Microwave-Assisted Homogeneous Acid Catalysis and Chemoenzymatic Synthesis of Dialkyl Succinate in a Flow Reactor

Laura Daviot 1, Thomas Len 1, Carol Sze Ki Lin 2 and Christophe Len 1,3,*

1 Centre de Recherche de Royallieu, Université de Technologie Compiègne, Sorbonne Universités, Cedex BP20529, F-60205 Compiègne, France; laura.daviot@gmail.com (L.D.); thomaslen@orange.fr (T.L.)
2 School of Energy and Environment, City University of Hong Kong, Tat Chee Avenue, Kowloon Tong, Hong Kong, China; carollin@cityu.edu.hk
3 Institut de Recherche de Chimie Paris, PSL Research University, Chimie ParisTech, CNRS, UMR 8247, Cedex 05, F-75231 Paris, France
* Correspondence: christophe.len@chimieparistech.psl.eu; Tel.: +33-144-276-752

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Abstract: Two new continuous flow systems for the production of dialkyl succinates were developed via the esterification of succinic acid, and via the trans-esterification of dimethyl succinate. The first microwave-assisted continuous esterification of succinic acid with H2SO4 as a chemical homogeneous catalyst was successfully achieved via a single pass (ca 320 s) at 65–115 °C using a MiniFlow 200ss Sairem Technology. The first continuous trans-esterification of dimethyl succinate with lipase Cal B as an enzymatic catalyst was developed using a Syrris Asia Technology, with an optimal reaction condition of 14 min at 40 °C. Dialkyl succinates were produced with the two technologies, but higher productivity was observed for the microwave-assisted continuous esterification using chemical catalysts. The continuous flow trans-esterification demonstrated a number of advantages, but it resulted in lower yield of the target esters.

Keywords: continuous flow; dialkyl succinates; homogeneous catalysis; lipase Cal B; succinate

1. Introduction

With the depletion of oil-based resources, wood-based biomass and especially plant waste rich in lignocellulosic feedstocks appear to be the main alternatives for the production of platform molecules. Among them, succinic acid (SA) as a linear C-4 dicarboxylic acid is considered as one of the top 12 prospective building blocks derived from sugars by the US Department of Energy. SA is mainly produced via a chemical catalytic route starting from maleic acid and maleic anhydride. The use of furan-derived SA at laboratory-scale using chemical process, as well as via biotechnological process (i.e., by fermentation) have also been studied [1]. SA can be used as a precursor to produce different chemical intermediates [2], such as tetrahydrofuran [3], γ-butyrolactone [4], and 1,4-butanediol [5]. Particularly, SA ester products can be used in the chemical industry as a green solvent, or plastic and fuel additive, as well as in the pharmaceutical and cosmetic industries [6]. Different processes using chemical homogeneous catalysis [7–10], heterogeneous catalysis [11–21], and chemo-enzymatic reaction [22] have been reported in batch process, but few reports have described continuous flow dialkyl succinate synthesis [21,23]. Among the dialkyl succinates with value-added properties, dimethyl-, diethyl-, di-isobutyl-, and dioctyl succinates can be used as green solvents; dibutyl-, didecyl-, diamyl-, and diisoamyl succinates can be used as plastic and fuel additives; tocopherol, estriol, chloramphenicol, and hydrocortisone succinates as pharmaceutical ingredients; and dipropyl, diethoxyethyl, or diethylhexyl succinates in cosmetic application [1]. Processes for the production of dialkyl succinates in a batch
reactor were developed in 2010 (Table 1). Among them, the use of sulfonic acid was the most described [8–11,14–17], followed by carboxylic acid [18] and phosphoric acid [19]. It is difficult to compare each result since the processes were conducted in different conditions by different groups. Nevertheless, the use of alcohol as both solvent and reagent was often in excess at temperature in the range of 25–160 °C for 25 h. Dialky succinates were produced in yields higher than 66%. Al₂O₃ was described as a heterogeneous catalyst at 25 °C for 48 h for the synthesis of dimethyl ester 2b in 70% yield [20]. Among the recent reports, Zhang et al. described the continuous flow synthesis of diesters 2a, 2b, and 2d in the presence of “man-made” heterogeneous catalyst in quantitative yield [21]. Moreover, Fabian et al. described the use of batch microwave radiation as alternative tool for the esterification of SA [14]. To the best of our knowledge, the chemoenzymatic production of diesters 2a–i using both pure SA (1) and pure dimethylester 2b as reactants has never been reported. Nevertheless, Delhomme et al. used crude fermentation broths produced from recombinant Escherichia coli for the synthesis of 2h in the presence of lipase Cal B [22].

