Available online at www.sciencedirect.com
SCIENCE DIRECT
Bioorganic & Medicinal Chemistry Letters
Bioorganic & Medicinal Chemistry Letters 16 (2006) 44–48

Synthesis and biological activity of novel 4-phenyl-1,8-naphthyridin-2(1*H*)-on-3-yl ureas: Potent acyl-CoA:cholesterol acyltransferase inhibitor with improved aqueous solubility

Hitoshi Ban,* Masami Muraoka, Katsuhisa Ioriya and Naohito Ohashi

Research Division, Sumitomo Pharmaceuticals Co., Ltd 1-98, Kasugadenaka 3-chome, Konohana-ku, Osaka 554-0022, Japan

Received 11 July 2005; revised 8 August 2005; accepted 21 September 2005
Available online 18 October 2005

Abstract—4-Aryl-1,8-naphthyridin-2(1*H*)-on-3-yl urea derivatives with hydrophilic groups were synthesized in order to improve aqueous solubility and pharmacokinetic property. SMP-797 possessing (4-aminophenyl)ureido and 3-(hydroxypropoxyphenyl) moieties showed potent ACAT inhibitory activity and excellent oral efficacy.

An enzyme acyl-CoA:cholesterol acyltransferase (ACAT), which catalyzes the intracellular cholesterol esterification,[1] plays important roles in several physiological processes: (1) absorption of dietary and biliary cholesterol in the intestines[2]; (2) determination of cholesteryl ester content and the secretion of hepatic very low density lipoprotein (VLDL)[3]; (3) accumulation of cholesteryl esters in macrophage in the arterial wall.[4] Inhibition of ACAT, therefore, is expected to reduce plasma lipid levels by inhibiting intestinal cholesterol absorption and hepatic VLDL secretion, and to prevent progression of atherosclerotic lesions by inhibiting the accumulation of cholesteryl esters in macrophage. For this reason, ACAT inhibitor is an attractive target for the treatment of hypercholesterolemia and atherosclerosis[5] (Fig. 1).

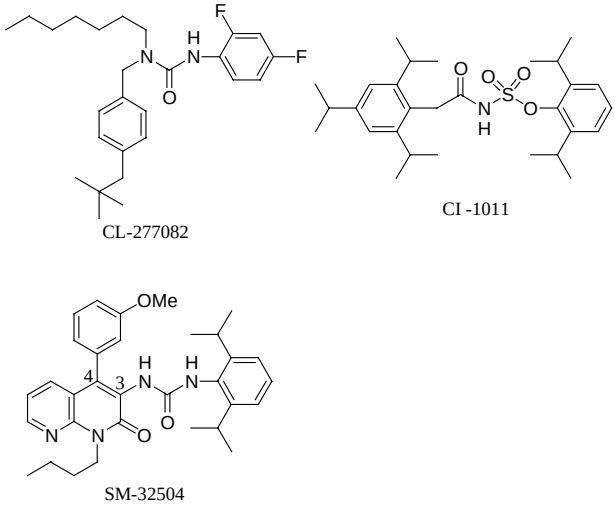


Figure 1.

Classical ACAT inhibitors[6] such as CL-277082,[7] which are considered to inhibit absorption of cholesterol in the intestines, failed in the clinical trials for their insufficient efficacy on hypercholesterolemia and atherosclerosis. The main reason should be their low systemic bioavailabilities expected from their lipophilic structural features. Therefore, recent interest in the development of ACAT inhibitors is switching the target from the intestinal ACAT to the liver and the arterial wall ACAT.[6] CI-1011, an ACAT inhibitor currently in clinical trial, has been found to show high efficacy not only in a variety of cholesterol-fed animal models but also in noncholesterol-fed animal models such as a rabbit model fed a casein-rich diet,[8] and is considered to inhibit ACAT in both the intestines and the liver.

