Accepted Manuscript

Title: Targeting a Mirabegron Precursor by BH_3-Mediated Continuous Flow Reduction Process

Authors: Sonia De Angelis, Claudia Carlucci, Modesto de Candia, Gabriele Rebuzzini, Paolo Celestini, Massimiliano Riscazzi, Renzo Luisi, Leonardo Degennaro

PII: S0920-5861(17)30673-9
DOI: https://doi.org/10.1016/j.cattod.2017.09.058
Reference: CATTOD 11063

To appear in: *Catalysis Today*

Received date: 27-7-2017
Revised date: 18-9-2017
Accepted date: 29-9-2017

Please cite this article as: Sonia De Angelis, Claudia Carlucci, Modesto de Candia, Gabriele Rebuzzini, Paolo Celestini, Massimiliano Riscazzi, Renzo Luisi, Leonardo Degennaro, Targeting a Mirabegron Precursor by BH3-Mediated Continuous Flow Reduction Process, Catalysis Today https://doi.org/10.1016/j.cattod.2017.09.058

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

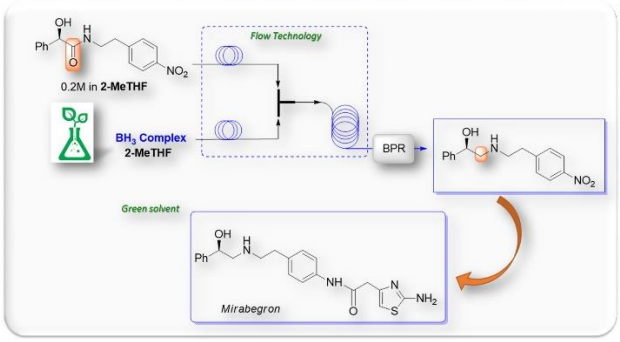
ACCEPTED MANUSCRIPT

Targeting a Mirabegron Precursor by BH_3-Mediated Continuous Flow Reduction Process

Sonia De Angelis,[a] Claudia Carlucci,[a] Modesto de Candia,[a] Gabriele Rebuzzini,[b] Paolo Celestini,[b] Massimiliano Riscazzi[b] Renzo Luisi,*[,a] and Leonardo Degennaro[a]

[a] *FLAME-Lab - Flow Chemistry and Microreactor Technology Laboratory; Department of Pharmacy — Drug Sciences, University of Bari "A. Moro" Via E. Orabona 4, Bari 70125, Italy.*
[b]*COSMA S.p.A, Via Colleoni 15/17, Ciserano (BG) 24040, Italy*
E mail: renzo.luisi@uniba.it

Graphical abstract



CATTOD Highlight

- Flow chemistry set up for pharmaceutical applications
- Continuous flow reduction using BH3 solutions
- Straightforward and green preparation of Mirabegron precursor

Abstract

A continuous-flow reduction of (*R*)-2-hydroxy-N-[2-(4-nitrophenyl)ethyl]-2-phenylacetamide, involved in the synthetic pathway of Mirabegron, has been developed. This study demonstrated the possibility to safely handling BH_3 complexes within microfluidic reactors using 2-MeTHF as greener alternative to traditional solvents, and without requiring any additive such as DMI. In addition, NMR and HPLC purity analysis revealed that the sole by-product of this process is the diamine **3**, which wouldn't affect the following synthetic steps towards Mirabegron.

1. Introduction

The phenylethanolamine core occurs in important molecules such as neurotransmitter noradrenaline and it is also contained in many naturally occurring molecules such as several alkaloids and compounds of pharmaceutical interest with different activities ranging from bronchodilation to antifungal activity (Fig. 1). [1,2]

