.5, 127.9, 128.1, 128.2, 128.6, 128.8, 128.9, 130.5, 143.2, 149.8, 173.3.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(Fur-2-ylmethyl)icosa-5,8,11,14-tetraenamide, 2. Method A. R_f = 0.80 (chloroform/methanol, 95:5). Yield = 57%. IR ($CHCl_3$, cm^{-1}): 3450, 2927, 1676, 1512, 1425, 1149, 1014, 927. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.87 (t, 3H, J = 6.8 Hz, 3H-20), 1.24−1.28 (m, 6H, 2H-17, 2H-18, 2H-19), 1.75 (quint, 2H, J = 7.1 Hz, 2H-3), 1.97−2.22 (m, 6H, 2H-4, 2H-16, 2H-2), 2.78−2.83 (m, 6H, 2H-7, 2H-10, 2H-13), 4.41 (d, 2H, J = 5.4 Hz, 2H-1″), 5.35−5.38 (m, 8H, vinylic-H), 5.83 (br s, 1H, NH), 6.20 (d, 1H, J = 7.4 Hz, H-3′), 6.29−6.31 (m, 1H, H-4′), 7.32−7.34 (m, 1H, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.5, 25.3, 25.6 (3C), 26.6, 27.2, 29.3, 31.5, 35.9, 36.4, 107.4, 110.4, 127.5, 127.8, 128.1, 128.2, 128.6, 128.8, 129.1, 130.5, 142.1, 151.4, 172.5.

***S*-(Fur-3-ylmethyl) (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenethioate, 3. Method B.** R_f = 0.17 (hexane/ethyl ether, 100:1). Yield = 59%. IR (CH_2Cl_2, cm^{-1}): 3013, 2957, 2928, 2856, 1693, 1502, 1456, 1248, 1151, 1072, 1011, 910. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.89 (t, 3H, J = 6.6 Hz, 3H-20), 1.25−1.40 (m, 6H, 2H-17, 2H-18, 2H-19), 1.75 (quint, 2H, J = 7.3 Hz, 2H-3), 2.01−2.16 (m, 4H, 2H-4, 2H-16), 2.58 (t, 2H, J = 7.8 Hz, 2H-2), 2.76−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 4.15 (s, 2H, 2H-1″), 5.27−5.47 (m, 8H, vinylic-H), 6.21 (dq, 1H, J = 3.2, 0.7 Hz, H-4′), 6.28 (dd, 1H, J = 3.2, 1.7 Hz, H-3′), 7.32 (dd, 1H, J = 1.7, 0.7 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 13.9, 22.6, 25.3, 25.6 (4C), 26.3, 27.2, 29.3, 31.5, 43.7, 107.7, 110.4, 127.5, 127.8, 128.1, 128.2, 128.5, 128.6, 129.1, 130.5, 142.0, 150.4, 197.9.

Fur-3-ylmethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 4. Method A. R_f = 0.15 (hexane/methylene chloride, 2:1). Yield = 55%. IR ($CHCl_3$, cm^{-1}): 3013, 2957, 2928, 2856, 1734, 1458, 1379, 1217, 1151, 1024, 874. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.6 Hz, 3H-20), 1.22−1.37 (m, 6H, 2H-17, 2H-18, 2H-19), 1.70 (quint, 2H, J = 7.5 Hz, 2H-3), 2.01−2.13 (m, 4H, 2H-4, 2H-16), 2.32 (t, 2H, J = 7.5 Hz, 2H-2), 2.80 (m, 6H, 2H-7, 2H-10, 2H-13), 4.97 (s, 2H, 2H-1″), 5.28−5.41 (m, 8H, vinylic-H), 6.41 (m, 1H, H-4′), 7.38 (m, 1H, H-2′), 7.45−7.46 (m, 1H, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.5, 24.7, 25.6 (2C), 25.7, 26.5, 27.2, 29.3, 31.5, 33.6, 57.5, 110.5, 120.5, 127.5, 127.8, 128.1, 128.2, 128.6, 128.9, 129.0, 130.5, 141.5, 143.4, 173.4.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(Fur-3-ylmethyl)icosa-5,8,11,14-tetraenamide, 5 (UCM707). Yield = 45% for method A. Yield = 65% for method B. R_f = 0.43 (hexane/ethyl acetate, 1:1). IR ($CHCl_3$, cm^{-1}): 3369, 3018, 2930, 2856, 1709, 1655, 1522, 1458, 1420, 1022. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.0 Hz, 3H-20), 1.22−1.35 (m, 6H, 2H-17, 2H-18, 2H-19), 1.73 (quint, 2H, J = 7.4 Hz, 2H-3), 2.00−2.23 (m, 6H, 2H-4, 2H-16, 2H-2), 2.76−2.85 (m, 6H, 2H-7, 2H-10, 2H-13), 4.28 (d, 2H, J = 5.6 Hz, 2H-1″), 5.32−5.47 (m, 8H, vinylic-H), 5.58 (br s, 1H, NH), 6.35−6.36 (m, 1H, H-4′), 7.37−7.38 (m, 2H, H-2′, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.5, 25.4, 25.6 (3C), 26.6, 27.2, 29.3, 31.5, 34.4, 36.0, 110.3, 122.0, 127,5, 127.8, 128.1, 128.2, 128.6, 128.8, 129.0, 130.5, 140.1, 143.5, 172.6.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(Fur-3-ylmethyl)-*N*-methylicosa-5,8,-11,14-tetraenamide, 6. Method B. R_f = 0.16 (hexane/diethyl ether, 1:1). Yield = 52%. IR ($CDCl_3$, cm^{-1}): 3011, 2928, 2856, 1628, 1460, 1408, 1273, 1161, 1068, 874, 733. ^{1}H NMR (300 MHz, CD_3CN, 70 °C, δ): 0.92 (t, 3H, J = 7.0 Hz, 3H-20), 1.33−1.43 (m, 6H, 2H-17, 2H-18, 2H-19), 1.69 (quint, 2H, J = 7.4 Hz, 2H-3), 2.07−2.26 (m, 4H, 2H-4, 2H-16), 2.35−2.37 (m, 2H, 2H-2), 2.84−2.92 (m, 9H, 2H-7, 2H-10, 2H-13, 3H−NCH_3), 4.38 (s, 2H, 2H-1″), 5.34−5.64 (m, 8H, vinylic-H), 6.37 (s, 1H, H-4′), 7.42−7.45 (m, 2H, H-2′, H-5′). ^{13}C NMR (75 MHz, CD_3CN, 70 °C, δ): 14.6, 23.6 (2C), 26.8 (3C), 28.0 (2C), 28.4 (2C), 30.5 (2C), 32.7, 123.7, 129.1, 129.2, 129.4, 129.5, 129.7, 129.8, 129.9, 131.0, 131.7, 142.0, 145.0, 173.8.

(±)-2-Tetrahydrofur-2-ylmethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 8. Method A. R_f = 0.11 (hexane/methylene chloride, 1:5). Yield = 48%. IR ($CHCl_3$, cm^{-1}): 3016, 2957, 2930, 2858, 1732, 1086, 1024, 926. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.86 (t, 3H, J = 6.9 Hz, 3H-20), 1.22−1.38 (m, 6H, 2H-17, 2H-18, 2H-19), 1.53−1.63 (m, 2H, 2H-3′ or 2H-4′), 1.70 (quint, 2H, J = 7.5 Hz, 2H-3), 1.83−1.95 (m, 2H, 2H-3′ or 2H-4′), 1.98−2.12 (m, 4H, 2H-4, 2H-16), 2.33 (t, 2H, J = 7.6 Hz, 2H-2), 2.76−2.82 (m, 6H, 2H-7, 2H-10, 2H-13), 3.71−3.78 (m, 1H, H-5′), 3.81−3.89 (m, 1H, H5′), 3.91−3.99 (m, 1H, H-2′), 4.05−4.20 (m, 2H, 2H-1″), 5.32−5.35 (m, 8H, vinylic-H). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6, 24.8, 25.6 (3C), 26.5, 27.2 (2C), 28.0, 29.3, 31.5, 33.6, 66.4, 68.4, 76.4, 127.5, 127.9, 128.2 (2C), 128.6, 128.8, 128.9, 130.5, 173.5.