Keywords: SMP-797; AACAT inhibitor; A4-Phenyl-1,8-naphthyridin-2(1*H*)-on-3-yl ureas.
* Corresponding author. Tel.: +8166 466 5227; fax: +8166 466 5287; e-mail: ban@sumitomopharm.co.jp

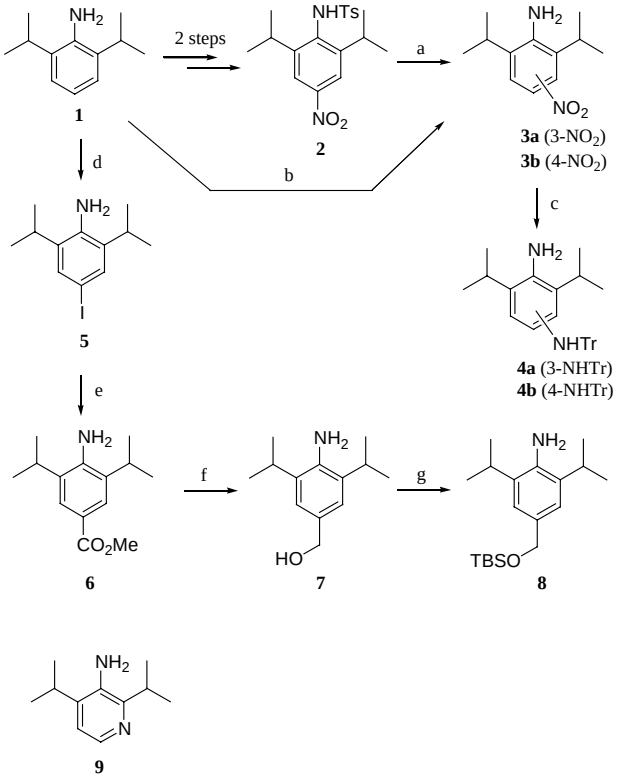
0960-894X/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2005.09.056

We previously found a naphthyridinyl urea SM-32504 as a potent inhibitor of ACAT,[9] which showed the cholesterol-lowering effect in cholesterol-fed animal models. Despite the excellent efficacy, however, this compound showed only poor oral absorption due to low aqueous solubility; its maximum drug concentration (C_{max}) value in mice was under the lowest detectable concentration (<0.01 ng/mL) after oral administration at the dosage of 30 mg/kg. The cholesterol lowering effect of SM-32504, therefore, may be due to ACAT inhibitory activity in the intestines and not to ACAT inhibitory activity in the liver. The results led us to modify SM-32504 and develop compounds possessing better pharmacokinetic properties.

We considered that the pharmacokinetic properties of SM-32504 would be improved by increasing its aqueous solubility, especially solubility under acidic conditions. Drugs administered orally are absorbed mainly in the stomach and/or the small intestine; pH values of gastric juices are 1.4–2.1 (fasting) or 4.3–5.4 (non-fasting); pH values of small intestinal fluids are 5.0–7.0.[10] We report herein the synthesis, aqueous solubility, and biological activity of hydrophilic derivatives of SM-32504.

We examined two modifications of SM-32504 to afford the hydrophilic derivatives: (i) introduction of an amino group on the 2,6-diisopropylphenyl moiety; (ii) replacement of the 3-methoxy group at the 4-phenyl moiety with a hydrophilic group. Although a number of ACAT inhibitors possessing 2,6-diisopropylphenyl moiety were reported, the effect of amino group on the aromatic moiety has not been examined.

The synthesis of 2,6-diisopropylanilines **4a**, **4b**, **8**, and **9** required in the present study is shown in Scheme 1. Deprotection of *N*-tosyl-4-nitroaniline **2**, readily available from 2,6-diisopropylaniline (**1**) in two steps, gave 4-nitroaniline **3b** according to the known procedures.[11] In contrast, it was found that the direct nitration of aniline **1** with concd HNO_3 in concd H_2SO_4 gave 3-nitroaniline **3a**.[12] Pd/C-catalyzed hydrogenation of **3a** and **3b** followed by the selective protection of the less hindered amino group with triphenylmethyl group gave **4a** and **4b**, respectively. 4-Iodoaniline **5**, readily prepared from **1** by a known iodination method,[13] was converted to **6** by Pd-catalyzed carbonylation in MeOH-DMF.[14] Reduction of **6** with $LiAlH_4$ followed by protection of the hydroxy group gave a silyl ether **8**. A pyridine intermediate **9** was prepared according to the literature.[15]