ACCEPTED MANUSCRIPT

Among pharmaceutical molecules containing the phenylethanolamine scaffold, 2-(2-aminothiazol-4-yl)-*N*-[4-(2-{[(2*R*)-2-hydroxy-2-phenylethyl]amino}ethyl)phenyl]acetamide, commonly known as Mirabegron®, is a β-3 adrenergic agonist for the treatment of overactive bladder (OAB) with symptoms of urge urinary incontinence and urinary frequency. The main synthetic pathways leading to Mirabegron, reported as patents, are summarized in Scheme 1. Whichever pathway is followed for the production of Mirabegron, a key building block is represented by amine **3** (Scheme 1). Amine **3** can be targeted by reduction of amide **1** to amine **2** using BH_3·THF in THF and 1,3-dimethyl-2-imidazolidinone (DMI) (Scheme 1, path a). [3] Alternatively, other reducing agents, such as BH_3·DMS, BH_3·THF, $NaBH_4$, $NaBH_4/BF_3Et_2O$, $LiBH_4$, $LiAlH_4$, in suitable solvents (alcohols, ethers and polar aprotic mixtures of solvents) have been proposed. [4] As different approach to amine **3**, the nucleophilic ring-opening of toxic and harmful stireneoxide (Scheme 1, path b), provides derivative **5** which in turn can be converted to derivative **3** upon reductive conditions.

Nevertheless, from a safety and green perspective, either the use of borane as reducing agent, or the use of toxic chemicals (i.e. stireneoxide) should be carefully evaluated when planning an industrial production. As far as safety and toxicity are concerns, it is worth pointing out that flow chemistry is emerging as a green technology for handling of toxic and potentially hazardous chemicals. [5-7] In fact, in recent years the "flow chemistry" and the development of new microfluidic devices are playing an important role, both in academic and industrial research, as viable alternative to batch processing, offering in many cases sustainable synthetic routes. [8-12] The continuous flow production attracted the interest of chemical and pharmaceutical industry, ensuring lower costs, and effectively tackling out safety concerns. [13-15] The use of continuous flow processing allowed also exploring reaction conditions not accessible using traditional batch chemistry. [16] The high surface area in a microflow reactor allows for very fast heat transfer, and precise temperature control, decreasing the risk for runaway reactions. Another important point is the higher efficiency in mixing, realized in flow microreactor compared to batch, thus eliminating or considerably reducing concentration gradients that could negatively affect a fast reaction.

Based on these premises, we report in this work the development of a continuous flow process for the preparation of intermediate **2** (Scheme 1) by reduction of (*R*)-2-hydroxy-*N*-[2-(4-nitrophenyl)ethyl]-2-phenylacetamide **1** using BH_3·DMS as reducing agent in a greener solvent such as 2-MeTHF. [17]

2. Material and methods

2.1 General (Standard techniques)

ACCEPTED MANUSCRIPT

Nuclear magnetic resonance spectra were recorded on 300 and 500 MHz spectrometers. Chemical shifts for ^{1}H NMR spectra were recorded in parts per million from tetramethylsilane with the solvent resonance as the internal standard (chloroform, d = 7.26 ppm).

2.2 General (HPLC analysis)

Each sample has been diluted to final concentration of 0.1 mM in HPLC grade MeOH, filtered, and injected for analysis. Samples were analyzed on Waters HPLC 1525 binary pump system, integrated with Water 2487 UV-Vis dual wavelength detector and an autosampler. Chromatographic conditions: UV detection, λ_1 = 290 nm; λ_2 = 270 nm; Column: Kinetex 5µ C18 100A, 150 × 4.6 mm (Phenomenex), equipped with C18 pre-column; column temperature: 25 °C; injection volume: 10 µL; flow rate 0.5 mL/min. Mobile phase composition: linear gradient in 10 min, from 90% 10 mM Ammonium formate (AF) (pH 5)/10% MeOH v/v to 30% 10 mM AF (pH 5)/70% MeOH v/v; then reset to start conditions in 5 min. Total analysis time 20 min.

2.3 Reagents

(*R*)-2-hydroxy-N-(4-nitrophenethyl)-2-phenylacetamide, BH_3·THF 1M in THF, BH_3·DMS 2M in THF, BH_3·DMS 1M in 2-MeTHF were commercially available (Sigma Aldrich, Merck, TCI Europe, Alfa Aesar). THF and 2-MeTHF solutions of BH_3·THF and BH_3·DMS complexes were prepared from commercial solutions and their concentrations were determined by ^{1}H NMR analysis using BF_3·Et_2O as internal standard. [18]