(±)-(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(Tetrahydrofur-2-ylmethyl)icosa-5,8,11,14-tetraenamide, 9. Method A. R_f = 0.18 (chloroform/ethyl acetate, 5:1). Yield = 83%. IR ($CHCl_3$, cm^{-1}): 3443, 3329, 3016, 2930, 2858, 1655, 1522, 1078. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.85 (t, 3H, J = 6.0 Hz, 3H-20), 1.22−1.35 (m, 6H, 2H-17, 2H-18, 2H-19), 1.46−1.55 (m, 1H, H-3′ or H-4′), 1.71 (quint, 2H, J = 7.8 Hz, 2H-3), 1.81−1.91 (m, 4H, 2H-4, 2H-16), 1.97−2.11 (m, 3H, H-3′ or H-4′), 2.16 (t, 2H, J = 7.2 Hz, 2H-2), 2.76−2.82 (m, 6H, 2H-7, 2H-10, 2H-13), 3.07 (ddd, 1H, J = 13.5, 7.5, 4.5 Hz, H-1″), 3.57 (ddd, 1H, J = 13.8, 6.6, 3.3 Hz, H-1″), 3.70 (q, 1H, J = 6.9 Hz, H-5′), 3.81 (q, 1H, J = 6.6 Hz, H-5′), 3.91 (qd, 1H, J = 7.5, 3.3 Hz, H-2′), 5.26−5.39 (m, 8H, vinylic-H), 5.81 (br s, 1H, NH). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.5, 25.5, 25.6 (3C), 25.8, 26.7, 27.2, 28.6, 29.3, 31.5, 36.0, 43.1, 67.9, 77.7, 127.5, 127.8, 128.2 (2C), 128.5, 128.7, 129.1, 130.4, 172.7.

(±)-Tetrahydrofur-3-ylmethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 10. Method A. R_f = 0.17 (chloroform). Yield = 38%. IR ($CHCl_3$, cm^{-1}): 3013, 2928, 2856, 1738, 1456,

1153, 1078, 916, 712. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.8 Hz, 3H-20), 1.19−1.32 (m, 6H, 2H-17, 2H-18, 2H-19), 1.54−1.77 (m, 3H, 2H-4, 1H-4′), 1.95−2.16 (m, 5H, 1H-4′, 2H-3, 2H-16), 2.32 (t, 2H, J = 7.8 Hz, 2H-2), 2.50−2.64 (m, 1H, H-3′), 2.78−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 3.56 (dd, 1H, J = 8.8, 5.6 Hz, H−C−O), 3.69−4.13 (m, 5H, H−C−O), 5.26−5.41 (m, 8H, vinylic-H). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.5, 24.7, 25.6 (3C), 26.5, 27.2, 29.0, 29.3, 31.5, 33.6, 38.3, 65.7, 67.7, 70.5, 127.5, 127.8, 128.1, 128.2, 128.6, 128.8, 128.9, 130.5, 173.4.

(±)-(Fur-2-yl)ethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 11. Yield = 30% for method A. Yield = 68% for method B. R_f = 0.16 (hexane/chloroform, 3:2). IR (CH_2Cl_2, cm^{-1}): 3013, 2957, 2930, 2856, 1736, 1456, 1375, 1244, 1153, 1055, 1011. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.87 (t, 3H, J = 6.8 Hz, 3H-20), 1.24−1.36 (m, 6H, 2H-17, 2H-18, 2H-19), 1.56 (d, 3H, J = 6.9 Hz, CH_3−CH), 1.68 (quint, 2H, J = 7.5 Hz, 2H-3), 2.00−2.11 (m, 4H, 2H-4, 2H-16), 2.30 (t, 2H, J = 7.5 Hz, 2H-2), 2.74−2.83 (m, 6H, 2H-7, 2H-10, 2H-13), 5.28−5.42 (m, 8H, vinylic-H), 5.96 (q, 1H, J = 6.6 Hz, H-1″), 6.28−6.32 (m, 2H, H-3′, H-4′), 7.35−7.36 (m, 1H, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 18.2, 22.6, 24.8, 25.6 (3C), 26.5, 27.2, 29.3, 31.5, 33.8, 64.8, 107.6, 110.2, 127.5, 127.9, 128.2 (2C), 128.6, 128.8, 128.9, 130.4, 142.4, 153.6, 172.7.

Data of (+)-**12** and (−)-**13** were identical to those recorded for the racemic material (**11**) except for the optical rotation. (+)-**12**: $[\alpha]^{20}_D$ +29.6 (*c* 3.0, ethanol). (−)-**13**: $[\alpha]^{20}_D$ −31.7 (*c* 2.9, ethanol).

(±)-(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-[1-(Fur-2-yl)ethyl]icosa-5,8,11,-14-tetraenamide, 14. Method B. R_f = 0.22 (chloroform). Yield = 72%. IR ($CDCl_3$, cm^{-1}): 3327, 3013, 2959, 2930, 2856, 1662, 1506, 1452, 1375, 1227, 1151, 1096, 1072, 1013. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.90 (t, 3H, J = 6.8 Hz, 3H-20), 1.24−1.39 (m, 6H, 2H-17, 2H-18, 2H-19), 1.48 (d, 3H, J = 6.9 Hz, CH−CH_3), 1.68−1.79 (m, 2H, 2H-3), 2.03−2.22 (m, 6H, 2H-4, 2H-16, 2H-2), 2.78−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 5.23 (q, 1H, J = 7.3 Hz, CH−CH_3), 5.32−5.42 (m, 8H, vinylic-H), 5.68 (br d, 1H, J = 7.8 Hz, NH), 6.18 (dt, 1H, J = 3.3, 0.8 Hz, H-3′), 6.32 (dd, 1H, J = 3.3, 1.9 Hz, H-4′), 7.35 (dd, 1H, J = 1.8, 0.7 Hz, H-5′). ^{13}C NMR (75 MHz, $CDCl_3$, δ): 13.9, 19.5, 22.4, 25.2, 25.4 (2C), 25.5, 26.4, 27.0, 29.1, 31.3, 35.9, 42.6, 105.4, 110.0, 127.3, 127.7, 128.0 (2C), 128.4, 128.6, 128.9, 130.3, 141.7, 155.2, 171.6.

