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Research paper

Discovery of potent liver-selective stearoyl-CoA desaturase-1 (SCD1) inhibitors, thiazole-4-acetic acid derivatives, for the treatment of diabetes, hepatic steatosis, and obesity

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ABSTRACT

SCD1 is a rate-limiting enzyme in the conversion of saturated fatty acids to monounsaturated fatty acids. SCD1 inhibitors have potential effects on obesity, diabetes, acne, and cancer, but the adverse effects associated with SCD1 inhibition in the skin and eyelids are impediments to clinical development. To avoid mechanism-based adverse effects, we explored the compounds that selectively inhibit SCD1 in the liver in an *ex vivo* assay. Starting from a systemically active lead compound, we focused on the physicochemical properties tPSA and cLogP to minimize exposure in the off-target tissues. This effort led to the discovery of thiazole-4-acetic acid analog **48** as a potent and liver-selective SCD1 inhibitor. Compound **48** exhibited significant effects in rodent models of diabetes, hepatic steatosis, and obesity, with sufficient safety margins in a rat toxicology study with repeated dosing.

1. Introduction

Stearoyl-CoA desaturase (SCD) is a lipogenic enzyme involved in the *de novo* lipogenesis of monounsaturated fatty acids (MUFAs) and catalyzes insertion of the *cis* double bond at the delta-9 position (between carbons 9 and 10) of saturated fatty acids (SFAs) [1]. There are four isoforms of SCD (SCD1–4) in the mouse and two isoforms in the human (hSCD1 and hSCD5) [2–4]. Human SCD1 shows a high degree of homology with mouse SCD1 [5]. The products of SCD1, such as oleic acid and palmitoleic acid, are incorporated into the lipids, including phospholipids, triglycerides (TGs), cholesteryl esters, and wax esters. Maintaining the optimal balance of MUFA and SFA composition in lipids is important because an imbalance in the ratio affects membrane fluidity and lipoprotein metabolism [6]. In an epidemiological survey, the serum SCD1 activities were significantly higher in men who developed metabolic syndrome than those who did not [7]. SCD1 affects the ratio of SFAs to MUFAs, though dietary MUFAs also have an effect.

Mice with a targeted disruption in the SCD1 isoform ($SCD1^{-/-}$) are resistant to diet-induced accumulation of hepatic TGs and fat in their adipose tissues throughout their bodies, concomitantly preventing obesity due to the increased energy expenditure. The mice also exhibited improved insulin sensitivity because of the reduced adipose tissue mass [8]. From these findings, SCD1 inhibitor was considered to be a novel potential agent for the treatment of obesity and type-2 diabetes [9]. However, the adverse events in the eye and skin, such as eye abnormalities characterized by squinting and alopecia, were observed in mice with systemic SCD1 knockout [10,11]. Systemically distributed SCD1 inhibitors have also been reported to have mechanism-based adverse effects in eye and skin, as well as beneficial metabolic effects [12].

Although SCD1 is expressed ubiquitously, it is predominant in lipogenic tissues, especially hepatocytes and adipocytes [13]. SCD1 antisense oligonucleotide (ASO) treatment protects mice from high fat diet-induced weight gain and improves insulin resistance without any mechanism-based adverse effects because most ASO is distributed to the liver [14]. In addition, liver-specific SCD1 knockout mice are resistant to high carbohydrate diet-induced obesity and hepatic steatosis, without adverse effects [15]. Therefore, SCD1 inhibitors selectively distributed in the liver are hypothesized to circumvent mechanism-based adverse effects, and

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much effort has been made to discover liver-selective SCD1 inhibitors [16,17]. In particular, Merck disclosed **MK-8245** as a potent and liver-selective SCD1 inhibitor, proceeding to clinical trials (phase IIa). They screened their compounds using a cell-based assay and identified **MK-8245** as a substrate of liver-specific organic anion transporting polypeptides [18]. We took an *ex vivo* assay approach, measuring the activity of all SCD isozymes in both the liver and off-target tissues after oral administration of compounds [19]. This method could be expected to detect the liver-selective distribution of compounds for various reasons. First, based on a high-throughput screening (HTS) hit, we developed a potent but systemically distributed lead compound (**4**) and hypothesized that modifying its physicochemical properties could change the distribution pattern (Fig. 1). Lead optimization focused on the topological polar surface area (tPSA) and calculated logarithm of the partition coefficient (cLogP), leading to the discovery of highly potent, orally bioavailable thiazole-4-acetic acid analogs. The **48** was demonstrated to be liver-selective SCD1 inhibitor [20]. Here, we report the generation of potent and liver-selective small molecule inhibitors of SCD1 and the discovery of **48**.

2. Results and discussion

2.1. HTS hit to lead identification

We performed HTS with our small molecule compound library looking for inhibitors of human recombinant SCD1. The HTS compound library and synthesized compounds were screened in a biochemical assay measuring the production of tritiated water from the desaturation of radio-labeled stearoyl-CoA. We identified a novel 4-aminopyridine-based SCD1 inhibitor (**1**) with a moderate IC_{50} of 1400 nM (Fig. 1). The selectivity of other lipogenic enzymes and structural features amenable to the accelerated synthesis of derivatives made **1** an attractive starting point. Our first hit-to-lead exploration was performed on the pyridine moiety. Screening a small molecule library of various heterocyclic amide analogs revealed that a 2-amino-4-methyl thiazole of **2** (IC_{50} = 150 nM) was a preferred replacement for the 4-aminopyridine. A further increase in potency was obtained by replacing an oxygen atom in the ether linker group with a nitrogen atom. The IC_{50} value shifted approximately 16-times in amine analog **3** from ether **2** (Fig. 1). The lead identification chemistry culminated in compound **4** (IC_{50} = 6.4 nM), which had good ligand binding efficiency (IC_{50}/number of non-hydrogen atoms, LE = 0.5) and selectivity from other lipogenic enzymes [21]. As the pharmacokinetic (PK) data in mice indicated that **4** had moderate plasma exposure (10 mg/kg, po, AUC_{0-inf} = 2.4 μM h) with a resulting oral bioavailability (%F) of 15%, we examined the *in vivo* activity of **4** in mice. Anti-diabetic effects and reduced diet-induced body weight gains were observed in db/db mice at a dose of 10 mg/kg (data not shown). However, adverse effects, such as squinting and alopecia, were observed on day 9 and thereafter. Therefore, we assessed whether **4** inhibits SCD1 enzyme in off-target tissues 6 h and 24 h after oral dosing (0.1–10 mg/kg) in mice. In the *ex vivo* assay, compound **4** dose-dependently inhibited SCD1 in both the liver and eyelids at 6 h, and sustained inhibition of SCD1 was observed in the eyelids at 24 h at a dose of 10 mg/kg (Fig. 2, Supporting Information Table S1). Thus, the PK profiles of the lead compound had to be changed to the desired profile. The screening flow shown in Fig. 3 was set to guide the lead optimization. All compounds with IC_{50} < 20 nM were advanced to the *ex vivo* experiments. First, we screened SCD1 activities in the liver and eyelids of mice 6 h after oral administration at a dose of 1 mg/kg. Second, select compounds from the first step were investigated in the *ex vivo* assay at higher doses (10 and 100 mg/kg) 24 h after dosing.

2.2. Lead optimization

Physicochemical properties are one of the factors affecting the PK profiles [22]. We postulated that altering the physicochemical properties of lead compound **4** may result in different compound exposure between the liver and eyelids. Our lead optimization approach based on the hit-to-lead study is shown in Fig. 4. Compounds having different physicochemical properties were designed for each part of the molecule, synthesized, and tested with the SCD1 enzyme assay. The tPSA and cLogP were employed as the index physicochemical properties of the compounds [23]. Initially, we examined the substituents on the left-hand phenyl ring. As shown in Table 1, hydrophobic groups, including CF_3 (**6**) and *n*-propyl (**9**) on the ortho-position and Cl group on the meta-position (**7**), were well tolerated. However, removing the ortho substituent (**5**) or Cl group at the para-position (**8**) decreased the potency. Polar substituents, such as COOH (**10**) and $CONMe_2$ (**11**), were not

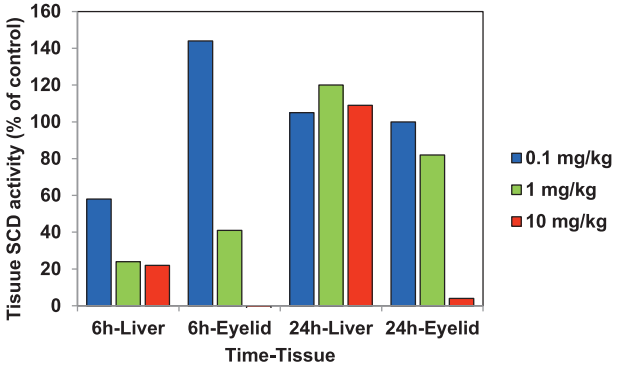


Fig. 2. Effects of **4** on liver and eyelid SCD activities in mice. Compound **4** suspended in vehicle (0.5% methylcellulose aqueous solution) was orally administered to C57BL/6 J mice. Six or 24 h after administration, liver and eyelid samples were collected and homogenized to measure tissue SCD activity. Results are expressed as percentage of control for remaining SCD activity.

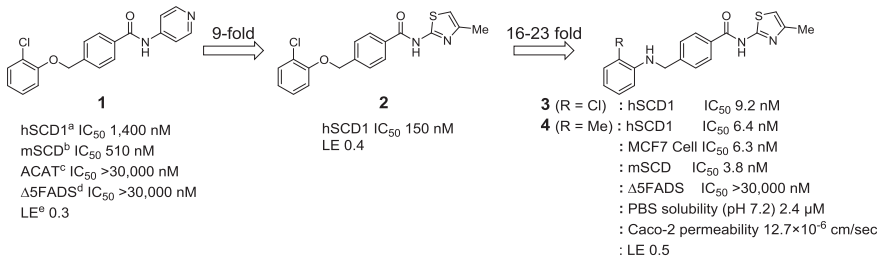


Fig. 1. Hit-to-lead identification and *in vitro* profiles of lead compound **4**.
[a]Recombinant human SCD1. [b]Mouse liver microsome SCD. [c]Acetyl-CoA acetyltransferase. [d]Δ-5 Fatty acid desaturase. [e]Ligand efficiency.

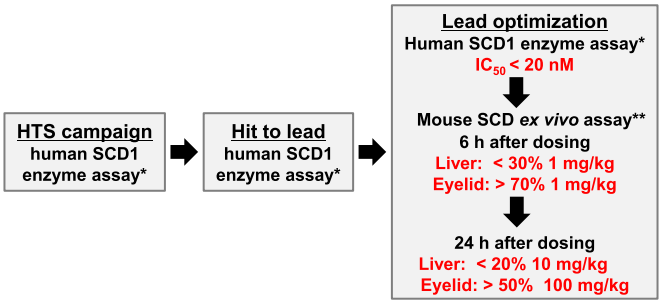


Fig. 3. Screening flow for identifying liver-selective SCD1 inhibitor.
*SCD1 enzyme activity was measured using recombinant human SCD1.
**Compounds were orally administered to mice and then the liver and eyelid collected 6 h (1 mg/kg) or 24 h (10 and 100 mg/kg) after dosing. Tissue homogenates were obtained and SCD activity measured. Results are expressed as percentage of control for remaining SCD activity.

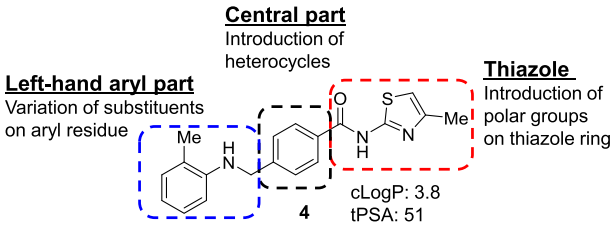
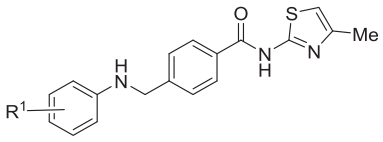


Fig. 4. Design strategies to modify the physicochemical properties of compound **4**.

Table 1
Exploring the left-hand phenyl ring with substituents.

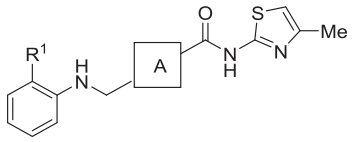


Compound	R^1	IC_{50} (nM)[a]	cLogP[b]	tPSA[c]
5	H	95	3.3	54
6	2-CF_3	4.1	4.7	50
3	2-Cl	9.2	4.3	52
7	3-Cl	18	4.3	54
8	4-Cl	760	4.3	54
9	2-*n*-Pr	8.9	4.9	51
10	2-CO_2H	3600	4.1	92
11	2-$CONMe_2$	2700	2.3	73

[a] Recombinant human SCD1 enzyme.
[b] Calculated logarithm of the partition coefficient.
[c] Topological polar surface area.

allowed. Next, we replaced the central phenyl ring with heterocyclic rings. Unfortunately, the pyridines (**12**, **13**) and piperidine (**14**) were less potent, whereas thiophene ring **15** was equipotent to phenyl ring **4** (Table 2). Finally, we tested polar functional groups at the thiazole moiety. As shown in Table 3, introduction of a COOH group at position 4 on the thiazole ring (**16**) resulted in a loss of activity. However, the loss in potency was recovered by inserting a methylene unit between the COOH group and thiazole ring (**17**). A higher potency was obtained with a trifluoromethyl group on the left-hand phenyl ring (**18**). Extending the methylene chain to $-(CH_2)_2COOH$ (**19**) maintained the single digit nanomolar potency, but further extension to $-(CH_2)_3COOH$ (**20**) decreased the potency by more than 400-fold. Alcohols with different methylene lengths

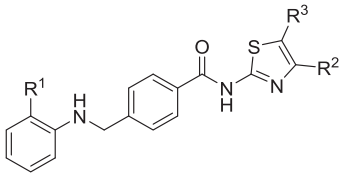
Table 2
Effects of replacing the central phenyl ring with heterocyclic rings.



Compound	R^1	A	IC_{50} (nM)[a]	cLogP[b]	tPSA[c]
4	Me		6.4	3.8	51
12	Me		46	3.0	65
13	Me		310	2.6	61
14	CF_3		30	4.1	49
15	CF_3		9.7	4.5	53

[a] Recombinant human SCD1 enzyme.
[b] Calculated logarithm of the partition coefficient.
[c] Topological polar surface area.

Table 3
Exploring the thiazole moiety with polar substituents.



Compound	R^1	R^2	R^3	IC_{50} (nM)[a]	cLogP[b]	tPSA[c]
4	Me	Me	H	6.4	3.8	51
16	Me	CO_2H	H	>10,000	3.7	96
17	Me	CH_2CO_2H	H	14	2.6	96
18	CF_3	CH_2CO_2H	H	5.1	3.5	95
19	CF_3	$(CH_2)_2CO_2H$	H	6.3	4.0	95
20	CF_3	$(CH_2)_3CO_2H$	H	2700	4.4	94
21	CF_3	CH_2OH	H	3.8	3.2	74
22	CF_3	$(CH_2)_2OH$	H	4.2	3.4	75
23	CF_3	$(CH_2)_3OH$	H	5.2	3.8	75
24	CF_3	H	CO_2H	8.1	4.4	95
25	CF_3	H	CH_2CO_2H	58	3.5	94
26	CF_3	H	$(CH_2)_2CO_2H$	570	4.0	94
27	CF_3	H	CH_2OH	2.8	3.2	74
28	CF_3	H	$(CH_2)_2OH$	3.8	3.4	73
29	CF_3	H	$(CH_2)_3OH$	260	3.8	75
30	CF_3	CH_2OH	CO_2H	22	3.0	117
31	CF_3	$(CH_2)_3OH$	$(CH_2)_2OH$	31	3.0	99

[a] Recombinant human SCD1 enzyme.
[b] Calculated logarithm of the partition coefficient.
[c] Topological polar surface area.

(**21**–**23**) had single digit nanomolar activities. Introduction of these polar substituents at position 5 on the thiazole ring resulted in a structure-activity relationship (SAR) different from that of substituents at position 4. Single digit nanomolar activity was found for compounds having COOH (**24**), $-CH_2OH$ (**27**), and $-(CH_2)_2OH$ (**28**), whereas other carboxylic acids with a methylene chain (**25**, **26**) and an alcohol with a longer methylene chain (**29**) decreased the potency. Combination of the alcohols and COOH at positions 4 and 5 (**30**, **31**) resulted in slightly decreased potency. All compounds with an $IC_{50} < 20$ nM were tested in the *ex vivo* experiments 6 h after

dosing with 1 mg/kg, but only compound **21** demonstrated the first sign of liver selectivity (SCD activity in the liver 20% and in the eyelid 73%). This compound had a higher tPSA value of 74 and lower cLogP value of 3.2 compared to lead compound **4** (tPSA = 51, cLogP = 3.8). In light of this result, further investigation focused on the combination of polar functional groups, which had highly potent activities in the SCD1 enzyme assay at position 4 on the thiazole ring and hydrophobic groups at the ortho-position on the left-hand phenyl ring (Table 4). A benzene ring and thiophene ring were selected for the central part. This combination resulted in compounds with tPSA values of 73–98, and cLogP values of 2.6–6.3. The compounds exhibited potent SCD1 inhibitory activities (IC_{50} < 20 nM) and advanced to the *ex vivo* assay in mice. Eleven compounds (**21**, **34**, **40**, **43**, **45**, **46**, **48**, **49**, **50**, **51**, and **52**) met the criteria for the *ex vivo* assay 6 h after dosing with 1 mg/kg (liver SCD activity < 30%, eyelid SCD activity > 70%). To further examine the tissue selectivity and duration of SCD1 inhibitory activity in these 11 compounds, we performed a higher dose *ex vivo* assay 24 h after dosing mice with 10 and 100 mg/kg (Fig. 5, Supporting Information Table S2). Among all tested compounds, five thiophene analogs (**46**, **48**, **49**, **50**, and **52**) significantly inhibited SCD1 in the liver at 10 mg/kg. However, central-part phenyl analogs (**21**, **34**, **40**, and **43**) had largely diminished activities in the liver at 10 mg/kg. Among five thiophene analogs, **48** and **49**, which bear an acetic acid moiety at position 4 on the thiazole ring and alkyl chain (*n*-butyl and *n*-hexyl, respectively) on the left-hand phenyl ring, achieved liver selectivity and efficacy in the liver (SCD activity in the liver < 20% at 10 mg/kg and in the eyelid > 50% at 100 mg/kg). However, when the alkyl chain length on the left-hand phenyl ring was extended to *n*-octyl (**50**), liver selectivity markedly decreased in the higher dose *ex vivo* assay (SCD activity 11% in the liver at 10 mg/kg and 8% in the eyelid at 100 mg/kg).

We then analyzed the higher dose *ex vivo* assay results against the physicochemical properties cLogP and tPSA. The correlation plots indicated that compounds in the ranges of 3.9 ≤ cLogP ≤5.0 and 90 ≤ tPSA ≤98 (**43**, **48**, **49**, **51**, and **52**) were highly selective for the liver (Fig. 6A and B). Conversely, systemically distributed compound **4** (tPSA = 51, cLogP = 3.8) exhibited strong SCD1 inhibitory activity in the eyelid 24 h after dosing mice with 10 mg/kg, and it lost its SCD1 inhibitory activity in the liver (Fig. 5, Supporting Information Table S2). Other compounds (**21**, **34**, **40**, **45**, **46**, and **50**) outside the ranges of 3.9 ≤ cLogP ≤ 5.0 and 90 ≤ tPSA ≤ 98 also had no liver selectivity. These results indicate that altering the physicochemical properties of lead compound **4** led to a difference in compound exposure between the liver and eyelid. In addition to physicochemical properties, liver selectivity can be caused by the functionality of the compounds. Hepatocellular compound concentrations can be affected by multiple factors, such as passive diffusion of the compounds across membranes, uptake/efflux transporters, and metabolism in the hepatocyte [24]. Hepatic uptake of acidic compounds with relatively large molecular weights (>400 Da) through liver-specific OATPs (1B1, 1B3) is known [25]. All of the liver selective compounds identified in this study are carboxylic acids; however, compound **50**, which is also a carboxylic acid, was not liver selective. Thus, the liver/eyelid tissue selectivity of our compounds may be due, at least in part, by the incorporation of a carboxylic acid substituent into the chemical structure.

