

A Novel Synthesis Method of Meriolin1

Jie Ren^a, Pengcheng Ding^b, Jichao Zhou^c, Pengchao Yu^d, Xinyue Wu^e and Kun Hu^f

School of Pharmacy, Changzhou University, 213164, Changzhou, China.

^arenjie2006@163.com, ^b2595131130 @qq.com, ^c820250784@qq.com, ^d2754915088@qq.com, ^e954687726@qq.com, ^fhukun1979@163.com

Abstract: Meriolins, a compound designed and synthesized by the similar structure of natural marine products Meridianins and Variolins, has been proved to be a new kinase inhibitor that can enhance protein kinase selectivity and cell death. It has stronger inhibitory activity, relative selectivity to kinases and antitumor effect. As the first compound in the Meriolins series, Meriolin1 has been studied the most. In this paper, the synthesis methods of Meriolin1 at home and abroad were summarized and analyzed. On this basis, a more economical and effective synthesis route was designed, that is, Meriolin1 was finally obtained from 7-azaindole through Friedel-Crafts acylation, protection, aldol condensation and cyclization reaction. This route avoids the disadvantages of the existing routes and has the advantages of mild reaction conditions, simple operation and low cost. The structures of the compounds were confirmed by ¹H NMR and ¹³C NMR.

Keywords: Meriolins, Synthesis, Anti-cancer, Kinase inhibitors, Marine organism.

1. Introduction

Variolins is a kind of marine alkaloid isolated from Antarctic sponge *Kirkpatrickia variolosa* [1-2], which has strong apoptosis-promoting activity [3]. Meridianins is isolated from Ascidian *Aplidium meridianum* [4], and also has antitumor activity. Meridianins is considered to be a mild non selective kinase inhibitor, which has been widely studied because of its strong antitumor effect and the ability to induce cell death. However, although marine natural products are very useful from a pharmacological point of view, it is very

difficult to obtain these compounds from a technical and biological point of view. At present, the chemical synthesis of the effective components of marine products has become the current research hotspot [5]. After analyzing the overlapping structure of natural marine products Meridianins and Variolins, marine biologists designed and synthesized Meriolins, which has been proved to be a new kinase inhibitor that can enhance protein kinase selectivity and cell death [6-7]. Meriolins showed stronger inhibitory activity, relative selectivity to kinase and antitumor effect than its lead compounds [8]. The structures are shown in Fig. 1.

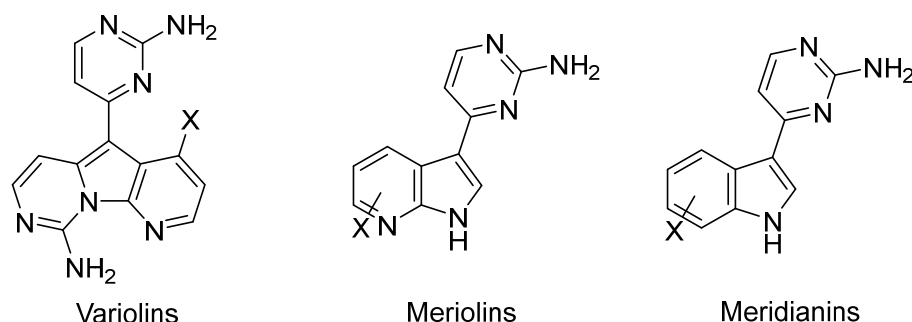


Figure 1. Structure of three compounds

Meriolin1, as the first compound in Meriolins series to be synthesized and named, has been studied most. The synthetic methods can be summarized as follows.