Thien-2-ylmethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 15. Method A. R_f = 0.66 (chloroform). Yield = 49%. IR ($CHCl_3$, cm^{-1}): 3013, 2926, 2858, 2856, 1736, 1439, 1230, 1146, 702. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.9 Hz, 3H-20), 1.25−1.37 (m, 6H, 2H-17, 2H-18, 2H-19), 1.70 (quint, 2H, J = 7.5 Hz, 2H-3), 2.01−2.13 (m, 4H, 2H-4, 2H-16), 2.33 (t, 2H, J = 7.8 Hz, 2H-2), 2.75−2.84 (m, 6H, 2H-7, 2H-10, 2H-13), 5.26 (s, 2H, 2H-1″), 5.28−5.42 (m, 8H, vinylic-H), 6.97 (dd, 1H, J = 5.1, 3.6 Hz, H-4′), 7.08 (d, 1H, J = 3.3 Hz, H-3′), 7.30 (d, 1H, J = 5.4 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6, 24.7, 25.6 (3C), 26.5, 27.2, 29.3, 31.5, 33.6, 60.4, 126.7, 126.8, 127.5, 127.9, 128.0, 128.1, 128.2, 128.6, 128.8, 128.9, 130.5, 138.1, 173.2.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(Thien-2-ylmethyl)icosa-5,8,11,14-tetraenamide, 16. Method A. R_f = 0.39 (methylene chloride/diethyl ether, 95:5). Yield = 72%. IR (CH_2Cl_2, cm^{-1}): 3319, 2930, 2858, 1713, 1651, 1545, 1529, 1458, 1431, 1367, 1223, 1159, 1043, 852, 702. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.92 (t, 3H, J = 6.6 Hz, 3H-20), 1.22−1.33 (m, 6H, 2H-17, 2H-18, 2H-19), 1.75 (quint, 2H, J = 7.4 Hz, 2H-3), 2.05−2.17 (m, 4H, 2H-4, 2H-16), 2.22 (t, 2H, J = 7.5 Hz, 2H-2), 2.82−2.85 (m, 6H, 2H-7, 2H-10, 2H-13), 4.61 (d, 2H, J = 5.4 Hz, 2H-1″), 5.32−5.39 (m, 8H, vinylic-H), 6.08 (br s, 1H, NH), 6.96 (m, 2H, H-3′, H-4′), 7.22−7.24 (m, 1H, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6, 25.4, 25.6 (3C), 26.6, 27.2, 29.3, 31.5, 35.9, 38.3, 125.2, 126.0, 126.9 (2C), 127.5, 127.8, 128.1, 128.2, 128.6, 129.0, 130.5, 141.0, 176.0.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*,*N*-Bis(thien-2-ylmethyl)icosa-5,8,11,-14-tetraenamide, 17. Method A. R_f = 0.21 (chloroform/hexane, 4:1). Yield = 27%. IR (CH_2Cl_2, cm^{-1}): 3055, 2961, 2930, 2856, 1713, 1636, 1420, 1364, 1094. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.8 Hz, 3H-20), 1.25−1.29 (m, 6H, 2H-17, 2H-18, 2H-19), 1.76−1.89 (m, 2H, 2H-3), 2.02−2.16 (m, 4H, 2H-4, 2H-16), 2.46 (t, 2H, J = 7.6 Hz, 2H-2), 2.79−2.82 (m, 6H, 2H-7, 2H-10, 2H-13), 4.61 (s, 2H, CH_2−N), 4.74 (s, 2H, CH_2−N), 5.30−5.46 (m, 8H, vinylic-H), 6.93−6.99 (m, 4H, 2H-3′, 2H-4′), 7.25−7.27 (m, 2H, 2H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6 (2C), 25.1, 25.6 (3C), 26.6, 27.2, 29.3, 31.5, 32.4, 33.6, 127.5, 127.9 (2C), 128.1 (2C), 128.2, 128.3, 128.4, 128.6, 128.7, 129.3 (2C), 130.5 (2C), 139.8 (2C), 176.1.

Thien-3-ylmethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 18. Method A. R_f = 0.22 (hexane/ethyl acetate, 25:1). Yield = 89%. IR ($CHCl_3$, cm^{-1}): 3016, 2957, 2930, 2856, 1730, 1439, 1251, 1157. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 7.2 Hz, 3H-20), 1.25−1.37 (m, 6H, 2H-17, 2H-18, 2H-19), 1.71 (quint, 2H, J = 7.5 Hz, 2H-3), 2.01−2.13 (m, 4H, 2H-4, 2H-16), 2.34 (t, 2H, J = 7.5 Hz, 2H-2), 2.75−2.84 (m, 6H, 2H-7, 2H-10, 2H-13), 5.11 (s, 2H, 2H-1″), 5.30−5.41 (m, 8H, vinylic-H), 7.07 (dd, 1H, J = 4.8, 1.2 Hz, H-4′), 7.28−7.31 (m, 2H, H-2,′ H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6, 24.7, 25.6 (3C), 26.5, 27.2, 29.3, 31.5, 33.6, 61.0, 124.2, 126.2, 127.5, 127.6, 127.9, 128.1, 128.2, 128.6, 128.9 (2C), 130.5, 136.9, 173.3.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(Thien-3-ylmethyl)icosa-5,8,11,14-tetraenamide, 19. Method B. Yield = 40%. R_f = 0.48 (chloroform/diethyl ether, 9:1). IR ($CHCl_3$, cm^{-1}): 3352, 3018, 2930, 2858, 1718, 1655, 1522, 1458, 1420, 1049, 908. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.6 Hz, 3H-20), 1.25−1.39 (m, 6H, 2H-17, 2H-18, 2H-19), 1.74 (quint, 2H, J = 7.1 Hz, 2H-3), 2.00−2.24 (m, 6H, 2H-4, 2H-16, 2H-2), 2.76−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 4.44 (d, 2H, J = 5.6 Hz, 2H-1″), 5.26−5.46 (m, 8H, vinylic-H), 5.68 (br s, 1H, NH), 7.02 (dd, 1H, J = 4.9, 1.2 Hz, H-4′), 7.12−7.15 (m, 1H, H-2′), 7.29 (dd, 1H, J = 4.9, 2.9 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.5, 25.5, 25.6 (3C), 26.7, 27.2, 29.3, 31.5, 36.0, 38.7, 122.3, 126.4, 127.3, 127.5, 127.8, 128.1, 128.2, 128.6, 128.8, 129.1, 130.5, 139.1, 172.5.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-[(1-Methyl-1*H*-pyrrol-2-yl)methyl]icosa-5,8,11,14-tetraenamide, 20. Method A. R_f = 0.34 (hexane/ethyl acetate, 3:2). Yield = 48%. IR ($CHCl_3$, cm^{-1}): 3018, 2926, 2854, 1655, 1508, 1425, 930. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.0 Hz, 3H-20), 1.25−1.39 (m, 6H, 2H-17, 2H-18, 2H-19), 1.72 (quint, 2H, J = 7.3 Hz, 2H-3), 2.00−2.24 (m, 6H, 2H-4, 2H-16, 2H-2), 2.79−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 3.57 (s, 3H, N−CH_3), 4.43 (d, 2H, J = 5.1 Hz, 2H-1″), 5.30−5.41 (m, 8H, vinylic-H), 5.43 (br s, 1H, NH), 6.06 (d, 2H, J = 2.4 Hz, H-3′, H-4′), 6.61 (t, 1H, J = 2.2 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.5, 25.5, 25.6 (3C), 26.7, 27.2, 29.3, 31.5, 33.7, 35.1, 36.0, 107.0, 108.7, 123.1, 127,5, 127.8, 128.1, 128.2, 128.6 (2C), 128.8, 129.0, 130.5, 172.0.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-[2-(Fur-2-yl)ethyl]icosa-5,8,11,14-tetraenamide, 21. Method B. R_f = 0.60 (methylene chloride/ethyl acetate, 9:1). Yield = 29%. IR (CH_2Cl_2, cm^{-1}): 3350, 2924, 2854, 2361, 2343, 1641, 1545, 1439, 1377, 1068, 1026. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.82 (t, 3H, J = 6.7 Hz, 3H-20), 1.18−1.36 (m, 6H, 2H-17, 2H-18, 2H-19), 1.62 (quint, 2H, J = 7.3 Hz, 2H-3), 1.93−2.12 (m, 6H, 2H-4, 2H-16, 2H-2), 2.71−2.80 (m, 8H, 2H-7, 2H-10, 2H-13, NH−CH_2−CH_2), 3.47 (q, 2H, J = 6.3 Hz, NH−CH_2), 5.20−5.37 (m, 8H, vinylic-H), 5.54 (br s, 1H, NH), 6.00 (dd, 1H, J = 3.0, 0.7 Hz, H-3′), 6.24 (dd, 1H, J = 3.0, 1.9 Hz, H-4′), 7.27 (dd, 1H, J = 1.5, 0.7 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.1, 22.6, 25.5, 25.6 (3C), 26.6, 27.2, 28.2, 29.3, 31.5, 36.1, 38.0, 106.4, 110.3, 127.5, 127.8, 128.1, 128.2, 128.6, 128.7, 129.1, 130.5, 141.6, 153.0, 174.0.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-[2-(Fur-3-yl)ethyl]icosa-5,8,11,14-tetraenamide, 22. Method B. R_f = 0.38 (chloroform/diethyl ether, 9:1). Yield = 42%. IR (CH_2Cl_2, cm^{-1}): 3296, 3011, 2955, 2928, 2856, 1643, 1553, 1502, 1454, 1439, 1161, 1066, 1026, 874. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.89 (t, 3H, J = 6.8 Hz, 3H-20), 1.25−1.39 (m, 6H, 2H-17, 2H-18, 2H-19), 1.62 (m, 2H, 2H-3), 2.00−2.18 (m, 6H, 2H-4, 2H-16, 2H-2), 2.63 (t, 2H, J = 6.8 Hz, NH−CH_2−CH_2), 2.78−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 3.46 (q, 2H, J = 6.8 Hz, NH−CH_2), 5.33−5.41 (m, 9H, vinylic-H, NH), 6.28−6.29 (m, 1H, H-4′), 7.26−7.27 (m, 1H, H-2′), 7.38 (t, 1H, J = 1.7 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$,