2.4 Flow set up

All experiments were planned by Vapourtec Flow Commander Software in order to set-up solution flow rates and residence time of each experiment (Table 3). The following solutions were prepared and loaded in two different 2 mL PTFE loops: **solution A**: solution of (*R*)-2-hydroxy-N-(4-nitrophenethyl)-2-phenylacetamide **1** (0.2M in dry 2-MeTHF) and **solution B**: solution of BH_3·DMS in dry 2MeTHF (Table 3, 0.6M, entry 7, 1M, entry 2), obtained by diluting commercially available 2M solutions. The solutions were pumped in a 10 mL PTFE coil reactor at a flow rate of 125 µL/min (entry 7) at 90 °C with a residence time of 40 minutes, and at a flow rate of 167 µL/min (Table 3, entry 2) at 90 °C with a residence time of 30 minutes. The resulting solution was collected under steady state conditions, after 40 minutes and after 30 minutes (Table 3, entry 7 and 2 respectively). The output was quenched with H_2O and extracted with ethyl acetate.

3. Results and Discussion

ACCEPTED MANUSCRIPT

Inspired by our previous work concerning the safe handling of BH_3 in a microfluidic reactor, we decided to investigate the reduction step of amide **1** to amine **2** directly in a flow system. [19-27]
The first set of reactions were conducted in a flow reactor consisting of a 4 mL coil reactor, a 5 mL coil loop fed with HPLC pumps. Table 1 collects the output of this investigation.

Initially, the effect of temperature and residence time was evaluated (Table 1, entries 1-3). Modest yields were observed with higher residence time using BH_3·DMS as the reducing agent. Similar results were observed using BH_3·THF as reducing agent (Table 1, entry 4), whereas increasing the temperature up to 60 °C (Table 1, entry 5) a doubling of the yield was obtained, highlighting a key role of the temperature. However, using BH_3·DMS at 60 °C, furnished only 25% yield of the desired amine **2** (Table 1, entry 6). Interestingly, reintroducing the solution obtained under the conditions of entry 6 into the loop, and reacted with additional 5 equivalents of BH_3·DMS resulted in 70% yield of **2** (Table 1, entry 7). This last result suggested that longer residence times and higher loading of reducing agent would provide better yields. Eventually, temperature could be considered as critical parameter. Nevertheless, the decomposition of the reducing agent need to be taken into account working at higher temperatures.

3.1 Analysis of the reduction process under batch conditions

Building on our previous ^{11}B NMR experiments [18] on the stability of ethereal solutions of BH_3 complexes, we planned to test the reduction of **1** to **2** using BH_3·DMS and BH_3·THF complexes in batch conditions, and with different solvents (Table 2). Under batch conditions, full conversion was observed in THF after 90 and 60 minutes using BH_3·THF and BH_3·DMS respectively (Table 2, entries 1, 2). According to our NMR, evidences suggesting a higher stability of BH_3 complexes in 2-MeTHF, [18] a faster reaction was observed in this solvent with both BH_3·THF and BH_3·DMS complexes (Table 2, entries 3,4). Nicely, the use of the more stable BH_3·DMS complex in 2-MeTHF provide full conversion in 15 min reaction time.

With these evidences in hand, the reduction process was reinvestigated using a different flow reactor, and different reaction conditions. In particular, 2-MeTHF was employed as the solvent for two main reasons: a) its higher boiling point; b) the higher stability of the BH_3 complexes. In addition, it is worth pointing out that 2-MeTHF is a greener alternative to other ethereal solvents; it has a higher boiling point with respect to THF (bp 2-MeTHF = 80 °C; bp THF = 67 °C), a lower solubility in water (14 g per 100 g at 23 °C), and it is obtained by renewable feedstocks. [28,29] The new flow set-up used two 2 mL loops, a 10 mL PTFE coil reactor, and a back pressure regulator all connected to a Vapourtec system (Fig. 2). This flow system allowed using temperatures close or even higher than the boiling point for the reaction solvent.

ACCEPTED MANUSCRIPT

By using this new flow set-up (Table 3, entries 1, 2), high conversions were observed with both reducing agents (87% with BH_3·THF, and 92% with BH_3·DMS respectively) even if the reaction with BH_3·DMS proceeded to full conversion using 2-MeTHF as the solvent, 5 equiv of BH_3 complex, at 90 °C and with 30 minutes residence time. Interestingly, under these conditions, the product of double reduction **3** was also observed though in very low yields (Table 3, entries 1, 2). Moreover, being derivative **2** just the precursor of diamine **3**, its presence in the reaction mixture wouldn't represent a problem for further processing, according to Scheme 1. In order to further improve the process, after optimizing the reaction conditions in terms of temperature, solvent and reducing agent, we tackled the problem of the lowest number of equivalents of BH_3·DMS required for full conversion (Table 3, entries 3-8).

