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Technical Note

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A Novel Process for Antimalarial Drug Pyronaridine Tetraphosphate

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Supporting Information

ABSTRACT: A novel process for preparation of pyronaridine tetraphosphate, an antimalarial drug substance, is reported. The overall yields are 54% and >99.8% (including five chemical steps). Formation and control of possible impurities are also described.

INTRODUCTION

Pyronaridine tetraphosphate (Figure 1) was first synthesized in the 1970s and has been extensively used in China as an antimalarial monotherapy, showing good activity and tolerability. Owing to the drug's high antimalarial activity, safety, tolerability, stability, and excellent water solubility, the World Health Organization plans to complete preclinical and clinical trials with the aim of replacing chloroquine with pyronaridine as the first line of treatment for malaria, particularly in Africa.[1]

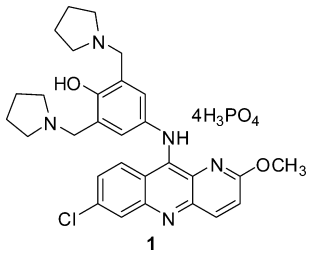

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Figure 1. Structure of pyronaridine tetraphosphate **1**.

The reported synthesis route is a linear synthetic strategy (as shown in Scheme 1).[2] The synthesis starts from the reaction of two compounds, 2,4-dichlorobenzoic acid (**1**) and 6-methoxypyridin-3-amine (**2**), coupled under the mediation of CuO to obtain 4-chloro-2-(6-methoxypyridin-3-ylamino)benzoic acid (**3**).[3] Compound **3** undergoes cyclization and chlorination by refluxing in $POCl_3$ to produce 7,10-dichloro-2-methoxybenzo-[*b*][1,5]naphthyridine (**4**), followed by SN_{Ar} reaction with aminophenol, Mannich reaction, and salt with orthophosphoric acid to obtain the final product.[4] However, no data on the purity of the final product have been revealed in the literature. Further duplication of these experiments at our laboratory resulted in the formation of impurity I (Figure 2) due to the overreaction of the Mannich reaction, which is very difficult to eliminate from the final product without affecting the yield.

Thus, developing a process for the preparation of pyronaridine tetraphosphate that can control the formation of impurities and does not require tedious purification techniques in obtaining the desired purity is necessary.

RESULTS AND DISCUSSION

An improved process for preparation of pyronaridine tetraphosphate, which is free from associated impurities and does not make use of column chromatography or other tedious purification methods, is reported.

A convergent synthesis route was designed as shown in Scheme 2, wherein the two key building blocks, 4-amino-2,6-bis(pyrrolidin-1-ylmethyl) phenol **12** and 7,10-dichloro-2-methoxybenzo[*b*][1,5]naphthyridine **4**, are combined in a one-step reaction.[5]

Preparation of 7,10-Dichloro-2-methoxybenzo[*b*]-[1,5]naphthyridine (4). The Ullman reaction mediated by CuO proceeded smoothly with very high regioselectivity. However, an impurity was produced during the steps of cyclization and chlorination (Scheme 3).

Impurity IV could further react with **12**, and the resulting impurity in the next step cannot be removed effectively due to the close R_f values and the relative retention time of 0.95. Different conditions were tried to eliminate the impurity but failed. We concluded that the impurity was produced via an intermediate (as shown in Scheme 4) under acidic conditions. Thus, a base had to be used to scavenge the HCl formed in the $POCl_3$ and in the reaction, and Et_3N was chosen as the most suitable base in the process. Qualified compound **4** was obtained after the reaction was worked up, filtered, and purified in toluene.

Preparation of 4-Amino-2,6-bis(pyrrolidin-1-ylmethyl)phenol (12). The temperature and sequence of the start of material addition are very important in Mannich reactions. Primarily, the reaction was refluxed in ethanol, where compound **10** remained, and a single substituted impurity was produced in the reaction. Additional amounts of pyrrolidine and paraformaldehyde did not resolve the problem.