We chose compound **48** for further investigation of biological activities. Compound **48** maintained single digit nanomolar potency in rats, and there were no differences in SCD enzyme inhibitory activities in rodents (mouse liver microsome SCD IC_{50} = 5.4 nM, rat liver microsome SCD IC_{50} = 6.5 nM). Prior to the pharmacological studies, the ADME profiles of the compound were explored. As summarized in Table 5, compound **48** was orally absorbed moderately in both rats and mice. There was relatively little difference in exposure between mice and rats. Compound **48** was highly permeable in the Caco-2 permeability assay and had good solubility in PBS buffer (pH 7.2). Against hERG channels, **48** had no significant measurable activity (14.7% inhibition at 10 μM). In addition, **48** had an IC_{50} > 50 μM against the six CYP 450 isoforms tested (3A4, 2C9, 2D6, 1A2, 2A6, and 2C19).

2.3. Pharmacological effects of compound *48*

We assessed anti-diabetic and anti-obesity effects in mice fed a high fat diet (HFD). Compound **48** was administered orally as a dietary mixture for 43 days. As shown in Fig. 7, **48** improved glucose tolerance in a dose-dependent manner and had a significant effect at a dose of 10 mg/kg. As shown in Fig. 8, a mild dose-dependent decrease in body weight was measured (6% body weight reduction at 10 mg/kg vs. vehicle-treated mice on day 43).

Compound **48** exhibited liver-selective SCD1 inhibition not only in mice, but also in rats. As shown in Fig. 9, dose-dependent SCD1 inhibitory activity was observed in epididymal adipose tissue and the liver 6 h after oral administration, whereas the inhibitory activity in the eyelid was milder. Confirming the selectivity, we measured the concentration of compound **48** in select tissues 6 h after dosing rats with 10 mg/kg of the compound (plasma = 0.552 μM, liver = 1.002 μM, and eyelid = 0.160 μM; n = 3). The liver to eyelid ratio was 6.3:1, which indicates a liver/eyelid selective tissue distribution profile. To assess the efficacy of hepatic TG content, **48** was administered orally to rats fed a high sucrose very low fat (HSVLF) diet for 8 days. As shown in Fig. 10, rats fed a HSVLF diet had hepatic TG content of approximately 80 mg/g tissue and compound **48** significantly attenuated hepatic TG accumulation in a dose-dependent manner. Similar results have been reported in liver-specific SCD1 knockout (LKO) mice [15]. In this report, the LKO mice fed a high-carbohydrate diet were protected from liver TG accumulation and hepatic steatosis observed in the control mice fed a high-carbohydrate diet. Therefore, compound **48** could be effective for hepatic steatosis.

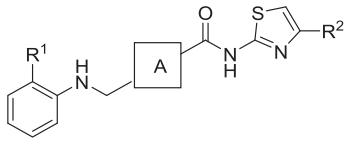
2.4. Preclinical toxicology study

To evaluate adverse effects, compound **48** was orally administered to male rats once daily at 100 mg/kg and 1000 mg/kg for 2 weeks. As noted above, systemically distributed compound **4** had adverse effects in the eyelid at therapeutic doses in db/db mice. In contrast, liver-selective SCD1 inhibitor **48** did not have significant adverse events during the treatment period at 100 mg/kg, and only very slight loss of fur in one out of five rats from day 9 and thereafter at a dose of 1000 mg/kg. Comparing plasma exposure at the no-observed adverse effect level (NOAEL) of 100 mg/kg (179 μM h) with the plasma exposure at therapeutic doses, **48** had a favorable safety margin (10–81-fold, AUC levels). This result was consistent with our *ex vivo* assay, which demonstrated different exposures between the liver and off-target tissues, such as the eyelid and skin.

3. Chemistry

The synthesis of HTS hit compound **1** and its analog **2** is outlined in Scheme 1. Commercially available methyl 4-(bromomethyl) benzoate (**53**; CAS 2417-72-3) reacted with 2-chlorophenol in the presence of K_2CO_3 to provide **54**, followed by alkaline hydrolysis of ester **54** to give **55**. Carboxylic acid **55** reacted with the corresponding amine using EDC·HCl in pyridine-mediated amide coupling to give **1** and **2**. The synthesis of **3**–**11** to explore the substituents on the left-hand phenyl ring is outlined in Scheme 2. Coupling commercially available **56** (CAS 1642-81-5) with 2-amino-4-methylthiazole (**65**; CAS 1603-91-4) in the presence of

Table 4
In vitro and *ex vivo* assays in mice 6 h after dosing with thiazole-2-carboxyamide derivatives.



Compound	R^1	A	R^2	IC_{50} (nM)[a]	*Ex vivo*[b] 1 mg/kg 6 h % of control		cLogP[c]	tPSA[d]
					Liver	Eyelid		
4	Me		Me	6.4	24	41	3.8	51
21	CF_3		CH_2OH	3.8	20	73	3.2	74
32	Cl		CH_2OH	4.3	12	37	2.8	75
33	*n*-Bu		CH_2OH	3.2	10	29	3.9	73
22	CF_3		$(CH_2)_2OH$	4.2	33	136	3.4	75
34	Cl		$(CH_2)_2OH$	6.4	29	74	3.0	77
35	*n*-Bu		$(CH_2)_2OH$	3.0	61	94	4.1	75
36	*iso*-Pen		$(CH_2)_2OH$	5.3	26	63	4.5	75
23	CF_3		$(CH_2)_3OH$	5.2	64	75	3.8	75
17	Me		CH_2CO_2H	14	NT	NT	2.6	97
18	CF_3		CH_2CO_2H	5.1	98	107	3.5	95
37	Cl		CH_2CO_2H	13	72	132	3.1	97
38	*n*-Bu		CH_2CO_2H	5.5	76	74	4.2	95
39	*n*-Hex		CH_2CO_2H	3.0	31	119	5.2	96
40	*n*-Oct		CH_2CO_2H	4.5	15	127	6.3	95
41	*iso*-Pen		CH_2CO_2H	4.8	19	68	4.6	96
42	Ph		CH_2CO_2H	8.7	74	187	4.0	95
43	Bn		CH_2CO_2H	5.5	18	76	4.2	93
44	CF_3		CH_2OH	5.1	9	33	2.9	76
45	CF_3		$(CH_2)_2OH$	3.9	10	171	3.1	77
46	*n*-Hex		$(CH_2)_2OH$	4.5	12	104	4.9	77
47	CF_3		CH_2CO_2H	9.0	51	124	3.2	98
48	*n*-Bu		CH_2CO_2H	8.8	13	97	3.9	98
49	*n*-Hex		CH_2CO_2H	4.8	12	88	5.0	98
50	*n*-Oct		CH_2CO_2H	17	7	77	6.0	98
51	*iso*-Pen		CH_2CO_2H	6.7	19	160	4.3	98
52	Bn		CH_2CO_2H	7.5	22	128	3.9	96

[a] Recombinant human SCD1 enzyme.
[b] Compounds (1 mg/kg) were orally administered to mice and then the liver and eyelid collected 6 h later. Tissue homogenates were obtained and SCD activity measured. Results are expressed as percentage of control for remaining SCD activity.
[c] Calculated logarithm of the partition coefficient.
[d] Topological polar surface area.

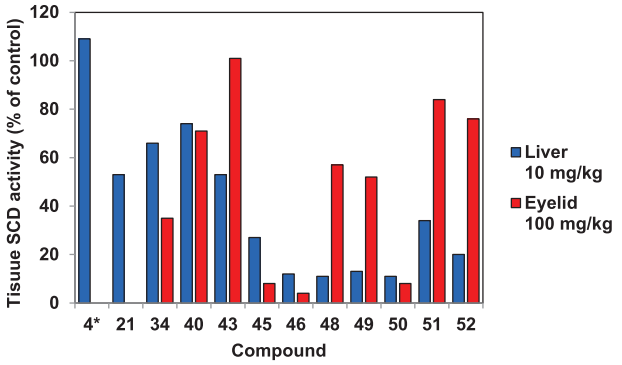


Fig. 5. Higher dose *ex vivo* assay in mice. Compounds **4**, **21**, **34**, **40**, **43**, **45**, **46**, and **48**–**52** (10 or 100 mg/kg) suspended in vehicle (0.5% methylcellulose aqueous solution) were orally administered to C57BL/6 J mice. Twenty-four hours after administration, liver and eyelid samples were collected and homogenized to measure tissue SCD activity. The results are expressed as percentage of control for remaining SCD activity. *Dose was 10 mg/kg.

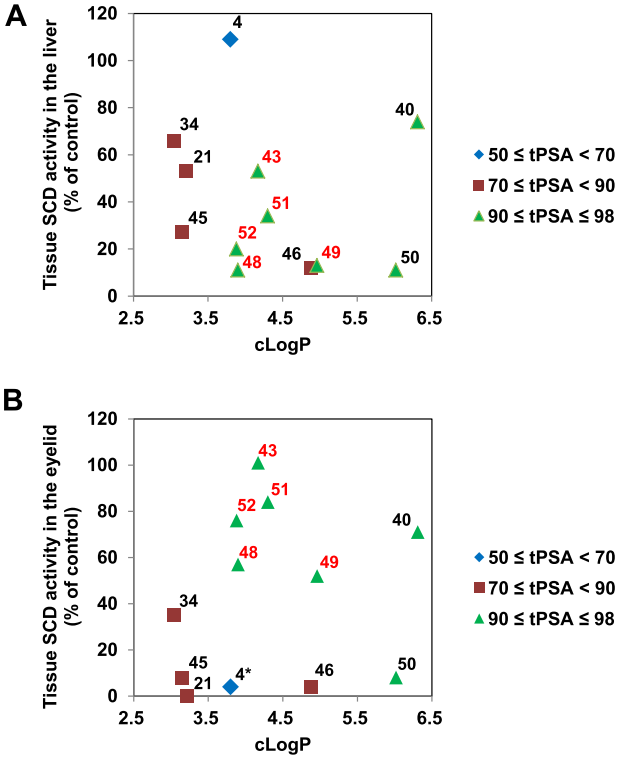


Fig. 6. Plot of higher dose *ex vivo* assay results against physicochemical properties. **A**. Liver SCD activity in higher dose *ex vivo* assays (y-axis, mice, 10 mg/kg, 24 h after dosing, Fig. 5) and the cLogP of the compounds (x-axis). Points are color-coded by tPSA (blue, brown, green) and labeled with compound identifiers. **B**. Eyelid SCD activity in higher dose *ex vivo* assays (y-axis, mice, 100 mg/kg, 24 h after dosing, Fig. 5) and the cLogP of the compounds (x-axis). Points are color-coded by tPSA (blue, brown, green) and labeled with compound identifiers. *Dose was 10 mg/kg.

HATU yielded 4-(chloromethyl)phenyl carboxamide (**57**), followed by nucleophilic substitution of the benzylchloride with corresponding 2-alkylaniline analogs in the presence of KI, provided **3**–**9**. Carboxylic acid analog **10** was obtained from the basic hydrolysis reaction of ester intermediate **58**. Compound **10** was then transformed into dimethylamide derivative **11** via PyBOP coupling with dimethylamine.

Compounds **12** and **13**, which have a pyridine ring in the central part, were prepared according to Scheme 3. The reaction of commercially available carboxylic acids **59a** (CAS 1154575-21-9) and **59b** (CAS 1154323-05-3) with borane-THF at appropriate temperatures provided corresponding alcohols **60a** and **60b**. The primary alcohols were converted to 4-chloromethylpyridines **61a** and **61b**, and alkylation with 2-methylaniline in the presence of KI provided compounds **12** and **13**.

Piperidinecarboxamide **14** was obtained as outlined in Scheme 4. 2-amino-4-methylthiazole (**65**) was converted to imidazole amide **66**. Subsequently, a coupling reaction was carried out with 4-substituted piperidine hydrochloride (**64**) synthesized by the palladium-assisted coupling of **62** (CAS 144222-22-0) with 2-bromobenzotrifluoride, followed by treatment with HCl.

The general synthesis of phenyl carboxamide analogs **16**–**43** to explore the polar functional groups at the thiazole moiety is described in Schemes 5 and 6. As shown in Scheme 5, benzyl bromide **53** was reacted with the corresponding 2-alkylaniline analogs in the presence of NaI to yield the corresponding N-alkylated compounds (**67**–**75**), which were subsequently hydrolyzed to the corresponding carboxylic acids (**76**–**84**). Compounds **27**–**29** were synthesized by coupling of carboxylic acid **77** with the corresponding amines. Compounds **16**–**20**, **24**–**26**, **30**, and **37**–**43** were synthesized by amide coupling, followed by hydrolysis of corresponding carboxylic esters **88**–**92**, **100**–**102**, **103**, and **93**–**99**. Hydroxyl compounds **22**, **23**, and **34**–**36** were synthesized by sodium borohydride reduction of corresponding carboxylic esters **90**, **91**, and **93**–**95**. Alkaline hydrolysis of acetates **85**, **86**, and **87** produced hydroxyl compounds **21**, **32**, and **33**.

The synthesis of 4,5-disubstituted thiazole **31** was performed as described in Scheme 6. Commercially available diester **104** (CAS 6317-49-3) was converted to 2-amino-4,5-disubstitued thiazole **106** in two steps: bromination with bromine in $CHCl_3$, followed by cyclization with thiourea. Amide coupling between **106** and carboxylic acid **77**, followed by ester reduction with lithium borohydride, resulted in the formation of **31**.

The general synthesis of thiophene carboxamide analogs **15** and **44**–**52** is described in Scheme 7. Imines **109**–**114** were prepared by reaction of commercially available 5-formyl-2-thiophenecarboxylic acid (**108**; CAS 4565-31-5) with corresponding 2-alkyl anilines under Dean-Stark conditions. Subsequent reduction with sodium borohydride yielded amines **115**–**120**. Compound **115** was then transformed into amide derivative **15** via EDC·HCl coupling in pyridine with 2-amino-4-methylthiazole (**65**). Amide coupling between **115**–**120** and ethyl (2-amino-4-thiazolyl) acetate, followed by alkaline hydrolysis of esters **124**–**129**, afforded carboxylic acids **47**, **51**, **52**, and **130**–**132**. Compounds **48**–**50** were obtained as a sodium salt by treatment of **130**–**132** with aqueous NaOH in EtOH. Analogously, compounds **44**–**46** were prepared from the coupling of carboxylic acid **115** or **117** with the corresponding amine in the presence of EDC·HCl and DMAP, followed by hydrolysis of the ester **121** or deprotection of the TBS ethers **122** and **123**.

4. Conclusion

In conclusion, novel thiazole-4-acetic acid analog **48** was identified as a highly potent, orally active, and liver-selective SCD1 inhibitor in *ex vivo* assay screening starting from systemically distributed lead compound **4**. Lead optimization focused on the physicochemical properties tPSA and cLogP, which was effective for achieving tissue selectivity. Exposure in eyelids was minimized by introducing a carboxylic acid group on the thiazole ring and

Table 5
PK data for **48**.

species	po						iv	
	Dose (mg/kg)	C_{max}[c] (μM)	t_{max}[d] (h)	$t_{1/2}$[e] (h)	$AUC_{0\text{-}inf}$[f] (μM·h)	BA[g] (%)	Dose (mg/kg)	Cl_{tot}[h] (L/h/kg)
Rat[a]	10	8.0	0.5	4.7	16.5	16	1	0.21
Mouse[b]	10	1.7	1.0	5.2	10.2	28	1	0.61

Metabolic stability in liver S9 (remaining %, 1 h)			Caco2 Papp ($cm/sec \cdot 10^{-6}$)	Solubility in PBS buffer (pH 7.2) (μM)
Human	Mouse	Rat		
91	90	87	43.60	82

[a] Based on a per os (po) dose of 10 mg/kg and intravenous (iv) dose of 1 mg/kg in two male Sprague Dawley (SD) rats.
[b] Based on a per os (po) dose of 10 mg/kg and intravenous (iv) dose of 1 mg/kg in two male C57BL/6 J mice.
[c] Maximum observed concentration of the compound in plasma.
[d] Time of maximum observed concentration of the compound in plasma.
[e] Apparent half-life of the terminal phase of elimination of the compound from plasma.
[f] Area under the concentration-time curve from the time of dosing to infinity.
[g] Bioavailability.
[h] Total body clearance of compound.

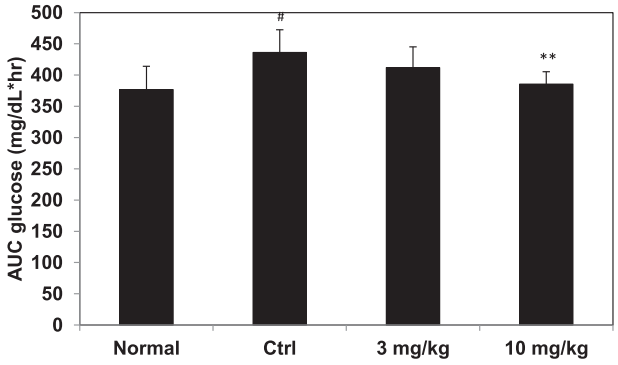


Fig. 7. Effects of **48** on glucose tolerance in C57BL/6 J mice fed a high fat diet (HFD). Compound **48** was administered with food admixture for 43 days. On day 22 of administration, a glucose tolerance (1 g/kg) test was performed with the mice fasted overnight. Area under the curve of plasma glucose level (AUC glucose) was calculated from plasma glucose levels 0, 0.5, 1, and 2 h after glucose loading. Data are presented as the mean ± standard deviation (n = 5–8). #p < 0.05 vs. Normal group (Student's t-test), **p < 0.01 vs. HFD group (Dunnett's test).

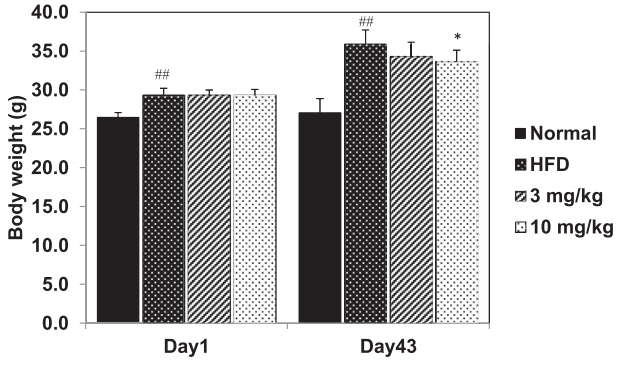


Fig. 8. Effects of **48** on body weight in C57BL/6 J mice fed a high fat diet for 43 days. Compound **48** was administered with food admixture for 43 days. Data are presented as the mean ± standard deviation (n = 5–8). ##p < 0.01 vs. Normal group (Student's t-test), *p < 0.05 vs. Ctrl group (Dunnett's test).

hydrophobic groups on the left-side phenyl ring with a central thiophene ring. Compound **48** ameliorated diet-induced obesity and hepatic TG accumulation, and improved glucose tolerance in diabetic mouse and rat models in the absence of adverse skin or eye effects. Based on a preliminary toxicology study, compound **48** could have a sufficient therapeutic window for chronic treatment.

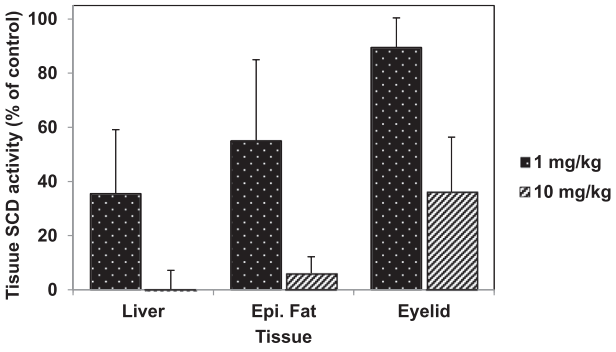


Fig. 9. Effects of **48** on the liver, epididymal adipose tissue, and eyelid SCD activities in Sprague Dawley rats fed a high sucrose (HS) diet. Samples were collected and homogenized 6 h after administration. Results are expressed as percentage of control for remaining SCD activity. Data are presented as the mean ± standard deviation (n = 3).

