



Enzymatic synthesis of biobased poly(1,4-butylene succinate-*ran*-2,3-butylene succinate) copolyesters and characterization. Influence of 1,4- and 2,3-butanediol contents

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ARTICLE INFO

Keywords:

Enzymatic polymerization
2,3-butanediol
1,4-butanediol
Crystallinity
Copolyester
Thermal stability

ABSTRACT

Different (co)polyesters, poly(1,4-butylene succinate-*ran*-2,3-butylene succinate) (PBB'S), were enzymatically copolymerized at different 1,4-butanediol (1,4-BDO)/2,3-butanediol (2,3-BDO) molar ratios, with immobilized *Candida antarctica* lipase B (CALB). The synthesized macromolecular architectures were characterized by NMR, SEC, FTIR, WAXS, MALDI-TOF MS, DSC, TGA and specific optical rotation. Based on an optimized process, PBB'S copolyesters were synthesized in high yield. A random distribution between both BDOs was obtained along the chain. \overline{M}_w decreased with increasing 2,3-BDO content, from 26 to 6 kg/mol. The specific optical rotation decreased linearly with decreasing 2,3-BDO content, and can be used as a characterization technique to determine PBB'S composition. DSC results showed that an increase in 2,3-BDO content raised the Tg and reduced the degree of crystallinity, leading to amorphous materials for 2,3-BDO content higher than 47 mol.%. Synthesized copolyesters exhibited a good thermal stability, higher than 250 °C, with a degradation profile depending on the 1,4-BDO/2,3-BDO ratio.

1. Introduction

The research of renewable materials derived from biomass feedstocks is becoming an increasingly relevant topic in academic and industry fields, as such materials allow for the decrease of greenhouse gas emissions and permit the production of novel macromolecular architectures [1–4]. As an abundant carbon source, biomass stocks are produced from solar energy in a short cycle. The development of biorefinery throughout the world was a huge step in the production of a great number of monomers and macromers by biological pathways or chemical modifications [4–6].

Biobased 1,4-butanediol (1,4-BDO) is a short diol principally obtained via a chemical hydrogenation of biobased succinic acid [4,7]. However, recently Genomatica (USA) has genetically modified *E. Coli* for the direct bioproduction of 1,4-BDO from sugars [8]. Produced at an industrial level by different companies (Novamont, BioAmber, Genomatica-BASF), biobased 1,4-BDO is largely used, for instance, as chain extender in the polyurethane synthesis [9,10] or as building block for the synthesis of various polyesters and polyamides [11,12].

2,3-butanediol (2,3-BDO) is also a very interesting building block, with secondary hydroxyl functions, mainly used as a precursor in the synthesis of a wide range of chemical products [13]. 2,3-BDO is a very conventional fermentation product from many species of bacteria. It can be directly produced from different bio-resources such as xylose, sugar cane molasses, glucose and corn [14].

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However, it still has a limited use in macromolecular architectures as a monomer. Watson et al. [15,16], pioneered the work of 2,3-BDO-based polyesters by the esterification of many diacids with 2,3-BDO in 1950, followed by different studies on the synthesis of aliphatic-aromatic polyesters or resins containing 2,3-BDO as main or co-diol [17–20]. In spite of its expected low reactivity, properties caused by the usage of 2,3-BDO such as a drop in crystallinity or a rise of the glass transition temperature can be interesting for some applications such as adhesives [11,17–20].

The use of 2,3-BDO as monomer for polyester synthesis have been mainly studied using organometallic catalysts, in melt and under harsh reaction conditions [11,17–20]. For instance, our group has also recently investigated the kinetics and properties of the organometallic synthesis of copolyesters based on 1,4-BDO and 2,3-BDO [11]. However, conventional organometallic catalysts favored undesirable side reactions during the synthesis at temperatures above 200 °C, like decomposition and discoloration [21]. Besides, metal residues from the catalysts may induce some toxicity (which could be detrimental for e.g., implants or tissue engineering) and environmental pollution [22]. Therefore, it is necessary to develop a more eco-friendly pathway by replacing for instance, the metal catalyst with biocatalysts such as enzymes.

Lipases (E.C. 3.1.1.3) are non-toxic enzymes and more specifically serine hydrolases, which can be found in most organisms [23]. In aqueous media, they catalyze ester bond cleavage, but are also able to catalyze the reverse reaction (i.e., ester bonds formation) in organic media [24]. Enzymatic polymerization can thus be considered as an environment friendly process, and one of the best examples of “green polymer chemistry”. Lipases have already been proven to be versatile in polyesters synthesis under mild conditions [25], with a very good reaction control of enantio-, chemo-, regio-, and stereo-selectivity. Owing to these benefits, enzymatic processes could provide precise control of polymer architectures and increased energy savings [24]. Among lipases, *Candida antarctica* lipase B (CALB) and more particularly the immobilized form (Novozym 435 or N435) is the working horse in biocatalytic polyester synthesis, as it shows several advantages such as broad substrate specificity, high reaction yields, excellent thermal stability and commercial availability [26]. Moreover, CALB is able to form ester bonds with secondary hydroxyl (OH) groups even if their reactivity is much lower in comparison to primary OH groups [27]. CALB enantioselectivity towards secondary OH depends strongly on reactants, decreases by decreasing acyl donor chain length and decreases with the temperature [28–30]. Rottici et al. showed that the CALB-catalyzed esterification of 3-methyl-2-butanol, (which has a close structure to 2,3-BDO) exhibited a higher preference for the R configuration of the secondary OH compared to the S configuration [30]. By analogy, the 2R,3R configuration of 2,3-BDO should be the most adequate stereoisomer of 2,3-BDO for CALB.