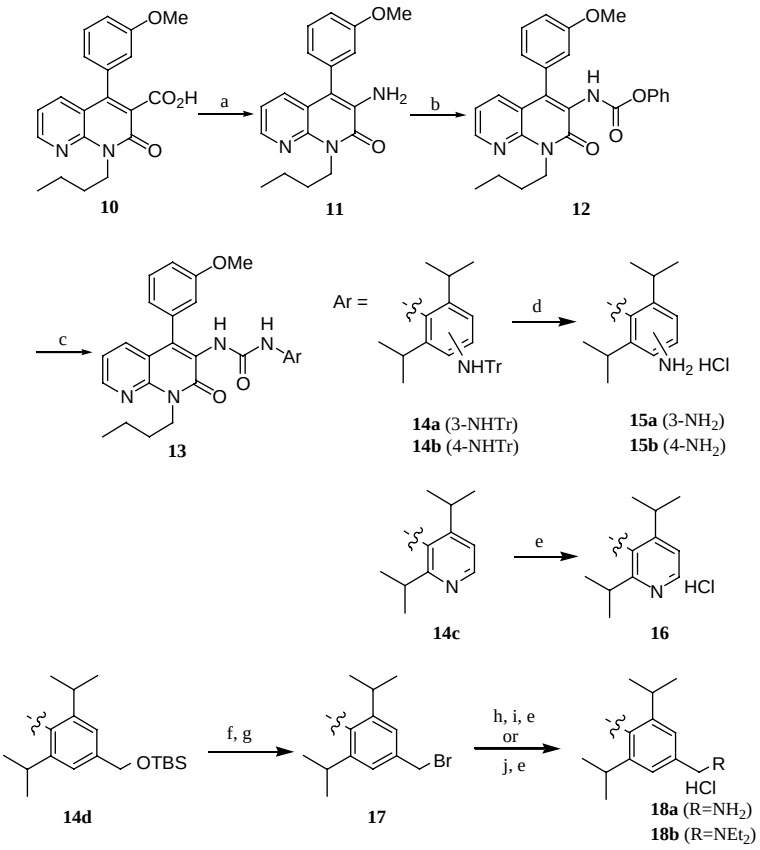


Scheme 1. Reagents and conditions: (a) concd H_2SO_4, 70 °C, 3 h, 96%; (b) concd HNO_3, concd H_2SO_4, −10 °C, 1 h; 96%; (c) H_2, cat. Pd/C, MeOH, rt, 2–12 h, then TrCl, Et_3N, CH_2Cl_2, rt, overnight; **4a** 64% and **4b** 44%; (d) HIO_3, I_2, AcOH–H_2SO_4–H_2O (150:1:4), 80 °C, 5 h, 91%; (e) CO, cat. $Pd(OAc)_2$, DPPP, Et_3N, MeOH, DMF, 80 °C, 4 h, 83%; (f) $LiAlH_4$, THF, 60 °C, 30 min, 98%; (g) TBSCl, DMAP, Et_3N, CH_2Cl_2, rt, overnight, 83%.

Syntheses of naphthyridinyl ureas **15a**, **15b**, **16**, **18a**, and **18b** are shown in Scheme 2. Curtius rearrangement[16] of acid **10**[9] with diphenylphosphoryl azide gave amine **11**,[17] which was converted to phenyl carbamate **12**. Then, **12** was treated with **4a**, **4b**, **8**, and **9** in the presence of DMAP in DMF and was converted to the corresponding naphthyridinyl ureas **14a–d**, respectively. Deprotection of **14a** and **14b** was conducted with 10% HCl in MeOH at room temperature, and **15a** and **15b** were obtained. Treatment of **14c** with 1 M HCl in ether gave the corresponding salt **16**. **17** was synthesized by the desilylation of **14d** followed by bromination with PBr_3. Ammonium salt **18a** was obtained by reaction of **17** with potassium phthalimide, cleavage of the phthaloyl group, and treatment with 1 M HCl in ether. Amination of **17** with *N*,*N*-diethylamine followed by treatment with 1 M HCl in ether gave **18b**.

