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SYNTHESIS OF HANTZSCH 1,4-DIHYDROPYRIDINES IN A CONTINUOUS FLOW MICROREACTOR

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Abstract – A simple and efficient synthesis of Hantzsch 1,4-dihydropyridines from the condensation of benzaldehydes with alkyl acetoacetates/benzoylacetate and aqueous ammonia in the absence of catalyst has been developed by using a continuous flow microreactor. Under optimized condition, various 1,4-dihydropyridines were obtained in 80-93% yield with high atom efficiency. Compared with using batch system, the present smooth procedure greatly accelerated the reaction due to the excellent mixing and mass transfer of reactant in micro flow system.

Hantzsch 1,4-dihydropyridines (1,4-DHPs) scaffold exist in the core structure of several biologically active compounds such as vasodilator, calcium channel blocker, bronchodilator, antiatherosclerotic, antitumor, geroprotective, hepatoprotective and antidiabetic agents.[1] Furthermore, it has been demonstrated that 1,4-DHPs could exhibit other medicinal applications including radioprotection and HIV protease inhibition, in addition to acting as a cocaine dependent regulator and as a TGFβ signal inhibitors.[2] In recent year, Hantzsch reaction[3] also have been used as a powerful method for one-pot multicomponent synthesis of polyhydroquinolines[4a-b] and hexa-substituted 1,4-dihydropyridines[4c] which possess potential biological activities. Moreover, oxidative aromatization of Hantzsch 1,4-DHPs is of great interest to synthetic chemists for it could generate corresponding pyridine derivatives.[5]

It is well know that 1,4-DHPs have been achieved by using the classical Hantzsch procedure and the modified Hantzsch conditions, which involves a one-pot condensation of aldehyde with ethyl acetoacetate, and ammonia either in acetic acid or refluxing in alcohol.[3] Although various catalysts, nitrogen sources

Dedicated with respect to Professor Dr. Lutz F. Tietze's 75th birthday

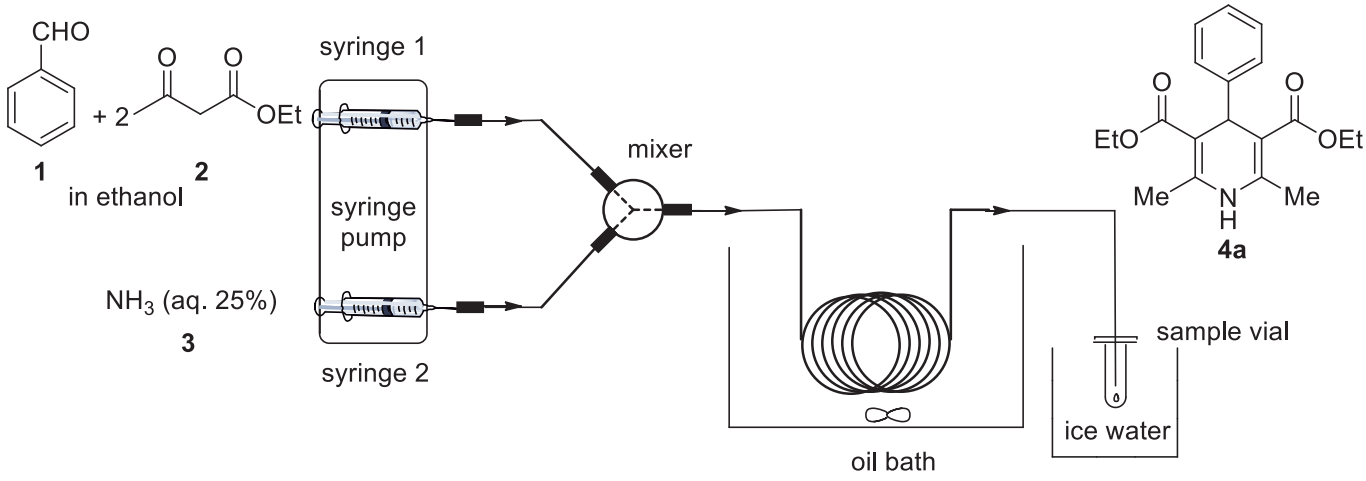
and techniques have been introduced into this reaction process, most of these protocols suffer from moderate yields, long reaction times, and/or low atom efficiency.[6] Therefore, it is highly desirable to develop efficient synthetic methods for this class of compound and expand their structure diversity for current medicinal chemistry and organic synthesis chemistry demands.