In recent years, enzymatic syntheses of sustainable polyesters from biobased building blocks have attracted much attention. Different biobased polyesters have been prepared via enzymatic polymerization, e.g. poly(lactic acid), succinate-, glycerol-, fatty acids-, sorbitol-, 1,4:3,6-dianhydrohexitol-, 2,5-furandicarboxylate-based polyesters, etc. [24,26,31–35]. Until now, to the best of our knowledge, only two researches mentioned the synthesis of enzyme-catalyzed polyesters based on 2,3-BDO. Jiang et al. reported the synthesis of small oligomers (500 g/mol) resulting from the CALB-catalyzed reaction between 2,3-BDO and dimethyl-2,5-furandicarboxylate [36], whereas Yoon et al. reported a CALB-catalyzed polycondensation between 2,3-BDO and sebacic acid without details on the achieved molar mass [37].

The aim of this study was to investigate the CALB-catalyzed transesterification of 1,4-BDO, 2,3-BDO and diethyl succinate in solution after optimization of the reaction parameters (solvent type and amount, temperature, catalyst amount). To the best of our knowledge, the present study is the first to investigate in detail the lipase-catalyzed copolyesters synthesis with 2,3-BDO as a building block. Chemical and physicochemical properties of the different synthesized macromolecular architectures were fully investigated by ¹H, ¹³C, HSQC 2D NMR, SEC, FTIR, MALDI-TOF MS and specific optical rotation. The thermal stability, crystalline structure and thermal properties were analyzed by TGA, WAXS and DSC, respectively. The effect of the 1,4-BDO/2,3-BDO molar ratio on the physicochemical properties was particularly discussed.

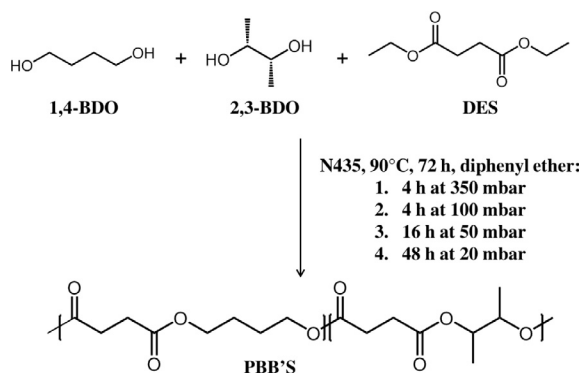
2. Experimental part

2.1. Materials

Novozym®435 (N435) (about 10 wt.% of immobilized *Candida antarctica* lipase B on an acrylic carrier (> 5000 U·g⁻¹)), chloroform (99.0–99.4%) and deuterated chloroform (CDCl₃) were purchased from Sigma-Aldrich. 1,4-butanediol (1,4-BDO) (99%), 2R,3R-(–)-butanediol (2,3-BDO) and benzyl methyl ether (97%) were purchased from Alfa Aesar. Diethyl succinate (DES) (99%), diphenyl ether (99%), dibenzyl ether (99%), toluene (99.85%, extra dry, AcroSeal), *n*-dibutyl ether (99 + %, extra dry, AcroSeal), phenetole (99%), dodecane (99%) and anhydrous anisole (99%, extra dry, AcroSeal) were supplied by Acros. Xylene (99.9%) was supplied by Fisher Chemicals. 4 Å molecular sieves and petroleum ether were purchased from VWR. N435 and molecular sieves were dried at 25 and 120 °C, respectively, for 24 h under vacuum before use. All other reactants were used without further purification. All solvents used for the analytical methods were of analytical grade.

2.2. Enzymatic synthesis of PBB'S copolyesters

An equimolar proportion of DES and 1,4-BDO/2,3-BDO diol mixture, N435 (10 wt.% vs. total mass of monomers) and diphenyl ether (150 wt.% vs. total mass of monomers) were introduced in a Schlenk reactor. The schlenk reactor was immersed in a hot oil bath at 90 °C. The pressure inside the reactor was decreased stepwise at 350, 100, 50 and 20 mbar for 4, 4, 16 and 48 h, respectively. The global reaction scheme and conditions are summarized in Scheme 1. After 72 h of reaction, 10 mL of chloroform was introduced in the reaction mixture in order to quench the reaction. The diluted reaction mixture was filtered. The filtrate was concentrated in a



Scheme 1. Reaction pathway for the CALB-catalyzed synthesis of PBB'S.

rotavapor. Thereafter, the synthesized copolyester was precipitated into a large volume of vigorously stirred cold petroleum ether, recovered by filtration and dried under reduced pressure in an oven at 50 °C for 24 h.