The ACAT inhibitory activity and solubility of **15a**, **15b**, **16**, **18a**, and **18b** are shown in Table 1. All compounds exhibited enhanced aqueous solubility compared to that of SM-32504, and **15b** and **16** showed more potent ACAT inhibitory activity than SM-32504. However, **16** was not examined further, since gradual degradation of the urea moiety took place in an aqueous solution at pH 2.5 at room temperature.

We next focused on the synthesis and activity of **15** derivatives possessing hydrophilic substituents on the 4-phenyl group. The syntheses of **22a–c** and **26a–c** are summarized in Scheme 3. Treatment of **11** with BBr_3 in CH_2Cl_2 gave **19**, which was treated with several ω-acetoxyalkyl bromides in the presence of K_2CO_3 and KI in DMF to afford **20a–c** and **23**. The naphthyridine ureas **21a–d** were prepared by the reaction of **20a–c** with **4a** or **4b** in the presence of DMAP in DMF at room temperature. Deacylation of **21a–d** with NaOMe in MeOH followed by



Scheme 2. Reagents and conditions: (a) DPPA, Et_3N, DMF, 50 °C, 2 h, then 6 M NaOH, 50 °C, 3 h, 97%; (b) phenyl chloroformate, THF, 50 °C, 5 h, 84%; (c) **4a**, **4b**, **8**, or **9**, DMAP, DMF, rt, overnight; **14a**, 79%; **14b**, 77%; **14c**, 64%; **14d**, 73%; (d) 10% HCl/MeOH, rt, overnight; **15a**, 94%; **15b**, 97%; (e) 1 M HCl/ether, AcOEt, rt, 30 min; **16**, 80%; **18a**, 63%; **18b**, 80%; (f) concd HCl, EtOH, rt, 3 h, 89%; (g) PBr_3, CH_2Cl_2, rt, 3 h, 45% (h) potassium phthalimide, DMF, rt, overnight, 88%; (i) $H_2NNH_2{\cdot}H_2O$, EtOH, rt, 3 days, 81%; (j) Et_2NH, K_2CO_3, DMF, rt, 1 h, 62%.

Table 1. ACAT inhibitory activity and solubility of **15a**, **15b**, **16**, **18a**, and **18b**

Compound	ACAT inhibitory activity[a] IC_{50} (nM)	Solubility[b] (mg/mL)		
		pH 7.4	pH 5.5	pH 2.5
SM-32504	11	<0.001	<0.001	<0.001
15a	14	0.001	0.001	0.70
15b	8.3	<0.001	0.001	0.022
16	5.4	0.005	0.019	>1.0
18a	382	0.036	0.023	0.22
18b	680	0.062	1.0	>1.0