δ): 14.1, 22.6, 25.0, 25.5, 25.6 (3C), 26.6, 27.2, 29.3, 31.5, 36.2, 39.4, 110.8, 121.8, 127.6, 127.9, 128.2, 128.3, 128.6, 128.8, 129.1, 130.6, 139.6, 143.3, 172.8.

2-Thien-2-ylethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 23. Method A. R_f = 0.18 (hexane/ethyl acetate, 20:1). Yield = 40%. IR (CH_2Cl_2, cm^{-1}): 3055, 2957, 2932, 2860, 1732, 1421, 1173, 897. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.89 (t, 3H, J = 6.6 Hz, 3H-20), 1.22−1.40 (m, 6H, 2H-17, 2H-18, 2H-19), 1.70 (quint, 2H, J = 7.4 Hz, 2H-3), 2.01−2.16 (m, 4H, 2H-4, 2H-16), 2.34 (t, 2H, J = 7.6 Hz, 2H-2), 2.78−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 3.15 (td, 2H, J = 6.7, 0.7 Hz, O−CH_2−CH_2), 4.30 (t, 2H, J = 6.7 Hz, O−CH_2), 5.27−5.47 (m, 8H, vinylic-H), 6.84−6.87 (d, 1H, J = 3.4 Hz, H-4′), 6.92−6.96 (dd, 1H, J = 5.1, 3.4 Hz, H-3′), 7.16 (dd, 1H, J = 5.1, 1.2 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6, 24.7, 25.6 (2C), 26.5, 27.2, 29.3 (2C), 30.9, 31.5, 33.6, 64.6, 124.0, 125.5, 126.8, 127.5, 127.8, 128.1, 128.2, 128.6, 128.8, 128.9, 130.5, 140.0, 173,5.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(2-Thien-2-ylethyl)icosa-5,8,11,14-tetraenamide, 24. Method A. R_f = 0.26 (chloroform/diethyl ether, 95:5). Yield = 42%. IR (CH_2Cl_2, cm^{-1}): 3292, 3011, 2955, 2928, 2856, 1645, 1553, 1435, 1032, 850, 822, 694. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.82 (t, 3H, J = 6.6 Hz, 3H-20), 1.21−1.42 (m, 6H, 2H-17, 2H-18, 2H-19), 1.71 (quint, 2H, J = 7.4 Hz, 2H-3), 2.05−2.19 (m, 6H, 2H-4, 2H-16, 2H-2), 2.80−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 3.04 (t, 2H, J = 6.6 Hz, NH−CH_2−CH_2), 3.54 (q, 2H, J = 6.3 Hz, NH−CH_2), 5.25−5.47 (m, 8H, vinylic-H), 5.49 (br s, 1H, NH), 6.82−6.84 (m, 1H, H-3′), 6.95 (dd, 1H, J = 5.1, 3.4 Hz, H-4′), 7.17 (dd, 1H, J = 5.1, 1.2 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6, 25.5, 25.6 (3C), 26.7, 27.2, 29.3, 30.0, 31.5, 36.1, 40.7, 123.9, 125.3, 127.0, 127.5, 127.8, 128.2 (3C), 128.6, 128.8, 129.1, 130.5, 173.0.

(±)-(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-[(1-Methyl-2-thien-2-yl)ethyl]icosa-5,8,11,14-tetraenamide, 25. Method B. R_f = 0.30 (chloroform). Yield = 40%. IR ($CDCl_3$, cm^{-1}): 3300, 3013, 2957, 2928, 2856, 1645, 1545, 1514, 1454, 1377, 1258, 1159, 1034, 976. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.6 Hz, 3H-20), 1.14 (d, 3H, J = 6.8 Hz, CH−CH_3), 1.25−1.39 (m, 6H, 2H-17, 2H-18, 2H-19), 1.62 (m, 2H, 2H-3), 2.00−2.18 (m, 6H, 2H-4, 2H-16, 2H-2), 2.78−2.86 (m, 6H, 2H-7, 2H-10, 2H-13), 2.90−3.10 (m, 2H, NH−CH−CH_2), 4.21−4.34 (m, 1H, NH−C*H*), 5.23−5.38 (m, 9H, vinylic-H, NH), 6.81 (dd, 1H, J = 3.4, 1.0 Hz, H-3′), 6.94 (dd, 1H, J = 5.1, 3.4 Hz, 1H, H-4′), 7.15 (dd, 1H, J = 5.1, 1.0 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 19.8, 22.5, 25.5, 25.6 (3C), 26.7, 27.2, 29.3, 31.5, 36.1, 36.3, 45.5, 124.1, 126.3, 126.9, 127.5, 127.8, 128.2 (2C), 128.6, 128.7, 129.1, 130.5, 139.4, 171.9.

2-Thien-3-ylethyl (5*Z*,8*Z*,11*Z*,14*Z*)-Icosa-5,8,11,14-tetraenoate, 26. Method A. R_f = 0.18 (hexane/chloroform, 3:2). Yield = 73%. IR ($CHCl_3$, cm^{-1}): 3018, 2957, 2932, 2860, 1728, 1458, 1414, 1381, 1175. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.86 (t, 3H, J = 6.8 Hz, 3H-20), 1.23−1.37 (m, 6H, 2H-17, 2H-18, 2H-19), 1.65 (quint, 2H, J = 7.4 Hz, 2H-3), 1.99−2.10 (m, 4H, 2H-4, 2H-16), 2.28 (t, 2H, J = 7.6 Hz, 2H-2), 2.75−2.82 (m, 6H, 2H-7, 2H-10, 2H-13), 2.93 (t, 2H, J = 6.9 Hz, O−CH_2−CH_2), 4.25 (t, 2H, J = 6.9 Hz, O−CH_2), 5.26−5.41 (m, 8H, vinylic-H), 6.93 (dd, 1H, J = 4.8, 1.2 Hz, H-4′), 6.98 (d, 1H, J = 2.7 Hz, H-2′), 7.23 (dd, 1H, J = 4.6, 3.0 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.2, 22.6, 24.8, 25.6 (2C), 26.5, 27.2, 29.3, 29.6, 31.5, 33.7, 34.5, 64.3, 121.5, 125.6, 127.6, 127.9, 128.1, 128.2 (2C), 128.6, 128.8, 128.9, 130.5, 138.2, 173.6.