As reported in Table 3, reducing the amount of BH_3·DMS from 4 equiv. to 2 equiv., while maintaining the same residence time, resulted in lower conversion and lower yield of amine **2** (Table 3, entries 2-6). The best compromise was obtained using 3 equiv of BH_3·DMS with 40 minutes of residence time (Table 3, entry 7). Under these conditions, a 94% yield of amine **2** was obtained jointly to 4.6% of diamine **3**, so reaching 98.6% of useful compounds for further processing. Increasing the residence time, and reducing the amount of BH_3·DMS resulted in lower yield and conversion (Table 3, entry 8).

3.2 NMR monitoring of the flow reduction

The reaction mixtures obtained under flow conditions reported in Table 3, were analyzed by ^{1}H NMR in order to evaluate the amount of compounds **1**, **2** and **3** in such mixture (Fig. 3). By ^{1}H NMR analysis, clear signals for the aromatic protons of **1**, **2** and **3** could be recognized allowing for a quantitative evaluation. According to Table 3, entries 2 and 7 gave the best results in terms of conversions and yields.

Moreover, in the case of entry 7, where a lower amount of reducing agent was used, 1.4% of starting material could be detected, we found that acidic work up allowed for complete removal of amide **1** and full recover of reduction products **2** (96.6%) and **3** (3.4%) (see Supplementary material). [30]
It is worth mentioning that such conclusions were confirmed by a HPLC quantitative analysis (see Supplementary material).

4. Conclusions

In conclusion, a continuous-flow reduction of (*R*)-2-hydroxy-*N*-[2-(4-nitrophenyl)ethyl]-2-phenylacetamide, involved in the synthetic pathway of Mirabegron, has been developed. This study demonstrated the possibility to safely handly BH_3 complexes within microfluidic reactors using 2-MeTHF as greener alternative to traditional solvents, and without requiring any additive such as DMI. In addition, NMR and

ACCEPTED MANUSCRIPT

HPLC purity analysis revealed that the sole by-product of this process is the diamine **3**, which wouldn't affect the following steps towards Mirabegron.

Acknowledgements

We thank the University of Bari and Laboratorio SISTEMA, (Code PONa300369) financed by Italian Miur.

ACCEPTED MANUSCRIPT

References

[1] Patent WO2010147631 A1.

[2] Patent WO200842898 A2.

[3] Patent WO 2003037881 A1.

[4] Patent WO 2014132270 A2.

[5] B. Gutmann, D. Cantillo, C. O. Kappe, Angew. Chem. Int. Ed. 54 (2015) pp. 6688-6729.

[6] R. Singh, H.-J. Lee, A. Kumar Singh, D.-P. Kim, Korean J. Chem. Eng. 33 (2016) pp. 2253-2267.

[7] M. B. Plutschack, B. Pieber, K. Gilmore, P. H. Seeberger, Chem. Rev. (2017) DOI 10.1021/acs.chemrev.7b00183.

[8] W. Reschetilowski, Microreactors in Preparative Chemistry Ed., Wiley-VCH: Weinheim, 2013.

[9] T. Wirth, Microreactors in Organic Synthesis and Catalysis, 2nd ed., Ed., Wiley-VCH: Weinheim, 2013.

[10] V. Hessel, J. C. Schouten, A. Renken, Y. Wang, J.-i. Yoshida, Handbook of Micro Reactors, Eds., Wiley-VCH: Weinheim, 2009.

[11] S. V. Luis, E. Garcia-Verdugo, Chemical Reactions and Processes under Flow Conditions, Eds., RSC Green Chemistry, Royal Society of Chemistry: Cambridge, 2010.

[12] D. Dallinger, C. O. Kappe, Current Opinion in Green and Sustainable Chemistry 7 (2017) pp. 6-12.

[13] L. Pellegatti, J. Sedelmeier, Org. Process Res. Dev. 19 (2015) pp. 551-557.