In 2005, A.S. Karpov et al [9] synthesized Meriolin1 in only four steps by iodization reaction, protecting nitrogen on indole ring, introducing alkyne ketone under catalyst conditions, and cyclization reaction. In the third step, when Pd (dppf) Cl₂ was used alone, alkyne ketone became the secondary product and TMS ethynyl indole was the main product. So far, the mechanism of the effect of this special catalyst mixture on the Alkynylation selectivity of carbonyl

compounds is not clear. Then, in the last step of cyclization reaction, the reagent is particularly important. After many failures, the effect of using tert-butyl alcohol and acetonitrile as mixed solvent was the most unexpected. This not only contributed to the Michael addition of methanol and successfully synthesized the pyrimidine ring, but also cleaved and removed the TMS and -BOC groups at the same time, eliminating the deprotection step. However, in the third step, palladium catalyst, CO gas and highly flammable trimethylsilylene are used, which makes the condition control more stringent. The specific conditions are shown in Fig. 2.

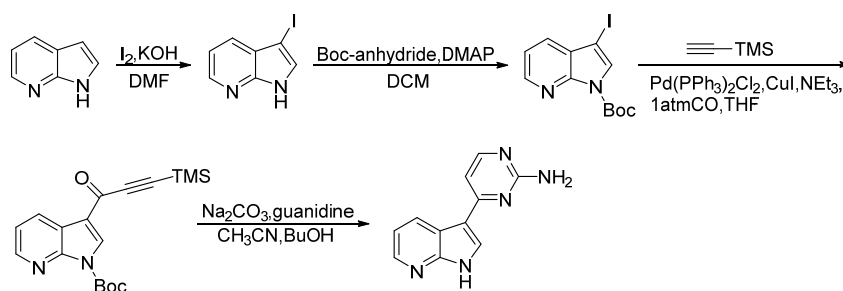


Figure 2. Synthetic route of Meriolin1 (A.S. Karpov et al)

In 2011, E. Merkul et al. [10] synthesized Meriolin1 in only three steps. On the one hand, Cs₂CO₃ was used to promote the Suzuki-Miyaura reaction, and in this reaction solution, the protective group of tert butoxycarbonyl can be cracked to obtain the product directly without additional deprotection steps. As shown in Fig. 3, azaindolyl iodide [11] protected by tert butoxycarbonyl was easy to obtain and store, and can be used as an important part of cross coupling reaction. Since its direct coupling with heteroarylboric acid or ester would be

limited by the latter, they first converted the iodide into the corresponding pinacol ester [12], and then reacted with heterocyclic halide on the way, which makes it easier to obtain the final product. The difference from the previous route is that one pot borylation/ Suzuki-Miyaura reaction was used as the key step, and Meriolin1 was synthesized in the same four-step reaction. The price of 2-amino-4-chloropyrimidine and catalyst in the last step of this route is relatively high.

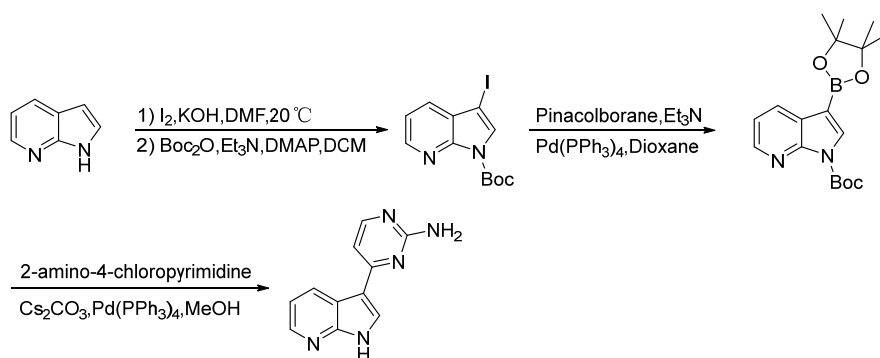


Figure 3. Synthetic route of Meriolin1 (E. Merkul et al)

In 2014, based on the synthesis of Meridianin, S.R. Walker et al [13] took 2-amino-3-iodopyridine as raw material, underwent acylation reaction, and then reacted with iodide under the action of metal catalyst to obtain the parent nuclear skeleton. Finally, the protective group was removed as a whole by one pot method to obtain compound Meriolin1. This

method is not only complex and cumbersome, but also the raw material price is six times that of the previous two routes. In addition, the metal catalyst is used, so the reaction conditions are relatively strict, which has no advantage over the previous two routes. The specific conditions are shown in Fig.4.