Combinatorial chemistry as a high-throughput synthetic technology has became a powerful tool for the discovery of new drugs and increasing interests have been drawn onto the application of microfluidic continuous flow technique in this field.[7] Automated microreactor-based (microfluidic chip) continuous flow systems have the potential to greatly accelerate the production of small molecule libraries of drug-like structures (primarily heterocycles).[8] Our previous work had demonstrated the use of microfluidic continuous flow microreactors could eliminate the formation of the corresponding byproduct and simultaneously accelerate the desired transformation with very high selectivity and efficiency.[9] In comparison with the classical batch systems, especially for the three-component cascade approach, the continuous micro flow technique enhances accurate temperature control and efficient mixing. As a result of ongoing research on develop new synthetic methods for the generation of heterocycle libraries and potential radiolabelling imaging probes based on microfluidic reactor,[10] we herein report the continuous flow synthesis of 1,4-DHPs.

The structure of the continuous flow microreactor system is illustrated in Scheme 1. This system is made from Longer LSP02-1B syringe pump, gas-tight syringes, oil bath, Y-shape mixer, stainless steel tubing and fittings. The end of inlet tube is connected to the syringe pump and the end of outlet tube is connected to a sample vial. The sample vial is cooled down by ice water to collect the final product solution.

Firstly, the model reaction for synthesis of 1,4-DHP was investigated in continuous flow system by varing the flow rate and the temperature. As shown in Scheme 1, during the experiment, the ethanol solution of benzaldehyde (**1**, 0.67 M) mixed with ethyl acetoacetate (**2**, 2 equiv.) in syringe 1 and equivoluminal aqueous ammonium (**3**, 25%) in syringe 2 were respectively introduced into the mixer by syringe pump at identical flow rates changed from 5 μL/min to 8.5 μL/min (residence times changed from 39.3 min to 23.1 min accordingly). The reaction mixture was then allowed to flow through a stainless steel tube reactor (ϕ = 500 μm, l = 1 m) which was dipped in a heated oil bath. The outlet solution of crude poduct was collected in a cold sample vial and then concentrated under vacuum. The residue was subjected to silica gel column chromatography with petroleum ether – ethyl acetale Pet-EA (5:1) as eluent to give pure product **4a**. Herein, flow rate and reaction temperature were preliminary optimized and the results are summarized in Table 1. At 100 ˚C, when the flow rates were respectively set up at 5, 6.7, 8.5 μL/min, with the increase of flow rate, the yields of product droped from 80% to 72% (Table 1, entries 1-3). At 110 ˚C, when flow rates set up at 5, 6.7, 8.5 μL/min respectively, the reactions afforded the product with yields between 93% and 90% (Table 1, entries 4-6). While, further raising the temperature to

120 ˚C give similar yields as that of 110 ˚C (Table 1, entry 7-9). In general, at 110 ˚C and 6.7 μL/min flow rate, the reaction could afford best yield. Notably, all the reactions were carried out with catalyst free. At same time, oxidative aromatization of 1,4-DHP did not perform because the corresponding pyridine was not observed by thin layer chromatography (TLC). In addition, no leakage of ammonia was detected from this semi-closed system even the reactant mixture was heated to about 120 ˚C. In comparison to continuous flow microreactor system, the same reaction could give a 75% yield after 10 h in a batch system. This result demonstrated the advantage of the micro system which could accelerate the reaction.



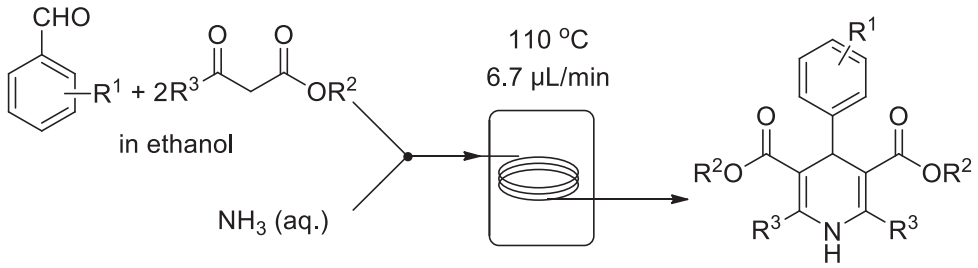
Scheme 1. Continuous flow microreactor system for the synthesis of 1,4-dihydropyridines

Table 1. Optimization of reaction conditions for the synthesis of 1,4-dihydropyridine in continuous flow microreactor

Entry	Temperature (˚C)	Flow rate (μL/min)	Residence time (min)	Yield (%)[a]
1	100	5	39.3	80
2	100	6.7	29.3	78
3	100	8.5	23.1	72
4	110	5	39.3	93
5	110	6.7	29.3	93
6	110	8.5	23.1	90
7	120	5	39.3	93
8	120	6.7	29.3	93
9	120	8.5	23.1	91

[a] Isolated yield.