[14] P. Poechlauer, J. Colberg, E. Fisher, M. Jansen, M. D. Johnson, S. G. Koenig, M. Lawler, T. Laporte, J. Manley, B. Martin, A. O'Kearney-McMullan, Org. Process Res. Dev. 17 (2013) pp. 1472-1478.

[15] S. G. Newman, K. F. Jensen, Green Chem. 15 (2013) pp. 1456-1472.

[16] K. F. Jensen, AIChE J., 63 (2017) pp. 858-869.

[17] A. D. Mamuye, S. Monticelli, L. Castoldi, W. Holzera, V. Pace, Green Chem. 17 (2015) pp. 4194-4197.

[18] De Angelis, S., Celestini, P., Rebuzzini, G., Degennaro, L., Luisi, R. Synthesis 49 (2017) pp. 1969-1971.

[19] L. Degennaro, M. Zenzola, P. Trinchera, L. Carroccia, A. Giovine, G. Romanazzi, A. Falcicchio, R. Luisi, Chem. Commun. 50 (2014) pp. 1698-1700.

[20] M. Zenzola, L. Degennaro, P. Trinchera, L. Carroccia, A. Giovine, G. Romanazzi, P. Mastrorilli, R. Rizzi, L. Pisano, R. Luisi, Chem. – Eur. J. 20 (2014) pp. 12190-12200.

[21] L. Degennaro, D. Maggiulli, C. Carlucci, F. Fanelli, G. Romanazzi, R. Luisi, Chem. Commun. 52 (2016) pp. 9554-9557.

[22] S. De Angelis, M. De Renzo, C. Carlucci, L. Degennaro, R. Luisi, Org. Biomol. Chem. 14 (2016) pp. 4304-4311.

[23] L. Degennaro, C. Carlucci, S. De Angelis, R. Luisi, J. Flow Chem. 6 (2016) pp. 136-166.

[24] L. Carroccia, B. Musio, L. Degennaro, G. Romanazzi, R. Luisi, J. Flow. Chem. 3 (2013) pp. 29-33.

[25] A. Giovine, B. Musio, L. Degennaro, A. Falcicchio, A. Nagaki, J.-i. Yoshida, R. Luisi, Chem. Eur. J. 19 (2013) pp 1872-1876.

ACCEPTED MANUSCRIPT

[26] L. Degennaro, F. Fanelli, A. Giovine, R. Luisi, Adv. Synth. Catal. 357 (2015) pp 21-24.

[27] L. Degennaro, A. Tota, S. De Angelis, M. Andresini, C. Cardellicchio, M. A. Capozzi, G. Romanazzi, R. Luisi Eur. J. Org. Chem. (2017) doi: 10.1002/ejoc.201700850.

[28] D. Prat, O. Pardigon, H.-W. Flemming, S. Letestu, V. Ducandas, P. Isnard, E. Guntrum, T. Senac, S. Ruisseau, P. Cruciani, P Hosek, Org. Process Res. Dev. 17 (2013) pp. 1517-1525.

[29] D. Prat, A. Wells, J. Hayler, H. Sneddon, C. R. McElroy, S. Abou-Shehadad, P. J. Dunne, Green Chem. 18 (2016) pp. 288-296.

[30] Attempts to induce, under batch conditions, complete reduction of amine **2** to diamine **3** failed even using a large excess of BH_3 DMS.

