HETEROCYCLES, Vol. 94, No. 4, 2017, pp. 702 - 715. © 2017 The Japan Institute of Heterocyclic Chemistry
Received, 30th January, 2017, Accepted, 16th March, 2017, Published online, 24th March, 2017
DOI: 10.3987/COM-17-13663

A CONVERGENT SCALE-UP SYNTHESIS OF A HER2/EGFR DUAL KINASE INHIBITOR

Osamu Yabe,* Akihiro Suzuki, and Tomomi Ikemoto

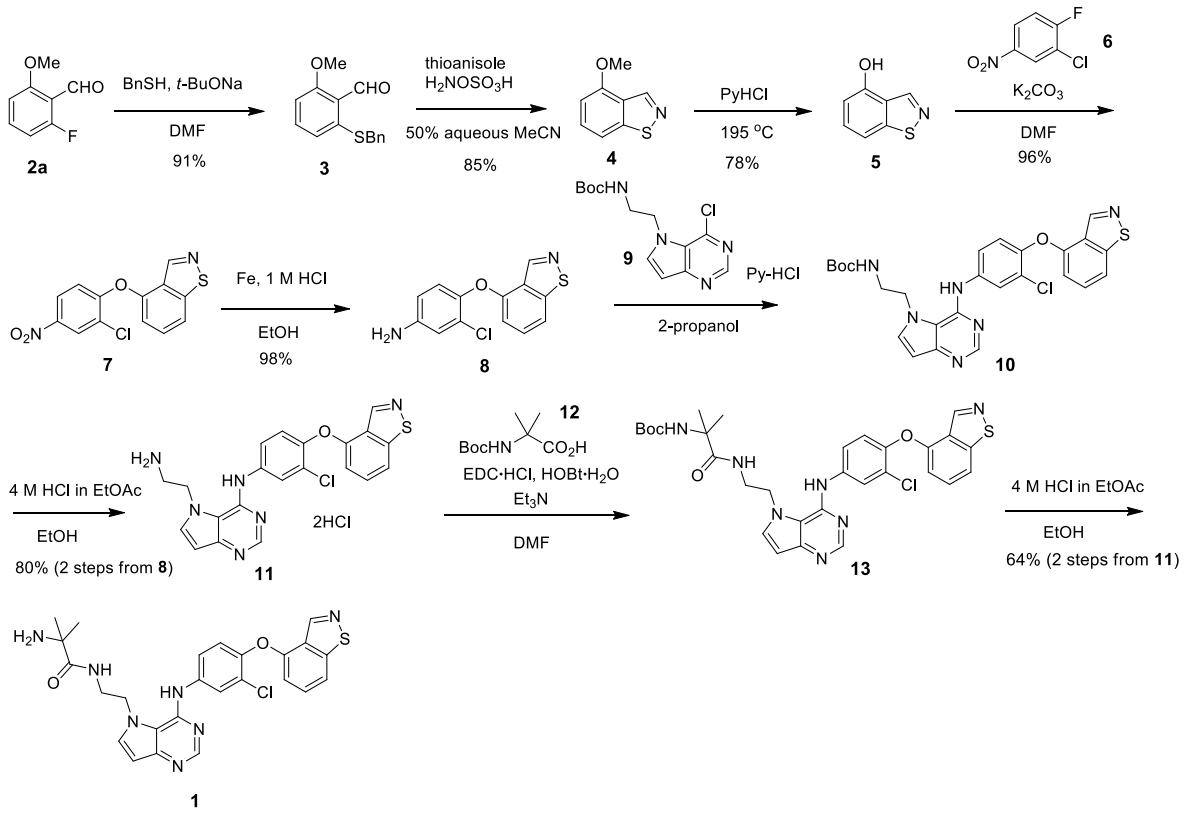
Process Chemistry, Pharmaceutical Sciences, Takeda Pharmaceutical Company Ltd., 17-85, Jusohonmachi 2-chome, Yodogawa-ku, Osaka 532-8686, Japan. E-mail: osamu.yabe@takeda.com

Abstract – A practical and scalable synthesis of the human epidermal growth factor receptor 2 (HER2)/epidermal growth factor receptor (EGFR) dual kinase inhibitor **1** has been developed. The key features of the process development include convenient construction of the benzisothiazole skeleton directly from commercially available materials in high yield, a practical *O*-demethylation utilizing an ethanethiol or octanethiol/aluminium chloride system without harsh conditions, and development of a more convergent alternative route that coupled two key intermediates in the final step. The novel synthesis allowed the manufacturing process to produce high quality API **1** (>99% purity (LCAP)) over 7 steps, compared to 9 steps in the original route, without chromatographic purification.

INTRODUCTION

Protein tyrosine kinases play a key role in signal transduction pathways that regulate numerous cellular functions, and these kinase inhibitors are attractive candidates for cancer therapy. The targeting of human epidermal growth factor receptor 2 (HER2 or ErbB-2/neu) and epidermal growth factor receptor (EGFR or HER1/ErbB-1) by tyrosine kinase inhibitors also represents one such therapeutic approach. Compound **1**, discovered in our company, shows potent tumor regressive efficacy against both HER2- and EGFR-over expressing tumors, and has the potential to cure HER2- and EGFR-overexpressing cancer.[1]