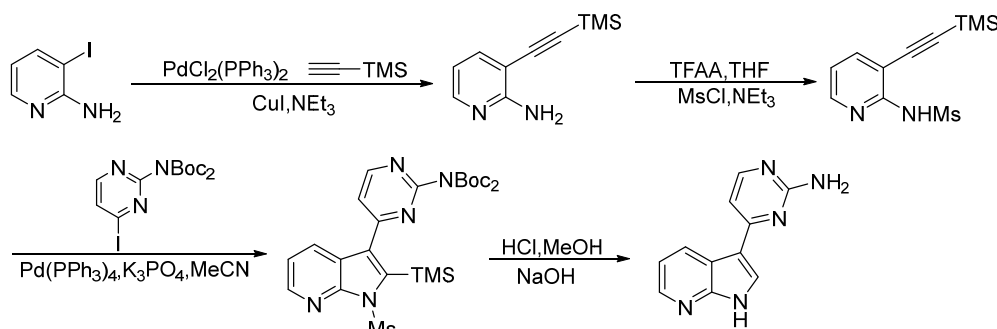


Figure 4. Synthetic route of Meriolin1 (S.R. Walker et al)

On this basis, adhering to the concept of innovation, we designed an economic and effective synthetic route.

2. Results and Discussion

Using 7-azaindole as raw material, the compound (2) was obtained by Friedel-Crafts acylation reaction in the first step. In the second step, potassium tert-butoxide was used to extract hydrogen. In order to avoid secondary amine being affected

in the follow-up reaction, potassium t-butoxide was used to protect the amino group, and then the compound (3) was obtained. Then the compound (4) was obtained by condensation with DMF-DMA [14-15], and Meriolin1 was obtained by cyclization of the compound (4) with guanidine hydrochloride in an overall yield of 27.58%. The specific conditions are shown in Fig.5.

In the process of synthesizing Meriolin1, in order to ensure innovation, we compared with the above synthetic route to

avoid some disadvantages in other routes, such as the use of expensive raw materials and metal catalysts, complex conditions and post-treatment, and reagents with strong irritation or toxicity, etc.

In the preparation of compound (2), when acetyl chloride was used as acylating agent, the reaction conversion was complete and the yield was 80.44%.

In the preparation of compound (3), the protection of BOC

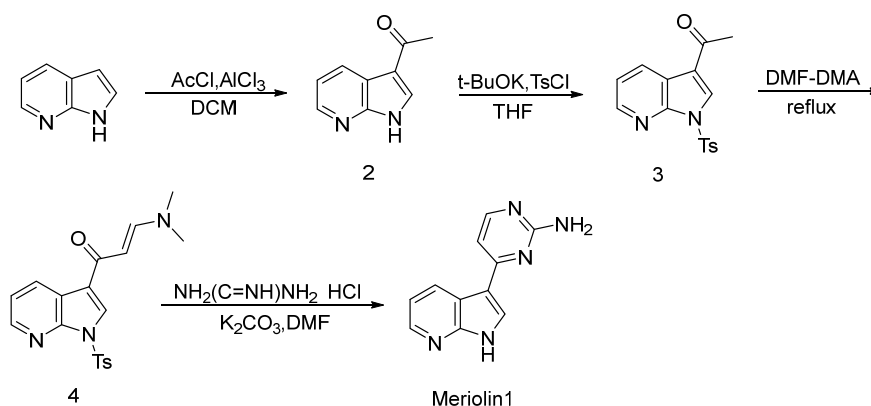


Figure 5. Synthetic route of Meriolin1

3. Experimental Section

3.1. Materials

All chemicals provided by commercial sellers were analytical grade, which can be used directly without purification. ¹H NMR and ¹³C NMR spectra were recorded in CDCl₃ or DMSO with TMS as internal standard using a Bruker 400 MHz spectrometer.