In the next step, the reaction proceeded in pyrrolidine without the solvent because *p*-nitrophenol is very soluble in pyrrolidine. The synthesis progressed completely, albeit with an intense uncontrolled exothermic reaction, which is deemed unsafe during scale up. Other solvents were screened, such as

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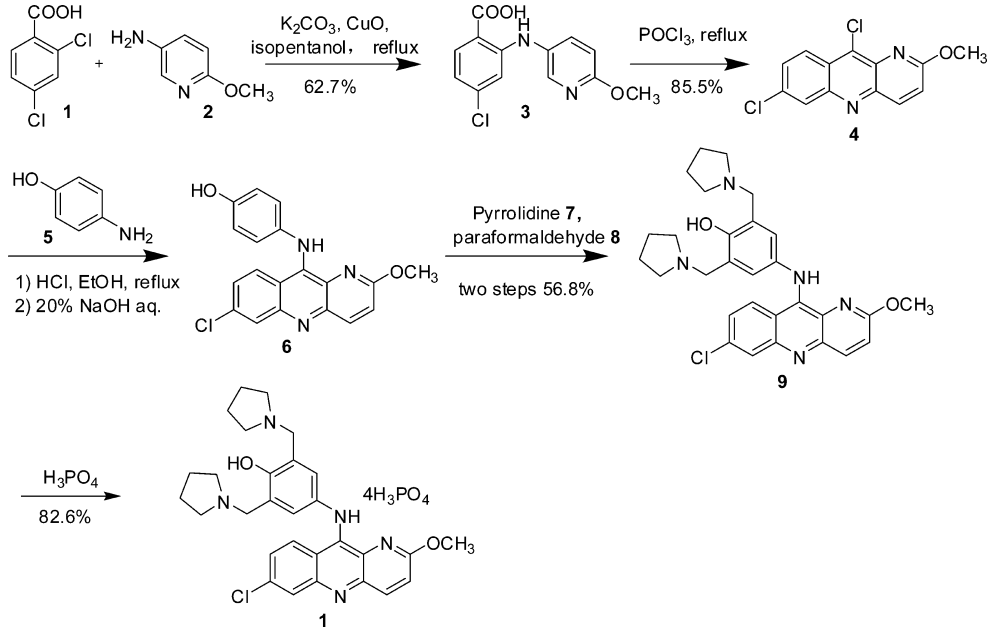
Scheme 1. Reported synthesis route



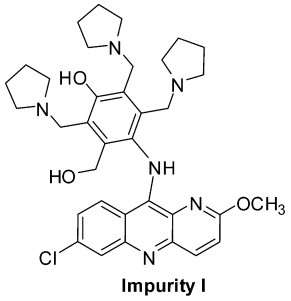
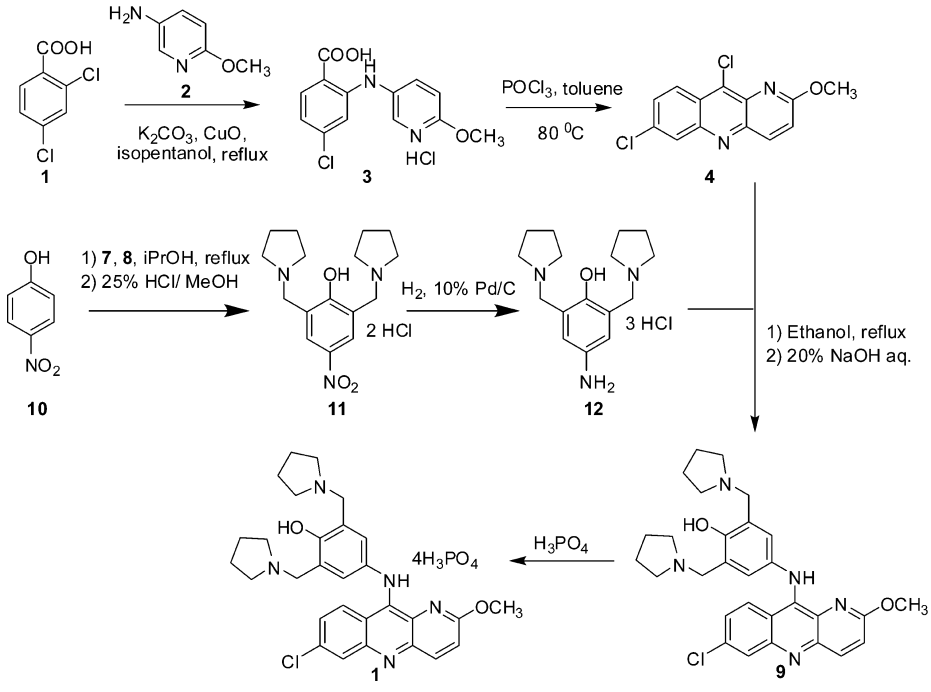


Figure 2. Formation impurity in the final product.

methanol and isopropanol. The reaction works smoothly in isopropanol without sudden heat production, and the starting material *p*-nitrophenol was totally consumed. Sequence of raw material was also considered. The addition of pyrrolidine to paraformaldehyde and nitrophenol suspension in isopropanol was regarded as the safest addition sequence.