Table 1. Selected catalysts reported for the conversion of succinic acid (1) to dialkyl succinates 2.

| Entry | Reactor | Catalyst | Reaction Conditions a | Yield of 2 (%) | Ref |
|-------|---------|----------|----------------------|----------------|-----|
| 1     | batch   | H₂SO₄    | nd:2:110:18          | 2g             | 69  | [6] |
| 2     | batch   | H₂SO₄    | nd:2:3:110:18        | 2f             | 78  | [9] |
| 3     | batch   | OPP-SO₂H-1 | 10:50:70:6         | 2h             | 70  | [9] |
| 4     | batch   | SS-0.010 | 10:2:100:6.5        | 2a             | 94  | [16]|
| 5     | batch   | Glu-TsOH | 100:80:80:4         | 2a             | 100 | [17]|
| 6     | batch   | CH₃SO₄H8@Al₂O₃ | 332,000:2:80:8 | 2b   | 97  | [14]|
| 7     | batch   | CH₃SO₄H8@Al₂O₃ | 332,000:2:80:8 | 2a   | 97  | [14]|
| 8     | batch   | CH₃SO₄H8@Al₂O₃ | 332,000:2:80:8 | 2c   | 97  | [14]|
| 9     | batch   | CH₃SO₄H8@Al₂O₃ | 332,000:2:80:8 | 2i   | 97  | [14]|
| 10    | batch   | C₆(Mim)₂SO₄ | 2:3:60:3  | 2b   | 76  | [10]|
| 11    | batch   | C₆(Mim)₂SO₄ | 2:3:60:3.5 | 2a   | 68  | [10]|
| 12    | batch   | C₆(Mim)₂SO₄ | 2:4:60:4  | 2c   | 74  | [10]|
| 13    | batch   | N-Butyl-2,4-dinitro-anilinium | 1:2:99:25 | 2h   | 93  | [7] |
| 14    | batch   | nano-SO₂ 2-/TiO₂ | 5:3:160:2  | 2g   | 97  | [11]|
| 15    | batch   | TSA₃/MCM-41 | 0.1:3:80:14     | 2a   | 66  | [18]|
| 16    | batch   | TSA₃/MCM-41 | 0.1:3:80:14     | 2a   | 66  | [18]|
| 17    | batch   | TPA₂/MCM-41 | 100:3:80:8     | 2d   | 68  | [19]|
| 18    | batch   | TPA₂/MCM-41 | 100:3:80:8     | 2f   | 68  | [19]|
| 19    | batch   | TPA₂/MCM-41 | 100:3:80:8     | 2f   | 68  | [19]|
| 20    | batch   | TPA₂/MCM-41 | 100:3:80:8     | 2h   | 73  | [19]|
| 21    | batch   | Al₂O₃    | 50:1.6:25:48     | 2b   | 70  | [20]|
| 22    | flow    | PIL-A    | 5:1:2:85:5       | 2b   | 100 | [21]|
| 23    | flow    | PIL-A    | 5:1:2:87:4       | 2a   | 100 | [21]|
| 24    | flow    | PIL-A    | 5:1:2:100:3.5    | 2d   | 100 | [21]|

a Reaction conditions: amount of catalysts (mg%), in some cases unit is mg), mole ratio of alcohol/succinic acid:reaction temperature (°C):reaction time (h). b Microwave-assisted esterification. OPP-SO₂H-1: organic knitted porous polyaromatics with pyrene; SS-0.001: silica-supported sulfate with sulfate loading 0.001 mol; TSA₃/MCM-41: [12]-tungstosilic acid (30 w%) anchored to MCM-41; TPA₂/MCM-41: terephthalic acid (20%) anchored to MCM-41; PIL-A: acidic polymeric liquid.

Recently, the use of homogeneous and heterogeneous flow systems in organic chemistry have been widely studied because of their highly efficient heat transfer compared with batch methodologies,
good temperature monitoring, and enhanced mass transfer [24–33]. This innovative approach also permits the time required to progress from research to pilot scale and production to be reduced. Due to our interest in the topic of green chemistry and alternative technologies, two continuous-flow systems for the production of dialkyl succinate were envisaged to develop an intensified process. Herein, we report an efficient extension of this work in order to establish a comparison between the homogeneous acid and the enzymatic continuous flow system for the production of selected dialkyl succinates.