Under the optimized continuous flow conditions, a series of 1,4-DHP derivatives were subsequently synthesized to study the scope and limitation of this procedure as shown in Scheme 2 and the results are summarized in Table 2. By altering the substituent group of benzaldehydes, such as electron-donating and electron-withdrawing groups, various substituted benzaldehydes and methyl/ethyl acetoacetate could afford corresponding products in good to excellent yields (85-92%). It was found that the electronic effect of substituted benzaldehydes in these reaction is not apparent. In addition, ethyl benzoylacetate was used to prepare more sterically congested 1,4-dihydropyridine which gave slightly lower yield (80%).



Scheme 2. Synthesis of 1,4-dihydropyridines in continuous flow microreactor

Table 2. Synthesis of 1,4-dihydropyridines in continuous flow microreactor

Entry	R^1	R^2	R^3	Product	Yield (%)[a]	Mp(˚C) Measured	Mp(˚C) Lit. reported
1	4-OMe	Et	Me	**4b**	90	157-159	158-160[6i]
2	4-Br	Et	Me	**4c**	91	162-164	162-164[6i]
3	4-NO_2	Et	Me	**4d**	90	129-131	130-132[6i]
4	4-Me	Et	Me	**4e**	90	136-138	136-138[6i]
5	4-Cl	Et	Me	**4f**	92	144-146	145-147[6i]
6	H	Me	Me	**4g**	92	197-198	197-198[6f]
7	4-OMe	Me	Me	**4h**	90	172-174	173-174[6f]
8	4-Cl	Me	Me	**4i**	91	195-197	195-196[6f]
9	4-OH	Me	Me	**4j**	90	198-199	198-199[6f]
10	2-NO_2	Me	Me	**4k**	85	170-172	171-172[6f]
11	H	Et	Ph	**4l**	80	135-136	136-137[6j]

[a] Isolated yield.

In conclusion, we have successfully developed a rapid and efficient process for the synthesis of 1,4-DHP derivatives using micro flow reactor. Under the optimized conditions, a series of 1,4-DHP derivatives were synthesized in 80-93% isolated yields with catalyst free. Compared with the classical batch systems,

in the present micro flow reactor, the reactions completed smoothly within half an hour taking the advantage of efficient reactant mixing and enhanced mass transfer. These advances will facilitate the rapid synthesis of biologically compounds library and radio labeled imaging probes. Therefore, further exploration of the reactivity features of this methodology and applications to molecule imaging probe and the drug discovery are in progress.

EXPERIMENTAL

All chemicals were reagent grade and used as purchased without further purification unless otherwise stated. Column chromatography was performed using silica gel 60 (300-400 mesh). Melting points were measured on a YANACO micro melting point apparatus and are uncorrected. ^{1}HNMR spectra were recorded in $CDCl_3$ using TMS as an internal reference with a Bruker Avance-400 spectrometer operating at 400 MHz.

Typical procedure for the synthesis of 1,4-dihydropyridines in continuous flow microreactor system.

Diethyl 2,6-dimethyl-4-phenyl-1,4-dihydropyridine-3,5-dicarboxylate (4a). EtOH solution (3 mL) of benzaldehyde (0.67 M) mixed with ethyl acetoacetate (1.34 M) and the 25% aqueous NH_3 (3 mL) were respectively transferred into gas-tight syringe 1 and syringe 2. The syringes were placed in a Longer LSP02-1B syringe pump which was set to deliver the reactants into the mixer at identical flow rate of 6.7 μL/min. The reaction mixture was then allowed to flow through a stainless steel tube reactor which was dipped in a 110 ˚C oil bath. The output mixture was collected in a cooled sample vial. After reaction completion, rinsed the reactor with ethanol to collect all of the reactant solution and then concentrated under vacuum. The residue was subjected to silica gel column chromatography with Pet-EA (5:1) as eluent to give **4a** (612mg, 93% yield); Mp 158-160 ˚C (lit.,[6i] Mp 158-160 ˚C); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.22 (t, J = 7.0 Hz, 6H, $2CH_3CH_2$), 2.33 (s, 6H, $2CH_3$), 4.07 (q, J = 7.0 Hz, 4H, $2CH_3CH_2$), 4.99 (s, 1H, CH), 5.56 (s, 1H, NH), 7.12-7.14 (m, 1H), 7.17-7.21 (m, 2H), 7.26-7.29 (m, 2H); ^{13}C NMR ($CDCl_3$, 100 Hz): δ 14.4, 19.2, 40.0, 60.0, 104.0, 126.1, 127.8, 128.4, 144.2, 147.8, 167.6. FT-IR (KBr) : 3342, 2982, 1688, 1651, 1489, 1323 cm^{-1}.