(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-(2-Thien-3-ylethyl)icosa-5,8,11,14-tetraenamide, 27. Method A. R_f = 0.24 (diethyl ether/hexane, 7:3). Yield = 60%. IR (CH_2Cl_2, cm^{-1}): 3327, 3053, 3013, 2957, 2930, 2858, 1713, 1668, 1518, 1439, 895. ^{1}H NMR (300 MHz, $CDCl_3$, δ): 0.86 (t, 3H, J = 6.7 Hz, 3H-20), 1.23−1.36 (m, 6H, 2H-17, 2H-18, 2H-19), 1.66 (quint, 2H, J = 7.4 Hz, 2H-3), 1.99−2.14 (m, 6H, 2H-4, 2H-16, 2H-2), 2.75−2.84 (m, 8H, 2H-7, 2H-10, 2H-13, NH−CH_2−CH_2), 3.49 (q, 2H, J = 6.5 Hz, NH−CH_2), 5.26−5.40 (m, 8H, vinylic-H), 5.47 (br s, 1H, NH), 6.92 (dd, 1H, J = 4.9, 1.3 Hz, 1H, H-4′), 6.97−6.98 (m, 1H, H-2′), 7.26 (dd, 1H, J = 4.8, 3.0 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 22.6, 25.5, 25.6 (3C), 26.6, 27.2, 29.3, 30.2, 31.5, 36.1, 39.8, 121.3, 126.0, 127.5, 127.8, 128.0, 128.1, 128.2, 128.6, 128.8, 129.1, 130.5, 139.3, 173.0.

(±)-(5*Z*,8*Z*,11*Z*,14*Z*)-*N*-[(1-Methyl-2-thien-3-yl)ethyl]icosa-5,8,11,14-tetraenamide, 28. Method B. R_f = 0.15 (chloroform). Yield = 55%. IR ($CDCl_3$, cm^{-1}): 3327, 3013, 2960, 2928, 1653, 1508, 1456, 1097, 1020. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.88 (t, 3H, J = 6.8 Hz, 3H-20), 1.12 (d, 3H, J = 6.6 Hz, CH−CH_3), 1.26−1.39 (m, 6H, 2H-17, 2H-18, 2H-19), 1.66 (quint, 2H, J = 7.3 Hz, 2H-3), 2.00−2.16 (m, 6H, 2H-4, 2H-16, 2H-2), 2.79−2.86 (m, 8H, 2H-7, 2H-10, 2H-13, NH−CH−CH_2), 4.20−4.34 (m, 1H, NH−C*H*), 5.25−5.46 (m, 8H, vinylic-H), 6.93 (dd, 1H, J = 4.9, 1.2 Hz, H-4′), 6.96−6.98 (m, 1H, H-2′), 7.26 (dd, 1H, J = 4.9, 2.9 Hz, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.0, 20.3, 22.6, 25.5, 25.6 (3C), 26.6, 27.2, 29.3, 31.5, 36.2, 36.8, 45.2, 121.9, 125.5, 127.5, 127.8, 128.2 (2C), 128.6, 128.7 (2C), 129.1, 130.5, 138.1, 172.0.

Synthesis of *N*-(Fur-3-ylmethyl)-*N*-[(5*Z*,8*Z*,11*Z*,14*Z*)-icosa-5,8,11,14-tetraenyl]amine, 7. To a suspension of lithium aluminum hydride (0.54 mmol) in THF (1.0 mL), a solution of amide **5** (0.18 mmol) in THF (1.0 mL) was added dropwise under argon atmosphere. The mixture was stirred for 10 min, refluxed for 3 h, and stirred for 6 additional hours at room temperature. Then the mixture was cooled in an ice/water (0.1 mL) bath, and 15% sodium hydroxide solution (0.05 mL) and water (0.25 mL) were added sequentially, dropwise, to destroy the excess of hydride reagent. After the mixture was stirred for 45 min at room temperature, salts were removed by filtration and washed with diethyl ether and the organic extracts were dried over Na_2SO_4. Finally, the drying agent was removed, the solvent was evaporated under reduced pressure, and the amine was purified by column cromatography to afford 45 mg (68% yield) of pure **7** as an oil. R_f = 0.31 (chloroform/methanol, 9:1). IR ($CDCl_3$, cm^{-1}): 3352, 3013, 2928, 2856, 1458, 1161, 1024. ^{1}H NMR (200 MHz, $CDCl_3$, δ): 0.89 (t, 3H, J = 6.8 Hz, 3H-20), 1.22−1.36 (m, 6H, 2H-17, 2H-18, 2H-19), 1.40−1.59 (m, 4H, 2H-2, 2H-3), 1.92 (br s, 1H, NH), 2.00−2.13 (m, 4H, 2H-4, 2H-16), 2.64 (t, 2H, J = 7.3 Hz, 2H-1), 2.78−2.84 (m, 6H, 2H-7, 2H-10, 2H-13), 3.66 (s, 2H, 2H-1″), 5.30−5.44 (m, 8H, vinylic-H), 6.39−6.41 (m, 1H, H-4′), 7.37−7.38 (m, 2H, H-2′, H-5′). ^{13}C NMR (50 MHz, $CDCl_3$, δ): 14.1, 22.6, 25.7 (4C), 27.2, 27.4, 27.5, 29.3, 31.5, 44.2, 49.2, 110.6, 123.5, 127.5, 127.9, 128.0, 128.1, 128.3, 128.5, 130.0, 130.5, 140.0, 143.2.

Biochemistry. Radioligand Binding Assays. CB_1 Receptor. Receptor binding studies were performed according to the procedure of Houston et al.[48] with slight modifications. Briefly, rat cerebellum membranes were homogenized in 5 mL of ice-cold Tris buffer (50 mM Tris-HCl, pH 7.4, at 4 °C) and centrifuged at 48000*g* and 4 °C for 10 min. The membrane pellet was washed twice by resuspension and centrifugation. The final pellet was resuspended in 20 volumes of incubation buffer (50 mM Tris-HCl, 1 mM EDTA, 3 mM $MgCl_2$, and 5 mg/mL BSA fatty acid free, pH 7.4). Fractions of the final membrane suspension (about 2 mg/mL of protein) were incubated at 30 °C for 90 min with 0.5 nM [^{3}H]-WIN552122 (180 Ci/mmol), in the presence or absence of several concentrations of the competing drug, in a final volume of 0.5 mL of assay buffer (50 mM Tris-HCl, 1 mM EDTA, 3 mM $MgCl_2$, and 5 mg/mL BSA fatty acid free, pH 7.4). Nonspecific binding was determined in the presence of 10 μM SR141716A. Silanized tubes were used throughout the procedure to minimize receptor binding loss due to tube adsorption. The reaction was terminated by rapid vacuum filtration with a Brandel harvester apparatus through Whatman GF/C filters presoaked in Tris buffer (50 mM Tris-HCl, 1 mg/mL BSA, pH 7.4). The filters were washed three times with 5 mL of ice-cold buffer (50 mM Tris-HCl, 1 mg/mL BSA, pH 7.4, 4 °C), and bound radioactivity was measured by placing filters in 4 mL of Ecolite scintillation cocktail followed by liquid scintillation spectroscopy. The binding assay showed the appropriate sensitivity to CB_1 ligands. Thus, WIN552122, CP55940, and anandamide (in the absence of an FAAH inhibitor) inhibited the binding with K_i values of 4.5 ± 0.4, 1.3 ± 0.4, and 285 ± 14 nM, respectively.[39]

CB_2 Receptor. The receptor binding studies were performed according to the procedure of Griffin et al.[49] using membrane fractions of human CB_2 receptor transfected cells purchased from Receptor Biology, Inc. (Beltsville, MD).