Although the purity of the reaction was greatly improved by reaction condition optimization, there was a multisubstituted impurity (Figure 3) in the reaction, which was further converted to impurity I. Removal of impurity I could not be achieved after its formation, so this impurity must be removed at this step in order to obtain the qualified API. Because

Scheme 2. Convergent synthesis route



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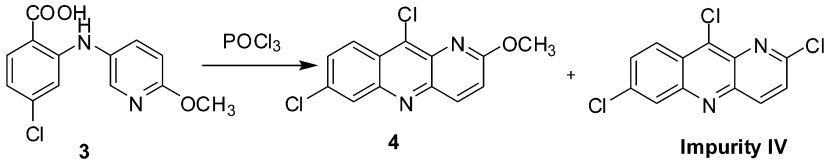
Scheme 3



Scheme 4

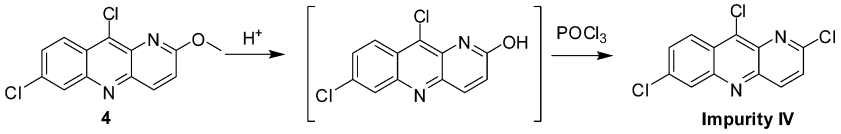


Table 1. Screening of Solvent and Reaction Temperature of the Mannich Reaction

entry	solvent	temp (°C)	impurity II[a] (%)	impurity III[a] (%)	product[a](%)	*p*-nitrophenol[a] (%)
1	MeOH	65	38.64	0.50	47.74	4.26
2	EtOH	75	8.00	0.67	81.76	0.41
3	pyrrolidine	85	3.98	0.11	62.40	ND
4	*i*-PrOH	75	2.53	1.21	75.15	1.25
5	*i*-PrOH	85	2.92	0.10	90.15	ND
6	*i*-PrOH	95	3.01	0.10	89.25	ND

[a]Purity measured by HPLC.

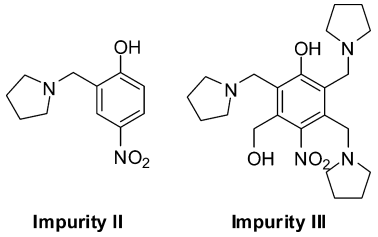


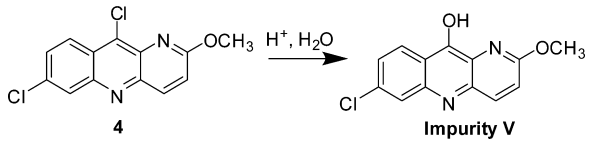
Figure 3. Formation impurities in the synthesis of compound **II**.

compound **11** is oil, we have to solidify it for the convenience of recrystallization. Several salts, such as phosphoric acid, hydrochloric acid, oxalic acid, and sulfuric acid, were screened in order to get a better solid. Finally we found that the HCl salt of compound **11** was a light-yellow solid, the purity of the HCl salt of compound **11** could be improved to >99.8%, and a single impurity could be reduced to <0.1% after recrystallization.

After high-quality compound **11** was obtained, we examined the catalytic hydrogenation conditions. 2,6-Substituted aminophenol **12** is sensitive to air, but its HCl salt is much more stable than its isolated base. Thus, an equimolar concentration of HCl aqua solution was added to the solution to substantially improve reaction rates. No impurity was produced in this step.

Preparation of Pyronaridine Tetraphosphate (1). The last step is SN_{Ar} reaction. Generally, this kind of reaction works under basic conditions. Interestingly, this reaction proceeds under acidic conditions. HCl or H_2SO_4 aqua solution addition to the solvent would produce an impurity (impurity V) (Scheme 5). We speculated that the HCl salt of compound **12** could promote the reaction work without additional acid, as we expected that the reaction worked well with a molecular ratio of 1:1 of the two starting materials. After the reaction was completed, the HCl salt of compound **9** was filtered and isolated with 20% NaOH aqua solution and then mixed with orthophosphoric acid to obtain pyronaridine tetraphosphate. The phosphoric acid salt was slurried in refluxed 75% ethanol

Scheme 5



to obtain a qualified active pharmaceutical ingredient (API) with purity of >99.8% and single impurity of less than 0.1%.