2. Results and Discussion

Initial batch diesterification was performed using SA (1, 2 M) and ethanol (10 mL) in the presence of H$_2$SO$_4$ (10 mol %) at 170 °C under microwave irradiation for the production of the corresponding diester 2a (Table 2). In the presented work, the reaction time was determined by HPLC monitoring either until no more conversion of the starting diacid 1 was observed, or within the maximum time of one hour with magnetic stirring (600 rpm). The optimization of the reaction conditions for the esterification of SA (1) with both acid catalysts and enzymes was first realized with a single-variable strategy, by varying one variable at a time while keeping the others constant. For the present work, error bars represent the standard deviation of five replicates. Different Bronsted acids, including H$_2$SO$_4$, H$_3$PO$_4$,  $p$-toulenesulfonic acid (PTSA) and 10-camphorsulfonic acid (CSA), were tested with a concentration of 10 mol % (Table 2, entries 1–4). CSA and H$_2$SO$_4$ gave identical yields, and for economic reasons, H$_2$SO$_4$ was selected for the following study. It should be pointed out that the use of PTSA and H$_3$PO$_4$ as acid catalysts resulted in a lower yield (77% and 50%) for the same reaction time (Table 2, entries 1 and 3). The experimental results with variation of H$_2$SO$_4$ (5–20 mol %) demonstrates that the maximum yield was obtained in the presence of 20 mol % of the acid (Table 2, entry 5). Using these conditions without catalyst, compound 2a was obtained in a low yield (9%). The acidity of the catalysts used were different (PTSA pKa $-6.5$; H$_2$SO$_4$ pKa $-3.0$; 1.9; CSA pKa 1.2 and H$_3$PO$_4$ pKa 2.1, 7.0 and 12.0). The lack of reactivity of H$_3$PO$_4$ can be related to its low acidity compared with H$_2$SO$_4$ while PTSA with a strong acidity may favor the saponification of the ester 2a.

| Entry | Acid  | [Acid] (mol %) | Yield of 2a (%) | Error Bar |
|-------|-------|----------------|-----------------|-----------|
| 1     | PTSA  | 10             | 77              | 1.48      |
| 2     | CSA   | 10             | 84              | 1.09      |
| 3     | H$_3$PO$_4$ | 10       | 50              | 1.52      |
| 4     | H$_2$SO$_4$ | 10       | 84              | 0.55      |
| 5     | H$_2$SO$_4$ | 20       | 87              | 0.84      |
| 6     | H$_2$SO$_4$ | 30       | 82              | 1.14      |
| 7     | H$_2$SO$_4$ | 5        | 70              | 3.36      |

* The diethyl succinate yield was calculated from gas chromatography analysis with a calibration curve. CSA: 10-camphorsulfonic acid; PTSA: $p$-toulenesulfonic acid.

Based on these previous results obtained in a batch reactor, the initial reaction using the microwave continuous flow was conducted with SA (1, 0.15–0.27 M) in the presence of H$_2$SO$_4$ (5–20 mol %) in ethanol. The molar concentration was more diluted in the flow device compared with the batch reactor due to viscosity. Starting from SA (1, 0.22 M) and H$_2$SO$_4$ (20 mol %), the temperature was fixed close to the boiling point of ethanol (75 °C) with a power input of 150 W, and the residence time was fixed at 100 s for this mixture. Conversion of SA (1) and the yield of diethyl succinate (2a) were 45% and
32%, respectively. In order to improve the process, residence times were increased from 100 to 400 s. The optimal residence time was obtained at 320 s with a quantitative conversion of SA (1) and yield of diethyl succinate (2a). Using a lower amount of H$_2$SO$_4$ (5 and 10 mol %) and variation of the amount of SA (1, 0.15 and 0.27 M) resulted in lower yields of diethyl succinate (Table 3, entries 5–8). The use of lower temperature (30 °C and 50 °C) did not lead to improvement in conversion and yield (Table 3, entries 9 and 10).

Table 3. Continuous flow microwave-assisted diethyl succinate (2a) synthesis by varying the amount of SA (1), H$_2$SO$_4$, the residence time, and the temperature at 150 W.

| Entry | 1 (mol L$^{-1}$) | H$_2$SO$_4$ (mol %) | Temperature (°C) | Residence Time (s) | Conversion (%) $^a$ | Yield of 2a (%) $^a$ | Error Bar |
|-------|----------------|---------------------|------------------|-------------------|-------------------|---------------------|-----------|
| 1     | 0.22           | 20                  | 75               | 100               | 45                | 32                  | 0.71      |
| 2     | 0.22           | 20                  | 75               | 320               | 100               | 99                  | 0.45      |
| 3     | 0.22           | 20                  | 75               | 320               | 100               | 99                  | 0.55      |
| 4     | 0.22           | 20                  | 75               | 400               | 100               | 99                  | 0.55      |
| 5     | 0.22           | 10                  | 75               | 320               | 85                | 82                  | 0.89      |
| 6     | 0.22           | 5                   | 75               | 320               | 75                | 68                  | 0.89      |
| 7     | 0.22           | 5                   | 75               | 320               | 95                | 90                  | 2.17      |
| 8     | 0.15           | 20                  | 75               | 320               | 82                | 78                  | 1.52      |
| 9     | 0.22           | 20                  | 50               | 320               | 73                | 68                  | 1.52      |
| 10    | 0.22           | 20                  | 30               | 320               | 35                | 16                  | 2.41      |

$^a$ The diethyl succinate yield was calculated from gas chromatography analysis with a calibration curve.

