Development of a Scalable Synthesis of a VEGFR Inhibitor

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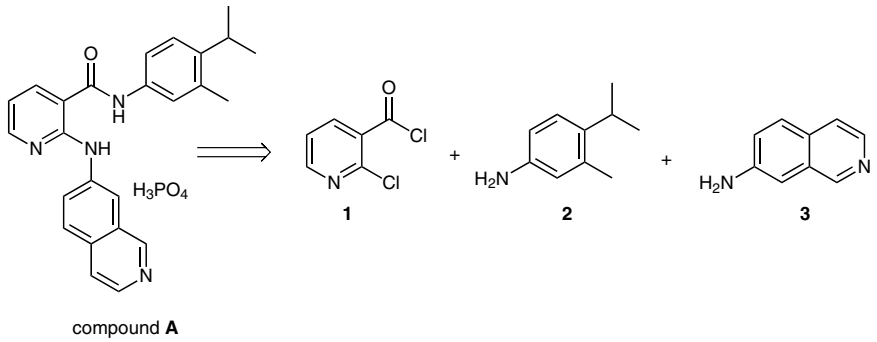
Abstract: Process development and salt selection for a novel VEGFR inhibitor are described. The overall convergent synthesis involved coupling of three key fragments, 2-chloronicotinoyl chloride, 4-isopropyl-3-methylaniline and 7-aminoisoquinoline. A cost-effective and scalable synthesis of 7-aminoisoquinoline was also achieved. A transition-metal-free S_NAr process enabled the final C–N coupling to afford the target molecule. A phosphate form of the drug substance with improved physical properties was selected for further development and the corresponding crystallization process was subsequently developed. Overall, a robust six-step route was developed and demonstrated on multikilogram scales affording the target compound in >30% yield and high purity (>99%).

Key words: amination, oxidation, amides, VEGFR inhibitor, 7-aminoisoqinoline

Vascular endothelial growth factor (VEGF) is an important signaling protein involved in both vasculogenesis (the formation of the circulatory system) and angiogenesis (the growth of blood vessels from pre-existing vasculature).[1] VEGFR inhibitors are in broad use for the treatment of metastatic renal-cell carcinoma, gastrointestinal stromal tumors and hepatocellular carcinoma and in development for a number of other oncology indications, including colorectal cancer, non-small-cell lung cancer, pancreatic cancer, thyroid malignancies, ovarian cancer, breast cancer and sarcomas.[2] Recent efforts at Amgen led to the discovery of the potent VEGFR inhibitor, compound **A**.[3] To supply toxicological and clinical studies, the development of a scalable and cost-effective synthesis of compound **A** was required.

The retrosynthetic analysis of **A** is outlined in Scheme 1. Amidation and amination were envisioned as the key late-stage steps, bringing together three fragments (**1**, **2** and **3**). 2-Chloronicotinoyl chloride (**1**) and 4-isopropyl-3-methylaniline (**2**) were both commercially available with reasonable cost. Our efforts initially focused on the development of a scalable and cost-effective process for key intermediate **3**. The coupling of these intermediates and controlling the level of process impurities posed a number of challenges for large-scale synthesis. This paper will describe how these challenges were addressed during the development of a robust process for preparing a pharmaceutically acceptable salt form of compound **A**.

The medicinal chemistry approach[3] to the key intermediate **3** is shown in Scheme 2. The synthesis began with protection of amino group of 4-nitrophenethylamine **4** as the corresponding amide **5**. Tetrahydroisoquinoline **6** was then prepared through a Pictet–Spengler[4] tetrahydroisoquinoline protocol by condensation of **5** with paraformaldehyde under strong acidic conditions. Cleavage of trifluoroacetamide protecting group followed by one-pot dehydrogenation and nitro group reduction under high temperature and microwave irradiation furnished fragment **3**. This route was successfully utilized to provide 10–20 grams of **3** to support early development work. However, it was recognized that this route would be unsuitable for long-term supply due to challenges associated with scaling up a microwave reaction and high cost of the starting material 4-nitrophenethylamine **4** at the outset of this development work. Moreover, not all the literature