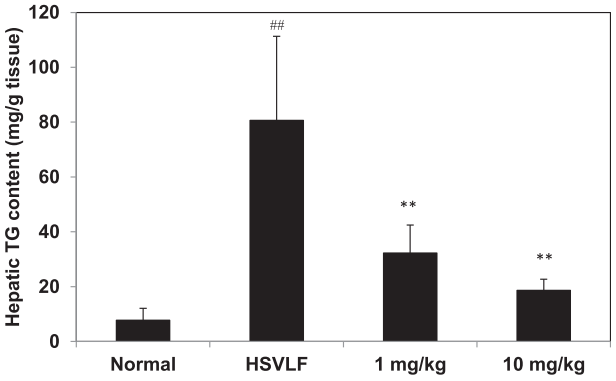
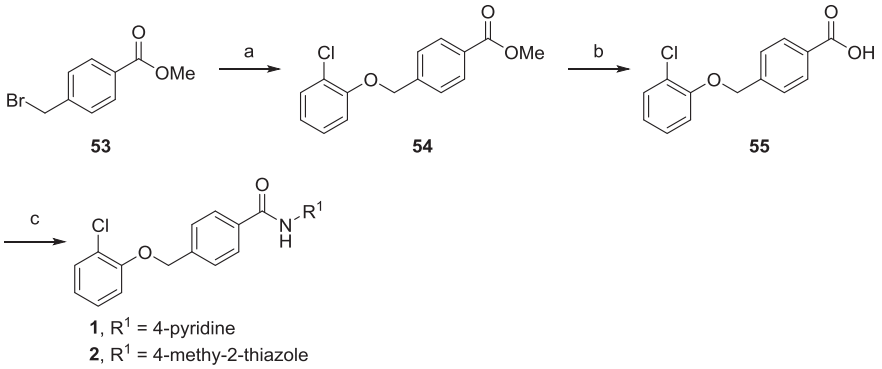


Fig. 10. Effects of **48** on liver triglyceride (TG) contents in Sprague Dawley rats fed a high sucrose very low fat (HSVLF) diet. Samples were collected after 8 days of treatment. Data are presented as the mean ± standard deviation (n = 6). ##p < 0.01 vs. Normal group (Student's t-test), **p < 0.01 vs. HSVLF group (Dunnett's test).

5. Experimental section

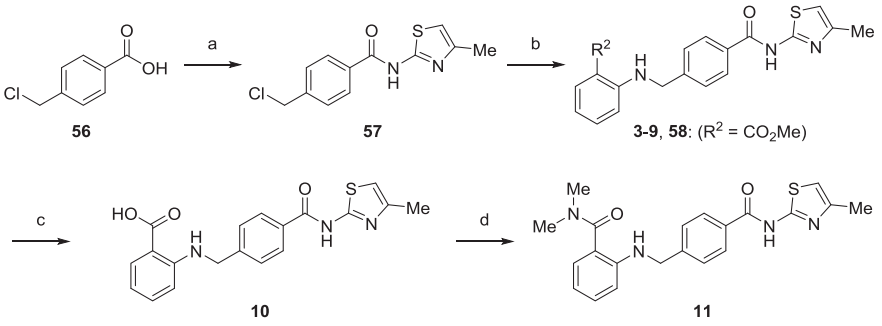
5.1. General

All reagents and solvents obtained from commercial sources were used without further purification or drying. TLC was performed using silica gel 60 F_{254} plates purchased from Merck. Silica gel chromatography was performed using either silica gel (Kanto



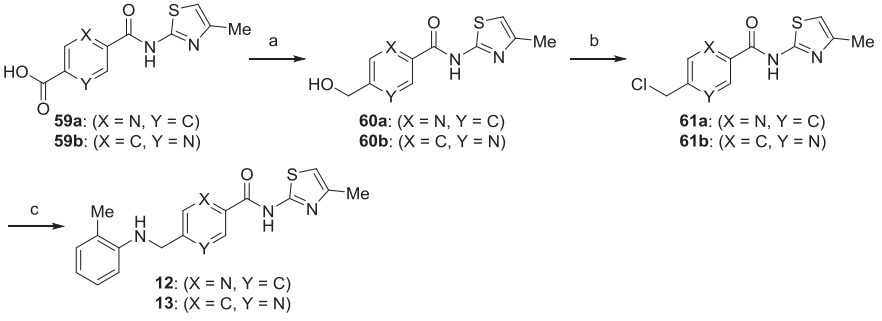
Reagents and conditions: (a) 2-chlorophenol, K_2CO_3, DMSO, 65°C; (b) 4N NaOH, MeOH, THF, reflux; (c) R^1-NH_2, EDC·HCl, pyridine, rt

Scheme 1. Synthesis of phenyl carboxamide **1** and **2**.



Reagents and conditions: (a) 2-amino-4-methylthiazole **65**, HATU, DIPEA, DMF, rt; (b) 2-R^2-aniline, KI, DMF, 60°C; (c) 4N NaOH, MeOH, THF, rt; (d) Me_2NH, PyBOP, rt

Scheme 2. Synthesis of phenyl carboxamide **3**–**11**.

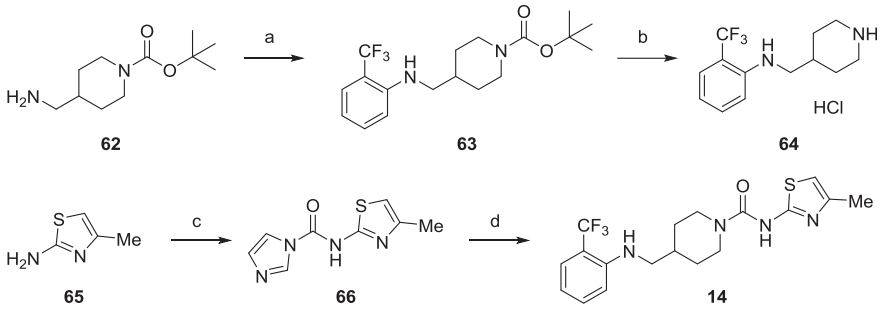


Reagents and conditions: (a) BH_3·THF, THF, 55°C; (b) $SOCl_2$, $CHCl_3$, 50°C; (c) 2-Methylaniline, KI, THF, 65°C

Scheme 3. Synthesis of picolinamide **12** and nicotinamide **13**.

Chemical, 40–100 μm) with standard techniques or prepacked SNAP Ultra C18 and the Biotage Isora One flash purification system. Analytic HPLC was performed on a SHIMAZU Prominence instrument. Melting points were determined using a Yanagimoto micro melting point apparatus or BüCHI B-540 melting point instrument and were uncorrected. ^{1}H NMR was recorded on a JEOL JNM-A300W, Bruker AMX-300, or JEOL JNM-AL400 spectrometer in the indicated solvent. Chemical shifts are reported in parts per million (ppm) relative to internal standard tetramethylsilane in δ-units, and coupling constants (*J*-values) are given in hertz (Hz). Elemental analysis was performed with an Elementar Analysensysteme GmbH vario EL III Element Analyzer. Mass spectra (ESI+) were recorded on a ThermoQuest LCQ mass spectrometer. All compounds that appear in SAR tables in the manuscript were determined to be ≥ 90% pure by NMR. The purity of compounds **48**–**50** was determined by HPLC (4.6 × 150 mm, 5 μm SHISEIDO CAPCELL PAK C18 UG120 column; mobile phase A: 0.1% formic acid in water; mobile phase B: 0.1% formic acid in MeCN; gradient: 50% B to 90% B from 0 to 17 min, 90% B from 17 to 20 min, 90% B to 50% B from 20 to 30 min; flow rate: 0.5 ml/min; detection wavelength: 250 nm).

The following section comprises the synthetic methods and analytical data for all compounds reported in this publication.



Reagents and conditions: (a) 2-bromobenzotrifluoride, $Pd_2(dba)_3$, X-Phos, *t*-BuONa, Toluene, 80°C; (b) 4N HCl, Dioxane, MeOH, rt; (c) CDI, $CHCl_3$, rt; (d) **64**, TEA, $CHCl_3$, rt

Scheme 4. Synthesis of piperidinecarboxamide **14**.

*5.1.1. 4-((2-Chlorophenoxy)methyl)–N-(pyridin-4-yl)benzamide (**1**)*

Step 1. Mixture of methy 4-(bromomethyl)benzoate (**53**; 2.3 g, 10 mmol), K_2CO_3 (4.1 g, 30 mmol), and 2-chlorophenol (1.2 ml, 12 mmol) in DMSO (23 ml) was stirred at 65 °C for 3 h under argon atmosphere. After cooling to room temperature, water and EtOAc were added and the water layer extracted twice with EtOAc. The combined organic layer was washed with brine and dried over anhydrous $MgSO_4$. Filtration and concentration afforded the title compound methyl 4-((2-chlorophenoxy)methyl)benzoate (**54**; 2.57 g, 98% yield). ^{1}H NMR (DMSO-d_6) δ: 3.86 (s, 3H), 5.32 (s, 2H), 6.96–7.00 (m, 1H), 7.22 (dd, J = 8.3, 1.4 Hz, 2H), 7.30 (dd, J = 8.6, 7.0 Hz, 2H), 7.46 (dd, J = 7.9, 1.4 Hz, 1H), 7.61 (d, J = 8.3 Hz, 1H), 8.00 (dd, J = 6.7, 1.9 Hz, 1H).

Step 2. To a solution of **54** (2.57 g, 9.78 mmol) in MeOH (20 ml) and THF (6 ml) was added 4 N NaOH (3.8 ml, 15 mmol) at room temperature. The reaction mixture was heated to reflux for 3 h. After cooling to 0 °C, water, followed by 2 N HCl, was added portion-wise and the reaction mixture allowed to stir at room temperature. The slurry was filtered and the filter cake washed with water, followed by drying in a vacuum to afford 4-((2-chlorophenoxy)methyl)benzoic acid (**55**; 2.43 g, 95%) as a white solid. ^{1}H NMR (DMSO-d_6) δ: 5.31 (s, 2H), 6.98 (td, J = 7.5, 1.2 Hz, 1H), 7.22 (dd, J = 8.3, 1.2 Hz, 1H), 7.28–7.32 (m, 1H), 7.46 (dd, J = 7.9, 1.6 Hz, 1H), 7.59 (d, J = 8.3 Hz, 2H), 7.98 (d, J = 8.3 Hz, 2H), 12.99 (br s, 1H).

Step 3. To a solution of **55** (50 mg, 0.19 mmol) in pyridine (1 ml) was added EDC·HCl (44 mg, 0.23 mmol), followed by 4-aminopyridine (18 mg, 0.19 mmol) at room temperature. The reaction mixture was stirred at room temperature overnight. Water was added to the reaction, and extraction with EtOAc performed twice. The combined organic layer was washed with brine and dried over anhydrous $MgSO_4$. After filtration and concentration, the residue was purified by preparative TLC ($CHCl_3$/MeOH = 9/1) to afford the title compound 4-((2-chlorophenoxy)methyl)–N-(pyridin-4-yl)benzamide (**1**; 8 mg, 12% yield). MS ESI m/e: 339 (M + H), 337 (M − H); ^{1}H NMR (DMSO-d_6) δ: 5.34 (s, 2H), 6.98 (td, J = 7.6, 1.5 Hz, 1H), 7.24 (dd, J = 8.3, 1.6 Hz, 1H), 7.29–7.33 (m, 1H), 7.46 (dd, J = 7.9, 1.4 Hz, 1H), 7.64 (d, J = 8.3 Hz, 2H), 7.80 (d, J = 6.5 Hz, 2H), 7.99 (d, J = 8.3 Hz, 2H), 8.49 (dd, J = 5.0, 1.5 Hz, 2H), 10.64 (br s, 1H); Anal. Calcd for $C_{19}H_{15}ClN_2O_2 \cdot 0.5H_2O$: C, 65.62; H, 4.64; N, 8.05. Found: C, 65.87; H, 4.49; N, 7.93.

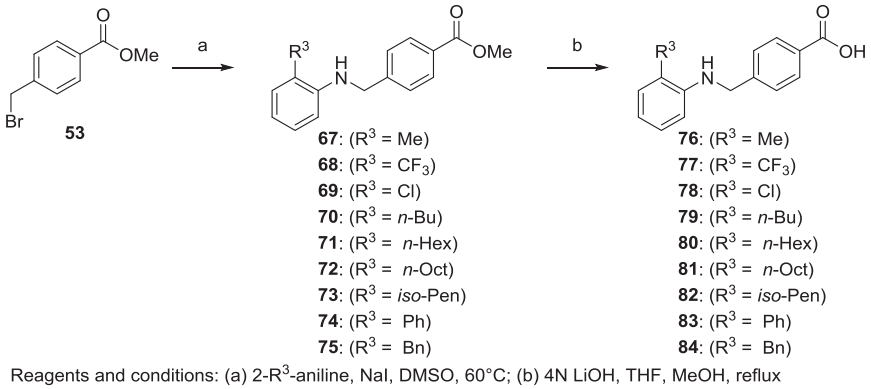
*5.1.2. 4-((2-Chlorophenoxy)methyl)–N-(4-methylthiazol-2-yl) benzamide (**2**)*

To a solution of 4-((2-chlorophenoxy)methyl)benzoic acid (**55**; 50 mg, 0.19 mmol) and 2-amino-4-methylthiazole (**65**; 22 mg, 0.19 mmol) in pyridine (0.5 ml) was added EDC·HCl (44 mg, 0.23 mmol) at room temperature. After stirring at room temperature overnight, water was poured into the reaction and the mixture extracted with EtOAc. The organic layer was washed with brine and dried over anhydrous $MgSO_4$. After filtration and concentration, the residue was purified by preparative TLC ($CHCl_3$/MeOH = 9/1) to afford the title compound 4-((2-chlorophenoxy)methyl)–N-(4-methylthiazol-2-yl)benzamide (**2**; 30 mg, 44% yield). MS ESI m/e: 357 (M + H), 359 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.31 (d, J = 0.9 Hz, 3H), 5.32 (s, 2H), 6.83 (s, 1H), 6.98 (td, J = 7.6, 1.5 Hz, 1H), 7.24 (dd, J = 8.3, 1.4 Hz, 1H), 7.31 (d, J = 17.3 Hz, 1H), 7.46 (dd, J = 7.9, 1.6 Hz, 1H), 7.62 (d, J = 8.3 Hz, 2H), 8.11 (d, J = 8.6 Hz, 2H), 12.56 (br s, 1H); Anal. Calcd for $C_{18}H_{15}ClN_2O_2S$: C, 60.25; H, 4.21; N, 7.81. Found: C, 60.19; H, 4.20; N, 7.79.

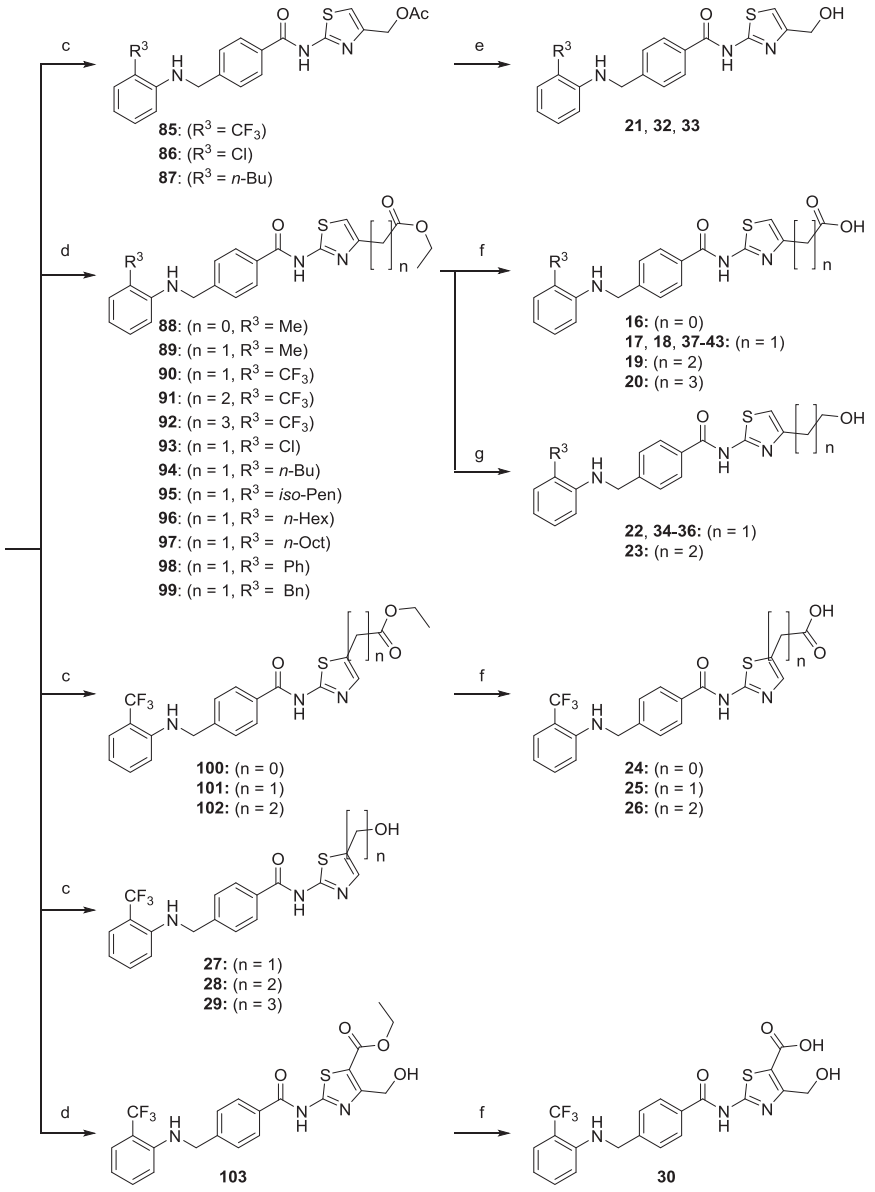
*5.1.3. N-(4-methyl-thiazol-2-yl)-4-(o-tolylamino-methyl)-benzamide (**4**)*

Step 1. To a solution of 4-(chloromethyl)benzoic acid (**56**; 1.71 g, 10 mmol) in DMF (17 ml) was added DIPEA (2.6 ml, 15 mmol), followed by 2-amino-4-methylthiazole (**65**; 1.14 g, 10 mmol) at room temperature. HATU (4.56 g, 12 mmol) was then added and the reaction mixture stirred at room temperature overnight. To the reaction was added saturated $NaHCO_3$ (10 ml), followed by extraction with EtOAc twice. The combined organic layer was washed with brine, dried over anhydrous $MgSO_4$, filtered, and concentrated. The insoluble material was precipitated from the evaporated residue and filtered off. The filter cake was washed with EtOAc. The filtrate was concentrated and purified by reverse phase flash chromatography (MeCN/H_2O) to afford **57** (1.3 g, 47%). MS ESI m/e: 267 (M + H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.31 (s, 3H), 4.84 (s, 2H), 6.83 (s, 1H), 7.59 (d, J = 8.1 Hz, 2H), 8.08 (d, J = 10.0 Hz, 2H), 12.60 (br s, 1H).