On the basis of the optimized conditions, the batch was carried out on 50 kg scale, and the results were reproduced at the plant level.

CONCLUSION

We have successfully developed a novel, commercially viable manufacturing process for pyronaridine tetraphosphate with high purity and satisfactory yield. The impurities produced in this process were well controlled and removed.

EXPERIMENTAL DETAILS

General Methods. All reagents were purchased from Sinopharm, and the purities of these reagents are greater than 99.0%. NMR spectra of the intermediates were recorded on a Bruker 300 NMR and 400 NMR spectrometer using TMS as an internal standard. EI−MS spectra were obtained on a Finnigan MAT 95 mass spectrometer, and ESI−MS spectra were obtained on a Kratos MS 80 mass spectrometer. Purity analysis was carried out on an HPLC (Agilent 1200 Series RRLC, Agilent technologies) using an optima PAK C18 (5 mm; 4.6150 mm) column. HPLC solvents consisted of potassium phosphate buffer:acetonitrile (9:1) (solvent A) and potassium phosphate buffer:acetonitrile (2:8) (solvent B). One liter of the buffer (pH 2) consisted of KH_2PO4 (1.36 g) and 1-octanesulfonic acid sodium salt hydrate (2.163 g). The flow rate was maintained at 1 mL/min, and the column temperature was maintained at 40 °C.[6]

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4-Chloro-2-(6-methoxypyridin-3-ylamino)benzoic Acid (3). The suspension of 2,4-dichlorobenzoic acid (7.16 kg, 37.5 mol) **1**, 6-methoxypyridin-3-amine (5.0 kg, 40.3 mol) **2**, potassium carbonate (2.65 kg, 19.0 mol), and CuO (30.0 g, 0.38 mol) in 20 L isopentanol was refluxed at 128−130 °C, with the production of CO_2 gas. The reaction was cooled to 100 °C after 10 h and quenched with 7 L water. Then, the pH of the solvent was adjusted to 11 with 3 L of 10% NaOH aqua solution at the same temperature. The obtained solution was cooled to 40−50 °C and filtered to remove the copper salt. The filtrate was adjusted to pH 3 with 6 N HCl to obtain a grey precipitate. The precipitate was slurried with 30 L hexane; then the product was obtained by filtration and dried to a water content of less than 0.3% to afford a grey solid **3** (8.62 kg, 82.5% yield, 97.45% purity), mp 198−200 °C; ^{1}H NMR (300 MHz, DMSO) δ 13.11 (s, 1H, br), 9.48 (s, 1H), 8.13 (d, J = 2.3 Hz, 1H), 7.88 (d, J = 8.5 Hz, 1H), 7.70 (dd, J = 8.7, 2.7 Hz, 1H), 6.90 (d, J = 8.7 Hz, 1H), 6.76 (dd, J = 8.5, 2.0 Hz, 1H), 6.69 (d, J = 2.0 Hz, 1H), 3.88 (s, 3H). Anal. Calcd for $C_{13}H_{11}ClN_2O_3$·HCl: C, 49.54; H, 3.84; N, 8.89; Found: C, 49.60; H, 3.80; N, 8.95. MS (ESI, m/z) 279 ([M + H]$^+$, 100%), 235 ([M − 44] $^+$, 15%).

7,10-Dichloro-2-methoxybenzo[*b*][1,5]naphthyridine (4). $POCl_3$ (8.9 kg, 58.0 mol) was slowly added to the suspension of 4-chloro-2-(6-methoxypyridin-3-ylamino)benzoic acid **3** (4.0 kg, 14.3 mol) and Et_3N (6.4 kg, 63.4 mol) in 20 L toluene under N_2 atmosphere at 8−10 °C. Then, the reaction temperature was heated to reflux for 4 h. The solvent was cooled to 10 °C and slowly transferred to another vessel with 10 L ice water, while keeping the temperature below 10 °C. The pH of the solvent was adjusted to 8−9 with 25% NaOH aqua solution at 10 °C. Precipitates were collected and slurried in 5 L toluene at 70 °C; then after the solvent was cooled to 0−10 °C, the solid was filtered and dried at 65 °C for 4 h as a grey solid **4** (3.8 kg, 95% yield, 99.10% purity), mp 185−186 °C; ^{1}H NMR (400 MHz, DMSO) δ 8.43 (d, J = 9.3 Hz, 2H), 7.83 (dd, J = 9.2, 2.0 Hz, 1H), 7.51 (d, J = 9.2 Hz, 1H), 4.15 (s, 3H). Anal. Calcd for $C_{13}H_8Cl_2N_2O$: C, 55.94; H, 2.89; N, 10.04. Found: C, 55.90; H, 2.84; N, 10.10. MS (ESI, m/z) 279 ([M + H]$^+$, 100%).