Various primary and secondary alcohols having linear and branched carbon chains were subjected to the continuous esterification under our optimized conditions (Figure 1). Due to viscosity, butan-1-ol and alcohols with higher molecular weight were used at 0.18 M. Yields decreased proportionally with the increase in the number of carbons in the chain. Using primary alcohols, the conversion of SA (1) and yields of the selected dialkyl succinates (2a–e) were higher than 95% and 88%, respectively (Table 4, entries 1–5). For those primary alcohols with more than six carbon atoms, productivity decreased with yields between 65% and 80% (Table 4, entries 6–8). In contrast, the use of secondary alcohols gave similar conversion (98%) and lower yields (36% for 2i and 89% for 2j) (Table 4, entries 9 and 10). To the best of our knowledge, this is the first investigation which reports dialkyl succinates produced in a continuous flow. More parameters can be explored, but the present yields were similar to those obtained in the literature with batch process.
Table 4. Scope of the microwave-assisted continuous flow dialkyl succinate 2a-j synthesis at 75 °C.

| Entry | 1 (mol L⁻¹) | Temperature (°C) | Conversion (%) a | Diesters 2 | Yield of 2a-j (%) a | Error Bar |
|-------|-------------|------------------|------------------|------------|---------------------|-----------|
| 1     | 0.22        | 65               | 100              | 2b         | 100                 | 0.89      |
| 2     | 0.22        | 75               | 100              | 2a         | 99                  | 0.55      |
| 3     | 0.22        | 95               | 95               | 2c         | 92                  | 0.89      |
| 4     | 0.18        | 115              | 98               | 2d         | 89                  | 0.55      |
| 5     | 0.18        | 115              | 98               | 2e         | 88                  | 1.30      |
| 6     | 0.18        | 115              | 97               | 2f         | 78                  | 1.30      |
| 7     | 0.18        | 115              | 98               | 2g         | 80                  | 4.55      |
| 8     | 0.18        | 115              | 96               | 2h         | 65                  | 0.84      |
| 9     | 0.22        | 80               | 98               | 2i         | 89                  | 1.64      |
| 10    | 0.18        | 96               | 98               | 2j         | 36                  | 1.95      |

a The dialkyl succinate yield was calculated from gas chromatography analysis with a calibration curve.

For fair comparison, compounds 2f–h were obtained by Stuart et al. [8,9] starting with a molar ratio of alcohol:SA (2:1) in the presence of H₂SO₄ as a catalyst at 110 °C for 18 h in a batch reactor. The yields of compounds 2f–h were 78%, 69%, and 70%, respectively. In our optimized microwave-assisted flow synthesis, alcohols were used in large excess at similar temperature range (115 °C) for a residence time of 320 s. In this study, the yields of diesters 2f–h were similar. It is obvious that the decrease in residence time (18 h vs. 320 s) led to significant improvement in the synthesis of biobased chemicals via esterification.

In order to explore high selectivity and smooth reaction conditions, continuous flow and bioconversion with Novozymes® 435, the lipase B from Candida antarctica immobilized on acrylic resin (Cal B) were studied in batch and flow reactors. The optimization of the reaction conditions for the trans-esterification of dimethylester 2b with enzymes was realized as reported above with the acid catalysts. To probe the scope of the methodology, the influence of thermal heating, the amount of starting material 2b, and the amount of Cal B were examined (Table 5). Dimethyl ester 2b (50 mM) and Cal B (270 g) in ethanol were mixed in a batch reactor for 24 h by varying the temperature. Whatever the temperature used, the yield of diethyl succinate 2a was 60% except for temperature above 60 °C due to the instability of the enzyme at high temperature (Table 5, entries 1–4). For these reasons, temperature of 20 °C was chosen and variation of the amount of enzyme was studied. For a quantity of 200 mg and 270 mg, the yields of the diesters 2a were similar while for smaller quantities the yield of diethyl succinate 2a were too low (Table 5, entries 5–7). The use of concentrated solution of dimethylester 2b were tested at 20 °C in the presence of Cal B (200 mg), but the yield of diethyl succinate 2a decreased (Table 5, entries 8 and 9).

For the transfer of the enzymatic trans-esterification from batch to continuous flow, dimethyl ester 2b (50 mM) and Cal B (200 mg) were tested at 20 °C with different residence times (7, 2.3, and 1.2 min). The longer the time, the better the yield, regardless of the amount of the diester 2b, enzyme dosage, and temperature (Table 6). Only dimethyl ester 2b in the presence of a minimum amount of enzyme (200 mg) at 40 °C with a time of 7 min allowed the production of diethyl ester 2a with a yield higher than 20% (Table 6, entries 4 and 13). It should be noted that for a doubling time of 14 min, the diester 2a yield was 48% (Table 6, entry 23). In these optimized conditions, the use of Cal B (100 mg) resulted in only 34% (Table 6, entry 24).
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(115 °C) for a residence time of 320 s. In this study, the yields of diesters resulted in only 34% (Table 6, entry 24).