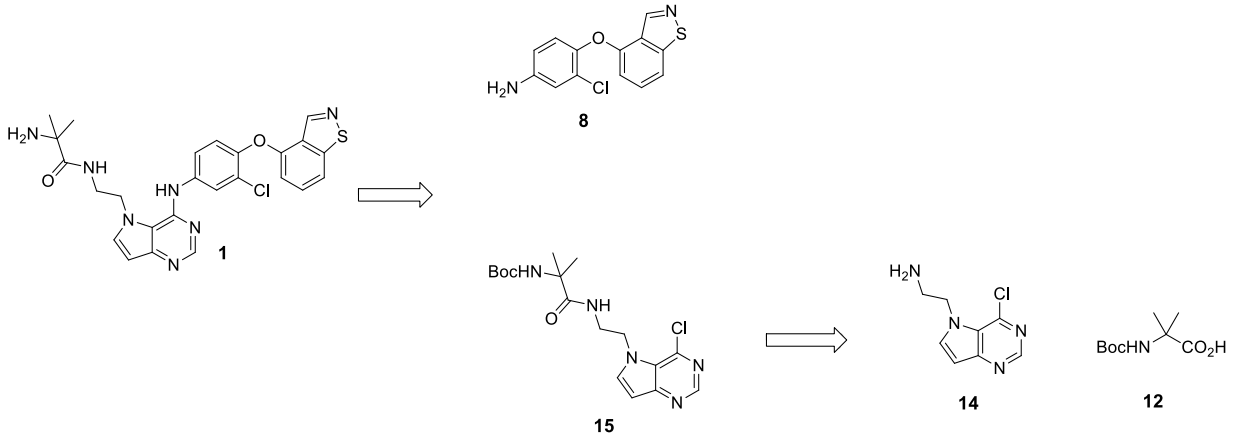
The initial medicinal chemistry route linearly led to **1**, as shown in Scheme 1.[2] This synthesis was effective to provide various derivatives for the early biological and toxicology evaluations. However, it had some drawbacks for large-scale manufacturing. Here, we disclose a convergent preparation of **1**, which has the potential to reduce the manufacturing cost and lead time.



Scheme 1. Medicinal chemistry synthesis of **1**

RESULTS AND DISCUSSION

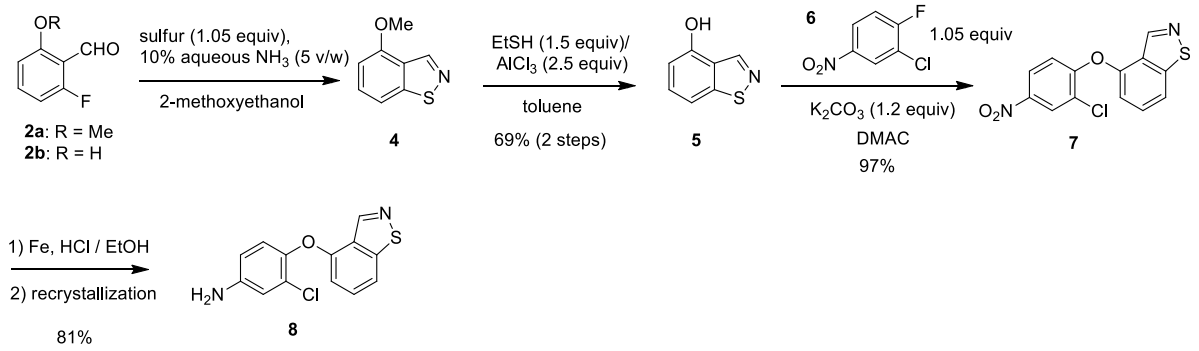
Our synthetic strategy is outlined in Scheme 2. To develop an efficient route to **1** we planned that a substitution reaction of aniline **8** and chloropyrrolopyrimidine **15**, prepared from amine **14** and amino acid **12**, could be conducted as the final step.



Scheme 2. Retrosynthetic analysis of **1**

The original synthesis started with the nucleophilic aromatic substitution of aldehyde **2a** with benzyl mercaptan to give thioether **3**. Compound **3** was then cyclized with hydroxylamine-*O*-sulfonic acid in the presence of thioanisole to afford benzisothiazole **4** along with a thioanisole derivative byproduct, which

was removed by chromatographic purification. To discover a simpler synthesis of **4**, a direct construction of the benzisothiazole ring from **2a** was examined in one-pot, as shown in Scheme 3. When **2a** with sulfur and aqueous ammonia in 2-methoxyethanol was treated at 100 ºC for 5 h in a sealed tube according to literature precedents, the desired **4** was obtained in 82% assay yield (Table 1, entry 1).[3] Reaction in an open flask instead of a sealed tube also converted **2a** to **4**, but a vigorous gas evolution was observed during the reaction (entry 2). The dropwise addition of 25% aqueous ammonia to the mixture of **2a** and sulfur in 2-methoxyethanol at 80 ºC afforded **4** in 81% yield with a controlled generation of gas (entry 3), and use of 10% aqueous ammonia controlled the gas evolution more moderately (entry 4). On the other hand, the change of reaction temperature to 70 ºC suppressed the reaction to lower the assay yield to 59% (entry 5). From these results, entry 4 was selected as the optimized condition for scale-up preparation. As the crystallization of **4** from *n*-heptane led to >20% loss to the mother liquid, crude **4** was used in the next step without further purification after the aqueous workup, extraction with EtOAc, and solvent switch to toluene from EtOAc. Also, the conversion of the phenolic compound **2b** produced a few unidentified byproducts and most **2b** remained without providing **5,** even when the same condition to entry 4 was used.



Scheme 3. Optimized synthesis of **8** from **2a**

Table 1. Construction of benzisothiazole ring **4**[a)]

entry	concentration of aqueous NH_3 (%)	temperature (ºC)	time (h)	assay yield (%)
1[b)]	25	100	5	82
2[c)]	25	80	4	N/A[d)]
3	25	80	4	81
4	10	80	4	80
5	25	70	4	59

a) Aqueous ammonia was added dropwise to a mixture of **2a** and sulfur in 2-methoxyethanol at the reaction temperature. b) The reaction was conducted in a sealed tube. c) A mixture of **2a**, sulfur and aqueous ammonia in 2-methoxyethanol was heated. Vigorous gas evolution was observed. d) Not available.

In the original route, the demethylation of solid **4** with solid pyridine hydrochloride was carried out without any solvents under fusion conditions (195 ºC). To improve on the procedure, various methods were investigated (Table 2). The use of 47% aqueous HBr in AcOH was not effective, and the combination of methionine and methanesulfonic acid gave only a low yield of the demethylated compound **5** (entries 1, 2).[4a,4b] The reaction with BBr_3 at 100 ºC in chlorobenzene almost consumed all of compound **4** to give **5** in 92% (LCAP), while the reaction with aluminium chloride and ethanethiol in toluene converted **4** to **5** at 25 ºC (entries 3, 4).[4a,4c] The resulting reaction mixture from entry 4 was poured into water to isolate **5** as crystals in 72% yield from **2a** with 97.0% purity (LCAP). The condition with ethanethiol/aluminium chloride at 25 ºC was selected for the first scale-up manufacturing, on a 4 kg scale, in light of a safety concern, and it gave 69% yield with 98.1% purity (LCAP). Although the odor concern of ethanethiol still remained, further optimization demonstrated that the odorless octanethiol also provided **5** in 71% isolated yield (entry 5).