ACCEPTED MANUSCRIPT

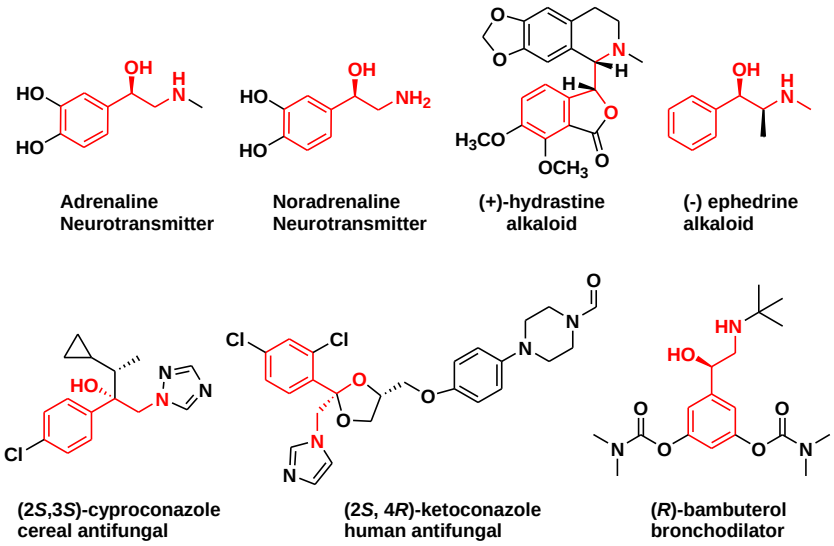


Fig. 1. Pharmaceutical molecules containing the phenylethanolamine core.