Table 5. Batch chemoenzymatic synthesis of diethyl succinate (2a) by varying the concentration and temperature.

| Entry | 2b (M) | Cal B (mg) | Temperature (°C) | Yield of 2a (%) a | Error Bar |
|-------|--------|------------|------------------|-------------------|-----------|
| 1     | 0.050  | 270        | 20               | 60                | 0.89      |
| 2     | 0.050  | 270        | 40               | 60                | 1.22      |
| 3     | 0.050  | 270        | 60               | 60                | 0.55      |
| 4     | 0.050  | 270        | 80               | 20                | 2.51      |
| 5     | 0.050  | 40         | 20               | 30                | 1.30      |
| 6     | 0.050  | 130        | 20               | 55                | 1.73      |
| 7     | 0.050  | 200        | 20               | 60                | 1.09      |
| 8     | 0.10   | 200        | 20               | 50                | 1.30      |
| 9     | 0.20   | 200        | 20               | 45                | 1.30      |

a The yield of diethyl succinate was calculated from gas chromatography analysis with a calibration curve.

Table 6. Flow chemoenzymatic synthesis of diethyl succinate (2a) by varying the concentration, temperature, and residence time.

| Entry | 2b (M) | Cal B (mg) | Residence Time (min) | Temperature (°C) | Conversion of 2b (%) a | Yield of 2a (%) a | Error Bar |
|-------|--------|------------|----------------------|------------------|------------------------|-------------------|-----------|
| 1     | 0.050  | 200        | 7                    | 20               | 90                     | 7                 | 1.00      |
| 2     | 0.050  | 200        | 2.3                  | 20               | 79                     | 1                 | 0.55      |
| 3     | 0.050  | 200        | 1.2                  | 20               | 76                     | 1                 | 0.45      |
| 4     | 0.050  | 200        | 7                    | 40               | 99                     | 23                | 1.30      |
| 5     | 0.050  | 200        | 2.3                  | 40               | 73                     | 2                 | 0.84      |
| 6     | 0.050  | 200        | 1.2                  | 40               | 73                     | 3                 | 0.89      |
| 7     | 0.050  | 200        | 7                    | 60               | 95                     | 18                | 1.22      |
| 8     | 0.050  | 200        | 2.3                  | 60               | 78                     | 5                 | 1.41      |
| 9     | 0.050  | 200        | 1.2                  | 60               | 73                     | 2                 | 1.00      |
| 10    | 0.050  | 100        | 7                    | 40               | 60                     | 14                | 1.09      |
| 11    | 0.050  | 100        | 2.3                  | 40               | 68                     | 6                 | 0.89      |
| 12    | 0.050  | 100        | 1.2                  | 40               | 63                     | traces            | 0.09      |
| 13    | 0.050  | 400        | 7                    | 40               | 100                    | 24                | 0.89      |
| 14    | 0.050  | 400        | 2.3                  | 40               | 97                     | 12                | 1.30      |
| 15    | 0.050  | 400        | 1.2                  | 40               | 91                     | 5                 | 0.89      |
| 16    | 0.025  | 200        | 7                    | 40               | 89                     | 10                | 1.30      |
| 17    | 0.025  | 200        | 2.3                  | 40               | 72                     | 3                 | 0.89      |
| 18    | 0.025  | 200        | 1.2                  | 40               | 75                     | 1                 | 0.27      |
| 19    | 0.100  | 200        | 7                    | 40               | 94                     | 14                | 1.41      |
| 20    | 0.100  | 200        | 2.3                  | 40               | 85                     | 5                 | 0.89      |
| 21    | 0.100  | 200        | 1.2                  | 40               | 78                     | 3                 | 0.27      |
| 22    | 0.050  | 200        | 28                   | 40               | 95                     | 18                | 1.30      |
| 23    | 0.050  | 200        | 14                   | 40               | 96                     | 48                | 1.52      |
| 24    | 0.050  | 100        | 14                   | 40               | 100                    | 34                | 1.09      |

a The diethyl succinate yield was calculated from gas chromatography analysis with a calibration curve.

In order to expand the array of substrates, dimethyl ester 2b was coupled with a variety of primary and secondary alcohols with linear and branched alkyl chains (Scheme 1). In general, the yields were twice as low as those obtained during esterification in the batch reactor. Exceptions were observed for diesters 2f, 2g, and 2h, which were obtained with much lower yields. Nevertheless, the variation in yields according to the alcohol used was similar.
In a typical experiment, a solution containing dimethyl succinate (2b, 50 mM, ROH (27 mL)) was pumped with a peristatic pump (5 tr/min). The solution was passed through a reactor under microwave activation (MiniFlow 200ss, Sairem®) at 40 °C with a residence time of 74 min. The selectivity of the chemoenzymatic synthesis of dialkyl succinates 2a and 2c–j was low using Cal B because the residence time was too low to have the second esterification. With a good conversion of the dimethylster 2b, the first trans-esterification was obtained to furnish the intermediate and then the second trans-esterification as a limiting step gave the target compounds 2a and 2c–j in low-to-moderate 13%–54% yields.