3.2. General Procedures

3.2.1. Preparation of compound 3-acetyl-7-azaindole (2)

Added anhydrous AlCl₃ (22.4 g, 167.98 mmol) and anhydrous CH₂Cl₂ (40 ml) into a 100 ml round bottomed flask. After stirring for 5 min, added 7-azaindole (10 g, 84.64 mmol) for N₂ protection. Slowly added acetyl chloride (19.92 g, 253.98 mmol) in an ice bath and moved it to room temperature for stirring. After 12 hours of reaction, the reaction was completed by TLC. The reaction solution was slowly dripped into 100 mL ice water to quench, a large number of solids were precipitated in the process. After the solid was precipitated, the filter cake was collected and dried, and then 10.91 g of light yellow solid was obtained with a yield of 80.44%. ¹H NMR (400 MHz, DMSO): δ 12.52 (bs, 1 H), 8.47 (dd, 2 H, J = 9.6 Hz), 8.32 (dd, 1 H, J = 6.4 Hz), 7.25 (dd, 1 H, J = 12.8 Hz), 2.47 (s, 3 H); ¹³C NMR (400 MHz, DMSO): δ 193.34, 149.50, 144.59, 135.21, 130.02, 118.47, 118.07, 115.92, 27.40.

3.2.2. Preparation of compound 1-p-toluenesulfonyl-3-acetyl-7-azaindole (3)

In a 100 ml round bottom flask, added compound (2) (10 g, 62.42 mmol) and THF (35 ml) to make it fully dissolve. Slowly added t-BuOK (14 g, 124.76 mmol) in an ice bath. After half an hour, added tosyl chloride (13.09 g, 68.66 mmol) to the flask and heated it for reflux. Two hours later, the reaction was found to be complete by TLC. Added ice water to the reaction solution and stirred it. A large amount of flocculent substances were precipitated. After standing for 10 minutes, filtered under reduced pressure and collected the filter cake. If the product was impure, added petroleum ether

was not conducive to the next reaction. In this step, tosyl was selected as the protective group, and the yield was 99.00%.

In the preparation of compound (4), DMF-DMA was directly used as solvent and reacted at room temperature and then at high temperature. The yield was 61.70%.

In the preparation of Meriolin1, the dosage of guanidine hydrochloride was selected. When the equivalent of guanidine hydrochloride was 5.0, the yield was 56.14%.

to dissolve the excess tosyl chloride, and filtered again. After the filter cake was dried, 19.42 g of light yellow solid was obtained with a yield of 99.00%. ¹H NMR (400 MHz, CDCl₃): δ 8.60 (dd, 1 H, J = 1.4 Hz), 8.48 (dd, 1 H, J = 1.2 Hz), 8.36 (s, 1 H), 8.17 (d, 2 H, J = 8.0 Hz), 7.33 (d, 2 H, J = 8.4 Hz), 7.29 (t, 1 H, J = 5.2 Hz), 2.58 (s, 3 H), 2.39 (s, 3 H); ¹³C NMR (400 MHz, CDCl₃): δ 193.24, 147.09, 146.13, 134.41, 131.81, 131.78, 129.89, 128.57, 120.47, 120.21, 118.66, 99.98, 27.27, 21.74.