HEK293EBNA membranes were resuspended in Tris buffer (50 mM Tris-HCl, 2.5 mM EGTA, 5 mM $MgCl_2$, 1 mg/mL BSA fatty acid free, pH 7.5). Fractions of the final membrane suspension (about 1.44 mg/mL of protein) were incubated at 30 °C for 90 min with 0.3 nM [^{3}H]-CP55940 (180 Ci/mmol), in the presence or absence of several concentrations of the competing drug, in a final volume of 0.2 mL of assay buffer (50 mM Tris-HCl, 2.5 mM EGTA, 5 mM $MgCl_2$, 1 mg/mL BSA fatty acid free, pH 7.5). Nonspecific binding was determined in the presence of 5 μM CP55940. Silanized tubes were used throughout the experiment to minimize receptor binding loss due to tube adsorption. The reaction was terminated by rapid vacuum filtration with a Brandel harvester apparatus through Whatman GF/C filters presoaked in 0.05% polyethylenimine (PEI). The filters were washed three times with 5 mL of ice-cold buffer (50 mM Tris-HCl, 1 mg/mL BSA, pH 7.4, 4 °C), and bound radioactivity was measured by placing filters in 4 mL of Ecolite scintillation cocktail followed by liquid scintillation spectroscopy. The binding assay showed the appropriate sensitivity to CB_2 ligands. Thus, WIN552122, CP55940, and anandamide (in the absence of an FAAH inhibitor) inhibited the binding with K_i values of 3.8 ± 0.8, 0.15 ± 0.05, and >1000 nM, respectively.[39]

VR_1 Receptor. The receptor binding studies were performed according to the procedure of Szallasi et al.[46] Briefly, rat spinal cord membranes were homogenized in HEPES buffer (10 mM HEPES, 5 mM KCl, 5.8 mM NaCl, 0.75 mM $CaCl_2$, 2 mM $MgCl_2$, 320 mM sucrose, pH 7.4) and centrifuged at 1000*g* at 4 °C for 10 min. The supernatant was removed, and samples were centrifuged again at 35000*g* at 4 °C for 30 min. Finally, the membrane pellet was resuspended in 10 volumes of HEPES buffer (10 mM HEPES, 5 mM KCl, 5.8 mM NaCl, 0.75 mM $CaCl_2$, 2 mM $MgCl_2$, 320 mM sucrose, pH 7.4). Fractions of the final membrane suspension (about 1 mg/mL of protein) were incubated at 37 °C for 60 min with 25 pM [^{3}H]-RTX (48 Ci/mmol), in the presence or absence of several concentrations of the competing drug, in a final volume of 0.5 mL of assay buffer (10 mM HEPES, 5 mM KCl, 5.8 mM NaCl, 0.75 mM $CaCl_2$, 2 mM $MgCl_2$, 320 mM sucrose, 0.25 mg/mL BSA fatty acid free, pH 7.4). Nonspecific binding was determined in the presence of 1 μM RTX. The reaction was stopped by chilling the assay mixture on an ice-cold water bath, and then 100 μg of bovine α_1-acid glycoprotein in 50 μL of buffer was added to reduce nonspecific binding. Bound and free [^{3}H]-RTX samples were separated by pelleting the membranes in a Beckman TJ-6 centrifuge. After the supernatant was removed by aspiration and the pellet carefully dried, the tip of the Eppendorf tube containing the pelleted membranes was cut off with a razor blade. Then an amount of 4 mL of Ecolite scintillation cocktail was added and bound radioactivity was determined by liquid scintillation spectroscopy. The binding assay showed the appropriate sensitivity to inhibition by nonradioactive RTX (K_i = 40 ± 1 pM).[39]

For all binding experiments, competition binding curves were analyzed by using an iterative curve-fitting procedure GraphPad (Prism), which provided IC_{50} values for test compounds. K_i values were determined by the method of Cheng and Prusoff.[50]

Endocannabinoid Transporter Assay. Human lymphoma U937 cells, maintained at 37 °C and 5% CO_2 wet atmosphere, were grown in RPMI 1640 culture medium containing 10% FBS previously heat-inactivated, 100 U/mL penicillin, 100 μg/mL streptomycin, and 2.5 mM sodium piruvate. For standard competition assays, an amount of 1 mL of U937 cells in RPMI 1640 culture medium (10^6 cells/mL) was preincubated at 37 °C for 10 min in the presence or absence of several concentrations of the tested inhibitors. Then a mixture of [^{3}H]-anandamide (0.45 nM) and cold anandamide at a final concentration of 100 nM was added and cells were incubated for 7 min. The reaction was stopped by rapid filtration over Whatman GF/C filters presoaked in 0.25% BSA. Filters were washed three times with 5 mL of ice-cold Krebs-HEPES buffer (118 mM NaCl, 4.7 mM KCl, 1.3 mM $CaCl_2$, 2.4 mM $MgSO_4$, 1 mM NaH_2PO_4, 20 mM $NaHCO_3$, 11.1 mM glucose, 3.98 μM Na_2EDTA, 110 mM ascorbic acid, 10 mM Na-HEPES, and 1% BSA fatty acid free, pH 7.4), and bound radioactivity was measured by placing filters in 4 mL of Ecolite scintillation cocktail followed by liquid scintillation spectroscopy.

Nonspecific reuptake was determined under the same experimental conditions at 4 °C. Silanized tubes were used throughout to minimize the influence of effects of compound adsorption. Competition curves were analyzed by using an iterative curve-fitting procedure GraphPad (Prism), which provided IC_{50} values for test compounds.

FAAH Inhibition Assay. FAAH activity was assayed in rat brain homogenates as described by Omeir et al.[51] and as adapted to the tritiated substrate by Fowler et al.[52] Briefly, test compounds or the ethanol carrier (10 μL) was incubated at 37 °C for 10 min with diluted homogenates (165 μL) of rat brain (minus cerebellum, in 10 mM Tris-HCl + 1 mM EDTA, pH 7.6) and 25 μL of a mixture of 16 μM (i.e., 2 μM final assay concentration) nonradioactive anandamide containing trace amounts of [^{3}H]anandamide with the label in the ethanolamine side chain (30−60 Ci/mmol) and 1% w/v fatty acid free BSA. The reaction was terminated by putting the samples on an ice bath followed by the addition of 0.4 mL of chloroform/methanol (1:1 v/v). After vigorous vortexing, aqueous and organic phases were separated by centrifugation, and the radioactivity (corresponding to the [^{3}H]ethanolamine produced as a result of the FAAH-catalyzed breakdown of [^{3}H]anandamide) found in aliquots (200 μL) of the aqueous phase was determined by liquid scintillation counting with quench correction. All compounds were tested in at least three experiments over a concentration range of 0.5−100 μM (eight concentrations were used). pI_{50}, and hence IC_{50} values, were determined as described previously.[53]

Acknowledgment. This work was supported by Ministerio de Ciencia y Tecnología (Grant BQU 2001-1459). The authors thank MECD and CAM for predoctoral grants to S.O.G. and E.L., respectively. C.J.F. and G.T. thank the Swedish Science Research Council (Grant No. 12158, medicine), Konung Gustav V and Drottning Victorias Foundation, the Swedish Asthma and Allergy Association's Research Foundation, the Swedish Psoriasis Association, and the Research Funds of the Medical Odontological Faculty, Umeå University, for financial support. The authors also thank Britt Jacobsson for her excellent technical assistence with the FAAH assays.