3. Experimental Methods

3.1. Materials

Substrate alcohols (MeOH, EtOH, PrOH, iso-PrOH, BuOH, iso-BuOH, sec-BuOH, HexOH, 2-Et-HexOH, and OctOH) and succinic acid were purchased from Fisher Scientific (Leicestershire, United Kingdom). Diethyl succinate (2a) was purchased from TCI Europe (Zwijndrecht, Belgium); dimethyl succinate (2b), dipropyl succinate (2c), dibutyl succinate (2d), and diisopropyl succinate (2i) were purchased from Sigma-Aldrich (Saint Louis, MO, USA). Diisobutyl succinate (2e) and di-sec-butyl succinate (2j) were purchased from AKos Consulting & Solutions GmbH (Steinen, Germany). Dihexyl succinate (2f), diethylhexyl succinate (2g), and dioctyl succinate (2h) were purchased from Hangzhou DayangChem Co. Ltd. (Hangzhou, China), BOC Sciences (Shirley, NY, USA), and Carbosynth Europe (Berkshire, United Kingdom), respectively. All materials were used without purification.

3.2. Microwave-Assisted Continuous Chemical Esterification

In a typical experiment, a 500-mL Erlenmeyer flask was first filled with succinic acid (1, 6.50 g, 55.1 mmol, 1 equiv.) and H2SO4 (1.08 g, 11.1 mmol, 0.2 equiv.) in alcohol (250–300 mL). The mixture was stirred at room temperature for 10 min, and it was pumped with a peristatic pump (5 tr·min⁻¹). The solution was passed through a reactor under microwave activation (MiniFlow 200ss, Sairem®) at 65–115 °C (150 W) with a residence time of 320 s. Among the outlet solution, one milliliter of mixture was collected, pH was adjusted to 7 by washing the mixture with 5% NaOH (0.5 mL), followed by water (0.5 mL) and saturated aqueous NaCl solution (0.5 mL). Then, the organic layer was dried over anhydrous Na2SO4 and the solvent was removed under reduced pressure. The aqueous phase was analyzed by HPLC in order to determine the remaining succinic acid concentration, and the organic phase was analyzed by gas chromatography to quantify the amount of esters produced.

3.3. Continuous Biochemical Trans-Esterification

In a typical experiment, a solution containing dimethyl succinate (2b, 200 mg, 1.37 mmol, 1 equiv.) in alcohol (27 mL) was pumped at 0.05 mL min⁻¹ using Syrris Asia equipment (Syrris, England). The solution was passed through a cartridge filled with Cal B (200 mg) at 40 °C, leading to a residence time of 14 min. Among the outlet solution, one milliliter of mixture was collected and saturated aqueous NaCl solution (1 mL) was added. Then, the organic layer was dried over anhydrous Na2SO4 and the solvent was removed under reduced pressure. The aqueous phase was analyzed by HPLC in order to determine the remaining succinic acid concentration, and the organic phase was analyzed by gas chromatography to quantify the amount of esters produced.
3.4. Gas Chromatography (GC) Analysis

Gas chromatography analyses of the organic phase were performed by a Perkin-Elmer gas chromatography instrument (Autosystem XL GC) (Perkin-Elmer, Singapore) using an Altech AT HT column with a detector at 300 °C, an injector at 340 °C, and a constant flow of nitrogen of 1 mL min⁻¹. The column was heated at 150 °C for 2 min, and the column temperature was then raised to 350 °C with a temperature gradient of 15 °C min⁻¹ before being held at this temperature for 4.67 min. Succinic esters were identified and quantified by comparing GC retention time and peak area with their respective calibration standards.

3.5. High-Performance Liquid Chromatography (HPLC) Analysis

Liquid chromatography analyses of the aqueous phase were performed by a Hewlett-Packard 1090 HPLC using a reversed phase C18 column (Novapak 3.9 mm × 150 mm) held at 40 °C. Water/acetonitrile (ACN) mixture was used as the mobile phase (0.8 mL min⁻¹) in a gradient mode (0% ACN at t = 0 min to 60% ACN at t = 20 min to 90% ACN at t = 25 min to 0% ACN at t = 28 min). The species were identified by UV detection (Hitachi L400H, San Diego, CA, USA) at a wavelength of 210 nm. Succinic acid was identified and quantified by comparing GC retention time and peak area with their respective calibration standard.

4. Conclusions

In this study, two clean, mild, reproducible, and scalable continuous flow process for the production of different dialkyl succinates using H₂SO₄ as the homogeneous catalyst and Cal B as the heterogeneous catalyst were developed. A scope of different linear and branched alcohols was successfully formulated based on the optimized protocols, leading to the target chemicals. The homogeneous protocol furnished excellent yields (≥88%) when alcohols containing less than six carbon atoms were used. One exception was observed with butan-2-ol as the reactant, which is probably due to the hindered or solubility effects. For alcohols with higher molecular weight, productivities decreased with yields between 65% and 80% even if the conversion of SA (1) was almost quantitative. In comparison with chemical homogenous catalysis, the chemoenzymatic protocol resulted in lower yields in the order of two at best with longer residence time (14 min vs. 5 min). The lack of reactivity must be due to the lower temperature, which is related to the low thermal stability of the enzyme. To the best of our knowledge, this is the first time that dialkyl succinates have been produced in continuous flow either by chemical catalysis or enzymatic catalysis. The first method gave excellent yields and it is possible on a larger scale. The second method requires optimization through the screening of more effective enzymes.