compound **A**

Scheme 1 General strategy towards the synthesis of compound **A**

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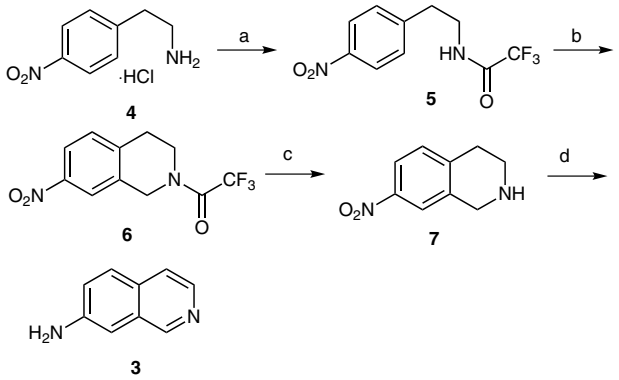
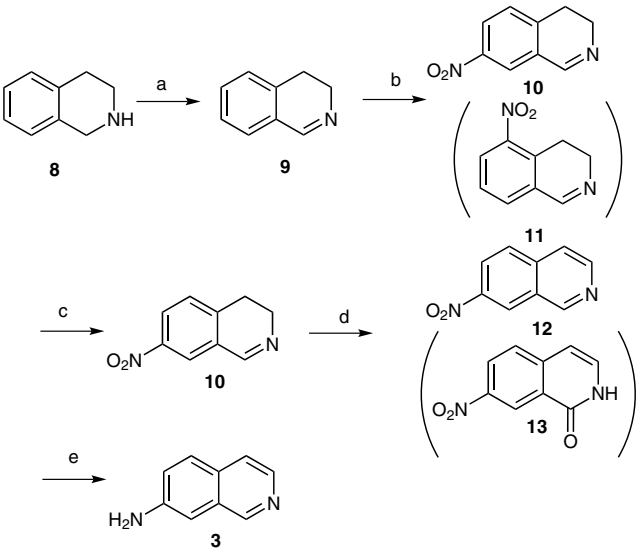
approaches[5] were deemed efficient and cost-effective since they suffered either expensive raw materials or multiple chromatography purifications, undesirable on large scale. With this in mind, we started to investigate a number of alternative approaches that could make use of economic raw materials and synthesize **3** by a more efficient route with an acceptable cost.



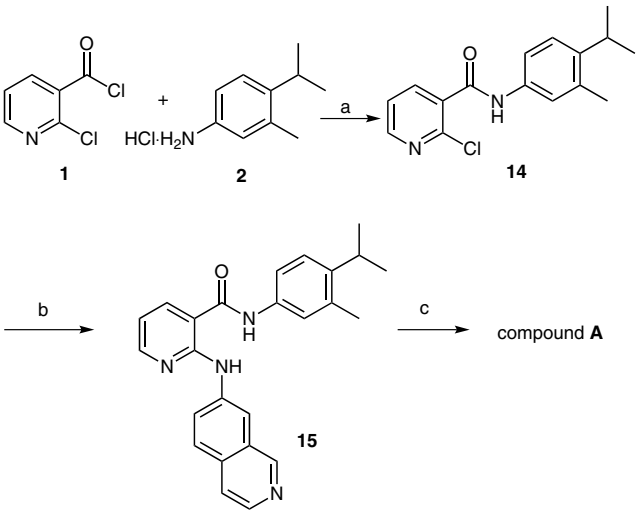
Scheme 2 Original synthesis of **3**. *Reagents and conditions*: (a) $(CF_3CO)_2O$–DIPEA, 99%; (b) $(CH_2O)_n$, AcOH, H_2SO_4, 90%; (c) LiOH, aq THF, 95%; (d) microwave, 220 °C, $(HOCH_2CH_2)_2O$, Pd/C, 76%.