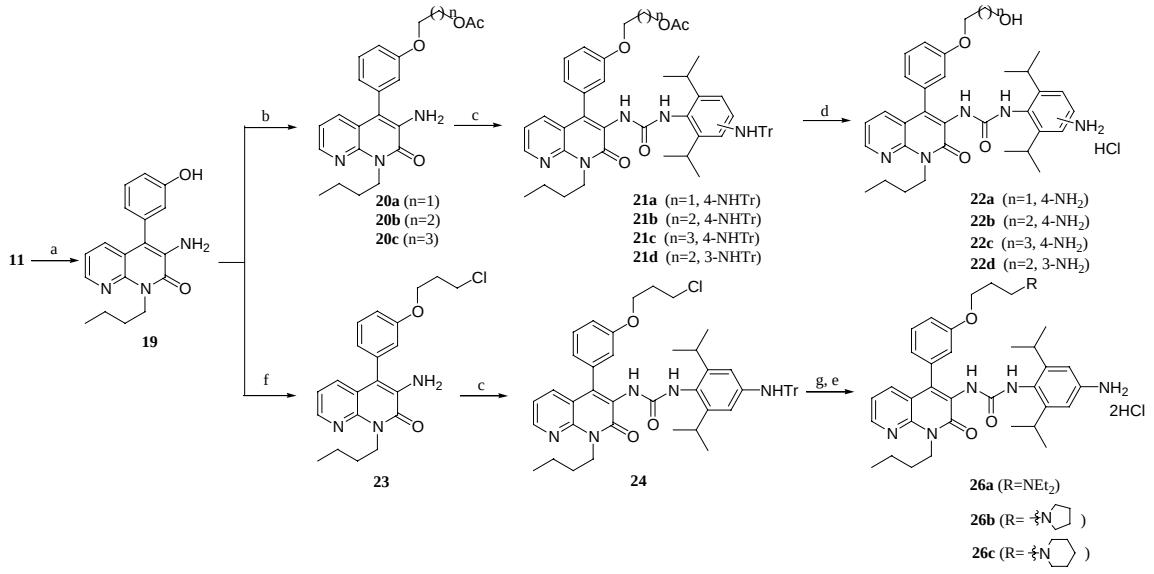
[a] ACAT inhibition in vitro measured in rat macrophages. See Ref. 18 for the detailed protocol.

[b] Solubility in phosphate buffer (5.53% aq citric acid/1.75% aq Na_2HPO_4) at rt was determined by HPLC analysis.

detritylation with 10% HCl in MeOH gave **22a**–**d**. Urea **24** prepared from **23** was coupled with *N*,*N*-diethylamine, pyrrolidine, or piperidine followed by deprotection with 10% HCl in MeOH, and naphthyridinyl ureas **26a**–**c**, respectively, were obtained.

The activity and aqueous solubility of **22a**–**d** and **26a**–**c** are summarized in Table 2. All the compounds showed enhanced solubility compared to SM-32504 and **15b**. Notably, the length of alkylene exhibits some effect on the activity as indicated by **22a**–**d** and **22b** (SMP-797)[19], with the propylene group turning out be the most potent (IC_{50}, 21 nM) with appreciable improvement of solubility at broad pH range (pH 2.5–7.4). The amino derivatives **26a**–**c** showed weak activity in spite of their higher solubility.

SMP-797 (**22b**) exhibited excellent pharmacokinetic properties in mice (C_{max}, 21.3 μg/mL; after oral administration at the dosage of 30 mg/kg) and rabbits (C_{max}, 16.0 ng/mL; area under the blood time curve (AU-$C_{0-24\ h}$), 39.8 ng h/mL after oral administration at the dosage of 1.0 mg/kg/day for 21 days). The results are highly contrasted to those of SM-32504, which was not absorbed orally at all in the same models. SMP-797 (**22b**) was then subjected to evaluation with a rabbit model fed a casein-rich diet, which was considered to exhibit the cholesterol-lowering effect due to the inhibitory of secretion from the liver. Rabbits were fed a casein-rich diet for 1 week, and then SMP-797 was orally administered at the dosage of 1.0 mg/kg/day for further 3 weeks with a casein-rich diet. Under these conditions SMP-797 decreased the serum total cholesterol level by 53% compared with control. It was reported that atorvastatin, the HMG-CoA reductase inhibitor with the most potent cholesterol lowering effect currently in clinical use, decreased the cholesterol level by 54% at a dose of 10 mg/kg/day when it was