Procedure for the synthesis of 1,4-dihydropyridine (4a) in batch system.

The EtOH solution (3 mL) of benzaldehyde (2 mmol) mixed with ethyl acetoacetate (4 mmol) and the 25% aqueous NH_3 (3 mL). The mixture was kept in a screw stoppered preasure bottle and heated for 10 h in a 110 ˚C oil bath. After cooling, the contents of the bottle were transferred to a round-bottomed flask and then concentrated under vacuum. The residue was subjected to silica gel column chromatography with Pet-EA (5:1) as eluent to give **4a** (494mg, 75% yield).

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REFERENCES

1. (a) V. Klusa, *Drugs Fut*., 1995, **20**, 135; (b) R. G. Bretzel, C. C. Bollen, E. Maeser, and K. F. Federlin, *Am. J. Kidney Dis*., 1993, **21**, 53; (c) R. G. Bretzel, C. C. Bollen, E. Maeser, and K. F. Federlin, *Drugs Fut*., 1992, **17**, 465; (d) R. Boer and V. Gekeler, *Drugs Fut*., 1995, **20**, 499. (d) A. Sausins and G. Duburs, *Heterocycles*, 1988, **27**, 269; (e) P. P. Mager, R. A. Coburn, A. J. Solo, D. J. Triggle, and H. Rothe, *Drug Design Discovery*, 1992, **8**, 273; (f) R. Mannhold, B. Jablonka, W. Voigdt, K. Schoenafinger, and K. Schravan, *Eur*. *J. Med*. *Chem*., 1992, **27**, 229.
2. (a) I. O. Donkor, X. Zhou, J. Schmidt, K. C. Agrawal, and V. Kishore, *Bioorg. Med. Chem.*, 1998, **6**, 563; (b) A. Hilgeroth and H. Lilie, *Eur. J. Med. Chem*., 2003, **38**, 495; (c) A. Kuzmin, S. Semenova, N. F. Ramsey, E. E. Zvartau, and J. M. Vanree, *Eur. J. Pharmacol.*, 1996, **295**, 19; (d) D. Schade, M. Lanier, E. Willems, K. Okolotowicz, P. Bushway, C. Wahlquist, G. Gilly, M. Mercola, and J. R. Cashman, *J. Med. Chem.*, 2012, **55**, 9946.
3. A. Hantzsch, *Justus Liebigs Ann*. *Chem*., 1882, **1**, 215.
4. (a) J. S. Yoo, T. J. Laughlin, J. J. Krob, and R. S. Mohan, *Tetrahedron Lett*., 2015, **56**, 4060; (b) L. M. Wang, J, Sheng, L, Zhang, J. W. Han, Z. Y. Fan, H. Tian, and C. T. Qian, *Tetrahedron*, 2005, **61**, 1539; (c) M. Li, Z. Q. Zuo, L. R. Wen, and S. W. Wang, *J. Comb. Chem.*, 2008, **10**, 436.
5. (a) F. Saikh, R. De, and S. Ghosh, *Tetrahedron Lett.*, 2014, **55**, 6171; (b) J. Zhang, M. Z. Jin, W. Zhang, L. Yang, and Z. L. Liu, *Tetrahedron Lett.*, 2002, **43**, 9687; (c) J. J. Xia and G. W. Wang, *Synthesis*, 2005, 2379; (d) N. Nakamichi, Y. Kawashita, and M. Hayashi, *Org. Lett.,* 2002, **4**, 3955.
6. (a) G. Sabitha, G. S. K. K. Reddy, Ch. S. Reddy, and J. S. Yadav, *Tetrahedron Lett*., 2003, **44**, 4129; (b) J. G. Breitenbucher and G. Figliozzi, *Tetrahedron Lett*., 2000, **41**, 4311; (c) R. K. Vohra, C. Bruneau, and J. L. Renaud, *Adv. Synth. Catal.*, 2006, **348**, 2571; (d) K. L. Bridgwood, G. E. Veitch, and S. V. Ley, *Org. Lett.*, 2008, **10**, 3627; (e) L. Singh, M. P. S. Ishar, M. Elango, V. Subramanian, V. Gupta, and P. Kanwal, *J. Org. Chem.*, 2008, **73**, 2224; (f) B. M. Khadilkar, V. G. Gaikar, and A. A. Chitnavis, *Tetrahedron Lett.*, 1995, **36**, 8083; (g) L. Shen, S. Cao, J. J. Wu, H. Li, J. Zhang, M. X. Wu, and X. H. Qian, *Tetrahedron Lett*., 2010, **51**, 4866; (h) Y. P. Liu, J. M. Liu, X. Wang, T. M. Cheng, and R. T. Li, *Tetrahedron*, 2013, **69**, 5242; (i) A. Debache, W. Ghalem, R. Boulcina, A. Belfaitah, S. Rhouati, and B. Carboni, *Tetrahedron Lett*., 2009, **50**, 5248; (j) B. J. Zhao, X. Q. Zhu, J. Q. He, C. Z. Xia, and J. P. Cheng. *Chem. J. Chinese Univ.*, 1999, **20**, 1733.