An improved and practical approach to 7-aminoisoquinoline (**3**) was developed. As shown in Scheme 3, the synthesis began with partial oxidation of a low-cost starting material, 1,2,3,4-tetrahydroisoqinoline (**8**) to provide dihydroisoquinoline **9** in 98% yield. The subsequent nitration was carried out with potassium nitrate in sulfuric acid to provide a mixture of regioisomers **10** and **11** in 13:1 ratio.[6] Since no improvement in the ratio was observed by optimizing the reaction conditions, we decided to explore an isolation process to reject the undesired isomer. To our delight, formation of the toluenesulfonic acid salt of **10** provided an effective control point for this purification purpose. In practice crystallization of TsOH salt of **10**, salt break and crystallization afforded **10** in 75% overall yield (steps b and c) with >99% purity. A screen of the subsequent oxidation conditions revealed that the highest assay yield and cleanest reaction profile was obtained using MnO_2 as the oxidant. However, 5% amide impurity **13** was consistently formed during this reaction and proved difficult to remove in downstream processing. Attention then turned to identifying a solvent system where **12** and **13** would have very different solubility and oxidation reaction can still proceed smoothly. Further studies showed trifluoromethylbenzene met our requirements. Thus, the insoluble amide impurity **13** produced in the reaction was conveniently removed via in-line filtration, and the product **12** was obtained with >98% purity and 85% yield. The final reduction of nitro group was carried out by hydrogenation in the presence of 1% Pd/C catalyst and the addition of 10% AcOH was critical to improve the substrate solubility and reaction kinetic rate. Ultimately this four-step synthesis was successfully demonstrated on multi-kilogram scales with 54% overall yield and >99% purity.



Scheme 3 Improved synthesis of **3**. *Reagents and conditions*: (a) NBS, NaOH, CH_2Cl_2; (b) KNO_3, H_2SO_4; (c) (1) TsOH, *i*-PrOH; (2) aq NaOH, 75%; (d) MnO_2, $PhCF_3$, 85%; (e) H_2, Pd/C, EtOH, 76%.

The final fragments assembling process began with amidation of 2-chloronicotinoyl chloride (**1**) with 4-isopropyl-3-methylaniline (**2**). Among all the solvents tested, dichloromethane provided the best assay yield (>99%) and full conversion was observed within 30 minutes even at 5–10 °C probably due to the higher solubility of both **1** and **2** in CH_2Cl_2. The coupling product **14** was isolated by crystallization from MTBE–heptanes with >99% purity and 98% yield (Scheme 4).



Scheme 4 Final coupling steps. *Reagents and conditions*: (a) Et_3N, CH_2Cl_2, 98%; (b) **3**, LiHMDS, THF, 79%; (c) H_3PO_4, EtOH, 86%.

The Pd-catalyzed cross-coupling of aryl chloride and aniline has rapidly emerged as a popular method for the construction of C–N bonds in contemporary organic synthesis.[7] Indeed, the original medicinal chemistry synthesis was carried out using 5 mol% $Pd_2(dba)_3$/2-dicyclohexylphosphino-2′-(*N*,*N*-dimethylamino)biphenyl