Step 2. To a solution of **57** (50 mg, 0.187 mmol) and KI (31 mg, 0.187 mmol) in DMF (0.5 ml) was added 2-methylaniline (0.06 ml, 0.562 mmol). The reaction mixture was stirred at 60 °C for 2 h, cooled to room temperature, and water (1 ml) added, followed by saturated $NaHCO_3$ (pH 8). The water layer was extracted with EtOAc and separated. The organic layer was washed with saturated $NaHCO_3$, followed by brine, then dried over Na_2SO_4. After filtration and removal of the solvent under reduced pressure, the residue was purified by flash chromatography ($CHCl_3$/EtOAc = 9/1) to afford N-(4-methyl-thiazol-2-yl)-4-(o-tolylamino-methyl)-benzamide (**4**; 45 mg, 67%) as an amorphous solid. MS ESI m/e: 338 (M + H), 336 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.18 (s, 3H), 2.30 (d, J = 0.9 Hz, 3H), 4.44 (d, J = 6.0 Hz, 2H), 5.72 (t, J = 6.0 Hz, 1H), 6.32 (d, J = 7.5 Hz, 1H), 6.47 (td, J = 7.3, 1.0 Hz, 1H), 6.80 (q, J = 1.1 Hz, 1H), 6.86–6.90 (m, 1H), 6.95–7.00 (m, 1H), 7.49 (d, J = 8.4 Hz, 2H), 8.03 (d, J = 8.4 Hz, 2H), 12.46 (br s, 1H); Anal. Calcd for $C_{19}H_{19}N_3OS \cdot 0.25H_2O$: C, 66.74; H, 5.75; N, 12.29. Found: C, 66.83; H,

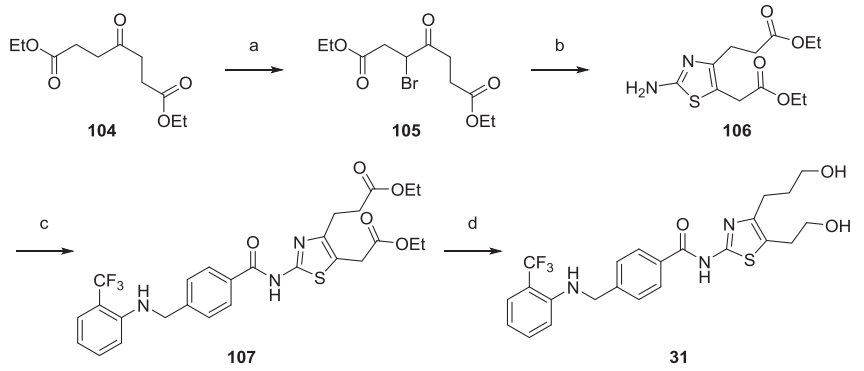


Reagents and conditions: (a) 2-R^3-aniline, NaI, DMSO, 60°C; (b) 4N LiOH, THF, MeOH, reflux



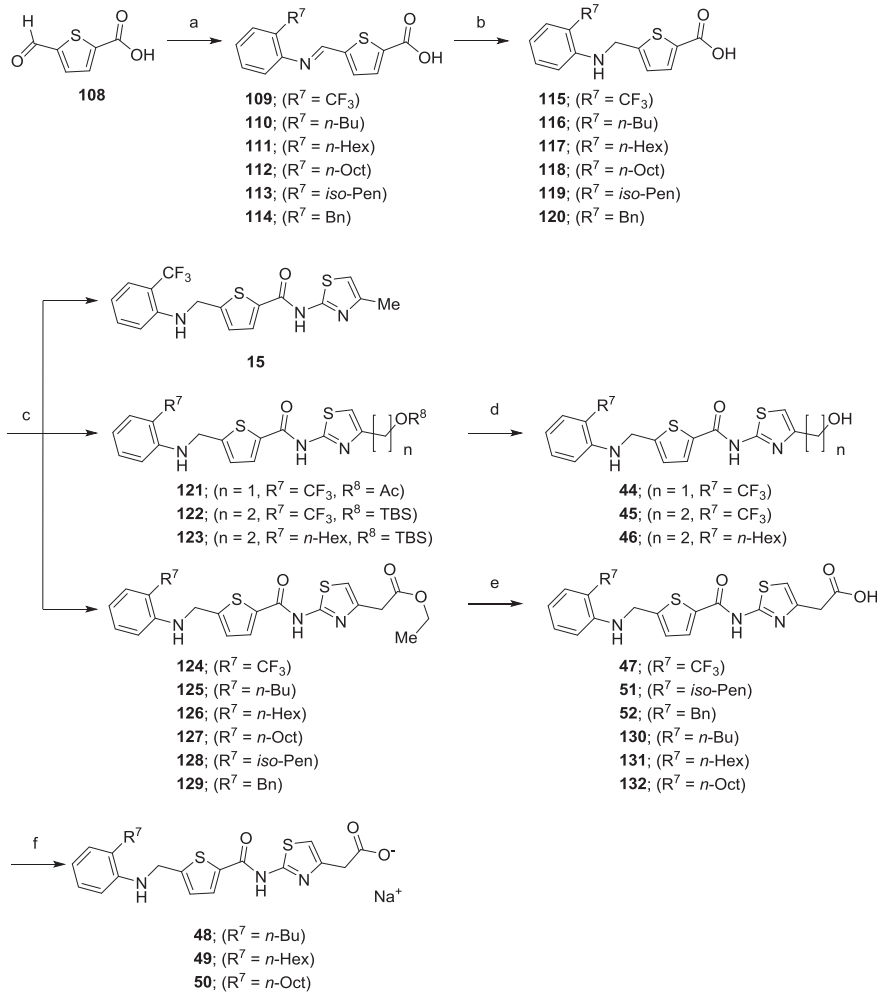
Reagents and conditions: (c) 2-Amino-4-R^4-5-R^5-thiazole, PyBOP, DIPEA, DMF, rt–65°C; (d) 2-Amino-4-R^6-thiazole, EDC·HCl, pyridine, 0°C–rt; (e) K_2CO_3, MeOH, H_2O, 65°C; (f) 4N NaOH, EtOH or MeOH, 0°C–reflux; (g) $NaBH_4$, t-BuOH, THF. MeOH, reflux.

Scheme 5. Synthesis of phenyl carboxamide **16**–**30**, **32**–**43**.



Reagents and conditions: (a) Br_2, $CHCl_3$, 0°C–rt; (b) thiourea, EtOH, 100°C; (c) **77**, PyBOP, DIPEA, DMF, 65°C; (d) $LiBH_4$, THF, rt–65°C

Scheme 6. Synthesis of phenyl carboxamide **31**.



Reagents and conditions: (a) 2-R^7-aniline, toluene, reflux; For **15**: 2-amino-CF_3-benzene, DME, AcOH, rt; (ii) $NaBH_4$, 0°C–rt; (b) $NaBH_4$, DME, 0°C–rt then AcOH, 0°C; (c) 2-Amino-4-R^2-thiazole, EDC·HCl, DMAP, $CHCl_3$, 0°C–rt; For **15** : 2-amino-4-methylthiazole, EDC·HCl, pyridine, rt; (d) For **44**: K_2CO_3, THF, MeOH, rt; For **45**, **46:** TBAF, THF, 50°C; (e) 2N NaOH, MeOH, THF, 0°C–rt; (f) 4N NaOH, EtOH, 80°C–rt

Scheme 7. Synthesis of thiophene carboxamide **15**, **44**–**52**.

5.72; N, 12.10; mp 157–159 °C.

*5.1.4. 4-[(2-Chloro-phenylamino)-methyl]–N-(4-methyl-thiazol-2-yl)-benzamide (**3**)*

This compound was prepared according to the synthetic protocols described for compound **4**, starting from 4-(chloromethyl)–N-(4-methylthiazol-2-yl)benzamide (**57**) and 2-chloroaniline. MS ESI m/e: 358 (M + H), 356 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.30 (d, J = 0.8 Hz, 3H), 4.49 (d, J = 6.4 Hz, 2H), 6.24 (t, J = 6.0 Hz, 1H), 6.48–6.58 (m, 2H), 6.80 (br s, 1H), 6.97–7.05 (m, 1H), 7.25 (dd, J = 7.9, 1.5 Hz, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.46 (br s, 1H); Anal. Calcd for $C_{18}H_{16}ClN_3OS$: C, 60.41; H, 4.51; N, 11.74. Found: C, 60.33; H, 4.56; N, 11.54.

*5.1.5. N-(5-methylthiazol-2-yl)-4-((phenylamino)methyl) benzamide (**5**)*

This compound was prepared according to the synthetic protocols described for compound **4**, starting from 4-(chloromethyl)–N-(4-methylthiazol-2-yl)benzamide (**57**) and aniline. MS ESI m/e: 324 (M + H), 322 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.52 (s, 1H), 4.35 (d, J = 6.2 Hz, 2H), 6.33 (t, J = 6.2 Hz, 1H), 6.49–6.57 (m, 3H), 6.81 (s, 1H), 7.04 (td, J = 6.9, 1.6 Hz, 2H), 7.49 (d, J = 8.3 Hz, 2H), 8.04 (d, J = 8.3 Hz, 2H), 12.47 (br s, 1H); Anal. Calcd for $C_{18}H_{17}N_3OS$: C, 66.85; H, 5.30; N, 12.99. Found: C, 66.80; H, 5.41; N, 13.02.

*5.1.6. N-(4-methyl-thiazol-2-yl)-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**6**)*

This compound was prepared according to the synthetic protocols described for compound **4**, starting from 4-(chloromethyl)–N-(4-methylthiazol-2-yl)benzamide (**57**) and 2-(trifluoromethyl) aniline. MS ESI m/e: 392 (M + H), 390 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.30 (s, 3H), 4.54 (d, J = 5.7 Hz, 2H), 6.38 (t, J = 6.2 Hz, 1H), 6.58–6.68 (m, 2H), 6.80 (br s, 1H), 7.28 (t, J = 7.3 Hz, 1H), 7.39–7.50 (m, 3H), 8.03 (d, J = 8.3 Hz, 2H), 12.47 (br s, 1H); Anal. Calcd for $C_{19}H_{16}F_3N_3OS$: C, 58.30; H, 4.12; N, 10.74. Found: C, 58.31; H, 3.97; N, 10.69; mp 159–161 °C.

*5.1.7. 4-(((3-Chlorophenyl)amino)methyl)–N-(5-methylthiazol-2-yl)benzamide (**7**)*

This compound was prepared according to the synthetic protocols described for compound **4**, starting from 4-(chloromethyl)–N-(4-methylthiazol-2-yl)benzamide (**57**) and 3-chloroaniline. MS ESI m/e: 358 (M + H), 356 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.32 (s, 3H), 4.37 (d, J = 6.0 Hz, 2H), 6.52 (dd, J = 8.0, 2.0 Hz, 2H), 6.57 (t, J = 2.1 Hz, 1H), 6.69 (t, J = 6.1 Hz, 1H), 6.81 (br s, 1H), 7.04 (t, J = 8.0 Hz, 1H), 7.48 (d, J = 8.3 Hz, 2H), 8.05 (d, J = 8.3 Hz, 2H), 12.49 (br s, 1H).; Anal. Calcd for $C_{18}H_{16}ClN_3OS$: C, 60.41; H, 4.51; N, 11.74. Found: C, 60.44; H, 4.57; N, 11.62.

*5.1.8. 4-(((4-Chlorophenyl)amino)methyl)–N-(5-methylthiazol-2-yl)benzamide (**8**)*

This compound was prepared according to the synthetic protocols described for compound **4**, starting from 4-(chloromethyl)–N-(4-methylthiazol-2-yl)benzamide (**57**) and 4-chloroaniline. MS ESI m/e: 358 (M + H), 356 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.30 (s, 3H), 4.35 (d, J = 6.2 Hz, 2H), 6.55–6.58 (m, 3H), 6.81 (s, 1H), 7.06 (dt, J = 9.7, 2.7 Hz, 2H), 7.47 (d, J = 8.3 Hz, 2H), 8.04 (d, J = 8.3 Hz, 2H), 12.48 (br s, 1H); Anal. Calcd for $C_{18}H_{16}ClN_3OS$: C, 60.41; H, 4.51; N, 11.74. Found: C, 60.28; H, 4.67; N, 11.62.

*5.1.9. N-(5-methylthiazol-2-yl)-4-(((2-propylphenyl)amino) methyl)benzamide (**9**)*

This compound was prepared according to the synthetic protocols described for compound **4**, starting from 4-(chloromethyl)–N-(4-methylthiazol-2-yl)benzamide (**57**) and 2-propylaniline. MS ESI m/e: 366 (M + H), 364 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.98 (t, J = 7.4 Hz, 3H), 1.61 (td, J = 15.0, 7.4 Hz, 2H), 2.30 (s, 3H), 2.48–2.55 (m, 2H), 4.43 (d, J = 6.0 Hz, 2H), 5.85 (t, J = 5.9 Hz, 1H), 6.30 (d, J = 8.1 Hz, 1H), 6.48 (dd, J = 7.3, 6.4 Hz, 1H), 6.81 (s, 1H), 6.86 (t, J = 6.9 Hz, 1H), 6.94 (dd, J = 7.3, 1.5 Hz, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.02 (d, J = 8.3 Hz, 2H), 12.46 (br s, 1H).; Anal. Calcd for $C_{21}H_{23}N_3OS$: C, 69.01; H, 6.34; N, 11.50. Found: C, 69.12; H, 6.31; N, 11.47.

*5.1.10. 2-((4-((5-Methylthiazol-2-yl)carbamoyl)benzyl)amino) benzoic acid (**10**)*

Step 1. To a solution of **57** (150 mg, 0.562 mmol) and KI (93 mg, 0.562 mmol) in DMF (1.5 ml) was added 2-aminobenzoic acid methyl ester (0.22 ml, 1.69 mmol). The reaction mixture was stirred at 60 °C for 2 h, cooled to room temperature, and water (3 ml) added, followed by saturated $NaHCO_3$ (pH 8). The water layer was extracted with EtOAc and separated. The organic layer was washed with saturated $NaHCO_3$, followed by brine, and then dried over $MgSO_4$. After filtration and removal of the solvent under reduced pressure, the residue was purified by reverse phase flash chromatography (MeCN/H_2O) to afford methyl 2-((4-((4-methylthiazol-2-yl)carbamoyl)benzyl)amino)benzoate (**58**; 89 mg, 42%) as an amorphous solid. MS ESI m/e: 382 (M + H), 380 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.30 (s, 3H), 3.83 (s, 3H), 4.59 (d, J = 6.0 Hz, 2H), 6.58–6.62 (m, 1H), 6.66 (d, J = 8.3 Hz, 1H), 6.81 (s, 1H), 7.30–7.34 (m, 1H), 7.48 (d, J = 8.1 Hz, 2H), 7.82 (dd, J = 7.9, 1.6 Hz, 1H), 8.05 (d, J = 8.3 Hz, 2H), 8.21 (t, J = 6.0 Hz, 1H), 12.51 (br s, 1H).

Step 2. To a solution of **58** (73 mg, 0.19 mmol) in THF (0.35 ml) and MeOH (0.35 ml) was added 4 N NaOH (144 μl, 0.57 mmol) at room temperature. The reaction mixture was stirred for 3 h at 60 °C. After cooling to 0 °C, water (5 ml), followed by 2 N HCl (0.2 ml), was added portion-wise and the reaction mixture allowed to stir at room temperature. The slurry was filtered and the filter cake washed with water (4 × 30 ml), followed by drying in a vacuum to afford 2-((4-((5-methylthiazol-2-yl)carbamoyl)benzyl)amino)benzoic acid (**10**; 50 mg, 71%) as a white solid. MS ESI m/e: 368 (M + H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.30 (s, 2H), 4.58 (s, 1H), 6.55–6.59 (m, 1H), 6.62 (d, J = 8.3 Hz, 1H), 6.81 (s, 1H), 7.26–7.31 (m, 1H), 7.48 (d, J = 8.3 Hz, 1H), 7.81 (dd, J = 7.9, 1.6 Hz, 1H), 8.05 (d, J = 8.3 Hz, 1H), 12.51 (br s, 1H); Anal. Calcd for $C_{19}H_{17}N_3O_3S$: C, 62.11; H, 4.66; N, 11.44. Found: C, 62.14; H, 4.71; N, 11.39.

*5.1.11. N,N-dimethyl-2-((4-((5-methylthiazol-2-yl)carbamoyl) benzyl)amino)benzamide (**11**)*

To a solution of **10** (50 mg, 0.135 mmol) in DMF (0.5 ml) was added a 2 M THF solution of dimethylamine (0.2 ml, 0.405 mmol), followed by PyBOP (77 mg, 0.149 mmol) at room temperature. The mixture was stirred at room temperature overnight under argon atmosphere. Water was added to the reaction mixture and the water layer extracted with EtOAc twice. The combined organic layer was washed with brine and dried over anhydrous $MgSO_4$. After filtration and removal of the solvent under reduced pressure, the residue was purified by flash chromatography (hexane/EtOAc = 4/1 to 2/1) to afford N,N-dimethyl-2-((4-((5-methylthiazol-2-yl)carbamoyl)benzyl)amino)benzamide (**11**; 39 mg, 73%) as an amorphous solid. MS ESI m/e: 395 (M + H), 393 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.32 (s, 3H), 2.96 (s, 6H), 4.43 (d, J = 6.0 Hz, 2H), 6.04 (t, J = 6.1 Hz, 1H), 6.49 (d, J = 8.1 Hz, 1H), 6.58 (td, J = 7.4, 0.9 Hz, 1H), 6.81 (s, 1H), 7.04 (dd, J = 7.5, 1.5 Hz, 1H), 7.06–7.10 (m, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.6 Hz, 2H), 12.47 (br s, 1H); Anal. Calcd for $C_{21}H_{22}N_4O_2S$: C, 63.94; H, 5.62; N, 14.20. Found: C, 63.55; H, 5.59; N, 14.04; mp 244–246 °C.

*5.1.12. N-(4-methylthiazol-2-yl)-5-((o-tolylamino)methyl) picolinamide (**12**)*

Step 1. To a stirred mixture of 6-((4-methylthiazol-2-yl)

carbamoyl)nicotinic acid (**59a**; 200 mg, 0.76 mmol) in THF (2 ml) was added borane tetrahydrofuran complex solution (1.0 M) in THF (1.5 ml, 1.5 mmol) via syringe at room temperature. The mixture was stirred at 55 °C for 1 h under argon atmosphere. The reaction was then cooled to 0 °C, followed by the careful addition of water and then K_2CO_3 (260 mg). The reaction mixture was stirred at room temperature for 30 min and extracted with EtOAc. The organic layer was washed with brine, dried over anhydrous $MgSO_4$, filtered, and concentrated. To the residue isopropyl ether was added and the slurry filtered. The filter cake was dried in a vacuum and purified by reverse phase preparative TLC (THF/EtOH/H_2O = 1/1/1) to afford the compound 5-(hydroxymethyl)−N-(4-methylthiazol-2-yl)picolinamide (**60a**; 90 mg, 48%) as a solid. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.38 (s, 3H), 4.70 (d, J = 4.2 Hz, 2H), 5.59 (s, 1H), 7.25 (s, 1H), 8.09 (d, J = 7.9 Hz, 1H), 8.24 (d, J = 7.9 Hz, 1H), 8.76 (s, 1H), 12.03 (br s, 1H).

Step 2. To a mixture of **60a** (90 mg, 0.361 mmol) in $CHCl_3$ (2 ml) was added thionyl chloride (0.067 ml, 1.08 mmol) at room temperature. The reaction mixture was stirred at 50 °C for 10 min and then catalyst DMF added. After the reaction mixture was stirred for 1 h at 50 °C, it was cooled to room temperature. The solvent was removed under reduced pressure and used directly in the next step without further purification.

Step 3. To a solution of 5-(chloromethyl)−N-(4-methylthiazol-2-yl)picolinamide (**61a**; 0.361 mmol, crude) and 2-methylaniline (0.154 ml, 1.44 mmol) in THF (5 ml) was added KI (60 mg, 0.361 mmol). After stirring for 1 h at 60 °C under argon atmosphere, the reaction was cooled to room temperature, water poured into it, and the mixture extracted with EtOAc. The organic layer was washed with brine, dried over anhydrous $MgSO_4$, filtered, and concentrated. The residue was purified by preparative TLC ($CHCl_3$/EtOAc = 2/1), hexane/Et_2O (20/1) added, and the slurry filtered. The filter cake was dried in a vacuum to afford the title compound N-(4-methylthiazol-2-yl)-5-((o-tolylamino)methyl)picolinamide (**12**; 15 mg, 12%) as a pale yellow solid: MS ESI m/e: 339 (M + H), 337 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.18 (s, 3H), 2.30 (s, 3H), 4.52 (d, J = 4.4 Hz, 2H), 5.79 (br s, 1H), 6.39 (d, J = 8.2 Hz, 1H), 6.49 (t, J = 7.3 Hz, 1H), 6.89−6.91 (m, 2H), 6.98 (d, J = 7.3 Hz, 1H), 8.01 (dd, J = 7.9, 2.0 Hz, 1H), 8.11 (d, J = 7.9 Hz, 1H), 8.73 (s, 1H), 11.76 (s, 1H); Anal. Calcd for $C_{18}H_{18}N_4OS$: C, 63.88; H, 5.36; N, 16.55. Found: C, 63.86; H, 5.29; N, 16.48; mp 117−120 °C.