Table 2. Optimization of demethylation reaction of **4**

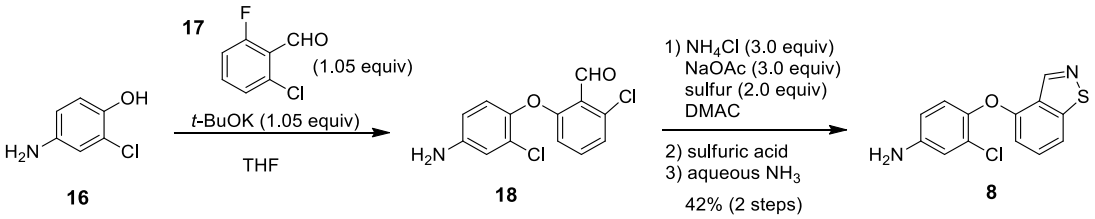
entry	solvent	demethylation reagent (equiv)	temperature (ºC)	time (h)	**4**[a)]	**5**[a)]
1	AcOH	47% aqueous HBr (10)	110	8	77	7
2	—	$MeSO_3H$ (11)/methionine (1.1)	130	8	62	22
3	PhCl	BBr_3 (5.0)	100	2	0.6	92
4	PhMe	ethanethiol (1.5)/$AlCl_3$ (2.5)	25	6	0.7	96 (72)[b)]
5	PhMe	octanethiol (1.5)/$AlCl_3$ (2.5)	25	6	0.8	95 (71)[b)]

a) LCAP. b) Isolated yield of **5** from **2a**.

The coupling reaction of **5** with **6** in DMAC in the presence of K_2CO_3 was conducted, and ether **7** was isolated as crystals in 96% yield and >99% purity (LCAP) by adding water directly to the reaction mixture. The treatment of **7** with an optimized amount of iron powder (8.2 equiv) and 1 M HCl quantitatively gave **8** at 70−80 ºC for 3 h in EtOH (20 v/w). However, since a large amount of EtOAc (42 v/w) as extraction solvent was needed, due to the low solubility of **8**, a different procedure was attempted. When the reaction mixture was neutralized with 2 M NaOH and THF was subsequently added to dissolve **8**, the insoluble materials were precipitated. After filtration of insoluble materials, the filtrate was concentrated to half volume under reduced pressure, and water was added into the resulting concentrated solution to crystallize crude **8**. The isolated crude **8** was then recrystallized from aqueous EtOH after treatment with activated carbon to afford purified **8** in 81% yield from **7** and >99% purity (LCAP).

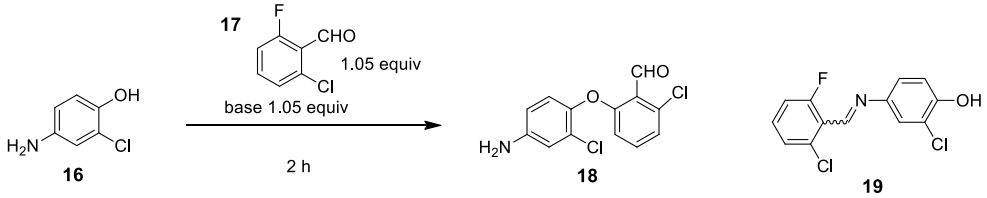
The alternative synthesis using readily available **16** and **17** as starting materials was also investigated, aiming to further shorten a number of manufacturing steps, as illustrated in Scheme 4. Initially, the solvents and bases for reaction of **16** with **17** were screened, and the results are described in Table 3. Use of K_2CO_3, $NaHCO_3$, Et_3N, and DBU mainly gave byproduct **19** (entries 1−4), whereas *t*-BuOK gave the

desired **18** in DMAC with good conversion (entry 5). The other solvents, such as DMF, MeCN, and 2-methoxyethanol in the presence of *t*-BuOK decreased the yield (entries 6−8). Interestingly, the reaction at lower concentration suppressed the generation of impurities to show better conversion than for higher concentration reactions (entries 5 vs 9). At the lower concentration, THF gave the highest conversion (89.2%, LCAP) when the limited solvent screening was conducted again (entries 9−11). Compound **18** could be isolated as its HCl salt in 83% yield with 89.9% purity (LCAP), by addition of 4 M HCl in EtOAc to the extraction solution after workup. However, crude **18** was used in the next step without salt formation because the purity was not improved by isolating as the HCl salt.



Scheme 4. Alternative synthesis of **8** from **16**

Table 3. Optimization of substitution reaction between **16** with **17**



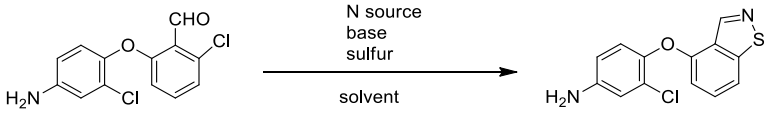
entry	solvent (v/w)	base	temperature (°C)	**18**[a)]	**19**[a)]
1	DMAC (10)	K_2CO_3	80	18.0	48.4
2	DMAC (10)	$NaHCO_3$	80	4.4	65.7
3	DMAC (10)	Et_3N	80	2.3	59.6
4	DMAC (10)	DBU	80	8.0	44.0
5	DMAC (10)	*t*-BuOK	80	81.6	0.1
6	DMF (10)	*t*-BuOK	80	46.7	11.9
7	MeCN (10)	*t*-BuOK	80	26.2	12.5
8	2-methoxyethanol (10)	*t*-BuOK	80	7.6	52.5
9	DMAC (20)	*t*-BuOK	80	86.8	0.1
10	DME (20)	*t*-BuOK	60	82.6	0.1
11	THF (20)	*t*-BuOK	60	89.2	0.1

a) LCAP.