Scheme 3. Reagents and conditions: (a) BBr_3, CH_2Cl_2, rt, overnight, 95%; (b) $BrCH_2(CH_2)_nOAc$, K_2CO_3, KI, DMF, rt, overnight; **20a** (n = 1), 75%; **20b** (n = 2), 91%; **20c** (n = 3), 92%; (c) phenyl chloroformate, THF, rt, overnight, then **4a** or **4b**, DMAP, DMF, rt, overnight; **21a**, 64%; **21b**, 72%; **21c**, 83%; **21d**, 78%; **24**, 49%; (d) NaOMe, MeOH, rt to reflux, 8 h to overnight, then 10% HCl/MeOH, rt, overnight; **22a**, 72%; **22b**, 83%; **22c**, 80%; **22d**, 85%; (e) 10% HCl/MeOH, rt, overnight; **26a**, quant.; **26b**, 84%; **26c**, 87%; (f) 1-bromo-3-chloropropane, K_2CO_3, KI, DMF, rt, overnight, 67%; (g) *N*,*N*-diethylamine, pyrrolidine, or piperidine, K_2CO_3, KI, DMF, 50 °C, 10 h; **25a**, 64%; **25b**, 73%; **25c**, 30%.

Table 2. ACAT inhibitory activity and solubility of **22** and **26**

Compound	ACAT inhibitory activity[a] IC_{50} (nM)	Solubility[b] (mg/mL)		
		pH 7.4	pH 5.5	pH 2.5
SM-32504	11	<0.001	<0.001	<0.001
15b	8.3	<0.001	0.001	0.022
22a	>1000	0.030	0.032	>1.0
22b (SMP-797)	21	0.010	0.014	>1.0
22c	61	0.005	0.007	0.68
22d	43	0.017	0.019	>1.0
26a	452	0.64	0.69	>1.0
26b	427	0.13	0.20	>1.0
26c	540	0.20	0.18	>1.0

[a] ACAT inhibition in vitro measured in rat macrophages. See Ref. 18 for the detailed protocol.

[b] Solubility in phosphate buffer (5.53% aq citric acid/1.75% aq Na_2HPO_4) at rt was determined by HPLC analysis.

administered to rabbits fed a casein diet for 6 weeks.[20] These results suggest that the cholesterol-lowering effect of SMP-797 at a dose of 1.0 mg/kg/day is almost the same as with that of atorvastatin at a dose of 10 mg/kg/day.

In summary, we have developed SMP-797 possessing a potent ACAT inhibitory activity and significantly enhanced aqueous solubility under acidic conditions. The compound is a promising agent for oral treatment of hypercholesterolemia. The details of pharmacological and toxicological studies will be discussed in due course.

Acknowledgments

We thank Y. Saitou and Y. Konya for evaluation of pharmacokinetic properties, and Ms. Y. Nakatsuji for her technical assistance.