(DavePhos) in THF at 65 °C affording compound **15** in 55% isolated yield after chromatography purification. The low yield was mainly attributed to incomplete reaction. Additionally a more critical issue identified was the Pd removal after the coupling reaction. The residual Pd in the final product **A** remained high (>100 ppm) even after several treatments, using different solid scavengers, during the workup of the coupling reaction. In addition, the Pd levels cannot be further reduced in the following salt formation step. With the concern of intrinsic binding capability of final product to Pd, we turned our attention to develop a transition-metal-free S_NAr reaction to build the C–N bond. Extensive base and solvent screening revealed that treatment of **3** with 3.0 equivalents of LiHMDS in THF, followed by addition of aryl chloride **14**, provided >99% conversion. However varied amounts (0.5–3%) of an impurity (**17**) with $m/z = 283$ (Scheme 5)[8] was found in many development lots and rejection of this impurity proved to be extremely difficult in this step and the following phosphate formation step under a variety of crystallization conditions. Therefore, in an attempt to completely avoid its formation during the reaction, we decided to investigate the mechanism of this impurity formation. On the basis of the literature example[9] that the imino radical can be formed in *tert*-butoxide-catalyzed autoxidation of 1(or 2)-aminonaphthalene, an oxidative coupling mechanism was proposed in Scheme 5. The fact that this impurity can be prepared in ca. 60% isolated yield by sparging air into the reaction mixture would serve as additional evidence in support of this mechanism. Ultimately the level of this impurity can be decreased to <0.1% by implementing a rigorous degassing procedure prior to adding the base LiHMDS. After quenching the reaction with water and solvent-switch to MeOH, the free base **15** was isolated by crystallization from MeOH–H_2O (2:1). Using this process, a multikilogram-scale preparation of free base **15** was achieved in 79% yield and >99% purity.

A free base was initially used by the medicinal chemistry team to enable screening and preliminary toxicology studies, but the poor stability, bioavailability and solubility of this form precluded its use for the further development. After extensive high throughput and subsequent manual salt and polymorph screenings, a more crystalline, stable and water-soluble phosphate salt was identified for the long-term development. A robust crystallization was subsequently designed on the basis of solubility of the free base and salt. In practice, 1.1 equivalents of H_3PO_4 was slowly added to a free-base solution in EtOH at 80 °C and 1% seed was added to the reaction mixture to induce crystallization. The resulting salt was isolated by filtration in 86% yield and >99% purity.

In summary, a non-chromatography and robust chemical process to synthesize a VEGFR inhibitor was developed to support the preclinical and clinical studies. Process research efforts also led to the development of a practical and cost-effective synthesis of 7-aminoisoquinoline (**3**). The whole process was demonstrated on multikilogram scales with >30% overall yield and >99% purity.

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Supporting Information for this article is available online at http://www.thieme-connect.com/ejournals/toc/synlett.

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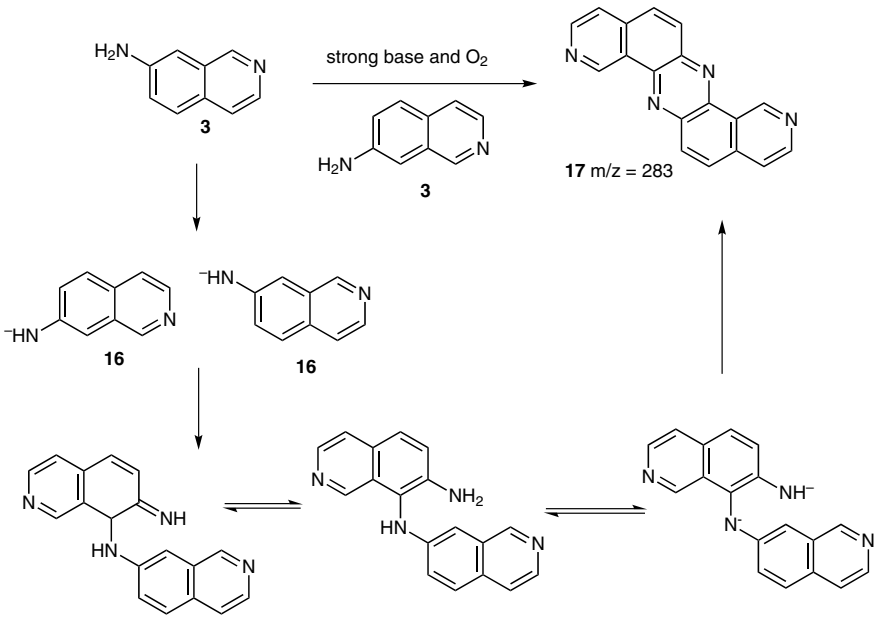

Scheme 5 Proposed mechanism for the formation of **17** with $m/z = 283$

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