3.2.3. Preparation of compound (E)-1-[1-p-toluenesulfonyl-7-azaindole]-3-(dimethylamino)-2-propene-1-one (4)

Added compound (3) (10 g, 79.50 mmol) and N, N-dimethylformamide dimethyl acetal (120 ml) into a 100 ml round bottom flask. The compounds were dissolved as fully as possible by ultrasound and refluxed after reacting at room temperature for 6 hours. The reaction was completed after 20 hours of TLC monitoring. After cooling to room temperature, ice water was slowly added to the flask. Then the mixture was extracted with EA (100ml × 4). The combined organic phases were washed with saturated NaCl solution (100ml × 4) and dried with anhydrous sodium sulfate. The solvent was evaporated by vacuum rotary evaporator to obtain 7.25 g of light yellow solid with a yield of 61.70%. ¹H NMR (400 MHz, CDCl₃): δ 8.64 (d, 1 H, J = 7.6 Hz), 8.42 (d, 2 H, J = 4.0 Hz), 8.22 (s, 1 H), 8.10 (d, 2 H, J = 8.0 Hz), 7.78 (d, 2 H, J = 12.4 Hz), 7.27 (d, 2 H, J = 8.4 Hz), 5.60 (d, 1 H, J = 12.4 Hz), 3.13 (d, 6 H), 2.34 (s, 3 H); ¹³C NMR (400 MHz, CDCl₃): δ 183.39, 153.12, 147.27, 145.63, 145.43, 134.85, 132.00, 129.74, 128.26, 127.95, 121.54, 120.52, 119.87, 92.44, 45.06, 37.43, 21.67.

3.2.4. Preparation of compound 3-(2-Aminopyrimidine)-7-azaindole (Meriolin1)

Compound (4) (2 g, 5.4 mmol), anhydrous K₂CO₃ (2.24 g, 16.20 mmol) and guanidine hydrochloride (2.56 g, 26.78 mmol) were added into a 50 ml round bottom flask, and then DMF was added to dissolve it and heat it at 130 °C. After 4 h, the reaction was found to be complete by TLC. The reaction solution was extracted with CH₂Cl₂ (100ml × 4), washed with

saturated NaCl solution (100ml × 4) and dried with anhydrous sodium sulfate. After the organic phase was combined, it was rotated and evaporated under reduced pressure to obtain light yellow solid powder. A small amount of ethyl acetate was added to the solid, heated by ultrasound, filtered under reduced pressure, the filter cake was taken and dried to obtain 1.28 g of light yellow solid with a yield of 56.14%. ¹H NMR (400 MHz, DMSO): δ 12.16 (bs, 1 H), 8.89 (dd, 1 H, *J* = 1.6 Hz), 8.30(s, 1 H), 8.25 (dd, 1 H, *J* = 1.4 Hz), 8.10 (d, 1 H, *J* = 5.2 Hz), 7.17 (dd, 1 H, *J* = 4.6 Hz), 7.02 (d, 1 H, *J* = 5.6 Hz), 6.44(s, 2 H); ¹³C NMR (400 MHz, DMSO): δ 163.95, 162.55, 157.75, 149.63, 143.90, 131.23, 128.93, 118.27, 117.20, 112.86, 105.46.

4. Conclusion

In this paper, a simple and novel route was used to synthesize Meriolin1 in an overall yield of 27.58%. The synthesis of Meriolin1 from 7-azaindole has not been reported before. This route avoids the disadvantages of the existing route, and Meriolin 1 can be synthesized in only four steps with cheap raw materials and simple post-treatment, which lays a foundation for the research of Meriolin series compounds in the future.

5. Conflicts of Interest

The authors declare no conflicts of interest.

Acknowledgment

We sincerely thank the financial support from the Postgraduate Research & Practice Innovation Program of Jiangsu Province (Grant No. SJCX22_1445 and Grant No. SJCX21_1239).