References and notes

1. (a) Spector, A. A.; Mathur, S. N.; Kaduce, T. L. *Prog. Lipid Res.* **1979**, *18*, 31; (b) Suckling, K. E.; Stange, E. F. *J. Lipid Res.* **1985**, *26*, 647.
2. (a) Field, F. J.; Salome, R. G. *Biochim. Biophys. Acta* **1982**, *712*, 557; (b) Heider, J. G.; Pickens, C. E.; Kelly, L. A. *J. Lipid Res.* **1983**, *24*, 1127.
3. (a) Drevon, C. A.; Engelhorn, S. C.; Steinberg, D. *J. Lipid Res.* **1980**, *21*, 1065; (b) Khan, B.; Wilcox, H. G.; Heimberg, M. *Biochem. J.* **1989**, *258*, 807; (c) Cianflone, K. M.; Yasruel, Z.; Rodriguez, M. A.; Vas, D.; Sniderman, A. D. *J. Lipid Res.* **1990**, *31*, 2045.
4. (a) Brecher, P. I.; Chobanian, A. V. *Circ. Res.* **1974**, *35*, 692; (b) Day, A. J.; Proudlock, J. W. *Atherosclerosis* **1974**, *19*, 253; (c) Hashimoto, S.; Dayton, S. *Atherosclerosis* **1977**, *28*, 447.
5. Sliskovic, D. R.; White, A. D. *Trends Pharmacol. Sci.* **1991**, *12*, 194.
6. For recent reviews of ACAT inhibitor, see: (a) Picard, J. A. *Curr. Opin. Ther. Pat.* **1993**, *3*, 151; (b) Matsuda, K. *Med. Res. Rev.* **1994**, *14*, 271; (c) Sliskovic, D. R.; Trivedi, B. K. *Curr. Med. Chem.* **1994**, *1*, 204; (d) Roark, W. H.; Roth, B. D. *Exp. Opin. Invest. Drug* **1994**, *3*, 1143.
7. Harris, W. S.; Dujoune, C. A.; von Bergmam, K.; Neal, J.; Akester, J.; Windsor, S. L.; Greene, D.; Look, Z. *Clin. Pharmacol. Ther.* **1990**, *48*, 189.
8. Lee, H. T.; Sliskovic, D. R.; Picard, J. A.; Roth, B. D.; Wierenga, W.; Hicks, J. L.; Bousley, R. F.; Hamelehle, K. L.; Homan, R.; Speyer, C.; Stanfield, R. L.; Krause, B. R. *J. Med. Chem.* **1996**, *39*, 5031.
9. Ohnuma, S.; Ioriya, K.; Muraoka, M.; Ohashi, N. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 1309.
10. Kararli, T. T. *Biopharm. Drug Dispos.* **1995**, *16*, 351.
11. Carver, F. J.; Hunter, C. A.; Livingstone, J. D.; McCabe, J. F.; Seward, E. M. *Chem. Eur. J.* **2002**, *8*, 2847.
12. Giumanini, A. G.; Verardo, G.; Polana, M. *J. Prakt. Chem.* **1988**, *330*, 161.
13. Merkushev, E. B. *Synthesis* **1988**, 923.

14. Donald Valentine, Jr.; Tilley, J. W.; LeMahieu, R. A. *J. Org. Chem.* **1981**, *46*, 4614.
15. Pascual, A.; Rindlisbacher, A. *Pestic. Sci.* **1994**, *42*, 253.
16. Curtius, T. *Chem. Ber.* **1902**, *35*, 3226.
17. Shioiri, T.; Ninomiya, K.; Yamada, S. *J. Am. Chem. Soc.* **1972**, *94*, 6203.
18. Ioriya, K.; Noguchi, T.; Muraoka, M.; Fujita, K.; Shimizu, H.; Ohashi, N. *Pharmacology* **2002**, *65*, 18.
19. Mp 196–197 °C (MeOH). IR (KBr): *v* 2966, 1636, 1584, 1546 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz), δ (ppm) 0.85–0.99 (15H, m), 1.41 (2H, qt, *J* = 7.3, 7.3 Hz), 1.70 (2H, tt, *J* = 7.3, 7.3 Hz), 1.87 (2H, tt, *J* = 6.1, 6.1 Hz), 2.95 (2H, tt, *J* = 7.3, 7.3 Hz), 3.54 (2H, t, *J* = 6.1 Hz), 4.03 (2H, t, *J* = 6.1 Hz), 4.50 (2H, t, *J* = 7.0 Hz), 6.88–6.89 (2H, m), 7.00–7.03 (3H, m), 7.25 (1H, dd, *J* = 4.6, 7.9 Hz), 7.40 (1H, t, *J* = 7.9 Hz), 7.60 (1H, dd, *J* = 1.5, 7.9 Hz), 7.79 (1H, s), 7.87 (1H, s), 8.61 (1H, dd, *J* = 1.5, 4.6 Hz), 9.88 (2H, br s); Anal. calcd for $C_{34}H_{43}N_5O_4 \cdot HCl \cdot H_2O$: C, 63.79; H, 7.24; Cl, 5.54; N, 10.94. Found: C, 63.85; H, 7.21; Cl, 5.74; N, 10.96.
20. Auerbach, B. J.; Krause, B. R.; Bisgaier, C. L.; Newton, R. S. *Atherosclerosis* **1995**, *115*, 173.