4-Nitro-2,6-bis(pyrrolidin-1-ylmethyl)phenol (11). Pyrrolidine **7** (10.0 kg, 148.0 mol) was added to a suspension of *p*-nitrophenol **10** (3.95 kg, 28.4 mol) and paraformaldehyde **8** (4.56 kg, 152.0 mol) in isopropanol (15 L) at 20−30 °C over 1 h. Then, the reaction temperature was raised to 50 °C until the solvent became clear, after which the temperature was raised to 85−90 °C for 3 h. The solvent was evaporated to dryness under reduced pressure, and the obtained yellow oil was dissolved in 10 L DCM. The organic solvent was washed with water (2 × 5 L), and the water phase was combined and extracted with DCM 5 L. The organic phases were combined and removed in vacuo to obtain a yellow oil. The yellow oil was dissolved in isopropanol (16 L) and cooled to 5 °C. A 2 L 23% HCl/MeOH solution was added to the isopropanol solution and stirred for 3 h until a light-yellow precipitate was produced from the solvent. The solid was filtered and washed with isopropanol (1 L) to obtain a light-yellow solid weighing 11.6 kg. The solid was recrystallized in 30 L methanol to furnish the compound as a light-yellow solid **11** (8.8 kg, 82.0% yield, 99.88% purity by HPLC), mp 220−221 °C; ^{1}H NMR (300 MHz, DMSO) δ 8.56 (s, 2H), 4.89 (s, 2H, br), 4.60 (s, 4H), 3.61 (m, 8H), 2.13 (m, 8H). Anal. Calcd for $C_{16}H_{23}N_3O_3$·2HCl: C, 50.80; H, 6.66; Cl, 18.74; N, 11.11. Found: C, 50.83; H, 6.72; N, 11.09. MS (ESI, m/z) 306 ([M + H]$^+$, 100%).

4-Amino-2,6-bis(pyrrolidin-1-ylmethyl)phenol (12). Up to 10% Pd/C (495.0 g, 70% water) was added to a solution of compound **11** (5.0 kg, 13.0 mol) and 250 mL concentrated HCl aqua solution in 25 L methanol. The mixture was stirred under H_2 atmosphere under 3 bar pressure at 25 °C. The reaction was terminated after 5.5 h. The reaction mixture was filtered, and the filtrate was concentrated to produce clear oil. Subsequently, the oil was solidified in the mixture solution (18.2 L of 95% ethanol and 60.7 L of ethyl acetate), followed by filtration and drying at 60 °C for 4 h to afford a white solid (4.98 kg, 97.9% yield, 98.68% purity by HPLC), mp 190−191 °C; ^{1}H NMR (400 MHz, DMSO) δ 10.57 (s, 2H, br), 7.62(s, 1H), 7.60(s, 1H), 4.49 (s, 4H), 3.45 (m, 8H), 1.95 (m, 8H). Anal. Calcd for $C_{16}H_{25}N_3O$. 3HCl: C, 49.94; H, 7.33; N, 10.92. Found: C, 49.90; H, 7.41; N, 10.89. MS (ESI, m/z) 276 ([M + H]$^+$, 100%).

Pyronaridine Tetraphosphate (1). The solution of 7,10-dichloro-2-methoxybenzo[*b*][1,5]naphthyridine (4.0 kg, 14.3 mol) **4** and 4-amino-2,6-bis(pyrrolidin-1-ylmethyl)phenol **12** (5.5 kg, 14.3 mol) in 45.8 L ethanol was stirred at 50 °C for 2.5 h. The solvent was removed under reduced pressure, and the obtained yellow solid was dissolved in 50 L water. Afterwards, the solvent was adjusted to pH 10−12 with 5 L of 20% NaOH water solution. The solid was collected by filtration and washed with water until neutral. The solid was suspended in 20 L water without drying; orthophosphoric acid was then added to the suspension to adjust the pH to 2−3 and stirred for 1 h. Isopropanol (40 L) was added to the solvent, after which the yellow solid was precipitated and collected by filtration. The solid was purified by refluxing the slurry containing 15 L of 75% ethanol to obtain the qualified product **1** (11.2 kg, 86% yield, and 99.8% purity), mp 180−182 °C; ^{1}H NMR (300 MHz, DMSO) δ 8.25 (d, J = 9.2 Hz, 1H), 8.00 (s, 1H), 7.90 (d, J = 9.3 Hz, 1H), 7.32 (t, J = 9.7 Hz, 2H), 7.13 (s, 2H), 4.02 (s, 3H), 3.87 (s, 2H), 2.91 (m, 6H), 1.85 (m, 6H), 1.25 (m, 6H). Anal. Calcd for $C_{29}H_{32}ClN_5O_2$·$4H_3PO_4$: C, 38.27; H, 4.87; N, 7.70. Found: C, 38.32; H, 4.80; N, 7.73. MS (ESI, m/z) 518 ([M + H]$^+$, 100%).