Next, the cyclization of **18** was performed (Table 4). The optimal cyclization condition for synthesis of **4** gave 12% assay yield of **8** from **16** with 30% (LCAP) of residual **18** (entry 1).[3] It was thought that the low solubility of **18** in 2-methoxyethanol caused the low yield. When the cyclization of **18** using ammonium

chloride (3.0 equiv) as nitrogen source, sodium acetate (3.0 equiv) and sulfur (5.0 equiv) at 80 ºC in DMF was attempted, the assay yield over 2 steps was 57% (entry 2).[5] To our knowledge, this was the first example to construct a benzisothiazole ring by the transformation of an *o*-halogeno-benzaldehyde with sulfur and NH_4Cl. The replacement of DMF with DMAC increased the assay yield to 65%, while the change of reaction temperature to 60 ºC decreased the assay yield to 30% (entries 3, 4). Although the reaction with 1.1 equiv of sulfur decreased the assay yield to 47% (entry 5), 2.0 equiv of sulfur provided **8** in 62% yield (entry 6). Since the excess amount of sulfur required a complicated workup procedure, entry 6 was selected as the optimized condition.

Table 4. Optimization of the cyclization of **18** to give **8**



18 **8**

entry	N source (equiv)	NaOAc (equiv)	sulfur (equiv)	solvent	temperature (ºC)	time (h)	assay yield of **8** (%)[a)]
1	aqueous NH_3 (8.0)	—	1.05	2-methoxyethanol	80	5	12
2	NH_4Cl (3.0)	3.0	5.0	DMF	80	4	57
3	NH_4Cl (3.0)	3.0	5.0	DMAC	80	4	65
4	NH_4Cl (3.0)	3.0	5.0	DMAC	60	4	30
5	NH_4Cl (3.0)	3.0	1.1	DMAC	80	4	47
6	NH_4Cl (3.0)	3.0	2.0	DMAC	80	4	62

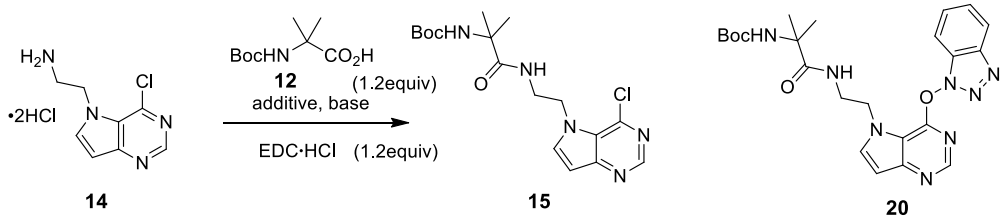
a) Assay yield from **16**.

After workup, the free base **8** was crystallized from aqueous ethanol. However, the obtained crystals were of 72.0% purity based on the analytical standard, and included a large amount of tarry products. In order to afford high quality **8**, purification by salt formation of **8** was examined. In the case of the monosulfate, a gum-like mass that interfered with smooth agitation was first precipitated in the solution and the salt was subsequently crystallized.[6] HPLC analysis showed that the gum-like mass contained many different byproducts in small amounts. Thus, when the gummy material was removed by decantation, the resulting solution led to crystallization of **8** as a monosulfate with 98.0% purity (LCAP).[6] Subsequent neutralization of the isolated sulfate salt in aqueous ethanol with 25% aqueous ammonia gave rise to crystallization, followed by filtration to afford **8** as a free base with 98.4% purity (LCAP) in 42% yield from **16** (98.0% purity based on analytical standard). Although a two-step synthesis of **8** from **16** was found, we finally selected the route to **8** from **2a** for the scale-up synthesis, considering both the operations and the overall yield.

Next, we turned our attention to the synthesis of **15** to accomplish the convergent synthesis. The Boc group of **9**[7a] was successfully removed using 4 M HCl in EtOAc, and the desired product **14** was isolated

directly from the reaction mixture as a dihydrochloride salt in quantitative yield (Scheme 5). An amide coupling of acid **12** with amine **14** using 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC·HCl) in the presence of 0.1 equiv of *N*-hydroxybenzotriazole hydrate (HOBt·H_2O) gave the desired **15** in 90% assay yield containing impurity **20** (4.6%, LCAP), which could not be removed by crystallization (Table 5, entry 1). When *N*,*N*-dimethylaminopyridine (DMAP) was used instead of HOBt·H_2O, **15** was obtained in 75−81% assay yield (entries 2, 3). Among the screened solvents, MeCN was found to be the most effective, affording **15** in 90% assay yield without generation of any notable impurities (entry 4). The pH control in the extraction was critical to give **15** after workup. The residual DMAP was difficult to remove by crystallization of **15**. On the other hand, a large amount of **15** was lost to the aqueous layer when the organic layer was washed with dilute aqueous HCl in order to completely remove DMAP. However, when the pH was adjusted to 2−3 with 2 M aqueous HCl, DMAP was effectively removed to the aqueous layer without loss of **15**. The obtained organic layer was concentrated and then crystallized from a toluene/*n*-heptane mixture to furnish **15** with >99% purity (LCAP) in 78% yield (Scheme 5).

Table 5. Amidation reaction of **14** with **12**

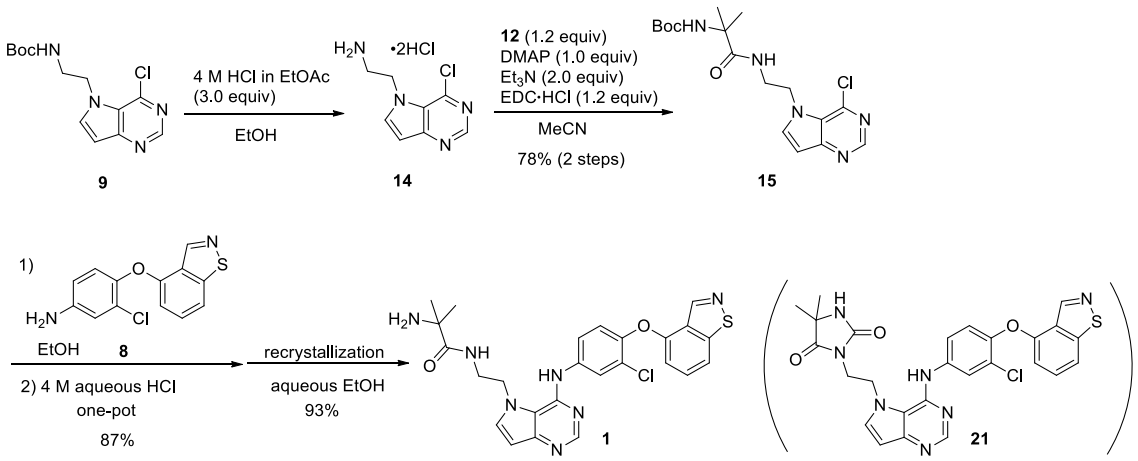


entry	base (equiv)	additive (equiv)	solvent	Temp (ºC)	time (h)	LCAP (%) **15**	**14**	**20**	yield[a] (%)
1	Et_3N (2.0)	HOBt·H_2O (0.1)	DMAC	25	2	90.3	2.7	4.6	90
2	Et_3N (2.0)	DMAP (1.0)	DMAC	50	5	75.7	17.2	ND[b]	75
3	—	DMAP (3.0)	DMAC	50	5	80.1	13.1	ND[b]	81
4	Et_3N (2.0)	DMAP (1.0)	MeCN	25	3	95.1	4.6	ND[b]	90