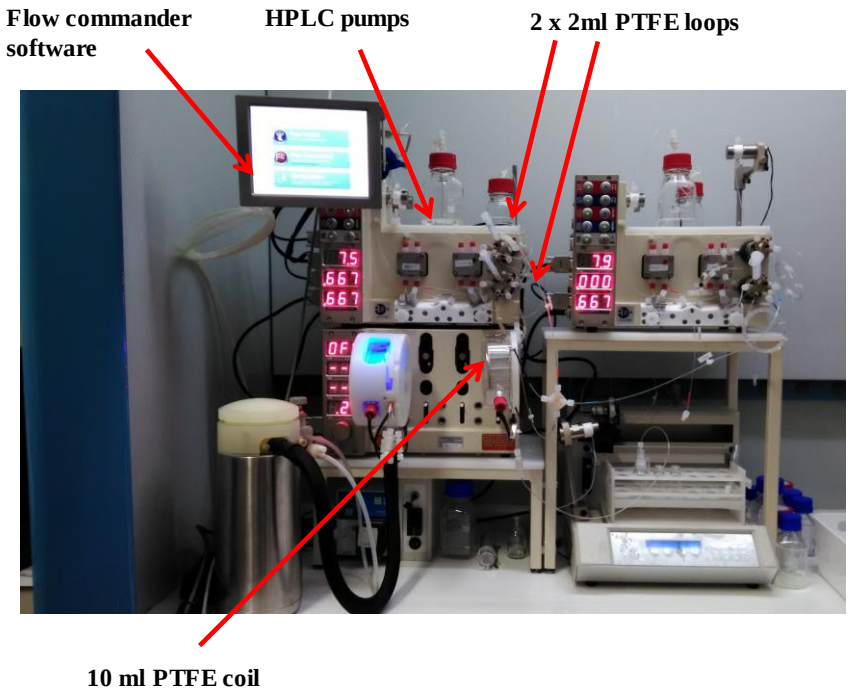


Fig. 2. Complete flow set-up Vapourtec

ACCEPTED MANUSCRIPT

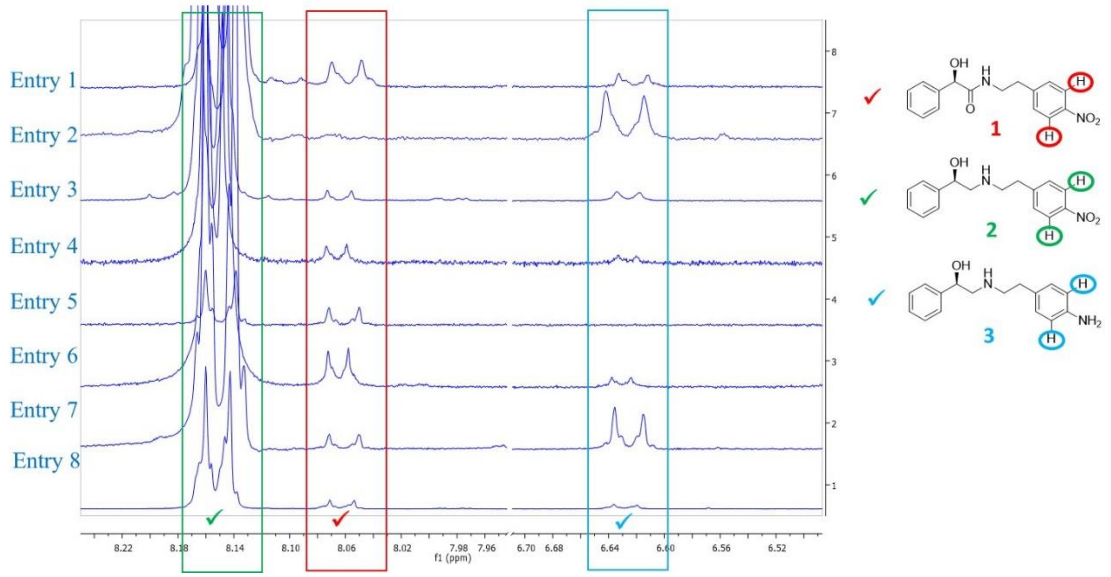
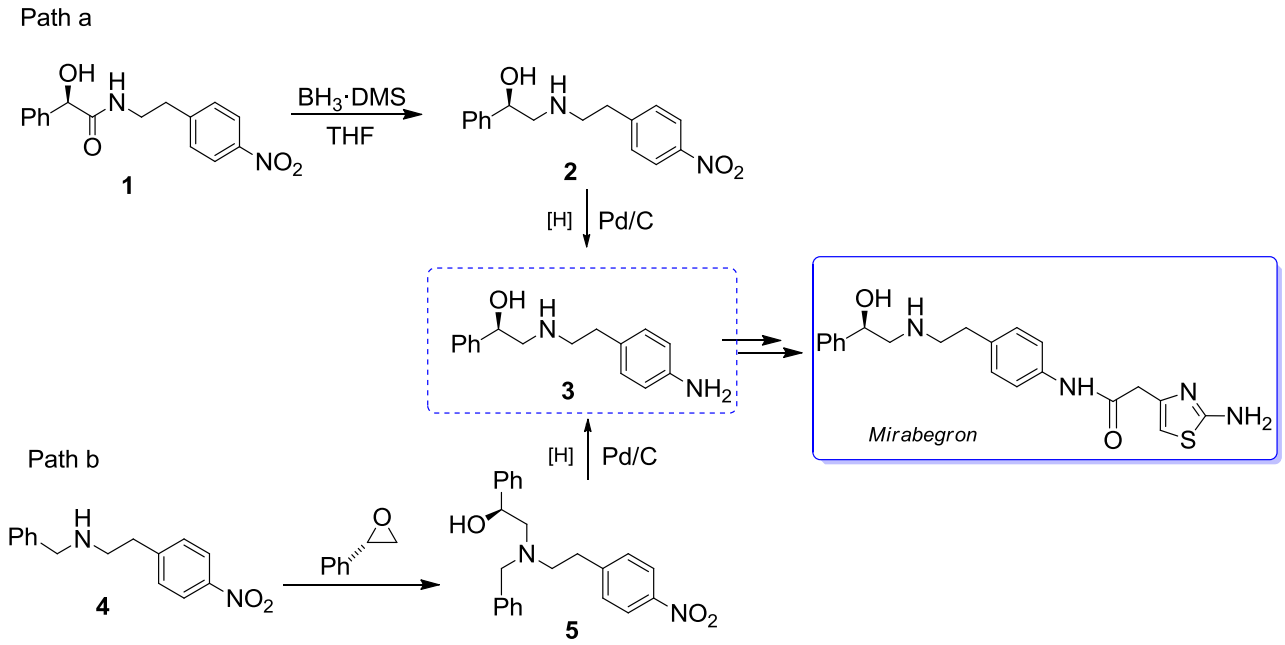


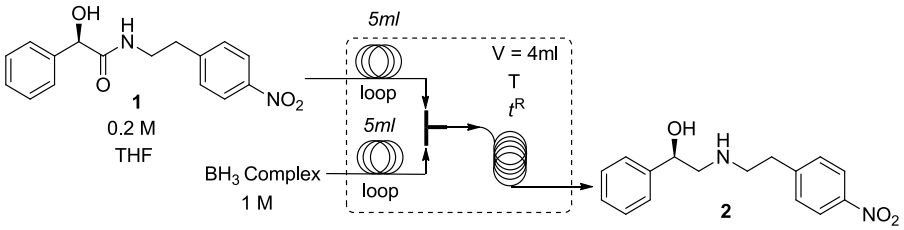
Fig. 3. ^{1}H-NMR monitoring of the flow reduction