5.1.13. N-(4-methylthiazol-2-yl)-6-((o-tolylamino)methyl) nicotinamide (*13*)

This compound was prepared according to the synthetic protocols described for compound **12**, starting from 6-(chloromethyl)-3-pyridinecarboxylic acid (**59b**). MS ESI m/e: 339 (M + H), 337 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.20 (s, 3H), 2.31 (s, 3H), 4.51 (d, J = 6.0 Hz, 2H), 5.82 (t, J = 5.8 Hz, 1H), 6.30 (d, J = 7.9 Hz, 1H), 6.50 (t, J = 7.3 Hz, 1H), 6.84 (s, 1H), 6.90 (t, J = 7.6 Hz, 1H), 7.00 (d, J = 7.1 Hz, 1H), 7.46 (d, J = 8.2 Hz, 1H), 8.33 (d, J = 8.3 Hz, 1H), 9.17 (s, 1H), 12.70 (s, 1H); Anal. Calcd for $C_{18}H_{18}N_4OS$: C, 63.88; H, 5.36; N, 16.55. Found: C, 63.77; H, 5.39; N, 16.17; mp 167−168 °C.

5.1.14. N-(4-methylthiazol-2-yl)-4-(((2-(trifluoromethyl)phenyl) amino)methyl)piperidine-1-carboxamide (*14*)

Step 1. To a mixture of 2-bromobenzotrifluoride (0.41 ml, 3 mmol), *tert*-butyl 4-(aminomethyl)piperidine-1-carboxylate (**62**; 0.77 g, 3.6 mmol), sodium *tert*-butoxide (0.41 g, 4.2 mmol), and 2-dicyclohexylphosphino-2′,4′,6′-triisopropylbiphenyl (0.14 g, 0.3 mmol) in toluene (10 ml) was added tris(dibenzylideneacetone) dipalladium(0) (0.14 g, 0.15 mmol). The mixture was stirred at 80 °C for 16 h under argon atmosphere. After cooling to room temperature, the mixture was passed through a celite pad. The solvent was removed under reduced pressure and the residue purified by silica gel column chromatography (hexane/EtOAc = 100/0−4/1) to give *tert*-butyl 4-(((2-(trifluoromethyl)phenyl)amino)methyl)piperidine-1-carboxylate (**63**; 0.64 g, 59%) as a pale yellow oil. ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 1.31−1.11 (m, 3H), 1.46 (s, 9H), 1.85−1.71 (m, 3H), 2.70 (t, J = 12.4 Hz, 2H), 3.08 (t, J = 6.0 Hz, 2H), 4.14 (s, 2H), 4.39 (s, 1H), 6.74−6.67 (m, 2H), 7.36 (t, J = 7.8 Hz, 1H), 7.43 (d, J = 7.9 Hz, 1H).

Step 2. To a solution of **63** (0.64 g, 1.79 mmol) in dioxane (1 ml) and MeOH (0.5 ml) was added 4 N HCl/dioxane solution (4 ml). After stirring at room temperature for 12 h, the solvent was removed under reduced pressure. The residue was purified by trituration with ethyl acetate to give *N*-(piperidin-4-ylmethyl)-2-(trifluoromethyl)aniline hydrochloride (**64**; 0.51 g, 97%) as a solid. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 1.42−1.28 (m, 2H), 1.86−1.72 (m, 2H), 1.97−1.86 (m, 1H), 2.88−2.72 (m, 2H), 3.15−3.06 (m, 2H), 3.30−3.20 (m, 2H), 4.99 (s, 1H), 5.58 (s, 1H), 6.66 (t, J = 7.5 Hz, 1H), 6.84 (d, J = 8.6 Hz, 1H), 7.43−7.33 (m, 2H), 8.62 (s, 1H).

Step 3. To a solution of 1,1′-carbonyldiimidazole (0.50 g, 3.08 mmol) in $CHCl_3$ (4 ml) was added a solution of 2-amino-4-methylthiazole (**65**; 0.39 g, 3.42 mmol) in $CHCl_3$ (1 ml). After stirring at room temperature for 3 h, precipitated solids were collected to give *N*-(4-methylthiazol-2-yl)-1*H*-imidazole-1-carboxamide (**66**; 0.57 g, 89%). ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 13.18 (s, 1H), 8.26 (s, 1H), 7.63 (s, 1H), 7.02 (s, 1H), 6.72 (s, 1H), 2.23 (s, 3H).

Step 4. To a solution of **64** (62 mg, 0.211 mmol) and *N*-(4-methylthiazol-2-yl)-1*H*-imidazole-1-carboxamide (**66**; 40 mg, 0.192 mmol) in $CHCl_3$ (10 ml) was added TEA (53 μl, 0.384 mmol). After stirring for 24 h at room temperature, the mixture was purified by silica gel column chromatography (hexane/EtOAc = 100/0−2/3) to give *N*-(4-methylthiazol-2-yl)-4-(((2-(trifluoromethyl)phenyl)amino)methyl)piperidine-1-carboxamide (**14**; 75 mg, 99%) as an oil. MS ESI m/e: 399 (M + H), 397 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 1.01−1.14 (m, 2H), 1.69 (d, J = 11.2 Hz, 2H), 1.79−1.92 (m, 1H), 2.19 (s, 3H), 2.76 (t, J = 11.7 Hz, 2H), 3.09 (t, J = 6.4 Hz, 2H), 4.19 (d, J = 12.1 Hz, 2H), 5.45 (t, J = 5.6 Hz, 1H), 6.51 (s, 1H), 6.65 (t, J = 7.4 Hz, 1H), 6.84 (d, J = 8.6 Hz, 1H), 7.36−7.42 (m, 2H), 10.83 (br s, 1H); Anal. Calcd for $C_{18}H_{21}F_3N_4OS \cdot 0.25H_2O$: C, 53.65; H, 5.38; N, 13.90. Found: C, 53.88; H, 5.50; N, 13.63.

5.1.15. N-(4-hydroxymethyl-thiazol-2-yl)-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (*21*)

Step 1. To a solution of methyl 4-(bromomethyl)benzoate (**53**; 25 g, 0.109 mol) in DMSO (250 ml) was added 2-(trifluoromethyl) aniline (40.6 ml, 0.327 mol) and NaI (16.3 g, 0.109 mol). After stirring at 60 °C for 3 h, the mixture was cooled to room temperature. Water (1 L) and EtOAc were added to the reaction and separated. The organic layer was washed with 5% sodium thiosulfate, followed by brine, and dried over $MgSO_4$, filtered, and the solvent removed under reduced pressure. The residue was purified by flash chromatography (hexane/EtOAc = 1/1) to afford methyl 4-(((2-(trifluoromethyl)phenyl)amino)methyl)benzoate (**68**; 19.34 g, 57%). ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 3.83 (s, 3H), 4.55 (d, J = 6.0 Hz, 2H), 6.41 (t, J = 6.0 Hz, 1H), 6.56 (d, J = 8.6 Hz, 1H), 6.65 (t, J = 7.5 Hz, 1H), 7.27 (t, J = 7.7 Hz, 1H), 7.42 (d, J = 7.8 Hz, 1H), 7.47 (d, J = 8.3 Hz, 2H), 7.93 (d, J = 8.3 Hz, 2H).

Step 2. To a solution of methyl ester **68** (19.34 g, 65.5 mmol) in MeOH (48 ml) and THF (48 ml) was added 4 N LiOH (25 ml, 98 mmol). The reaction mixture was heated to reflux for 4 h. After the solvent was removed under reduced pressure and the mixture acidified with 2 N HCl, it was extracted with EtOAc. The organic layer was washed with brine and dried over $MgSO_4$. After filtration and removal of the solvent under reduced pressure, hexane and EtOAc were added to the resulting residue and the slurry filtered. The filter cake was washed with hexane and dried in a vacuum to afford 4-(((2-(trifluoromethyl)phenyl)amino)methyl)benzoic acid

(**77**; 18.94 g, 98%) as a white solid. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 4.52 (d, J = 5.8 Hz, 2H), 6.39 (t, J = 5.8 Hz, 1H), 6.56 (d, J = 8.3 Hz, 1H), 6.64 (t, J = 7.5 Hz, 1H), 7.26 (t, J = 7.7 Hz, 1H), 7.42 (t, J = 7.5 Hz, 3H), 7.89 (d, J = 8.1 Hz, 2H), 12.84 (br s, 1H).

Step 3. To a solution of **77** (200 mg, 0.68 mmol), (2-amino-1,3-thiazol-4-yl)methyl acetate hydrochloride (213 mg, 1.02 mmol), and DIPEA (0.30 ml, 1.7 mmol) in DMF (2 ml) was added PyBOP (451 mg, 1.02 mmol). The reaction mixture was heated to 65 °C for 9 h. After cooling to room temperature, water and EtOAc were added and separated. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by flash chromatography (hexane/EtOAc = 4/1) to afford (2-(4-(((2-(trifluoromethyl)phenyl)amino)methyl)benzamido)thiazol-4-yl)methyl acetate (**85**; 26.9 mg, 66%) as a solid. MS ESI m/e: 450 (M + H), 448 (M − H). ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.06 (s, 3H), 4.54 (d, J = 6.0 Hz, 2H), 5.06 (s, 2H), 6.42 (t, J = 6.0 Hz, 1H), 6.59 (d, J = 8.3 Hz, 1H), 6.65 (t, J = 7.5 Hz, 1H), 7.25 (s, 1H), 7.28 (t, J = 7.7 Hz, 1H), 7.41–7.43 (m, 1H), 7.47 (d, J = 8.1 Hz, 2H), 8.05 (d, J = 8.3 Hz, 2H), 12.66 (br s, 1H).

Step 4. K_2CO_3 (27 mg, 0.2 mmol) was added to a solution of **85** (176 mg, 0.39 mmol) in water (0.24 ml) and MeOH (2.6 ml). The reaction mixture was stirred at room temperature overnight, and then heated at 65 °C for 1 h under N_2 atmosphere. After adding water to the reaction mixture, the slurry was filtered. The filter cake was washed with water and dried in a vacuum to afford N-(4-hydroxymethyl-thiazol-2-yl)-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**21**; 55.9 mg, 35%) as a white solid. MS ESI m/e: 408 (M + H), 406 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 4.50 (d, J = 5.8 Hz, 2H), 4.54 (d, J = 6.0 Hz, 2H), 5.23 (t, J = 5.7 Hz, 1H), 6.41 (t, J = 6.0 Hz, 1H), 6.58 (d, J = 8.3 Hz, 1H), 6.65 (t, J = 7.5 Hz, 1H), 6.98 (s, 1H), 7.28 (t, J = 7.8 Hz, 1H), 7.42 (d, J = 6.5 Hz, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.53 (br s, 1H); Anal. Calcd for $C_{19}H_{16}F_3N_3O_2S$: C, 56.01; H, 3.96; N, 10.31. Found: C, 56.28; H, 3.90; N, 10.07.

5.1.16. 4-[(2-Chloro-phenylamino)-methyl]–N-(4-hydroxymethyl-thiazol-2-yl)-benzamide (**32**)

This compound was prepared according to the synthetic protocols described for compound **21**, starting from methyl 4-(bromomethyl)benzoate (**53**) and 2-chloroaniline. MS ESI m/e: 374 (M + H), 372 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 4.49–4.51 (m, 4H), 5.22 (t, J = 5.8 Hz, 1H), 6.26 (t, J = 6.3 Hz, 1H), 6.50–6.57 (m, 2H), 6.97 (s, 1H), 6.99–7.03 (m, 1H), 7.24–7.27 (m, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.04 (d, J = 8.3 Hz, 2H), 12.52 (br s, 1H); Anal. Calcd for $C_{18}H_{16}ClN_3O_2S$: C, 57.83; H, 4.31; N, 11.24. Found: C, 57.72; H, 4.11; N, 11.20.

5.1.17. 4-[(2-Butyl-phenylamino)-methyl]–N-(4-hydroxymethyl-thiazol-2-yl)-benzamide (**33**)

This compound was prepared according to the synthetic protocols described for compound **21**, starting from methyl 4-(bromomethyl)benzoate (**53**) and 2-butylaniline. MS ESI m/e: 396 (M + H), 394 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.83 (t, J = 6.8 Hz, 6H), 1.40–1.47 (m, 8H), 3.01–3.03 (m, 1H), 3.54 (s, 2H), 4.48 (d, J = 5.7 Hz, 2H), 6.08 (t, J = 6.0 Hz, 1H), 6.45 (d, J = 7.9 Hz, 1H), 6.53 (t, J = 7.5 Hz, 1H), 6.92 (s, 1H), 7.04 (t, J = 7.5 Hz, 1H), 7.27 (d, J = 7.5 Hz, 1H), 7.44 (d, J = 7.9 Hz, 2H), 8.06 (d, J = 7.9 Hz, 2H); Anal. Calcd for $C_{22}H_{25}N_3O_2S$: C, 66.81; H, 6.37; N, 10.62. Found: C, 66.74; H, 6.34; N, 10.59; mp 181–182 °C.

5.1.18. {2-[4-(o-Tolylamino-methyl)-benzoylamino]-thiazol-4-yl}-acetic acid (**17**)

Step 1. To a solution of (2-amino-4-thiazolyl)acetic acid ethyl ester (77 mg, 0.413 mmol) and 4-((o-tolylamino)methyl)benzoic acid (**76**; 100 mg, 0.413 mmol) in pyridine (1 ml) was added EDC·HCl (95 mg, 0.500 mmol) at 0 °C. The reaction mixture was stirred at room temperature overnight under N_2 atmosphere. Water and EtOAc were added to the reaction and separated. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by flash chromatography (hexane/EtOAc = 4/1) to afford **89** (71 mg, 42%) as an oil. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 1.19 (t, J = 7.2 Hz, 3H), 2.18 (s, 3H), 3.73 (s, 2H), 4.09 (q, J = 7.1 Hz, 2H), 4.43 (d, J = 6.0 Hz, 2H), 5.73 (t, J = 6.0 Hz, 1H), 6.31 (d, J = 7.9 Hz, 1H), 6.47 (t, J = 6.9 Hz, 1H), 6.87 (t, J = 7.6 Hz, 1H), 6.97 (d, J = 6.9 Hz, 1H), 7.04 (s, 1H), 7.49 (d, J = 8.1 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.58 (br s, 1H).

Step 2. To a solution of **89** (73 mg, 0.178 mmol) in EtOH (0.7 ml) was added 4 N NaOH (0.13 ml, 0.534 mmol) at 0 °C. The reaction mixture was stirred at room temperature for 30 min. After cooling to 0 °C, 10% citric acid was added portion-wise and the reaction mixture was allowed to stir at room temperature. The slurry was filtered and the filter cake washed with water and dried in a vacuum to afford {2-[4-(o-tolylamino-methyl)-benzoylamino]-thiazol-4-yl}-acetic acid (**17**; 67 mg, 99%) as a white solid. MS ESI m/e: 382 (M + H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.18 (s, 3H), 3.64 (s, 2H), 4.44 (d, J = 4.9 Hz, 2H), 5.72–5.74 (br m, 1H), 6.32 (d, J = 8.2 Hz, 1H), 6.47 (t, J = 7.3 Hz, 1H), 6.88 (t, J = 7.7 Hz, 1H), 6.97 (d, J = 7.1 Hz, 1H), 7.01 (s, 1H), 7.49 (d, J = 8.2 Hz, 2H), 8.04 (d, J = 8.4 Hz, 2H), 12.48 (br s, 1H); Anal. Calcd for $C_{20}H_{19}N_3O_3S \cdot 0.25H_2O$: C, 62.24; H, 5.09; N, 10.89. Found: C, 62.39; H, 5.05; N, 10.76.

5.1.19. 2-[4-(o-Tolylamino-methyl)-benzoylamino]-thiazole-4-carboxylic acid (**16**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **76** and ethyl 2-aminothiazole-4-carboxylate. MS ESI m/e: 368 (M + H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.19 (s, 3H), 4.45 (s, 2H), 6.35 (d, J = 7.9 Hz, 1H), 6.49 (t, J = 7.2 Hz, 1H), 6.89 (t, J = 7.7 Hz, 1H), 6.98 (d, J = 7.1 Hz, 1H), 7.51 (d, J = 8.4 Hz, 2H), 8.03 (s, 1H), 8.07 (d, J = 8.4 Hz, 2H), 12.90 (s, 1H); Anal. Calcd for $C_{19}H_{17}N_3O_3S$: C, 62.11; H, 4.66; N, 11.44. Found: C, 61.97; H, 4.76; N, 11.47.

5.1.20. (2-{4-[(2-Trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-acetic acid (**18**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **77** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 436 (M + H), 434 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 3.60 (s, 2H), 4.54 (d, J = 6.0 Hz, 2H), 6.40 (t, J = 5.7 Hz, 1H), 6.59 (d, J = 8.2 Hz, 1H), 6.65 (t, J = 7.5 Hz, 1H), 6.97 (s, 1H), 7.28 (t, J = 7.7 Hz, 1H), 7.42–7.46 (m, 3H), 8.06 (d, J = 8.2 Hz, 2H); Anal. Calcd for $C_{20}H_{16}F_3N_3O_3S$: C, 55.17; H, 3.70; N, 9.65. Found: C, 55.33; H, 3.87; N, 9.72.

5.1.21. 3-(2-{4-[(2-Trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-propionic acid (**19**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **77** and methyl 3-(2-amino-1,3-thiazol-4-yl)propanoate hydrobromide. MS ESI m/e: 450 (M + H), 448 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.61 (t, J = 7.5 Hz, 2H), 2.86 (t, J = 7.5 Hz, 2H), 4.53 (d, J = 6.0 Hz, 2H), 6.39 (t, J = 6.0 Hz, 1H), 6.58 (d, J = 8.6 Hz, 1H), 6.64 (t, J = 7.5 Hz, 1H), 6.84 (s, 1H), 7.27 (t, J = 7.8 Hz, 1H), 7.40–7.43 (m, 1H), 7.45 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.18 (br s, 1H), 12.50 (br s, 1H); Anal. Calcd for $C_{21}H_{18}F_3N_3O_3S \cdot 0.5H_2O$: C, 55.02; H, 4.18; N, 9.17. Found: C, 54.95; H, 4.15; N, 9.01.

5.1.22. 4-(2-{4-[(2-Trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-butyric acid (**20**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **77** and ethyl 4-(2-

amino-1,3-thiazol-4-yl)butanoate. MS ESI m/e: 464 (M + H), 462 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 1.83–1.90 (m, 2H), 2.24 (t, J = 7.3 Hz, 2H), 2.64 (t, J = 7.7 Hz, 2H), 4.53 (d, J = 5.8 Hz, 2H), 6.39 (t, J = 6.0 Hz, 1H), 6.58 (d, J = 8.3 Hz, 1H), 6.64 (t, J = 7.5 Hz, 1H), 6.83 (s, 1H), 7.27 (t, J = 7.7 Hz, 1H), 7.40–7.43 (m, 1H), 7.45 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.10 (s, 1H), 12.47 (s, 1H); Anal. Calcd for $C_{22}H_{20}F_3N_3O_3S$: C, 57.01; H, 4.35; N, 9.07. Found: C, 57.20; H, 4.22; N, 8.76.