a) Assay yield. b) Not detected.

With the two stable and well-characterized building blocks **8** and **15** at hand, the stage was set for the convergent coupling to the API **1**. The solvent screening quickly identified ethanol as a suitable solvent providing good conversion without any additives. Condensation of **8** with **15** in ethanol at 70 ºC for 6 h, followed by the addition of 4 M aqueous HCl to remove the Boc group at the same temperature for 2 h, furnished the target product **1** in one-pot. In the original route, the condensation of **11** with **12** gave 1−5% of byproduct **21** under basic condition (Scheme 5). On the other hand, **21** was not identified at all in the new method. Finally, keeping the pH to 9.5 with 25% aqueous ammonia provided crystals of the free base

form **1**, which were recrystallized from ethanol/water to give pure **1** with >99% purity (LCAP) in 81% yield.



Scheme 5. Optimized synthesis of **1** from **9**

CONCLUSIONS

In summary, a chromarography-free and convergent preparation of **1** was achieved. The *ortho*-fluorobenzaldehyde **2a** was converted to benzisothiazole **4** using sulfur and aqueous ammonia in 2-methoxyethanol. Mild conditions utilizing an ethanethiol or octanethiol/aluminium chloride system facilitated the *O*-demethylation of **4** to give **5** without resort to a solventless condition at high temperature. In the final step, the condensation of two key intermediates, **8** derived from **5** and **15** formed from the corresponding intermediate, was performed and the product was deprotected by addition of aqueous HCl to give **1** in a one-pot sequence. The novel convergent synthesis, combined with some process improvements, allowed the manufacturing process to produce high quality API **1** (>99% purity (LCAP)) over 7 steps, compared to 9 steps in the original route. In addition, an alternative synthesis for key intermediate **8** was also developed, which gave the compound in only two steps in 42% yield.

EXPERIMENTAL

All materials were purchased from commercial suppliers and used without any additional purification. Melting points were determined by using the capillary method on a Büchi 545 apparatus and were uncorrected. ^{1}H NMR and ^{13}C NMR spectra were determined in $CDCl_3$ or DMSO-d_6 on a Bruker DPX-300 or Bruker Avance 500 and reported in δ ppm using tetramethylsilanes as the internal standard. The following abbreviations were used: s = singlet, d = doublet, t = triplet, m = multiplet, dd = doublets of doublet, br = broad. Elemental analyses and HRMS were carried out by Takeda Analytical Laboratories Ltd. All HPLC analyses were performed on a HPLC system equipped with a reversed-phase column (Inertsil ODS-3 (5 μm, 4.6 x 150 mm)) with a detection by UV at 230 nm. Purity was determined

by HPLC and presented as an area percentage of the compound peak relative to the total area of all the peaks integrated.

1,2-Benzisothiazol-4-ol (5).[2] To a solution of **2a** (6.00 kg, 38.9 mol) in 2-methoxyethanol (30 L) was added sulfur (1.31 kg, 40.9 mol) and the resulting mixture was heated to 85 ºC. 10% Aqueous ammonia (30 L) was slowly added to the mixture over 45 min while maintaining the temperature at 80−90 ºC and the resulting solution was stirred for 4 h at 80−90 ºC. After cooling to 30 ºC, EtOAc (48 L) and activated carbon (0.60 kg) were added to the reaction mixture and the resulting mixture was stirred for 0.5 h. The insoluble materials were filtered off and washed with EtOAc (12 L). The biphasic filtrate was separated and the aqueous layer was extracted with EtOAc (30 L). The combined organic layer was washed with 10% aqueous NaCl twice (36 L each) and concentrated to 11 kg under reduced pressure. The solution of residue in toluene (24 L) was concentrated to 12 kg under reduced pressure and these operations were repeated once again. Toluene (24 L) was added to the residue to give **4** as a solution, which was used for the next reaction without isolation. A mixture of anhydrous aluminium chloride (13.0 kg, 97.5 mol) in toluene (18 L) was cooled to 5 ºC. The solution of **4** in toluene was slowly added to the mixture over 2 h while maintaining the temperature at 5−20 ºC, followed by slow addition of ethanethiol (3.64 kg, 58.6 mol) at the same temperature over 0.5 h. The resulting mixture was warmed to 20 ºC and stirred for 3 h at 20−30 ºC. The reaction mixture was slowly added to water (60 L) and allowed to crystallize while maintaining the temperature at 20−30 ºC. The resulting slurry was then stirred for 2 h at 20−30 ºC. The crystals were filtered, washed successively with toluene (12 L) and water (24 L), and dried *in vacuo* at 50 ºC to give **5** as a pale-brown solid (4.06 kg, 69%). Purity: 98.1% (LCAP); ^{1}H NMR (300 MHz, DMSO-d_6): δ 6.81 (1H, d, J = 7.6 Hz), 7.42 (1H, t, J = 7.8 Hz), 7.58 (1H, d, J = 8.1 Hz), 9.07 (1H, s), 10.70 (1H, s); MS: m/z = 151 (M^+); Anal. Calcd for C_7H_5NOS: C, 55.61; H, 3.33; N, 9.26; S, 21.21. Found: C, 55.49; H, 3.30; N, 9.23; S, 21.11.