References

- [1] S.R. Walker, E.J. Carter, B.C. Huff, and J.C. Morris, "Variolins and related alkaloids", *Chemical Reviews*, 2009, Vol. 109(7), p3080.
- [2] M.F. Pilar, M. Pedro, D. Santiago, "Synthetic studies towards the 2-aminopyrimidine alkaloids variolins and meridianins from marine origin", *Tetrahedron Letters*, 2000, Vol. 41(24), p4777-4780.
- [3] M. Simone, E. Erba, G. Damia, F. Vikhanskaya and R. Riccardi, "Variolin B and its derivate deoxy-variolin B: new marine natural compounds with cyclin-dependent kinase inhibitor activity", *European Journal of Cancer*, 2005, Vol. 41(15), p2366-2377.
- [4] M. Gompel, M. Leost, E. Joffe, L. Puricelli, L.H. Franco and J. Palermo, "Meridianins, a new family of protein kinase inhibitors isolated from the Ascidian *Aplidium meridianum*", *ChemInform*, 2004, Vol. 35(7), p1703-1707.
- [5] S. Dan, W. Wenbin, W. Xinyue, L. Minyue, Y. Xue and H. Zhonghong, "Meriolin1 induces cell cycle arrest, apoptosis, autophagy and targeting the Akt/Mapks pathways in human neuroblastoma sh-sy5y cells", *The Journal of pharmacy and pharmacology*, 2019, Vol. 72(4), p561-574.
- [6] J. Dumas, "Protein kinase inhibitors : emerging pharmacophores 1997-2000", *Expert Opinion on Therapeutic patents*, 2001, Vol. 11(3), p405-429.
- [7] P. Traxler, P. Furet, "Strategies toward the design of novel and selective protein tyrosine kinase inhibitors", *Pharmacology therapeutics*, 1999, Vol. 82(2), p195-206.
- [8] E.K. Bettay, O.M. Tirado, L.S. Marionneau, Y. Ferandin, O. Lozach and J.C. Morris, "Meriolins, a new class of cell death inducing kinase inhibitors with enhanced selectivity for cyclin-dependent kinases", *Cancer Research*, 2007, Vol. 67(17), p8325-8334.
- [9] A.S. Karpov, E. Merkul, F. Rominger and T. Mueller, "Concise syntheses of meridianins by carbonylative alkynylation and a four-component pyrimidine synthesis", *ChemInform*, 2006, Vol. 44(42), p6951-6956.
- [10] E. Merkul, E. Schaefe, and T.J.J. Mueller, "Rapid synthesis of bis(hetero)aryls by one-pot masuda borylation-suzuki coupling sequence and its application to concise total syntheses of meridianins a and g", *Organic & Biomolecular Chemistry*, 2011, Vol. 9(9), p3139-3141.
- [11] W. Bernhard, B. Nicole, and B. Uwe, "Hydroboration and Suzuki-Miyaura coupling reactions with the electronically modulated variant of an Ynamine: the synthesis of (e)-β-Arylenamides", *Tetrahedron*, 2000, Vol. 56(43), p8473.
- [12] A.K. Venkata, S. Feng, and F. Sulagna, "Cheminform abstract: boc groups as protectors and directors for ir-catalyzed c-h borylation of heterocycles", *ChemInform*, 2010, Vol. 41(18), p9199-9201.
- [13] S.R. Walker, M.L. Czyz and J.C. Morris, "Concise syntheses of meridianins and meriolins using a catalytic domino amino-palladation reaction", *Organic Letters*, 2014, Vol. 16(3), p708.
- [14] A. Fathi, A. Ahmed, and M. Sayed, "Synthesis of 1,4-Diaryl-piperazine-2,5-diones : New Behavior of N, N-Dimethylformamide Dimethyl Acetal (DMFDMA)", *Synthetic Communications*, 2008, Vol. 38(3), p376-382.
- [15] T.M. Tarasiuk, A.V. Tatyana, and S.P. Kirill, "Reactions of 4,5-Dihydro-1,4-Benzothiazepin-3(2H)-one 1,1-Dioxide and 1,5-Dihydro-4,1-Benzothiazepin-2(3H)-one 4,4-Dioxide Derivatives with Vilsmeier Reagent and DMFDMA", *Journal of Heterocyclic Chemistry*, 2014, Vol. 51(3), p755-759.