5.1.23. (2-{4-[(2-Chloro-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-acetic acid (**37**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **78** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 402 (M + H), 400 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 3.64 (s, 2H), 4.49 (d, J = 6.2 Hz, 2H), 6.26 (t, J = 6.3 Hz, 1H), 6.50–6.57 (m, 2H), 6.99–7.04 (m, 2H), 7.25 (dd, J = 7.8, 1.4 Hz, 1H), 7.47 (d, J = 8.4 Hz, 2H), 8.04 (d, J = 8.4 Hz, 2H), 12.52 (br s, 1H); Anal. Calcd for $C_{19}H_{16}ClN_3O_3S$: C, 56.79; H, 4.01; N, 10.46. Found: C, 56.80; H, 4.12; N, 10.24.

5.1.24. (2-{4-[(2-Butyl-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-acetic acid (**38**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **79** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 424 (M + H), 422 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.94 (t, J = 7.4 Hz, 3H), 1.35–1.42 (m, 2H), 1.54–1.61 (m, 2H), 2.55 (t, J = 7.7 Hz, 2H), 3.58 (s, 2H), 4.43 (d, J = 5.7 Hz, 2H), 5.81 (t, J = 6.0 Hz, 1H), 6.31 (d, J = 7.9 Hz, 1H), 6.47–6.49 (m, 1H), 6.86 (t, J = 7.4 Hz, 1H), 6.93–6.95 (m, 2H), 7.46 (d, J = 8.2 Hz, 2H), 8.06 (d, J = 7.9 Hz, 2H); Anal. Calcd for $C_{23}H_{25}N_3O_3S$: C, 65.23; H, 5.95; N, 9.92. Found: C, 65.46; H, 5.86; N, 9.84.

5.1.25. (2-{4-[(2-Hexyl-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-acetic acid (**39**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **80** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 452 (M + H), 450 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.89 (t, J = 7.1 Hz, 3H), 1.34 (dt, J = 17.4, 7.0 Hz, 6H), 1.57 (dd, J = 15.0, 7.3 Hz, 2H), 2.49–2.56 (m, 2H), 4.43 (d, J = 6.0 Hz, 2H), 5.81 (t, J = 5.8 Hz, 1H), 6.31 (d, J = 7.9 Hz, 1H), 6.48 (t, J = 6.8 Hz, 1H), 6.86 (t, J = 7.8 Hz, 1H), 6.94 (d, J = 7.4 Hz, 1H), 7.01 (s, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.56 (br s, 1H); Anal. Calcd for $C_{25}H_{29}N_3O_3S$: C, 66.49; H, 6.47; N, 9.30. Found: C, 66.49; H, 6.36; N, 9.23.

5.1.26. (2-{4-[(2-Octyl-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-acetic acid (**40**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **81** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 480 (M + H), 478 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.86 (t, J = 6.8 Hz, 3H), 1.28–1.33 (m, 8H), 1.55–1.59 (m, 2H), 2.51–2.56 (m, 2H), 3.64 (s, 2H), 4.43 (d, J = 5.7 Hz, 2H), 5.79 (t, J = 6.2 Hz, 1H), 6.32 (d, J = 8.2 Hz, 1H), 6.46–6.50 (m, 1H), 6.84–6.87 (m, 1H), 6.93–6.95 (m, 1H), 7.01 (s, 1H), 7.47 (d, J = 8.4 Hz, 2H), 8.03 (d, J = 8.4 Hz, 2H); Anal. Calcd for $C_{27}H_{33}N_3O_3S$: C, 67.61; H, 6.93; N, 8.76. Found: C, 67.72; H, 6.76; N, 8.64.

5.1.27. [2-(4-{[2-(3-Methyl-butyl)-phenylamino]-methyl}-benzoylamino)-thiazol-4-yl]-acetic acid (**41**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **82** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 438 (M + H), 436 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.96 (d, J = 6.5 Hz, 6H), 1.49 (dd, J = 15.2, 7.3 Hz, 2H), 1.58–1.68 (m, 1H), 2.49–2.57 (m, 2H), 3.61 (s, 2H), 4.43 (d, J = 5.3 Hz, 2H), 5.77 (t, J = 5.8 Hz, 1H), 6.32 (d, J = 7.7 Hz, 1H), 6.49 (t, J = 7.5 Hz, 1H), 6.84–6.88 (m, 1H), 6.94 (dd, J = 7.2, 1.4 Hz, 1H), 6.98 (s, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.04 (d, J = 8.3 Hz, 2H); Anal. Calcd for $C_{24}H_{27}N_3O_3S$: C, 65.88; H, 6.22; N, 9.60. Found: C, 65.58; H, 6.19; N, 9.40.

5.1.28. {2-[4-(Biphenyl-2-ylaminomethyl)-benzoylamino]-thiazol-4-yl}-acetic acid (**42**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **83** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 444 (M + H), 442 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 3.63 (s, 2H), 4.39 (d, J = 5.8 Hz, 2H), 5.44 (t, J = 5.9 Hz, 1H), 6.46 (d, J = 8.3 Hz, 1H), 6.64 (t, J = 7.3 Hz, 1H), 6.98–7.00 (m, 2H), 7.03–7.07 (m, 1H), 7.36–7.41 (m, 1H), 7.46–7.52 (m, 6H), 8.04 (d, J = 8.3 Hz, 2H), 12.49 (br s, 1H); Anal. Calcd for $C_{25}H_{21}N_3O_3S$: C, 67.70; H, 4.77; N, 9.47. Found: C, 67.56; H, 4.80; N, 9.36.

5.1.29. (2-{4-[(2-Benzyl-phenylamino)-methyl]-benzoylamino}-thiazol-4-yl)-acetic acid (**43**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **84** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 458 (M + H), 456 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 3.64 (s, 2H), 3.94 (s, 2H), 4.41 (d, J = 5.7 Hz, 2H), 5.76 (t, J = 6.0 Hz, 1H), 6.38 (d, J = 7.9 Hz, 1H), 6.51 (t, J = 7.2 Hz, 1H), 6.86–6.96 (m, 2H), 7.01 (s, 1H), 7.20–7.36 (m, 7H), 7.99 (d, J = 8.3 Hz, 2H), 12.47 (br s, 2H); Anal. Calcd for $C_{26}H_{23}N_3O_3S \cdot 0.25H_2O$: C, 67.59; H, 5.13; N, 9.09. Found: C, 67.77; H, 5.02; N, 9.13.

5.1.30. 4-Hydroxymethyl-2-{4-[(2-trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazole-5-carboxylic acid (**30**)

This compound was prepared according to the synthetic protocols described for compound **17**, starting from **77** and methyl 2-amino-4-(hydroxymethyl)-1,3-thiazole-5-carboxylate. MS ESI m/e: 452 (M + H), 450 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 4.53 (d, J = 6.2 Hz, 2H), 4.57 (s, 2H), 6.38 (t, J = 6.1 Hz, 1H), 6.59 (d, J = 8.6 Hz, 1H), 6.65 (t, J = 7.7 Hz, 1H), 7.27 (t, J = 7.8 Hz, 1H), 7.39–7.48 (m, 3H), 8.02 (d, J = 8.4 Hz, 2H); Anal. Calcd for $C_{20}H_{16}F_3N_3O_4S \cdot 0.25H_2O$: C, 52.69; H, 3.65; N, 9.22. Found: C, 52.67; H, 3.66; N, 8.89.

5.1.31. N-[4-(2-hydroxy-ethyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**22**)

To a solution of ethyl 2-(2-(4-(((2-(trifluoromethyl)phenyl) amino)methyl)benzamido)thiazol-4-yl)acetate (**90**; 100 mg, 0.222 mmol) in *t*-BuOH (0.9 ml) and THF (0.9 ml) was added sodium borohydride (21 mg, 0.555 mmol) under argon atmosphere, followed by MeOH (0.2 ml) drop-wise while heating to reflux. The reaction mixture was heated to reflux for 4 h. After cooling to room temperature, the reaction mixture was quenched with water. The water layer was extracted with EtOAc and separated. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and the solvent removed under reduced pressure. After adding hexane/EtOAc (1/1) to the residue, the slurry was filtered. The filter cake was washed with hexane and dried in a vacuum to afford N-[4-(2-hydroxy-ethyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**22**; 4.1 mg, 4%) as a white solid. MS ESI m/e: 422 (M + H), 420 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.79 (t, J = 7.1 Hz, 2H), 3.70 (dd, J = 12.3, 7.0 Hz, 2H), 4.54 (d, J = 6.0 Hz, 2H), 4.65 (t, J = 5.3 Hz, 1H), 6.41 (t, J = 6.1 Hz, 1H), 6.59 (d, J = 8.6 Hz, 1H), 6.65 (t, J = 7.5 Hz, 1H), 6.86 (s, 1H), 7.28 (t, J = 7.7 Hz, 1H), 7.42 (d, J = 7.7 Hz, 1H), 7.46 (d, J = 8.1 Hz, 2H), 8.03 (d, J = 8.1 Hz, 2H), 12.50 (br s, 1H); Anal. Calcd for $C_{20}H_{18}F_3N_3O_2S$: C, 57.00; H, 4.31; N, 9.97. Found: C, 57.04; H, 4.40; N, 9.69.

*5.1.32. N-[4-(3-hydroxy-propyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**23**)*

This compound was prepared according to the synthetic protocols described for compound **22**, starting from **91**. MS ESI m/e: 436 (M + H), 434 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 1.75–1.82 (m, 2H), 2.66 (t, J = 7.5 Hz, 2H), 3.44 (dd, J = 11.7, 6.4 Hz, 2H), 4.47 (t, J = 5.2 Hz, 1H), 4.54 (d, J = 6.3 Hz, 2H), 6.41 (t, J = 6.0 Hz, 1H), 6.59 (d, J = 8.3 Hz, 1H), 6.65 (t, J = 7.4 Hz, 1H), 6.82 (s, 1H), 7.28 (t, J = 7.2 Hz, 1H), 7.42 (d, J = 6.7 Hz, 1H), 7.46 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.50 (br s, 1H); Anal. Calcd for $C_{21}H_{20}F_3N_3O_2S \cdot 0.25H_2O$: C, 57.33; H, 4.70; N, 9.55. Found: C, 57.29; H, 4.66; N, 9.54.

*5.1.33. 4-[(2-Chloro-phenylamino)-methyl]–N-[4-(2-hydroxy-ethyl)-thiazol-2-yl]-benzamide (**34**)*

This compound was prepared according to the synthetic protocols described for compound **22**, starting from **93**. MS ESI m/e: 388 (M + H), 386 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.79 (t, J = 7.1 Hz, 2H), 3.68–3.72 (br m, 2H), 4.49 (d, J = 6.2 Hz, 2H), 4.63 (br s, 1H), 6.24 (t, J = 6.2 Hz, 1H), 6.50–6.57 (m, 2H), 6.84 (s, 1H), 6.99–7.04 (m, 1H), 7.24–7.27 (m, 1H), 7.47 (d, J = 8.4 Hz, 2H), 8.04 (d, J = 8.2 Hz, 2H), 12.48 (br s, 1H); Anal. Calcd for $C_{19}H_{18}ClN_3O_2S$: C, 58.83; H, 4.68; N, 10.83. Found: C, 58.47; H, 4.79; N, 10.45.

*5.1.34. 4-[(2-Butyl-phenylamino)-methyl]–N-[4-(2-hydroxy-ethyl)-thiazol-2-yl]-benzamide (**35**)*

This compound was prepared according to the synthetic protocols described for compound **22**, starting from **94**. MS ESI m/e: 410 (M + H), 408 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.94 (t, J = 7.3 Hz, 3H), 1.35–1.44 (m, 2H), 1.54–1.61 (m, 2H), 2.53–2.57 (m, 2H), 2.79 (t, J = 7.0 Hz, 2H), 3.67–3.72 (m, 2H), 4.43 (d, J = 5.6 Hz, 2H), 4.62–4.65 (m, 1H), 5.80–5.83 (m, 1H), 6.31 (d, J = 7.9 Hz, 1H), 6.48 (t, J = 7.0 Hz, 1H), 6.84–6.88 (m, 2H), 6.93–6.95 (m, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.03 (d, J = 8.3 Hz, 2H), 12.47 (br s, 1H); Anal. Calcd for $C_{23}H_{27}N_3O_2S$: C, 67.45; H, 6.65; N, 10.26. Found: C, 67.40; H, 6.50; N, 10.24.

*5.1.35. N-[4-(2-hydroxy-ethyl)-thiazol-2-yl]-4-{[2-(3-methyl-butyl)-phenylamino]-methyl}-benzamide (**36**)*

This compound was prepared according to the synthetic protocols described for compound **22**, starting from **95**. MS ESI m/e: 424 (M + H), 422 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.96 (d, J = 6.7 Hz, 6H), 1.49 (dd, J = 15.5, 7.0 Hz, 2H), 1.60–1.67 (m, 1H), 2.54 (t, J = 8.2 Hz, 2H), 2.79 (t, J = 7.0 Hz, 2H), 3.70 (q, J = 6.5 Hz, 2H), 4.43 (d, J = 5.3 Hz, 2H), 4.64 (t, J = 5.3 Hz, 1H), 5.77 (t, J = 5.9 Hz, 1H), 6.32 (d, J = 7.9 Hz, 1H), 6.49 (t, J = 7.3 Hz, 1H), 6.86 (t, J = 7.5 Hz, 2H), 6.95 (d, J = 7.2 Hz, 1H), 7.48 (d, J = 8.1 Hz, 2H), 8.03 (d, J = 8.1 Hz, 2H), 12.48 (br s, 1H); Anal. Calcd for $C_{24}H_{29}N_3O_2S \cdot 0.25H_2O$: C, 67.34; H, 6.95; N, 9.82. Found: C, 67.35; H, 6.83; N, 9.71.

*5.1.36. (2-{4-[(2-Trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazol-5-yl)-acetic acid (**25**)*

Step 1. To a solution of **77** (80 mg, 0.271 mmol), methyl 2-(2-aminothiazol-5-yl)acetate hydrobromide (82 mg, 0.324 mmol) and DIPEA (0.123 ml, 0.706 mmol) in DMF (0.8 ml) was added PyBOP (169 mg, 0.325 mmol). The reaction mixture was stirred at 65 °C for 2 h. After cooling to room temperature, water and EtOAc were added and separated. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated. The residue was purified by preparative TLC ($CHCl_3$/MeOH = 12/1) to afford methyl 2-(2-(4-(((2-(trifluoromethyl)phenyl)amino)methyl)benzamido)thiazol-5-yl)acetate (**101**; 98 mg, 80%) as a solid. ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 3.66 (s, 3H), 3.93 (s, 2H), 4.54 (d, J = 6.0 Hz, 2H), 6.40 (t, J = 5.8 Hz, 1H), 6.58–6.67 (m, 2H), 7.27 (t, J = 7.5 Hz, 1H), 7.35 (s, 1H), 7.45 (dd, J = 14.7, 7.9 Hz, 3H), 8.02 (d, J = 7.9 Hz, 2H), 12.45 (br s, 1H).

Step 2. To a solution of **101** (70 mg, 0.156 mmol) in MeOH (0.7 ml) was added 2 N NaOH (0.234 ml, 0.467 mmol) at 0 °C and stirred at room temperature for 30 min 2 N HCl (0.234 ml) was added portion-wise. After adding water, the reaction mixture was allowed to stir at room temperature. The slurry was filtered and the filter cake washed by water and dried in a vacuum to afford (2-{4-[(2-trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazol-5-yl)-acetic acid (**25**; 63 mg, 93%) as a solid. MS ESI m/e: 436 (M + H), 434 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 3.78 (s, 2H), 4.54 (d, J = 6.0 Hz, 2H), 6.39 (t, J = 6.0 Hz, 1H), 6.59 (d, J = 8.7 Hz, 1H), 6.65 (t, J = 7.5 Hz, 1H), 7.26 (t, J = 7.9 Hz, 1H), 7.31 (s, 1H), 7.42 (d, J = 7.9 Hz, 1H), 7.47 (d, J = 8.3 Hz, 2H), 8.02 (d, J = 8.3 Hz, 2H); Anal. Calcd for $C_{20}H_{16}F_3N_3O_3S$: C, 55.17; H, 3.70; N, 9.65. Found: C, 54.86; H, 3.79; N, 9.49.

*5.1.37. 2-{4-[(2-Trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazole-5-carboxylic acid (**24**)*

This compound was prepared according to the synthetic protocols described for compound **25**, starting from **77** and ethyl 2-aminothiazole-5-carboxylate. MS ESI m/e: 422 (M + H), 420 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 4.52 (d, J = 5.6 Hz, 2H), 6.37 (t, J = 7.5 Hz, 1H), 6.58–6.68 (m, 2H), 7.28 (t, J = 12.1 Hz, 2H), 7.42–7.40 (m, 3H), 7.48 (s, 1H), 8.02 (d, J = 8.4 Hz, 2H); Anal. Calcd for $C_{19}H_{14}F_3N_3O_3S \cdot 0.5H_2O$: C, 53.58; H, 3.43; N, 9.87. Found: C, 53.20; H, 3.71; N, 9.55.

*5.1.38. 3-(2-{4-[(2-Trifluoromethyl-phenylamino)-methyl]-benzoylamino}-thiazol-5-yl)-propionic acid (**26**)*

This compound was prepared according to the synthetic protocols described for compound **25**, starting from **77** and methyl 3-(2-aminothiazol-5-yl)propanoate. MS ESI m/e: 450 (M + H), 448 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.57 (t, J = 7.2 Hz, 2H), 2.98 (t, J = 7.2 Hz, 2H), 4.54 (d, J = 5.7 Hz, 2H), 6.39 (t, J = 5.8 Hz, 1H), 6.55–6.70 (m, 2H), 7.23–7.32 (m, 2H), 7.38–7.51 (m, 3H), 8.01 (d, J = 7.9 Hz, 2H), 12.32 (s, 2H); Anal. Calcd for $C_{21}H_{18}F_3N_3O_3S$: C, 56.12; H, 4.04; N, 9.35. Found: C, 56.19; H, 3.84; N, 9.36.

*5.1.39. N-[5-(2-hydroxy-ethyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**28**)*

To a solution of **77** (80 mg, 0.271 mmol), 2-(2-aminothiazol-5-yl)ethan-1-ol (43 mg, 0.298 mmol), and DIPEA (0.057 ml, 0.325 mmol) in DMF (0.8 ml) was added PyBOP (155 mg, 0.298 mmol). The reaction mixture was stirred at room temperature overnight. Water and EtOAc were added to the reaction and separated. The organic layer was washed with saturated $NaHCO_3$, followed by brine, and dried over Na_2SO_4, filtered, and concentrated. The residue was purified by preparative TLC ($CHCl_3$/MeOH = 10/1) to afford N-[5-(2-hydroxy-ethyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**28**; 105 mg, 46%) as a solid. MS ESI m/e: 422 (M + H), 420 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.88 (t, J = 6.4 Hz, 2H), 3.60 (q, J = 6.0 Hz, 2H), 4.54 (d, J = 6.0 Hz, 2H), 4.85 (t, J = 5.3 Hz, 1H), 6.39 (t, J = 6.0 Hz, 1H), 6.55–6.69 (m, 2H), 7.23–7.31 (m, 2H), 7.38–7.50 (m, 3H), 8.02 (d, J = 8.3 Hz, 2H), 12.33 (s, 1H); Anal. Calcd for $C_{20}H_{18}F_3N_3O_2S$: C, 57.00; H, 4.31; N, 9.97. Found: C, 56.72; H, 4.22; N, 9.75.

*5.1.40. N-(5-hydroxymethyl-thiazol-2-yl)-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**27**)*

This compound was prepared according to the synthetic protocols described for compound **28**, starting from **77** and (2-aminothiazol-5-yl)methanol. MS ESI m/e: 408 (M + H), 406 (M − H). ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 4.54 (d, J = 6.0 Hz, 2H), 4.61 (d, J = 5.6 Hz, 2H), 5.39 (t, J = 5.7 Hz, 1H), 6.41 (t, J = 6.0 Hz, 1H),

6.58 (d, $J = 8.3$ Hz, 1H), 6.65 (t, $J = 7.5$ Hz, 1H), 7.27 (t, $J = 7.9$ Hz, 1H), 7.36 (s, 1H), 7.42 (d, $J = 6.7$ Hz, 1H), 7.47 (d, $J = 8.3$ Hz, 2H), 8.03 (d, $J = 8.3$ Hz, 2H), 12.41 (br s, 1H); Anal. Calcd for $C_{19}H_{16}F_3N_3O_2S$: C, 56.01; H, 3.96; N, 10.31. Found: C, 56.01; H, 4.03; N, 10.13.