1,2-Benzisothiazol-4-ol (5) prepared using octanethiol instead of ethanethiol.[2] A mixture of anhydrous aluminium chloride (21.6 g, 0.162 mol) in toluene (30 mL) was cooled to 5 ºC. A solution of **4** in toluene (0.0649 mol of **4** was contained) was slowly added to the mixture while maintaining the temperature at 5−20 ºC, followed by slow addition of octanethiol (16.9 mL, 0.0973 mol) at the same temperature. The resulting mixture was warmed to 20 ºC and stirred for 3 h at 20−30 ºC. To a water (100 mL) was slowly added the reaction mixture to crystallize while maintaining the temperature at 20−30 ºC and the resulting slurry was stirred for 2 h at 20−30 ºC. The crystals were filtered, washed successively with toluene (20 mL) and water (40 mL), and dried *in vacuo* at 50 ºC to give **5** as a pale-brown solid (6.94 g, 71%). Purity: 97.6% (LCAP).

4-(2-Chloro-4-nitrophenoxy)-1,2-benzisothiazole (7).[2] A mixture of **5** (4.02 kg, 26.6 mol), **6** (4.90 kg, 27.9 mol) and K_2CO_3 (4.41 kg, 31.9 mol) in *N*,*N*-dimethylacetamide (12.1 L) was heated to 50 ºC and stirred for 3 h at 47−53 ºC. EtOH (12.1 L) was slowly added to the reaction mixture while maintaining the temperature at 47−53 ºC, followed by slow addition of water (24.1 L) at the same temperature. After cooling to 30 ºC, the resulting slurry was stirred for 1 h at 20−30 ºC. The crystals were filtered, washed successively with EtOH (8.0 L) and water (16.1 L), and dried *in vacuo* at 50 ºC to give **7** as a pale-brown solid (7.94 kg, 97%). Purity: 99.7% (LCAP); ^{1}H NMR (300 MHz, $CDCl_3$): δ 6.89 (1H, d, *J* = 7.7 Hz), 7.05 (1H, d, *J* = 9.1 Hz), 7.53 (1H, t, *J* = 7.8 Hz), 7.82 (1H, d, *J* = 8.2 Hz), 8.11 (1H, dd, *J* = 9.0, 2.7 Hz), 8.45 (1H, d, *J* = 2.6 Hz), 8.95 (1H, s); MS: *m/z* = 306 (M^+); Anal. Calcd for $C_{13}H_7N_2O_3SCl$: C, 50.91; H, 2.30; N, 9.13; S, 10.45; Cl, 11.56. Found: C, 50.89; H, 2.35; N, 9.08; S, 10.36; Cl, 11.49.

4-(1,2-Benzisothiazol-4-yloxy)-3-chloroaniline (8).[2] To a mixture of **7** (3.97 kg, 12.9 mol) in EtOH (79.4 L) was added iron powder (5.93 kg, 106.1 mol) at room temperature and the resulting mixture was heated to 75 ºC. 1 M HCl (7.9 L) was slowly added to the mixture over 1 h while maintaining the temperature at 70−80 ºC and the resulting mixture was refluxed for 3 h. After cooling to 30 ºC, THF (19.9 L) and 2 M NaOH (11.9 L) were added to the reaction mixture and the mixture was stirred for 0.5 h at 20−30 ºC. The iron powder was filtered and washed with THF (11.9 L). The combined filtrate was concentrated to 40 kg under reduced pressure. Water (19.9 L) was slowly added to the residue at 20−30 ºC and the resulting slurry was stirred for 1 h at the same temperature. The crystals were filtered, and washed successively with EtOH/water (1:2, 7.9 L) and water (7.9 L). The wet cake was dissolved in EtOH (31.8 L) at 80 ºC, followed by addition of activated carbon (0.32 kg) at the same temperature. After stirring for 0.5 h at 75−80 ºC, insoluble materials were filtered and washed with EtOH (7.9 L). The combined filtrate was cooled to 30 ºC and water (39.7 L) was slowly added to the mixture at 20−30 ºC. After confirming the crystallization, the resulting slurry was cooled to 10 ºC and aged for 1 h at 0−10 ºC. The crystals were filtered, washed with cooled EtOH/water (1:1, 7.9 L), and dried *in vacuo* at 50 ºC to give **8** as a pale-brown solid (2.90 kg, 81%). Purity: 99.6% (LCAP); ^{1}H NMR (300 MHz, $CDCl_3$): δ 3.74 (1H, br), 6.49 (1H, d, *J* = 7.6 Hz), 6.62 (1H, d, *J* = 7.7 Hz), 6.82 (1H, s), 7.01 (1H, d, *J* = 8.5 Hz), 7.35 (1H, t, *J* = 7.8 Hz), 7.57 (1H, d, *J* = 7.6 Hz), 9.10 (1H, s); MS: *m/z* = 277 $[M + H]^+$; Anal. Calcd for $C_{13}H_9N_2OSCl$: C, 56.42; H, 3.28; N, 10.12; S, 11.59; Cl, 12.81. Found: C, 56.33; H, 3.40; N, 10.04; S, 11.30; Cl, 12.66.

***tert*-Butyl (2-{[2-(4-chloro-5*H*-pyrrolo[3,2-*d*]pyrimidin-5-yl)ethyl]amino}-1,1-dimethyl-2- oxoethyl)-carbamate (15).** To a mixture of **9**[7a] (2.50 kg, 8.42 mol) in EtOH (6.3 L) was added 4M HCl in EtOAc