References

(1) Gaoni, Y.; Mechoulam, R. Isolation, Structure, and Partial Synthesis of an Active Constituent of Hashish. *J. Am. Chem. Soc.* **1964**, *86*, 1646−1647.
(2) Martin, B. R. Identification of the Endogenous Cannabinoid System through Integrative Pharmacological Approaches. *J. Pharmacol. Exp. Ther.* **2002**, *301*, 790−796.
(3) Matsuda, L. A.; Lolait, S. J.; Brownstein, M. J.; Young, A. C.; Bonner, T. I. Structure of a Cannabinoid Receptor and Functional Expression of the Cloned cDNA. *Nature* **1990**, *346*, 561−564.
(4) Munro, S.; Thomas, K. L.; Abu-Shaar, M. Molecular Characterization of a Peripheral Receptor for Cannabinoids. *Nature* **1993**, *365*, 61−65.
(5) Devane, W. A.; Hanuš, L.; Breuer, A.; Pertwee, R. G.; Stevenson, L. A.; Griffin, G.; Gibson, D.; Mandelbaum, A.; Etinger, A.; Mechoulam, R. Isolation and Structure of a Brain Constituent That Binds to the Cannabinoid Receptor. *Science* **1992**, *258*, 1946−1949.
(6) Sugiura, T.; Kondo, S.; Sukagawa, A.; Nakane, S.; Shinoda, A.; Itoh, K.; Yamashita, A.; Waku, K. 2-Arachidonoylglycerol: A

Possible Endogenous Cannabinoid Receptor Ligand in Brain. *Biochem. Biophys. Res. Commun.* **1995**, *215*, 89−97.
(7) Hanuš, L.; Abu-Lafi, S.; Fride, E.; Breuer, A.; Vogel, Z.; Shalev, D. E.; Kustanovich, I.; Mechoulam, R. 2-Arachidonyl Glyceryl Ether, an Endogenous Agonist of the Cannabinoid CB_1 Receptor. *Proc. Natl. Acad. Sci. U.S.A.* **2001**, *98*, 3662−3665.
(8) Porter, A. C.; Sauer, J.-M.; Knierman, M. D.; Becker, G. W.; Berna, M. J.; Bao, J. Q.; Omikos, G. G.; Carter, P.; Bymaster, F. P.; Leese, A. B.; Felder C. C. Characterization of a Novel Endocannabinoid, Virodhamine, with Antagonist Activity at the CB_1 Receptor. *J. Pharmacol. Exp. Ther.* **2002**, *301*, 1020−1024.
(9) Deutsch, D. G.; Chin, S. A. Enzymatic Synthesis and Degradation of Anandamide, a Cannabinoid Receptor Agonist. *Biochem. Pharmacol.* **1993**, *46*, 791−796.
(10) Calignano, A.; La Rana, G.; Giuffrida, A.; Piomelli, D. Control of Pain Initiation by Endogenous Cannabinoids. *Nature* **1998**, *394*, 277−281.
(11) Walker, J. M.; Hohmann, A. G.; Martin, W. J.; Strangman, N. M.; Huang, S. M.; Tsou, K. The Neurobiology of Cannabinoid Analgesia. *Life Sci.* **1999**, *65*, 665−673.
(12) Fernández-Ruiz, J.; Berrendero, F.; Hernández, M. L.; Ramos, J. A. The Endogenous Cannabinoid System and Brain Development. *Trends Neurosci.* **2000**, *23*, 14−20.
(13) Hampson, R. E.; Deadwyler, S. A. Cannabinoids, Hippocampal Function and Memory. *Life Sci.* **1999**, *65*, 715−723.
(14) Wilson, R. I.; Nicoll, R. A. Endogenous Cannabinoids Mediate Retrograde Signalling at Hippocampal Synapses. *Nature* **2001**, *410*, 588−592.
(15) Di Marzo, V.; Goparaju, S. K.; Wang, L.; Liu, J.; Bátkai, S.; Járai, Z.; Fezza, F.; Miura, G. I.; Palmiter, R. D.; Sugiura, T.; Kunos, G. Leptin-Regulated Endocannabinoids Are Involved in Maintaining Food Intake. *Nature* **2001**, *410*, 822−825.
(16) Giuffrida, A.; Piomelli, D. The Endocannabinoid System: A Physiological Perspective on Its Role in Psychomotor Control. *Chem. Phys. Lipids* **2000**, *108*, 151−158.
(17) De Petrocellis, L.; Melck, D.; Bisogno, T.; Di Marzo, V. Endocannabinoids and Fatty Acid Amides in Cancer, Inflammation and Related Disorders.*Chem. Phys. Lipids* **2000**, *108*, 191−209.
(18) Pertwee, R. G. Cannabinoid Receptor Ligands: Clinical and Neuropharmacological Considerations, Relevant to Future Drug Discovery and Development. *Expert Opin. Invest. Drugs* **2000**, *9*, 1553−1571.
(19) Baker, D.; Pryce, G.; Croxford, J. L.; Brown, P.; Pertwee, R. G.; Huffman, J. W.; Layward, L. Cannabinoids Control Spasticity and Tremor in a Multiple Sclerosis Model. *Nature* **2000**, *404*, 84−87.
(20) Galve-Roperh, I.; Sánchez, C.; Cortés, M. L.; Gómez del Pulgar, T.; Izquierdo, M.; Guzmán, M. Anti-tumoral Action of Cannabinoids: Involvement of Sustained Ceramide Accumulation and Extracellular Signal-Regulated Kinase Activation. *Nat. Med.* **2000**, *6*, 313−319.
(21) Pertwee, R. G. Pharmacology of Cannabinoid Receptor Ligands. *Curr. Med. Chem.* **1999**, *6*, 635−664.
(22) Piomelli, D.; Giuffrida, A.; Calignano, A.; Rodríguez de Fonseca, F. The Endocannabinoid System as a Target for Therapeutic Drugs. *Trends Pharmacol. Sci.* **2000**, *21*, 218−224.
(23) Giuffrida, A.; Beltramo, M.; Piomelli, D. Mechanisms of Endocannabinoid Inactivation: Biochemistry and Pharmacology. *J. Pharmacol. Exp. Ther.* **2001**, *298*, 7−14.
(24) Baker, D.; Pryce, G.; Croxford, J. L.; Brown, P.; Pertwee, R. G.; Makriyannis, A.; Khanolkar, A.; Layward, L.; Fezza, F.; Bisogno, T.; Di Marzo, V. Endocannabinoids Control Spasticity in a Multiple Sclerosis Model. *FASEB J.* **2001**, *15*, 300−302.
(25) Lastres-Becker, I.; Hansen, H. H.; Berrendero, F.; de Miguel, R.; Pérez-Rosado, A.; Manzanares, J.; Ramos, J. A.; Fernández-Ruiz, J. J. Alleviation of Motor Hyperactivity and Neurochemical Deficits by Endocannabinoid Uptake Inhibition in a Rat Model of Huntington's Disease. *Synapse* **2002**, *44*, 23−35.
(26) Beltramo, M.; Stella, N.; Calignano, A.; Lin, S. Y.; Makriyannis, A.; Piomelli, D. Functional Role of High-Affinity Anandamide Transport, As Revealed by Selective Inhibition. *Science* **1997**, *277*, 1094−1097.
(27) Piomelli, D.; Beltramo, M.; Glasnapp, S.; Lin, S. Y.; Goutopoulos, A.; Xie, X.-Q.; Makriyannis, A. Structural Determinants for Recognition and Translocation by the Anandamide Transporter. *Proc. Natl. Acad. Sci. U.S.A.* **1999**, *96*, 5802−5807.
(28) Jarrahian, A.; Manna, S.; Edgemond, W. S.; Campbell, W. B.; Hillard, C. J. Structure−Activity Relationships among *N*-Arachidonylehanolamine (Anandamide) Head Group Analogues for the Anandamide Transporter. *J. Neurochem.* **2000**, *74*, 2597−2606.
(29) Muthian, S.; Nithipatikom, K.; Campbell, W. B.; Hillard, C. J. Synthesis and Characterization of a Fluorescent Substrate for the *N*-Arachidonoylethanolamine (Anandamide) Transmembrane Carrier. *J. Pharmacol. Exp. Ther.* **2000**, *293*, 289−295.
(30) Bisogno, T.; Melck, D.; Bobrov, M. Y.; Gretskaya, N. M.; Bezuglov, V. V.; De Petrocellis, L.; Di Marzo, V. *N*-Acyldopamines: Novel Synthetic CB_1 Cannabinoid-Receptor Ligands and Inhibitors of Anandamide Inactivation with Cannabimimetic Activity in Vitro and in Vivo. *Biochem. J.* **2000**, *351*, 817−824.
(31) De Petrocellis, L.; Bisogno, T.; Davis, J. B.; Pertwee, R. G.; Di Marzo, V. Overlap between the Ligand Recognition Properties of the Anandamide Transporter and the VR1 Vanilloid Receptor: Inhibitors of Anandamide Uptake with Negligible Capsaicin-like Activity. *FEBS Lett.* **2000**, *483*, 52−56.
(32) Hillard, C. J.; Edgemond, W. S.; Jarrahian, A.; Campbell, W. B. Accumulation of *N*-Arachidonoylehanolamine (Anandamide) into Cerebellar Granule Cells Occurs via Facilitated Diffusion. *J. Neurochem.* **1997**, *69*, 631−638.
(33) Deutsch, D. G.; Glaser, S. T.; Howell, J. M.; Kunz, J. S.; Puffenbarger, R. A.; Hillard, C. J.; Abumrad, N. The Cellular Uptake of Anandamide Is Coupled to Its Breakdown by Fatty Acid Amide Hydrolase. *J. Biol. Chem.* **2001**, *276*, 6967−6973.
(34) Day, T. A.; Rakhshan, F.; Deutsch, D. G.; Barker, E. L. Role of Fatty Acid Amide Hydrolase in the Transport of the Endogenous Cannabinoid Anandamide. *Mol. Pharmacol.* **2001**, *59*, 1369−1375.
(35) Patricelli, M. P.; Cravatt, B. F. Proteins Regulating the Biosynthesis and Inactivation of Neuromodulatory Fatty Acid Amides. *Vitam. Hormon.* **2001**, *62*, 95−131.
(36) Bisogno, T.; Hanuš, L.; De Petrocellis, L.; Tchilibon, S.; Ponde, D. E.; Brandi, I.; Moriello, A. S.; Davis, J. B.; Mechoulam, R.; Di Marzo, V. Molecular Targets for Cannabidiol and Its Synthetic Analogues: Effect on Vanilloid VR_1 Receptors and on the Cellular Uptake and Enzymatic Hydrolysis of Anandamide. *Br. J. Pharmacol.* **2001**, *134*, 845−852.
(37) Zygmunt, P. M.; Chuang, H.-h.; Movahed, P.; Julius, D.; Högestätt, E. D. The Anandamide Transport Inhibitor AM404 Activates Vanilloid Receptors. *Eur. J. Pharmacol.* **2000**, *396*, 39−42.
(38) López-Rodríguez, M. L.; Viso, A.; Ortega-Gutiérrez, S.; Lastres-Becker, I.; González, S.; Fernández-Ruiz, J. J.; Ramos, J. A. PCT/ES01/00305 (WO 02/12167 A1), 2001.
(39) López-Rodríguez, M. L.; Viso, A.; Ortega-Gutiérrez, S.; Lastres-Becker, I.; González, S.; Fernández-Ruiz, J.; Ramos, J. A. Design, Synthesis and Biological Evaluation of Novel Arachidonic Acid Derivatives as Highly Potent and Selective Endocannabinoid Transporter Inhibitors. *J. Med. Chem.* **2001**, *44*, 4505−4508.
(40) Winans, C. F. Hydrogenation of Aldehydes in the Presence of Ammonia. *J. Am. Chem. Soc.* **1939**, *61*, 3566−3567.
(41) Abdel-Magid, A. F.; Carson, K. G.; Harris, B. D.; Maryanoff, C. A.; Shah, R. D. Reductive Amination of Aldehydes and Ketones with Sodium Triacetoxyborohydride. Studies on Direct and Indirect Reductive Amination Procedures. *J. Org. Chem.* **1996**, *61*, 3849−3862.
(42) Hays, S. J.; Caprathe, B. W.; Gilmore, J. L.; Amin, N.; Emmerling, M. R.; Michael, W.; Nadimpalli, R.; Nath, R.; Raser, K. J.; Stafford, D.; Watson, D.; Wang, K.; Jaen, J. C. 2-Amino-4*H*-3,1-bezoxazin-4-ones as Inhibitors of C1r Serine Protease. *J. Med. Chem.* **1998**, *41*, 1060−1067.
(43) Arnoldi, A.; Bregante, G.; Caldirola, P.; Merlini, L.; Tamburini, B. A New Synthesis of 4,5,6,7-Tetrahydrofuro[2,3-*c*]pyridines and Furo[2,3-*c*]pyrrolidines. *J. Heterocycl. Chem.* **1990**, *27*, 1169−1171.
(44) Goodman, M. M.; Kabalka, G. W.; Marks, R. C.; Knapp, F. F., Jr.; Lee, J.; Liang, Y. Synthesis and Evaluation of Radioiodinated 2-(2(*RS*)-Aminopropyl)-5-iodothiophenes as Brain Imaging Agents. *J. Med. Chem.* **1992**, *35*, 280−285.
(45) Khanolkar, A. D.; Makriyannis, A. Structure−Activity Relationships of Anandamide, an Endogenous Cannabinoid Ligand. *Life Sci.* **1999**, *65*, 607−616.
(46) Szallasi, A.; Bíró, T.; Szabó, T.; Modarres, S.; Petersen, M.; Klusch, A.; Blumberg, P. M.; Krause, J. E.; Sterner, O. A Nonpungent Triprenyl Phenol of Fungal Origin, Scutigeral, Stimulates Rat Dorsal Root Ganglion Neurons via Interaction at Vanilloid Receptors. *Br. J. Pharmacol.* **1999**, *126*, 1351−1358.
(47) de Lago, E.; Fernández-Ruiz, J.; Ortega-Gutiérrez, S.; Viso, A.; López-Rodríguez, M. L.; Ramos, J. A. UCM707, a Potent and Selective Inhibitor of Endocannabinoid Uptake, Potentiates Hypokinetic and Antinociceptive Effects of Anandamide. *Eur. J. Pharmacol.* **2002**, *449*, 99−103.
(48) Houston, D. B.; Evans, D. M.; Howlett, A. C.; Melvin, I. S. [^{3}H]-CP55940 Binding to the Cannabinoid Receptor. *Du Pont Biotech. Update* **1991**, *6*, 21−27.