5.1.41. N-[5-(3-hydroxy-propyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**29**)

This compound was prepared according to the synthetic protocols described for compound **28**, starting from **77** and 3-(2-aminothiazol-5-yl)propan-1-ol. MS ESI m/e: 436 (M + H), 434 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 1.68–1.81 (m, 2H), 2.79 (t, $J = 7.5$ Hz, 2H), 3.45 (q, $J = 5.9$ Hz, 2H), 4.48–4.58 (m, 3H), 6.39 (t, $J = 6.2$ Hz, 1H), 6.55–6.69 (m, 2H), 7.23 (s, 1H), 7.27 (t, $J = 7.7$ Hz, 1H), 7.42 (d, $J = 7.9$ Hz, 1H), 7.46 (d, $J = 8.3$ Hz, 2H), 8.01 (d, $J = 7.9$ Hz, 2H), 12.33 (s, 1H); Anal. Calcd for $C_{21}H_{20}F_3N_3O_2S \cdot 0.25H_2O$: C, 57.33; H, 4.70; N, 9.55. Found: C, 57.14; H, 4.70; N, 9.41.

5.1.42. N-[5-(2-hydroxy-ethyl)-4-(3-hydroxy-propyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**31**)

Step 1. A solution of **104** (5 ml, 23.5 mmol) in $CHCl_3$ (32 ml) was cooled to 0 °C. A solution of Br_2 (1.2 ml, 23.5 mmol) in $CHCl_3$ (5 ml) was added drop-wise over 2 min and the reaction mixture allowed to stir at room temperature for 30 min $CHCl_3$ and 10% $Na_2S_2O_3$ were added and separated. The organic layer was washed with 10% $Na_2S_2O_3$, followed by brine, and dried over $MgSO_4$, filtered, and concentrated to afford crude diethyl 3-bromo-4-oxoheptanedioate (**105**) as a pale yellow oil, which was used directly in the next step without further purification.

Step 2. To a solution of **105** (23.5 mmol theoretical yield) in EtOH (70 ml) was added thiourea (1.97 g, 25.9 mmol) and the reaction mixture heated to 100 °C for 30 min under argon atmosphere. After cooling to room temperature, saturated $NaHCO_3$ and EtOAc were added and separated. The combined organic layer was washed with brine, dried over Na_2SO_4, filtered, and the solvent removed under reduced pressure. The residue was purified by flash chromatography (hexane/EtOAc = 5/3) to afford ethyl 3-(2-amino-5-(2-ethoxy-2-oxoethyl)thiazol-4-yl)propanoate (**106**; 4.69 g, 70%) as a pale yellow oil. ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 1.22–1.29 (m, 6H), 2.63 (t, $J = 6.1$ Hz, 2H), 2.77 (t, $J = 7.3$ Hz, 2H), 3.63 (s, 2H), 4.08–4.19 (m, 4H), 4.85 (s, 2H).

Step 3. To a solution of **106** (516 mg, 1.8 mmol), 4-(((2-(trifluoromethyl)phenyl)amino)methyl)benzoic acid (**77**; 532 mg, 1.8 mmol), and DIPEA (0.32 ml, 1.8 mmol) in DMF (5 ml) was added PyBOP (937 mg, 1.8 mmol). The reaction mixture was heated to 65 °C overnight. After cooling to room temperature, water and EtOAc were added and separated. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated. After adding EtOAc and IPE, the slurry was filtered and the filter cake washed with IPE and dried in a vacuum to afford ethyl 3-(5-(2-ethoxy-2-oxoethyl)-2-(4-(((2-(trifluoromethyl)phenyl)amino)methyl)benzamido)thiazol-4-yl)propanoate (**107**; 897 mg, 88%) as a white solid. ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 1.21–1.30 (m, 6H), 2.68 (t, $J = 7.4$ Hz, 2H), 2.91 (t, $J = 7.6$ Hz, 2H), 3.76 (s, 2H), 4.12 (q, $J = 7.1$ Hz, 2H), 4.19 (q, $J = 7.1$ Hz, 2H), 4.54 (d, $J = 5.5$ Hz, 2H), 4.92 (br s, 1H), 6.59 (d, $J = 8.4$ Hz, 1H), 6.75 (t, $J = 7.6$ Hz, 1H), 7.26–7.31 (m, 1H), 7.47–7.51 (m, 3H), 7.93 (t, $J = 4.2$ Hz, 2H).

Step 4. To a solution of **107** (123 mg, 0.219 mmol) in THF (1.2 ml) was added $LiBH_4$ (16 mg, 0.735 mmol). The reaction mixture was stirred at room temperature for 4 h under argon atmosphere. Additional $LiBH_4$ (17 mg, 0.781 mmol) was added and heated to 65 °C for 1 h. After cooling to room temperature, dilute HCl was added and the reaction stirred at room temperature overnight. EtOAc and water were added and separated. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated. After adding EtOAc and hexane, the slurry was filtered and the filter cake washed with EtOAc and hexane before drying in a vacuum to afford N-[5-(2-hydroxy-ethyl)-4-(3-hydroxy-propyl)-thiazol-2-yl]-4-[(2-trifluoromethyl-phenylamino)-methyl]-benzamide (**31**; 26 mg, 25%) as a white solid. MS ESI m/e: 480 (M + H), 478 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 1.69–1.76 (m, 2H), 2.59 (t, $J = 7.4$ Hz, 2H), 2.81 (t, $J = 6.6$ Hz, 2H), 3.37–3.42 (m, 2H), 3.55 (q, $J = 6.3$ Hz, 2H), 4.44 (t, $J = 5.2$ Hz, 1H), 4.53 (d, $J = 5.8$ Hz, 2H), 4.83 (t, $J = 5.3$ Hz, 1H), 6.39 (t, $J = 5.9$ Hz, 1H), 6.59 (d, $J = 8.6$ Hz, 1H), 6.65 (t, $J = 7.5$ Hz, 1H), 7.27 (t, $J = 7.8$ Hz, 1H), 7.41–7.46 (m, 3H), 8.02 (d, $J = 8.1$ Hz, 2H), 12.31 (br s, 1H); Anal. Calcd for $C_{23}H_{24}F_3N_3O_3S \cdot H_2O$: C, 55.52; H, 5.27; N, 8.45. Found: C, 55.76; H, 5.03; N, 8.13.

5.1.43. N-(4-methylthiazol-2-yl)-5-(((2-(trifluoromethyl)phenyl)amino)methyl)thiophene-2-carboxamide (**15**)

Step 1. A solution of commercially available 5-formylthiophene-2-carboxylic acid (**108**; 1 g, 6.40 mmol) and 2-(trifluoromethyl)aniline (0.8 ml, 6.40 mmol) in AcOH (5 ml) and DME (5 ml) was stirred at room temperature for 1 h under argon atmosphere. The reaction mixture was cooled to 0 °C before adding sodium borohydride (315 mg, 8.32 mmol) portion-wise over 10 min. The reaction mixture was then allowed to stir at room temperature overnight. After cooling to 0 °C, water was added drop-wise over 10 min and the reaction mixture allowed to stir at room temperature for 30 min. The slurry was filtered and the filter cake washed with water and dried in a vacuum to afford 5-(((2-(trifluoromethyl)phenyl)amino)methyl)thiophene-2-carboxylic acid (**115**; 1.58 g, 82%) as a solid. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 4.66 (d, $J = 5.8$ Hz, 2H), 6.39 (t, $J = 6.0$ Hz, 1H), 6.70 (t, $J = 7.5$ Hz, 1H), 6.77 (d, $J = 8.6$ Hz, 1H), 7.09 (d, $J = 3.7$ Hz, 1H), 7.34 (t, $J = 7.6$ Hz, 1H), 7.43 (dd, $J = 7.9$, 1.2 Hz, 1H), 7.57 (d, $J = 3.7$ Hz, 1H), 12.89 (br s, 1H).

Step 2. To a solution of **115** (50 mg, 0.166 mmol) and 2-amino-4-methylthiazole (**65**; 19 mg, 0.166 mmol) in pyridine (1 ml) was added EDC·HCl (38 mg, 0.200 mmol) at room temperature. After stirring at room temperature overnight, water was poured into the reaction and the mixture extracted with EtOAc. The organic layer was washed with brine and dried over anhydrous $MgSO_4$. After filtration and concentration, the residue was purified by preparative TLC (hexane/EtOAc = 1/1) to afford the title compound N-(4-methylthiazol-2-yl)-5-(((2-(trifluoromethyl)phenyl)amino)methyl)thiophene-2-carboxamide (**15**; 59 mg, 90% yield). MS ESI m/e: 398 (M + H), 396 (M − H); ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 2.28 (s, 3H), 4.67 (d, $J = 6.0$ Hz, 2H), 6.42 (t, $J = 6.0$ Hz, 1H), 6.70 (t, $J = 7.5$ Hz, 1H), 6.78 (d, $J = 8.3$ Hz, 2H), 7.13 (d, $J = 3.7$ Hz, 1H), 7.34 (t, $J = 7.7$ Hz, 1H), 7.43 (t, $J = 3.9$ Hz, 1H), 8.04 (br s, 1H), 12.54 (br s, 1H); Anal. Calcd for $C_{17}H_{14}F_3N_3OS_2$: C, 51.38; H, 3.55; N, 10.57. Found: C, 51.27; H, 3.64; N, 10.24.

5.1.44. Sodium 2-(2-(5-(((2-butylphenyl)amino)methyl)thiophene-2-carboxamido)thiazol-4-yl)acetate (**48**)

Step 1. A solution of 2-butylbenzenamine (101 ml, 640.4 mmol) in toluene (100 ml) was added drop-wise to a mixture of 5-formylthiophene-2-carboxylic acid (**108**; 100 g, 640.4 mmol) in toluene (600 ml) and the resulting mixture heated to reflux for 3 h. After cooling to 90 °C, *n*-heptane (500 ml) was added and the resulting solution seeded with small crystals of the desired product at 68 °C. After cooling to room temperature, the precipitated solid was filtered and washed with *n*-heptane (1 L). The filter cake was dried in a vacuum to afford (E)-5-(((2-butylphenyl)imino)methyl)thiophene-2-carboxylic acid (**110**; 171.5 g, 93%) as a pale yellow solid. ^{1}H NMR ($CDCl_3$, 400 MHz) δ: 0.93 (t, $J = 7.3$ Hz, 3H), 1.33–1.42 (m, 2H), 1.58 (tt, $J = 8.6$, 3.6 Hz, 2H), 2.80 (t, $J = 7.8$ Hz, 2H), 6.99–7.02 (m, 1H), 7.18–7.27 (m, 3H), 7.47 (d, $J = 3.9$ Hz, 1H), 7.90 (d, $J = 3.9$ Hz, 1H), 8.53 (s, 1H).

Step 2. A solution of **110** (171.3 g, 596.1 mmol) in DME (580 ml)

was cooled to 0 °C, and sodium borohydride (16.9 g, 447.1 mmol) was added portion-wise over 30 min with DME (105 ml). The reaction mixture was allowed to stir at room temperature for 2 h. After cooling to 0 °C, AcOH (136.5 ml, 2.384 mol) was added drop-wise over 20 min, and the reaction mixture was allowed to stir for 40 min. Subsequently, water (260 ml) was added drop-wise over 15 min and the reaction mixture allowed to stir at room temperature for 15 min. Next, MeOH (377 ml) was added drop-wise over 10 min and the reaction mixture allowed to stir at room temperature for 5 min, followed by the drop-wise addition of water (500 ml) over 20 min. The resulting solution was seeded with small crystals of the desired product at room temperature, and the mixture was allowed to stir at room temperature for 20 min. Additional water (350 ml) was added drop-wise over 45 min, and the mixture was allowed to stir at room temperature for 30 min. After 690 ml of water was added drop-wise over 60 min, the mixture was allowed to stir at room temperature overnight. The precipitated solid was filtered and washed with water (1.8 L). The filter cake was dried in a vacuum to give 5-(((2-butylphenyl)amino) methyl)thiophene-2-carboxylic acid (**116**; 166.1 g, 96%) as a yellow solid. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.92 (t, J = 7.3 Hz, 3H), 1.32–1.41 (m, 2H), 1.50–1.58 (m, 2H), 2.48–2.52 (m, 2H), 4.54 (s, 2H), 5.83 (br s, 1H), 6.47–6.54 (m, 2H), 6.90–6.95 (m, 2H), 7.08 (d, J = 3.7 Hz, 1H), 7.57 (d, J = 3.5 Hz, 1H), 12.83 (br s, 1H).

Step 3. To a solution of ethyl (2-amino-4-thiazolyl)acetate (106.2 g, 570.1 mmol) and **116** (150.0 g, 518.3 mmol) in $CHCl_3$ (1500 ml) was added EDC·HCl (148.6 g, 777.5 mmol), followed by DMAP (31.7 g, 259.2 mmol) with $CHCl_3$ (250 ml) at 0 °C. The reaction mixture was stirred for 30 min under N_2 atmosphere at 0 °C, and then at room temperature for 4 h. The solvent was removed under reduced pressure. To the resulting residue was added 750 ml of EtOAc and the solvent removed under reduced pressure. EtOAc (750 ml) and additional water (300 ml) were then added and the solvent removed under reduced pressure. EtOAc (900 ml), water (300 ml), and 5% $KHSO_4$ (600 ml) were added to the resultant residue and the layers separated. After additional *n*-hexane (600 ml) was added, the organic portion was washed with 5% $KHSO_4$, brine, 5% $NaHCO_3$, and brine, and dried over $MgSO_4$. Silica gel (45 g) and activated charcoal (4 g) were added and the mixture stirred at room temperature for 1 h. The mixture was filtered through a celite pad and the filtrate concentrated to about half its initial volume. After adding EtOAc (59 ml) and EtOH (75 ml), the mixture was warmed to 80 °C and IPA (2100 ml) added carefully. The resulting solution was cooled to 45 °C and small crystals of the desired product seeded before stirring at room temperature overnight. The precipitated solid was filtered and washed with IPE (300 ml), followed by IPE/EtOH (50/1; 765 ml). The filter cake was dried in a vacuum to afford ethyl 2-(2-(5-(((2-butylphenyl)amino)methyl)thiophene-2-carboxamido)thiazol-4-yl)acetate (**125**; 182.1 g, 77%) as a pale yellow solid. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.93 (t, J = 7.3 Hz, 3H), 1.18 (t, J = 7.1 Hz, 3H), 1.33–1.42 (m, 2H), 1.51–1.59 (m, 2H), 2.49–2.53 (m, 2H), 3.71 (s, 2H), 4.08 (q, J = 7.1 Hz, 2H), 4.55 (d, J = 6.0 Hz, 2H), 5.86 (t, J = 5.8 Hz, 1H), 6.48–6.55 (m, 2H), 6.90–6.96 (m, 2H), 7.01 (s, 1H), 7.12 (d, J = 3.5 Hz, 1H), 8.07 (s, 1H), 12.62 (s, 1H).

Step 4. To a solution of **125** (35 g, 76.5 mmol) in THF (175 ml) and MeOH (350 ml) was added 2 N NaOH (115 ml, 229 mmol) at 0 °C. The reaction mixture was stirred at room temperature for 3 h. After cooling to 0 °C, water (700 ml), followed by 2 N HCl (115 ml), was added portion-wise and the reaction mixture allowed to stir at room temperature. After adding water (700 ml), the slurry was filtered and the filter cake washed with water (4 × 300 ml) and dried in a vacuum to afford 2-(2-(5-(((2-butylphenyl)amino) methyl)thiophene-2-carboxamido)thiazol-4-yl)acetic acid (**130**; 32.3 g, 98%) as a white solid. ^{1}H NMR (DMSO-d_6, 400 MHz) δ: 0.93 (t, J = 7.4 Hz, 3H), 1.33–1.42 (m, 2H), 1.51–1.59 (m, 2H), 2.48–2.54 (m, 2H), 3.62 (s, 2H), 4.55 (d, J = 5.7 Hz, 2H), 5.86 (t, J = 6.1 Hz, 1H), 6.48–6.55 (m, 2H), 6.90–6.98 (m, 3H), 7.12 (d, J = 4.0 Hz, 1H), 8.07 (d, J = 4.0 Hz, 1H), 12.54 (br s, 1H).

Step 5. A solution of 4 N NaOH (6.1 ml, 24.50 mmol) was added drop-wise to a stirred suspension of **130** (10 g, 23.30 mmol) in EtOH (350 ml) and water (150 ml) over 30 min while heating to 80 °C, and the reaction mixture was allowed to stir at room temperature overnight. The slurry was filtered and the filter cake washed with EtOH (1 L) and dried in a vacuum to afford sodium 2-(2-(5-(((2-butylphenyl)amino)methyl)thiophene-2-carboxamido)thiazol-4-yl)acetate (**48**; 8.18 g, 78%) as a white solid. MS ESI m/e: 452 (M + H), 450 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 0.93 (t, J = 7.3 Hz, 3H), 1.30–1.45 (m, 2H), 1.48–1.61 (m, 2H), 2.47–2.55 (m, 2H), 3.36 (s, 2H), 4.53 (d, J = 5.7 Hz, 2H), 5.83 (t, J = 5.7 Hz, 1H), 6.46–6.56 (m, 2H), 6.72 (s, 1H), 6.87–6.98 (m, 2H), 7.03 (d, J = 3.8 Hz, 1H), 8.59 (s, 1H); Anal. Calcd for $C_{21}H_{22}N_3O_3S_2$·Na: C, 55.86; H, 4.91; N, 9.31. Found: C, 55.79; H, 4.83; N, 9.24; Purity by HPLC 99.9%; mp 243–249 °C.

5.1.45. [2-({5-[(2-Trifluoromethyl-phenylamino)-methyl]-thiophene-2-carbonyl}-amino)-thiazol-4-yl]-acetic acid (*47*)

This compound was prepared according to the synthetic protocols described for compound **48**, starting from **115** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 442 (M + H), 440 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 3.62 (s, 2H), 4.67 (d, J = 6.0 Hz, 2H), 6.40 (t, J = 6.0 Hz, 1H), 6.70 (t, J = 7.3 Hz, 1H), 6.78 (d, J = 8.7 Hz, 1H), 6.98 (s, 1H), 7.13 (d, J = 3.8 Hz, 1H), 7.34 (t, J = 7.7 Hz, 1H), 7.43 (d, J = 7.9 Hz, 1H), 8.06 (d, J = 4.1 Hz, 1H), 12.48 (br s, 2H); Anal. Calcd for $C_{18}H_{14}F_3N_3O_3S_2$: C, 48.97; H, 3.20; N, 9.52. Found: C, 49.23; H, 3.49; N, 9.45.

5.1.46. Sodium [2-({5-[(2-hexyl-phenylamino)-methyl]-thiophene-2-carbonyl}-amino)-thiazol-4-yl]-acetate (*49*)

This compound was prepared according to the synthetic protocols described for compound **48**, starting from **117** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 480 (M + H), 456 (M − Na); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 0.88 (t, J = 7.0 Hz, 3H), 1.23–1.42 (m, 6H), 1.49–1.62 (m, 2H), 2.46–2.54 (m, 2H), 3.34 (s, 2H), 4.53 (d, J = 5.7 Hz, 2H), 5.80 (t, J = 5.7 Hz, 1H), 6.47–6.56 (m, 2H), 6.68 (s, 1H), 6.88–6.97 (m, 2H), 7.02 (d, J = 3.8 Hz, 1H), 8.41 (s, 1H); Anal. Calcd for $C_{23}H_{26}N_3O_3S_2$·Na: C, 57.60; H, 5.46; N, 8.76. Found: C, 57.42; H, 5.42; N, 8.68; Purity by HPLC 99.6%; mp 236–242 °C.