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References

1. Mazière, A.; Prinsen, P.; Garcia, A.; Luque, R.; Len, C. A review of progress in (bio)catalytic routes from/to renewable succinic acid. Biofuels Bioprod. Bioref. 2017, 11, 908–931. [CrossRef]

2. Delhomme, C.; Weuster-Botz, D.; Kuhn, F.E. Succinic acid from renewable resources as a C-4 building-block chemical—A review of the catalytic possibilities in aqueous media. Green Chem. 2009, 11, 13–26. [CrossRef]
3. Hong, U.G.; Kim, J.K.; Lee, J.; Lee, J.K.; Song, J.H.; Yi, J.; Song, I.K. Hydrogenation of succinic acid to tetrahydrofuran (THF) over rhenium catalyst supported on H2SO4-treated mesoporous carbon. J. Ind. Eng. Chem. 2014, 20, 3834–3840. [CrossRef]
4. Shao, Z.; Li, C.; Di, X.; Xiao, Z.; Liang, C. Aqueous-phase hydrogenation of succinic acid to g-butyl lactone and tetrahydrofuran over Pd/C, Re/C, and Pd-Re/C catalysts. Ind. Eng. Chem. Res. 2014, 53, 9638–9645. [CrossRef]
5. Kang, K.H.; Hong, U.G.; Bang, Y.; Choi, J.H.; Kim, J.K.; Lee, K.L.; Han, S.J.; Song, I.K. Hydrogenation of succinic acid to 1,4-butanediol over Re-Ru bimetallic catalysts supported on mesoporous carbon. Appl. Catal. A 2015, 490, 153–162. [CrossRef]
6. Delhomme, C. Process Integration of Fermentation and Catalysis for the Production of Succinic Acid Derivatives. Ph.D. Thesis, Technische Universitat München, Institute of Biochemical Engineering, Munich, Germany, 2011.
7. Sattenapally, N.; Wang, W.; Liu, H.; Gao, Y. i-Butyl-2,4-dinitro-anilinium p-toluenesulfonate as a highly active and selective esterification catalyst. Tetrahedron Lett. 2013, 54, 6665–6668. [CrossRef] [PubMed]
8. Stuart, A.; LeCaptain, D.J.; Lee, C.Y.; Mohanty, D.K. Poly(vinyl chloride) plasticized with mixtures of succinate di-esters—Synthesis and characterization. Eur. Polym. J. 2013, 49, 2785–2791. [CrossRef]
9. Stuart, A.; McCallum, M.M.; Fan, D.; LeCaptain, D.J.; Lee, C.Y.; Mohanty, D.K. Poly(vinyl chloride) plasticized with succinate esters: Synthesis and characterization. Polym. Bull. 2010, 65, 589–598. [CrossRef]
10. Zhao, D.; Liu, M.; Zhang, J.; Li, J.; Ren, P. Synthesis, characterization, and properties of imidazole dicaticionic ionic liquids and their application in esterification. Chem. Eng. J. 2013, 221, 99–104. [CrossRef]
11. Ji, X.; Chen, Y.; Shen, Z. Nano-SO2-/TiO2 catalyzed eco-friendly esterification of dicarboxylic acids. Asian J. Chem. 2014, 26, 5769–5772. [CrossRef]
12. Budarin, V.L.; Clark, J.H.; Luque, R.; Macquarrie, D.J. Versatile mesoporous carbonaceous materials for acid catalysis. Chem. Commun. 2007, 634–636. [CrossRef] [PubMed]
13. Clark, J.H.; Budarin, V.; Dugmore, T.; Luque, R.; Macquarrie, D.J.; Strelko, V. Catalytic performance of carbonaceous materials in the esterification of succinic acid. Catal. Commun. 2008, 9, 1709–1714. [CrossRef]
14. Fabian, L.; Gomez, M.; Kuran, J.A.C.; Molttrasio, G.; Moglioni, A. Efficient microwave-assisted esterification reaction employing methanesulfonic acid supported on alumina as catalyst. Synth. Commun. 2014, 44, 2386–2392. [CrossRef]
15. Varyambath, A.; Kim, M.R.; Kim, I. Sulfonic acid-functionalized organic knitted porous polyaromatic microspheres as heterogeneous catalysts for biodiesel production. New J. Chem. 2018, 42, 12745–12753. [CrossRef]
16. Yang, Z.W.; Niu, L.Y.; Jia, X.J.; Kang, Q.X.; Ma, Z.H.; Lei, Z.Q. Preparation of silica-supported sulfate and its application as a stable and highly active solid acid catalyst. Catal. Commun. 2011, 12, 798–802. [CrossRef]
17. Zhang, B.; Ren, J.; Liu, X.; Guo, Y.; Guo, Y.; Lu, G.; Wang, Y. Novel sulfonated carbonaceous materials from p-toluenesulfonic acid/glucose as a high-performance solid-acid catalyst. Catal. Commun. 2010, 11, 629–632. [CrossRef]
18. Brahmkhatri, V.; Patel, A. Synthesis and characterization of 12-tungstosilicic acid anchored to MCM-41 as well as its use as environmentally benign catalyst for synthesis of succinate and malonate diesters. Ind. Eng. Chem. Res. 2011, 50, 13693–13702. [CrossRef]
19. Brahmkhatri, V.; Patel, A. Esterification of bioplatform molecules over 12-tungstophosphoric acid anchored to MCM-41. J. Porous Mater. 2013, 20, 209–217. [CrossRef]
20. Santacrose, V.; Bigi, F.; Casnati, A.; Maggi, R.; Storaro, L.; Moretti, E.; Vaccaro, L.; Maestri, G. Selective monomethyl esterification of linear dicarboxylic acids with bifunctional alumina catalysts. Green Chem. 2016, 18, 5764–5768. [CrossRef]
21. Zhang, J.; Zhang, S.; Han, J.; Hu, Y.; Yan, R. Uniform acid poly ionic liquid based large particle and its catalytic application in esterification reaction. Chem. Eng. J. 2015, 271, 269–275. [CrossRef]
22. Delhomme, C.; Goh, S.L.M.; Kuhn, F.E.; Weuster-Botz, D. Esterification of bio-based succinic acid in biphasic systems: Comparison of chemical and biological catalysts. J. Mol. Catal. B Enz. 2012, 80, 39–47. [CrossRef]
23. Orjuela, A.; Kolah, A.; Lira, C.T.; Miller, D.J. Mixed succinic acid/acetic acid esterification with ethanol by reactive distillation. Ind. Eng. Chem. Res. 2011, 50, 9209–9220. [CrossRef]
24. Gerardy, R.; Emmanuel, N.; Toupy, T.; Kassin, V.E.; Tshibalonza, N.N.; Schmitz, M.; Monbaliu, J.C.M. Continuous flow organic chemistry: Successes and pitfalls at the interface with current societal challenges. *Eur. J. Org. Chem.* 2018, 20–21, 2301–2351. [CrossRef]