(49) Griffin, G.; Tao, Q.; Abood, M. E. Cloning and Pharmacological Characterization of the Rat CB_2 Cannabinoid Receptor. *J. Pharmacol. Exp. Ther.* **2000**, *292*, 886−894.
(50) Cheng, Y. C.; Prusoff, W. H. Relationship between the Inhibiton Constant (K_i) and the Concentration of Inhibitor Which Causes 50 Per Cent Inhibition (IC_{50}) of an Enzymatic Reaction. *Biochem. Pharmacol.* **1973**, *22*, 3099−3108.
(51) Omeir, R. L.; Chin, S.; Hong, Y.; Ahern, D. G.; Deutsch, D. G. Arachidonoyl Ethanolamide-[1,2-^{14}C] as a Substrate for Anandamide Amidase. *Life Sci.* **1995**, *56*, 1999−2005.
(52) Fowler, C. J.; Tiger, G.; Stenström, A. Ibuprofen Inhibits Rat Brain Deamidation of Anandamide at Pharmacologically Relevant Concentrations. Mode of Inhibition and Structure−Activity Relationship. *J. Pharmacol. Exp. Ther.* **1997**, *283*, 729−734.
(53) Jonsson, K.-O.; Vandevoorde, S.; Lambert, D. M.; Tiger, G.; Fowler, C. J. Effects of Homologues and Analogues of Palmitoylethanolamide upon the Inactivation of the Endocannabinoid Anandamide. *Br. J. Pharmacol.* **2001**, *133*, 1263−1275.

JM0210818