(6.3 L, 25.2 mol) while maintaining the temperature at below 40 ºC. After stirring for 2 h at 50 ºC, the resulting suspension was cooled to 25−30 ºC and stirred for 1 h. The solids were filtered, washed with EtOAc (5.0 L), and dried at 50 ºC under reduced pressure to give 2-(4-chloro-5*H*-pyrrolo[3,2-*d*]pyrimidin-5-yl)ethanamine **14** as a yellow solid. Triethylamine (2.35 L, 16.9 mol) and *N*,*N*-dimethylaminopyridine (1.03 kg, 8.43 mol) were added to the mixture of isolated **14** in MeCN (11.4 L) at room temperature. After stirring for 0.5 h at room temperature, **12** (2.06 kg, 10.1 mol) and EDC·HCl (1.94 kg, 10.1 mol) were successively added, and the reaction mixture was stirred at room temperature for 3 h. EtOAc (15.9 L) and water (6.8 L) were added to the reaction mixture, and the resulting biphasic mixture was adjusted pH to 2.5 with 2 M HCl (4.6 L). The organic layer was separated, and the aqueous layer was further extracted successively with EtOAc (11.4 L, 6.8 L). The combined organic layers were washed with 10% aqueous NaCl twice (11.4 L each). To the organic layer was added activated carbon (0.23 kg) and the resulting suspension was stirred for 0.5 h at room temperature. Activated carbon was removed by filtration and washed with EtOAc (4.5 L). The combined filtrate was concentrated under reduced pressure to remove the EtOAc. The residue was diluted with toluene (11.4 L), and concentrated under reduced pressure. After toluene (15.9 L) was added to the residue, *n*-heptane (9.1 L) was added dropwise to the slurry at room temperature. The resulting slurry was cooled to 0−10 ºC and aged for 1 h. The solids were filtered, washed with toluene/*n*-heptane (1:1, 4.5 L), and dried at 50 ºC under reduced pressure to give **15** as a white solid (2.51 kg, 78% over 2 steps from **9**).

14: Purity: 98.2% (LCAP); ^{1}H NMR (300 MHz, DMSO-d_6): δ 3.29−3.35 (2H, m), 4.85 (1H, t, *J* = 6.3 Hz), 6.82 (1H, d, *J* = 3.2 Hz), 8.22 (1H, d, *J* = 3.2 Hz), 8.56 (2H, br), 8.70 (1H, s), 11.52 (1H, br); ^{13}C NMR (126 MHz, DMSO-d_6): δ 45.82, 46.26, 102.31, 123.99, 140.05, 141.90, 149.26, 151.94; HRMS: *m/z* = calcd for $C_8H_9ClN_4$ $[M + H]^+$ 197.0589, found 197.0585; mp 237 ºC.

15: Purity: 99.4% (LCAP); ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.42 (9H, s), 1.44 (6H, s), 3.66−3.71 (2H, m), 4.66 (2H, t, *J* = 5.6 Hz), 6.68 (1H, d, *J* = 3.2 Hz), 6.73 (1H, br), 7.61 (1H, d, *J* = 3.1 Hz), 8.67 (1H, s); ^{13}C NMR (126 MHz, DMSO-d_6): δ 25.70, 28.26, 40.99, 48.26, 56.05, 78.41, 101.69, 123.41, 139.99, 141.49, 149.46, 152.75, 154.63, 175.22; MS: *m/z* = 382 $[M + H]^+$; Anal. Calcd for $C_{17}H_{24}N_5O_3Cl$: C, 53.47; H, 6.33; N, 18.34; Cl, 9.28. Found: C, 53.55; H, 6.49; N, 18.36; Cl, 9.14; mp 146 ºC.

***N*-[2-(4-{[4-(1,2-Benzisothiazol-4-yloxy)-3-chlorophenyl]amino}-5*H*-pyrrolo[3,2-*d*]pyrimidin-5-yl)-ethyl]-2-methylalaninamide (1).**[2] A mixture of **8** (1.15 kg, 4.16 mol), **15** (1.75 kg, 4.58 mol) and EtOH (5.8 L) was stirred at 70 ºC for 6 h. Then 4 M HCl (2.08 L, 8.32 mol) was added dropwise over 0.5 h while maintaining the temperature at 65−70 ºC. After stirring for 2 h at 70 ºC, to the resulting solution was added 25% aqueous ammonia (2.7 L) dropwise. Water (1.0 L) was added to the solution, and cooled to room temperature. The resulting slurry was stirred at room temperature for 1 h. The solids were filtered

with suction, washed with 50% aqueous ethanol (3.5 L) and dried at 50 ºC under reduced pressure to give crude **1** as a white solid (1.89 kg, 87%).

Recrystallization of 1.[2] Crude **1** (1.89 kg) was dissolved in EtOH/water (4:1, 11.3 L) at 80 ºC. The insoluble materials were removed by filtration and washed with EtOH/water (4:1, 3.8 L). To the filtrate was added water (9.1 L) dropwise while maintaining the temperature at 60 ºC. The resulting slurry was stirred at 60 ºC for 1 h and 25−30 ºC for 1 h. The solids were filtered, washed with EtOH/water (1:1, 3.8 L), and dried at 50 ºC under reduced pressure to give **1** as a white solid (1.75 kg, 93%). Purity 99.5% (LCAP); ^{1}H NMR (300 MHz, DMSO-d_6): δ 1.13 (6H, s), 1.84 (2H, br), 3.46 (2H, t, J = 6.2 Hz), 4.55 (2H, t, J = 6.2 Hz), 6.51 (1H, d, J = 2.9 Hz), 6.62 (1H, d, J = 7.7 Hz), 7.39 (1H, d, J = 8.9 Hz), 7.54 (1H, t, J = 8.0 Hz), 7.61 (1H, d, J = 2.5 Hz), 7.88−7.93 (2H, m), 8.13 (1H, s), 8.19 (1H, br), 8.36 (1H, s), 8.89 (1H, br), 9.25 (1H, s); Anal. Calcd for $C_{25}H_{24}N_7O_2ClS$: C, 57.52; H, 4.63; N, 18.78; Cl, 6.79; S, 6.14. Found: C, 57.33; H, 4.69; N, 18.75; Cl, 6.75; S, 6.05.