7. (a) B. Gutmann, D. Cantillo, and C. O. Kappe, *Angew*. *Chem*. *Int*. *Ed*., 2015, **54**, 6688; (b) I. R. Baxendale, J. J. Hayward, S. Lanners, S. V. Ley, and C. D. Smith, 'Microreactors in Organic Synthesis and Catalysis,' ed. by T. Wirth, Wiley-VCH, Weinheim, 2008; (c) W. Ehrfeld, V. Hessel, and H. Lowe, 'Microreactors: New Technology for Modern Chemistry,' Wiley, Weinheim, 2000; (d) H. Kim, A. Nagaki, and J. Yoshida, *Nature Commun*., 2011, **2**, 264.
8. (a) B. P. Mason, K. E. Price, J. L. Steinbacher, A. R. Bogdan, and D. T. McQuade, *Chem. Rev.*, 2007, **107**, 2300; (b) L. Kang, B. G. Chung, R. A. Langer, and A. Khademhosseini, *Drug Discovery Today*, 2008, **13**, 1; (c) C. Wiles and P. Watts, *Expert Opin. Drug Discovery*, 2007, **11**, 1487; (d) C. M. Griffiths-Jones, M. D. Hopkin, D. Jönsson, S. V. Ley, D. J. Tapolczay, E. Vickerstaffe, and M. Ladlow, *J. Comb. Chem.*, 2007, **9**, 422.
9. (a) R. J. Hu., M. Lei, H. S. Xiong, X. Mu, Y. G. Wang, and X. F. Yin, *Tetrahedron Lett.*, 2008, **49**, 387; (b) M. Lei, W. Tian, R. J. Hu. W. Li, and H. Zhang, *Synthesis*, 2012, **44**, 2519; (c) M. Lei, W. Li, R. J. Hu., W. Tian, Y. G. Wang, and H. Zhang, *Heterocycles*, 2012, **86**, 777; (d) M. Lei, R. J. Hu, Y. G. Wang, and H. Zhang, *Heterocycles* 2014, **88**, 1511; (e) M. Lei, W. Li, and H. Zhang, CN201310541031.3.
10. (a) M. W. Wang, W. Y. Lin, K. Liu, M. Masterman-Smith, and C. K.-F. Shen, *Mol. Imaging*, 2010, **9**, 175; (b) C.-C. Lee, G. D. Sui, A. Elizarov, C. J. Shu, Y.-S. Shin, A. N. Dooley, J. Huang, A. Daridon, P. Wyatt, D. Stout, H. C. Kolb, O. N. Witte, N. Satyamurthy, J. R. Heath, M. E. Phelps, S. R. Quake, and H.-R. Tseng, *Science*, 2005, **310**, 1793; (c) S. P. Chen, M. R. Javed, H.-K. Kim, J. Lei, M. Lazari, G. J. Shah, R. M. van Dam, P.-Y. Keng, and C.-J. Kim, *Lab. Chip*, 2014, **14**, 902.