25. Gerardy, R.; Morodo, R.; Estager, J.; Luis, P.; Debecker, D.P.; Monbaliu, J.C.M. Sustaining the transition from petro- to biobased chemical industry with flow chemistry. *Top. Curr. Chem.* 2019, 377, 1. [CrossRef]

26. Cherkasov, N.; Bao, Y.; Rebrev, E. Process intensification of alkynol semihydrogenation in a tube reactor coated with a Pd/ZnO catalyst. *Catalysts* 2017, 7, 358. [CrossRef]

27. Bai, Y.; Cherkasov, N.; Huband, S.; Walker, D.; Walton, R.I.; Rebrev, E. Highly selective continuous flow hydrogenation of cinnamaldehyde to cinnamyl alcohol in a Pt/SiO₂ coated tube reactor. *Catalysts* 2018, 8, 58. [CrossRef]

28. Kovalenko, G.A.; Perminova, L.V.; Beklemishev, A.B.; Parmon, V.N. Heterogeneous biocatalysts prepared by immuring enzymatic active components inside silica Xerogel and nanocarbons-in-silica composites. *Catalysts* 2018, 8, 177. [CrossRef]

29. Carvalho, F.; Marques, M.P.C.; Fernandes, P. Sucrose hydrolysis in a bespoke capillary wall-coated microreactor. *Catalysts* 2017, 7, 42. [CrossRef]

30. Sotto, N.; Cazorla, C.; Villette, C.; Billamboz, M.; Len, C. Toward the sustainable synthesis of biosourced divinylglycol from glycerol. *ACS Sustain. Chem. Eng.* 2016, 4, 6996–7003. [CrossRef]

31. Garcia-Olmo, A.J.; Yepez, A.; Balu, A.M.; Prinsen, P.; Garcia, A.; Mazière, A.; Len, C.; Luque, R. Activity of continuous flow synthesized Pd-based nanocatalysts in the flow hydroconversion of furfural. *Tetrahedron* 2017, 73, 5599–5604. [CrossRef]

32. Galy, N.; Nguyen, R.; Blach, P.; Sambou, S.; Luart, D.; Len, C. Glycerol oligomerization in continuous flow reactor. *J. Ind. Eng. Chem.* 2017, 51, 312–318. [CrossRef]

33. Len, C.; Bruniaux, S.; Delbecq, F.; Parmar, V.S. Palladium-catalyzed Suzuki-Miyaura cross-coupling in continuous flow. *Catalysts* 2017, 7, 146. [CrossRef]