■ ASSOCIATED CONTENT

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Notes

The authors declare no competing financial interest.

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■ REFERENCES

(1) (a) Kurth, F.; Beelard, S.; Basra, A.; Ramharter, M. *Expert Rev. Anti-Infect. Ther.* **2011**, *9*, 393−396. (b) Croft, S. L.; Duparc, S.; Arbe-Barnes, S. J.; Craft, J. C.; Shin, C.-S.; Fleckenstein, L.; Borghini-Fuhrer, I.; Rim., H.-J. *Malaria J.* **2012**, *11*, 270. (c) Guy, R. K.; Zhu, F.; Clark,

J. A.; Guiguemde, W. A.; Floyd, D.; Knapp, S.; Stein, P.; Castro, S. *PCT Int. Appl.* WO/2013/027196, 2013; (d) Haynes, R. K.; Cheu, K.-W.; Chan, H.-W.; Wong, H.-N.; Li, K.-Y.; Tang, M. M.-K.; Chen, M.-J.; Guo, Z.-F.; Guo, Z.-H.; Sinniah, K.; et al. *ChemMedChem* **2012**, *7*, 2204−2226.

(2) (a) Zheng, X-Y; Xia, Y.; Gao, F.-H.; Chen, C. *Acta Pharmaceut. Sin.* **1979**, *14*, 736−7. (b) Ni, Y.-C.; Xu, Y.-Q.; Shao, B.-R. *Acta pharmacologica Sinica* **1982**, *3* (1), 51−5. (c) Shao, B.-R. *Chin. Med. J.* **1990**, *103*, 428−34.

(3) (a) Kang, M. S. Repub. Korean Kongkae Taeho Kongbo KR/2009/114029, 2009; (b) Breytenbach, J. C.; N'Da, D. D.; Ashton, M. *PCT Int. Appl.* WO/2010/032165, 2010.

(4) (a) Liqiang, F.; Xin, L.; Jianjun, C.; Huili, H.; Zhan, C.; Xingsheng, Guo; Shi, D.; Yushe, Y. *Org. Process Res. Dev.* **2010**, *14* (4), 815−819. (b) Yu, L.; Xu-Feng, C.; Xin, L.; Yong-Bing, C.; Wen-Jing, C.; Yu-She, Y. *Chin. Chem. Lett.* **2013**, *24*, 321−324. (c) Park, S. H.; Pradeep, K.; Jang, S. H. *J. Labelled Cmpd. Radiopharm.* **2007**, *50*, 1248−1254. (d) Purfield, A. E.; Tidwell, R. R.; Meshnick, S. R. *Antimicrob. Agents Chemother.* **2008**, *52*, 2253−2255.

(5) (a) Yu, L.; Zining, L.; Xufeng, Cao; Xin, L.; Huili, He; Yushe, Y. *Bioorg. Med. Chem. Lett.* **2011**, *21*, 4779−4783. (b) Neelakandan, K.; Manikandan, H.; Santosha, N.; Prabhakaran., B. *Org. Process Res. Dev.* **2013**, *17*, 981−984. (c) Yong-Yong, Z.; Peng, X.; Long-Xuan, X.; Jing-Yu, W.; Jian-Qi, Li. *Org. Process Res. Dev* **2012**, *16*, 1921−1926.

(6) (a) Babalola, C. P.; Scriba, G. K. E.; Sowunmi, A.; Alawode, O. A. *J. Chromatogr. B* **2003**, *795* (2), 265−272. (b) Lee, J.; Son, J.; Chung, S.-J.; Lee, E.-S.; Kim, D.-H. *J. Mass Spectrom.* **2004**, *39*, 1036−1043.