4-(1,2-Benzisothiazol-4-yloxy)-3-chloroaniline (8, alternative synthesis).[2] To a solution of **16** (5.0 g, 34.8 mmol) in THF (90 mL) was added *t*-BuOK (4.10 g, 36.5 mmol) at room temperature. After heating to 60 ºC, a solution of **17** (5.80 g, 36.6 mmol) in THF (10 mL) was added to the mixture at 60 ºC, and stirred for 2 h at 60 ºC. The resulting solution was cooled to room temperature, diluted with toluene (50 mL), and washed successively with water (50 mL) and 10% aqueous NaCl (50 mL). The organic layer was concentrated under reduced pressure. To the solution of residue in DMAC (75 mL) were added sulfur (2.23 g, 69.6 mmol), NaOAc (8.58 g, 104.6 mmol) and NH_4Cl (5.58 g, 104.3 mmol) at room temperature. After stirring for 4 h at 80 ºC, the resulting solution was cooled to room temperature, diluted with EtOAc (75 mL), and washed successively with 5% aqueous Na_2CO_3 (75 mL), water (75 mL × 2). The organic layer was concentrated under reduced pressure. To the solution of residue in EtOH (40 mL) was added activated carbon (0.5 g) and the mixture was stirred for 0.5 h. The insoluble material was filtered and washed with EtOH (10 mL). The filtrate was heated to 50 ºC and sulfuric acid (1.86 mL, 34.9 mmol) was added dropwise at 50 ºC. After removing generated gum-like mass by decantation, the solution was cooled to precipitate out. The resulting suspension was stirred at room temperature for 1 h and at 5 ºC for 1 h. The solid was filtered, and washed with cooled EtOH (10 mL). The suspension of the resulting wet cake in EtOH (46.2 mL) and water (19.8 mL) was heated to 80 ºC. To the resulting solution was added activated carbon (0.66 g) and the resulting mixture was stirred for 0.5 h at 80 ºC. The insoluble materials were filtered and washed with EtOH/water (2:1, 10 mL). The filtrate was heated to 50 ºC and adjusted pH to 7.0 with 25% aqueous ammonia. To the resulting slurry was added water (28.6 mL) dropwise at 50 ºC and the slurry was aged at room temperature for 1 h. The solids were collected by filtration, washed with

EtOH/water (1:1, 13.2 mL), and dried at 50 ºC under reduced pressure to give **8** as a white solid (4.15 g, 42%). Purity 98.4% (LCAP); ^{1}H NMR (300 MHz, $CDCl_3$): δ 3.74 (1H, br), 6.49 (1H, d, *J* = 7.6 Hz), 6.62 (1H, d, *J* = 7.7 Hz), 6.82 (1H, s), 7.01 (1H, d, *J* = 8.5 Hz), 7.35 (1H, t, *J* = 7.8 Hz), 7.57 (1H, d, *J* = 7.6 Hz), 9.10 (1H, s).

ACKNOWLEDGEMENTS

We thank Dr. David Cork for his advice on the manuscript.

REFERENCES AND NOTES

1. (a) K. G. Petrov, Y. -M. Zhang, M. Carter, G. S. Cockerill, S. Dickerson, C. A. Gauthier, Y. Guo, R. A. Mook, Jr., D. W. Rusnak, A. L. Walker, E. R. Wood, and K. E. Lackey, *Bioorg. Med. Chem. Lett.*, 2006, **16**, 4686; (b) A. Ooi, T. Takehana, X. Li, S. Suzuki, K. Kunitomo, H. Iino, H. Fujii, Y. Takeda, and Y. Dobashi, *Mod. Pathol.*, 2004, **17**, 895; (c) H.-R. Tsou, E. G. Overbeek-Klumpers, W. A. Hallett, M. F. Reich, M. B. Floyd, B. D. Johnson, R. S. Michalak, R. Nilakantan, C. Discafani, J. Golas, S. K. Rabindran, R. Shen, X. Shi, Y.-F. Wang, J. Upeslacis, and A. Wissner, *J. Med. Chem.*, 2005, **48**, 1107; (d) F. A. L. M. Eskens, C. H. Mom, A. S. T. Planting, J. A. Gietema, A. Amelsberg, H. Huisman, L. van Doorn, H. Burger, P. Stopfer, J. Verweij, and E. G. E. de Vries, *Br. J. Cancer*, 2008, **98**, 80.
2. (a) Y. Kawakita, H. Banno, T. Ohashi, T. Tamura, T. Yusa, A. Nakayama, H. Miki, H. Iwata, H. Kamiguchi, N. Tanaka, N. Habuka, S. Sogabe, Y. Ohta, and T. Ishikawa, *J. Med. Chem.*, 2012, **55**, 3975; (b) T. Ishikawa and Y. Kawakita, WO/2009/113560 A1, 2009.
3. S. L. Boulet, S. A. Filla, P. T. Gallagher, K. J. Hudziak, A. M. Johansson, R. E. Karanjawala, J. J. Masters, V. Matassa, B. M. Mathes, R. E. Rathmell, M. A. Whatton, and C. N. Wolfe, WO2004/043903 A1, 2004.
4. (a) T. W. Greene and P. G. M. Wuts, 'Protective Groups in Organic Synthesis', 4th ed., Wiley, New York, 2007, pp. 372−382; (b) D. G. Melillo, R. D. Larsen, D. J. Mathre, W. F. Shukis, A. W. Wood, and J. R. Colleluori, *J. Org. Chem.*, 1987, **52**, 5143; (c) T. A. Grese, M. D. Adrian, D. L. Phillips, P. K. Shetler, L. L. Short, A. L. Glasebrook, and H. U. Bryant, *J. Med. Chem.*, 2001, **44**, 2857.
5. A. Y. Solovyev, D. A. Androsov, and D. C. Neckers, *J. Org. Chem.*, 2007, **72**, 3122.
6. The dihydrochloride salt did not improve the quality, and the yield of monooxalate salt was lower due to its higher solubility in common solvents. Solubilities of **8** monooxalate and **8** monosulfate in common solvents at 25 ºC are as follows: monooxalate, EtOH, 5.1 wt%, EtOH/water (95:5), 3.4 wt%, 2-propanol, 2.7 wt%, acetone, 5.5 wt%, THF, 11.9 wt%; monosulfate, EtOH 0.3 wt%, EtOH/water (95:5), 0.8 wt%, 2-propanol, 0.1 wt%, acetone, 0.1 wt%, THF, <0.1 wt%.

7. (a) T. Ishikawa, M. Seto, H. Banno, Y. Kawakita, M. Oorui, T. Taniguchi, Y. Ohta, T. Tamura, A. Nakayama, H. Miki, H. Kamiguchi, T. Tanaka, N. Habuka, S. Sogabe, J. Yano, K. Aertgeerts, and K. Kamiyama, *J. Med. Chem.*, 2011, **54**, 8030; (b) T. Doi, H. Takiuchi, A. Ohtsu, N. Fuse, M. Goto, M. Yoshida, N. Dote, Y. Kuze, F. Jinno, M. Fujimoto, T. Takubo, N. Nakayama, and R. Tsutsumi, *Br. J. Caner*, 2012, **106**, 666.