Scheme 1 Synthetic pathways leading to Mirabegron

ACCEPTED MANUSCRIPT

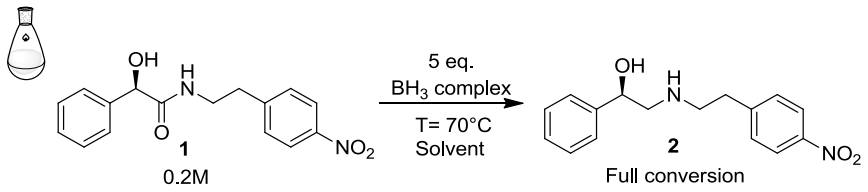
Table 1 Optimization study of the reduction of **1** with BH_3 in a continuous flow microreactor.



Entry	Reducing agent [mol/L]	Reducing agent (equiv)	Flow rate [a] (µL/min)	T (°C)	t_R (min)	Yield [b] (%)
1	BH_3·DMS [1M]	5	66.67	25	30	14
2	BH_3·DMS [1M]	5	66.67	50	30	14
3	BH_3·DMS [1M]	5	33.33	25	60	22
4	BH_3·THF [1M]	5	66.67	25	30	20
5	BH_3·THF [1M]	5	66.67	60	30	38
6	BH_3·DMS [1M]	5	66.67	60	30	25
7	BH_3·DMS [1M]	10 (2 runs)[c]	66.67	60	60 (2 runs)[c]	70

[a] Flow rates for solutions of **1** and BH_3. [b] Determined by ^{1}H-NMR using the internal standard. [c] The solution resulting from entry 6 was reintroduced into the loop and reacted with additional 5 equiv. of BH_3·DMS with a t_R of additional 30 min.

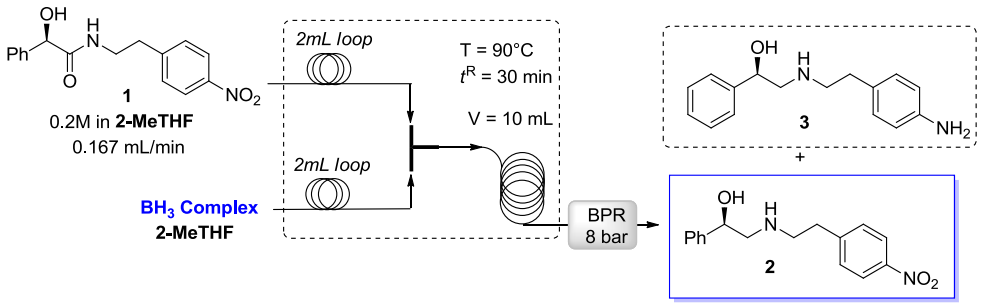
Table 2 Batch study of the reduction of amide **1** to amine **2**.



Entry	BH_3 Complex	T (min)	Solvent
1	BH_3·THF	90	THF
2	BH_3·DMS	60	THF
3	BH_3·THF	30	2-MeTHF
4	BH_3·DMS	15	2-MeTHF

Table 3 Optimization study of the reduction of **1** (0.2 M) in 2-MeTHF and BH_3DMS (2M in THF)

ACCEPTED MANUSCRIPT



Entry	BH_3 Complex	BH_3 (equiv)	Flow rate (µL/min)	t^R (min)	**1** (%)[a]	**2** (%)[a]	**3** (%)[a]
1	BH_3·THF	5	167	30	10.4	86.6	3
2	**BH_3·DMS**	**5**	**167**	**30**	**0**	**91.7**	**8.3**
3	BH_3·DMS	4	167	30	2	96	2
4	BH_3·DMS	3	167	30	6.3	89.2	4.5
5	BH_3·DMS	2	167	30	23	77	0
6	BH_3·DMS	2.5	167	30	4.7	94	1.3
7	**BH_3·DMS**	**3**	**125**	**40**	**1.4**	**94**	**4.6**
8	BH_3·DMS	2.5	100	50	10	84	6

[a] Flow rate of **1** and BH_3 Complex [b] Determined by ^{1}H-NMR using internal standard