5.1.47. Sodium [2-({5-[(2-octyl-phenylamino)-methyl]-thiophene-2-carbonyl}-amino)-thiazol-4-yl]-acetate (*50*)

This compound was prepared according to the synthetic protocols described for compound **48**, starting from **118** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 508 (M + H), 506 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 0.85 (t, J = 7.0 Hz, 3H), 1.26–1.36 (m, 10H), 1.51–1.59 (m, 2H), 2.47–2.49 (m, 2H), 3.32 (s, 2H), 4.52 (d, J = 5.6 Hz, 2H), 5.79 (t, J = 6.1 Hz, 1H), 6.50–6.53 (m, 2H), 6.65 (s, 1H), 6.90–6.94 (m, 2H), 7.01 (d, J = 3.7 Hz, 1H), 8.24 (br s, 1H); Anal. Calcd for $C_{25}H_{30}N_3O_3S_2$·Na: C, 59.15; H, 5.96; N, 8.28. Found: C, 59.09; H, 5.88; N, 8.20; Purity by HPLC 99.8%; mp 226–230 °C.

5.1.48. {2-[(5-{[2-(3-Methyl-butyl)-phenylamino]-methyl}-thiophene-2-carbonyl)-amino]-thiazol-4-yl}-acetic acid (*51*)

This compound was prepared according to the synthetic protocols described for compound **48**, starting from **119** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 444 (M + H), 442 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 0.95 (d, J = 6.4 Hz, 6H), 1.40–1.52 (m, 2H), 1.54–1.69 (m, 1H), 2.47–2.55 (m, 2H), 3.62 (s, 2H), 4.56 (d, J = 5.7 Hz, 2H), 5.81 (t, J = 5.7 Hz, 1H), 6.46–6.57 (m,

2H), 6.88–7.00 (m, 3H), 7.13 (d, $J = 3.8$ Hz, 1H), 8.07 (d, $J = 3.8$ Hz, 1H), 12.49 (br s, 2H); Anal. Calcd for $C_{22}H_{25}N_3O_3S_2$: C, 59.57; H, 5.68; N, 9.47. Found: C, 59.61; H, 5.77; N, 9.51; mp 218–223 °C.

5.1.49. [2-({5-[(2-Benzyl-phenylamino)-methyl]-thiophene-2-carbonyl}-amino)-thiazol-4-yl]-acetic acid (**52**)

This compound was prepared according to the synthetic protocols described for compound **48**, starting from **120** and (2-amino-4-thiazolyl)acetic acid ethyl ester. MS ESI m/e: 464 (M + H), 462 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 3.62 (s, 2H), 3.89 (s, 2H), 4.55 (d, $J = 5.7$ Hz, 2H), 5.90 (t, $J = 5.8$ Hz, 1H), 6.51–6.59 (m, 2H), 6.88 (d, $J = 6.4$ Hz, 1H), 6.93–7.01 (m, 2H), 7.05 (d, $J = 3.8$ Hz, 1H), 7.16–7.33 (m, 5H), 8.05 (d, $J = 3.8$ Hz, 1H), 12.51 (br s, 2H); Anal. Calcd for $C_{24}H_{21}N_3O_3S_2$: C, 62.18; H, 4.57; N, 9.06. Found: C, 62.23; H, 4.39; N, 9.28; mp 188–191 °C.

5.1.50. 5-[(2-Trifluoromethyl-phenylamino)-methyl]-thiophene-2-carboxylic acid (4-hydroxymethyl-thiazol-2-yl)-amide (**44**)

Step 1. To a solution of 5-(((2-(trifluoromethyl)phenyl)amino) methyl)thiophene-2-carboxylic acid (**115**; 70 mg, 0.232 mmol) and (2-amino-1,3-thiazol-4-yl)methyl acetate hydrochloride (58 mg, 0.280 mmol) in $CHCl_3$ (1.4 ml) was added EDC·HCl (53 mg, 0.280 mmol), followed by DMAP (65 mg, 0.530 mmol) at room temperature. The reaction mixture was stirred for 3 h under N_2 atmosphere. EtOAc and saturated $NaHCO_3$ were added to the reaction mixture and the layers separated. The organic layer was washed with brine and dried over Na_2SO_4. The mixture was filtered and concentrated. The residue was purified by preparative TLC (hexane/EtOAc = 1/1) to afford (2-(5-(((2-(trifluoromethyl)phenyl) amino)methyl)thiophene-2-carboxamido)thiazol-4-yl)methyl acetate (**121**; 90 mg, 85%) as a solid. ^{1}H NMR ($CDCl_3$, 300 MHz) δ: 2.11 (s, 3H), 4.66 (d, $J = 6.0$ Hz, 2H), 4.95 (br s, 1H), 5.08 (s, 2H), 6.71 (d, $J = 8.3$ Hz, 1H), 6.80 (t, $J = 7.7$ Hz, 1H), 6.95 (s, 1H), 7.06 (d, $J = 3.8$ Hz, 1H), 7.34 (t, $J = 7.7$ Hz, 1H), 7.49 (d, $J = 7.5$ Hz, 1H), 7.57 (d, $J = 4.1$ Hz, 1H), 9.47 (br s, 1H).

Step 2. To a solution of **121** (90 mg, 0.198 mmol) in THF (0.45 ml) and MeOH (0.9 ml) was added K_2CO_3 (55 mg, 0.396 mmol) at room temperature. The reaction mixture was stirred for 1 h under N_2 atmosphere. EtOAc and saturated $NaHCO_3$ were added to the reaction mixture and the layers separated. The organic layer was washed with brine and dried over Na_2SO_4. The mixture was filtered and concentrated. After adding hexane and EtOAc to the resulting residue, the slurry was filtered. The filter cake was washed with hexane and dried in a vacuum to afford 5-[(2-trifluoromethyl-phenylamino)-methyl]-thiophene-2-carboxylic acid (4-hydroxymethyl-thiazol-2-yl)-amide (**44**; 70 mg, 86%) as a solid. MS ESI m/e: 414 (M + H), 412 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 4.49 (d, $J = 5.7$ Hz, 2H), 4.67 (d, $J = 6.0$ Hz, 2H), 5.20 (t, $J = 5.7$ Hz, 1H), 6.41 (t, $J = 6.2$ Hz, 1H), 6.70 (t, $J = 7.5$ Hz, 1H), 6.78 (d, $J = 8.3$ Hz, 1H), 6.94 (s, 1H), 7.13 (d, $J = 3.8$ Hz, 1H), 7.34 (t, $J = 7.5$ Hz, 1H), 7.43 (d, $J = 7.5$ Hz, 1H), 8.05 (d, $J = 3.0$ Hz, 1H), 12.59 (s, 1H); Anal. Calcd for $C_{17}H_{14}F_3N_3O_2S_2$: C, 49.39; H, 3.41; N, 10.16. Found: C, 49.40; H, 3.08; N, 10.17; mp 183–190 °C.

5.1.51. 5-[(2-Trifluoromethyl-phenylamino)-methyl]-thiophene-2-carboxylic acid [4-(2-hydroxy-ethyl)-thiazol-2-yl]-amide (**45**)

Step 1. To a solution of 5-(((2-(trifluoromethyl)phenyl)amino) methyl)thiophene-2-carboxylic acid (**115**; 70 mg, 0.232 mmol) and 4-(2-((tert-butyldimethylsilyl)oxy)ethyl)thiazol-2-amine (72 mg, 0.280 mmol) in $CHCl_3$ (1.4 ml) was added EDC·HCl (53 mg, 0.280 mmol), followed by DMAP (31 mg, 0.254 mmol) at room temperature. The reaction mixture was stirred for 2 h under N_2 atmosphere. EtOAc and saturated $NaHCO_3$ were added to the reaction mixture and the layers separated. The organic layer was washed with brine and dried over Na_2SO_4. The mixture was filtered and concentrated. The residue was purified by preparative TLC (hexane/EtOAc = 3/1) to afford N-(4-(2-((tert-butyldimethylsilyl) oxy)ethyl)thiazol-2-yl)-5-(((2-(trifluoromethyl)phenyl)amino) methyl)thiophene-2-carboxamide (**122**; 110 mg, 87%) as a solid. ^{1}H NMR ($CDCl_3$, 300 MHz) δ: 0.00 (s, 6H), 0.86 (s, 9H), 2.84 (t, $J = 6.4$ Hz, 2H), 3.88 (t, $J = 6.6$ Hz, 2H), 4.65 (d, $J = 5.7$ Hz, 2H), 4.94 (br s, 1H), 6.64 (s, 1H), 6.72 (d, $J = 8.3$ Hz, 1H), 6.79 (t, $J = 7.5$ Hz, 1H), 7.05 (d, $J = 3.8$ Hz, 1H), 7.34 (t, $J = 7.7$ Hz, 1H), 7.48 (d, $J = 7.5$ Hz, 1H), 7.56 (d, $J = 3.8$ Hz, 1H).

Step 2. To a solution of **122** (110 mg, 0.203 mmol) in THF (0.55 ml) was added 1 M THF solution of TBAF (0.609 ml, 0.609 mmol) at room temperature. The reaction mixture was stirred at 50 °C for 1 h under N_2 atmosphere. EtOAc and saturated $NaHCO_3$ were added to the reaction mixture and the layers separated. The organic layer was washed with brine and dried over Na_2SO_4. The mixture was filtered and concentrated. The residue was purified by preparative TLC ($CHCl_3$/MeOH = 9/1). After adding hexane and EtOAc to the resulting residue, the slurry was filtered. The filter cake was washed with hexane and dried in a vacuum to afford 5-[(2-trifluoromethyl-phenylamino)-methyl]-thiophene-2-carboxylic acid [4-(2-hydroxy-ethyl)-thiazol-2-yl]-amide (**45**; 82 mg, 94%) as a solid. MS ESI m/e: 428 (M + H), 426 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.77 (t, $J = 6.8$ Hz, 2H), 3.62–3.75 (m, 2H), 4.56–4.71 (m, 3H), 6.40 (t, $J = 5.7$ Hz, 1H), 6.70 (t, $J = 7.5$ Hz, 1H), 6.74–6.85 (m, 2H), 7.12 (d, $J = 3.8$ Hz, 1H), 7.34 (t, $J = 7.7$ Hz, 1H), 7.43 (d, $J = 7.5$ Hz, 1H), 8.04 (s, 1H), 12.55 (s, 1H); Anal. Calcd for $C_{18}H_{16}F_3N_3O_2S_2$: C, 50.58; H, 3.77; N, 9.83. Found: C, 50.55; H, 3.50; N, 9.77; mp 182–184 °C.

5.1.52. 5-[(2-Hexyl-phenylamino)-methyl]-thiophene-2-carboxylic acid [4-(2-hydroxy-ethyl)-thiazol-2-yl]-amide (**46**)

This compound was prepared according to the synthetic protocols described for compound **45**, starting from **117** and 4-(2-((tert-butyldimethylsilyl)oxy)ethyl)thiazol-2-amine. MS ESI m/e: 444 (M + H), 442 (M − H); ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 0.88 (t, $J = 5.7$ Hz, 3H), 1.20–1.43 (m, 6H), 1.48–1.62 (m, 2H), 2.46–2.54 (m, 2H), 2.77 (t, $J = 6.8$ Hz, 2H), 3.69 (dd, $J = 11.7$, 6.8 Hz, 2H), 4.55 (d, $J = 5.7$ Hz, 2H), 4.62 (t, $J = 4.9$ Hz, 1H), 5.85 (t, $J = 5.8$ Hz, 1H), 6.46–6.57 (m, 2H), 6.82 (s, 1H), 6.87–6.98 (m, 2H), 7.11 (d, $J = 3.8$ Hz, 1H), 8.05 (s, 1H), 12.53 (s, 1H); Anal. Calcd for $C_{23}H_{29}N_3O_2S_2 \cdot 0.25H_2O$: C, 61.65; H, 6.64; N, 9.38. Found: C, 61.80; H, 6.37; N, 9.29; mp 108–111 °C.

5.2. Inhibitory effects on SCD1 enzyme activity

SCD1 enzyme activity was evaluated by measuring the production of [^{3}H] H_2O from [9, 10-^{3}H] stearoyl-CoA catalyzed by SCD1 enzyme. The microsomal fraction (recombinant human SCD; rhSCD1) of cells over-expressing human SCD1 via a baculovirus expression system was used as an enzyme source. The rhSCD1 (0.5 μg), 0.12 μmol/L [9, 10-^{3}H] stearoyl-CoA (1.4 μCi, Perkin Elmer), and the compound solution dissolved in DMSO were added to 96-well plates with 80 μl of the reaction buffer containing 100 mmol/L Tris-HCl (pH 7.4), 2 mmol/L NADH, 6 mmol/L $MgCl_2$, 125 mmol/L sucrose, 0.005% (w/v) BSA, and 0.25 μg cytochrome b5. Thirty minutes after incubation at room temperature, the reaction was terminated by adding 100 μl of stopping buffer containing 100 mmol/L Tris-HCl (pH 7.4), 10 mmol/L EDTA, and 62.5 mg/ml activated charcoal. [^{3}H] H_2O (liquid phase) was separated by filtration (Unifilter, GE Healthcare) from the charcoal-adsorbed [9, 10-^{3}H] stearoyl-CoA and scintillation cocktail (MicroScint-40, Perkin Elmer) added. The radioactivity of the mixture was measured by a scintillation counter (TopCount, Perkin Elmer). Control values were measured by evaluating the radioactivity of a well containing DMSO instead of the compound solution. Background values were

measured by evaluating the radioactivity of a well to which stopping buffer was added before the reaction. The remaining SCD1 activity (%) was calculated for each well using the following formula:

SCD1 activity (%) = (Compound − Background)/(Control − Background)

The 50% inhibitory concentration (IC_{50}) values were calculated from two measurements of the remaining SCD1 activity crossing 50%.

5.3. Effects on SCD1 activity in the liver and eyelid

C57BL/6 J mice were obtained from Charles River Laboratories (Japan). Compounds were orally administered to the mice (male, 6 weeks of age). Six or 24 h after administration, the liver or eyelids were dissected from the mice and added to 9-fold (liver) or 4-fold (eyelid) homogenizing buffer (100 mmol/L Tris-HCl (pH 7.4), 0.25 mol/L sucrose), respectively. The tissues were sufficiently homogenized using one zirconia ball (YTZ® Ball, Nikkato Corp.) and a mixer mill (MM300, Retsch Co., Ltd.), followed by centrifugation at 10,000 × *g* at 4 °C for 5 min to obtain the S9 fraction. The SCD1 reaction was initiated by adding 40 μl of S9 fraction to the substrate solution containing 100 mmol/L Tris-HCl (pH 7.4), 4 mmol/L NADH, 12 mmol/L $MgCl_2$, 0.5 μg cytochrome b5, and 12 μmol/L [^{14}C] stearoyl-CoA. The mixtures were incubated at room temperature for 10 min (liver) or 30 min (eyelid). The reaction was terminated by adding 1 ml of HCl in methanol (Wako Pure Chemical Industries). Two milliliters of hexane was added and the mixtures incubated at 100 °C for 1 h to saponify the lipids. The mixtures were centrifuged at 1000 × *g* for 10 min and the upper phases (hexane phases) collected. Hexane was evaporated by N_2 gas and the residues resolved in 100 μl of hexane and separated on a 10% $AgNO_3$-impregnated TLC plate with hexane/diethyl ether (9:1). The TLC plate was exposed to an imaging plate (BAS-MS2040, Fuji film Corporation, Japan) to visualize and quantify [^{14}C]-incorporated stearic acid and oleic acid using an imaging analyzer (BAS2500, Fujifilm Corporation, Japan).

5.4. Effects of **48** in mice fed a high fat diet

C57BL/6 J mice were obtained from Charles River Laboratories (Yokohama, Japan) and individually housed under controlled temperature (23 ± 3 °C), humidity (55 ± 15%), and lighting (12 h light/dark cycle with lights on at 8:00 a.m.). The mice were provided a HFD (D12330, Research Diets, Inc., USA) and water ad libitum. After being fed the HFD for a month, the mice were divided into three groups: a control group and 3 and 10 mg/kg **48**. The mice in the **48**-treated groups were fed a HFD containing appropriate amounts of **48** (0.0040% at 3 mg/kg/day, 0.0133% at 10 mg/kg/day) to achieve a daily dose of approximately 3 or 10 mg/kg for 43 days from 10 to 16 weeks of age. A normal chow diet (CRF-1; Oriental Yeast, Osaka, Japan) was provided for the control group. Body weight was measured every 3 or 4 days during the experimental period. On day 22 of administration, a glucose tolerance test (1 g/kg) was performed to the mice fasted overnight. The area under the curve of plasma glucose level (AUC glucose) was calculated from plasma glucose levels 0, 0.5, 1, and 2 h after glucose loading.

5.5. Effects of **48** on SCD activity in the liver, epididymal adipose tissue, and eyelid in rats fed a high sucrose diet

Sprague Dawley (SD) rats were obtained from Charles River Laboratories (Yokohama, Japan) and fed a HS diet (D11725, Research Diets, Inc.). Compound **48** was administered orally at 1 or 10 mg/kg. Six hours after administration, liver, epididymal adipose tissue, and eyelid samples were collected and homogenized to measure tissue SCD activity as described above.

5.6. Effects of **48** on the liver triglyceride contents in rats fed a high sucrose very low fat diet

SD rats were obtained from Charles River Laboratories (Yokohama, Japan) and fed a HSVLF diet (D08030601, Research Diets, Inc.). Compound **48** was administered orally at 1 or 10 mg/kg once daily for 8 days and liver slices collected to measure the TG contents of the liver on day 8 of administration. Methanol (0.4 ml) was added to the liver slices before homogenization using one zirconia ball and a mixer mill. The homogenates were combined with 0.4 ml methanol and 0.8 ml chloroform, mixed, and centrifuged at 10,000 × *g* for 5 min. A total of 0.2 ml of supernatant was collected and dried with N_2 gas. The residues were resolved in 200 μl of 2-propanol and TG levels measured using an automatic analyzer (Hitachi7170S, Tokyo, Japan).

5.7. Safety studies

In order to investigate any toxicity after repeated dosing for 2 weeks in rats, **48** suspended in vehicle (0.5% methylcellulose aqueous solution) was administered orally once daily to 6-week-old SD rats (5 males per group) at 100 and 1000 mg/kg. The animals in the control group (5 males) were given the vehicle. During the dosing period, clinical observations and measurements were made of body weight and food consumption. Ophthalmoscopy was conducted in week 2. At the end of the dosing period, necropsy, hematology, clinical chemistry, organ weight measurements, and histopathological examination were performed. The plasma concentrations of **48** were determined 24 h after dosing on days 1, 5, and 10.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.ejmech.2018.09.003.

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ABBREVIATIONS USED

SCD: stearoyl-CoA desaturase
MUFA: monounsaturated fatty acid
SFA: saturated fatty acid
TG: triglyceride
ASO: antisense oligonucleotide
HTS: high-throughput screening
PK: pharmacokinetic
SAR: structure-activity relationship
cLogP: calculated logarithm of the partition coefficient
tPSA: topological polar surface area
LE: ligand efficiency
HFD: high fat diet
HSVLF: high sucrose very low fat
SD: Sprague Dawley
HS: high sucrose
NOAEL: no-observed adverse effect level
hERG: human ether a-go-go-related gene
CYP: cytochrome P450
NT: not tested
TLC: thin-layer chromatography
CDI: 1,1′-carbonyldiimidazole
TBS: tert-butyldimethylsilyl
EDC·HCl: 1-(3-(dimethylamino)propyl)-3-ethyl-carbodiimide hydrochloride
HOBt: 1-hydroxybenzotriazole monohydrate
PyBOP: 1H-benzotriazol-1-yloxy-tri(pyrrolidino)phosphonium hexafluorophosphate
TEA: triethylamine
DMAP: 4-dimethylaminopyridine
DMSO: dimethyl sulfoxide
DMF: dimethylformamide
DME: 1,2-dimethoxyethane
AcOH: acetic acid
MeOH: methanol
EtOH: ethanol
$CHCl_3$: chloroform
$Pd_2(dba)_3$: tris(dibenzylideneacetone)dipalladium(0)
X-Phos: 2-dicyclohexylphosphino-2′,4′,6′-triisopropylbiphenyl
rt: room temperature