Different poly(1,4-butylenesuccinate-co-2,3-butylenesuccinate) (PBB'S) copolyesters were synthesized varying the 1,4-BDO/2,3-BDO feed molar ratios (100/0, 75/25, 50/50, 25/75 and 0/100). The corresponding samples are named "PBxB'yS", with x and y as the molar proportion of 1,4-BDO and 2,3-BDO units determined by ¹H NMR, respectively. Both homopolyesters (PB₁₀₀B'₀S and PB₀B'₁₀₀S) are also named PBS and PB'S, respectively. Copolyesters syntheses were repeated at least three times.

In the case of PBS or PB'S syntheses in a solvent other than diphenyl ether, the vacuum was adapted to the boiling point of the solvents. For syntheses performed at atmospheric pressure, a short condenser filled with molecular sieves was placed on top of the Schlenk reactor and the reaction was performed under argon.

2.3. General methods and analysis

¹H and ¹³C NMR spectra of polyesters were obtained on a Bruker 400 MHz spectrometer. CDCl₃ was used as solvent to prepare solutions with concentrations of 8–10 and 30–50 mg/mL for ¹H NMR and ¹³C NMR, respectively. The number of scans was set to 128 for ¹H NMR and at least 5000 for ¹³C NMR. Calibration of the spectra was performed using the CDCl₃ peak ($\delta_{\text{H}} = 7.26$ ppm, $\delta_{\text{C}} = 77.16$ ppm).

Size exclusion chromatography (SEC) was performed to determine the number-average molar mass (\overline{M}_n), the mass-average molar mass (\overline{M}_w) and the dispersity (Đ) of the samples. A Shimadzu liquid chromatograph was equipped with PLGel Mixed-C and PLGel 100 Å columns and a refractive index detector. Chloroform was used as eluent at a flow rate of 0.8 mL/min. The instrument was calibrated with linear polystyrene standards from 162 to 1,650,000 g/mol.

Infrared spectroscopy (IR) was performed with a Nicolet 380 Fourier transformed infrared spectrometer (Thermo Electron Corporation) used in reflection mode and equipped with an ATR diamond module (FTIR-ATR). The FTIR-ATR spectra were collected at a resolution of 4 cm⁻¹ and with 64 scans per run.

MALDI-TOF MS analysis was performed using a MALDI-TOF-TOF Autoflex II TOF/TOF (Bruker Daltonics, Germany) equipped with a nitrogen laser ($\lambda = 337$ nm) and super DHB (9:1 mixture of DHB and 2-hydroxy-5-methoxybenzoic acid) as matrix. Scan accumulation and data processing were performed with FlexAnalysis 3.0 software.

Differential scanning calorimetry (DSC) was performed using a TA Instrument Q 200 under nitrogen (flow rate of 50 mL/min), calibrated with high purity standards. Samples of 2–3 mg were sealed in aluminum pans. A three-step procedure with a 10 °C/min ramp was applied that involved: (1) heating up from room temperature to 130 °C and holding for 3 min to erase the thermal history, (2) cooling down to -80 °C and holding for 3 min and (3) heating (second heating) from -80 to 130 °C. The degree of crystallinity (X_c) was calculated according to Eq. (1),

$$X_c (\%) = \frac{\Delta H_m}{\Delta H_m^0} \times 100 \quad (1)$$

where ΔH_m is the melting enthalpy and ΔH_m^0 is the melting enthalpy of a 100% crystalline polyester.

Thermal degradations were studied by thermogravimetric analyses (TGA). Measurements were conducted under helium atmosphere (flow rate of 25 mL/min) using a Hi-Res TGA Q5000 apparatus from TA Instruments. Samples (1–3 mg) were heated from room temperature up to 600 °C at a rate of 20 °C/min.

Wide angle X-ray Scattering (WAXS) data were recorded on a Siemens D5000 diffractometer using Cu K α radiation (1.5406 Å) at 25–30 °C in the range of $2\theta = 14$ –32° at 0.4° min⁻¹. Analyses are performed on compression-molded sheets.

The specific optical rotation $[\alpha]_D^{20}$ was measured by a polarimeter MPC 200 thermostated at 20.0 °C at $\lambda = 589$ nm and in a 100 mm long cell (diameter = 0.3 mm). Measured solution was prepared at a concentration between 1.0 and 2.0 g/100 mL in chloroform.

Table 1

CALB-catalyzed synthesis of PBS at 90 °C with 10 wt.% of N435 and 0.1 g/mL of molecular sieves in 200 wt.% of various solvents and at different pressures.

Entry	Experimental conditions			SEC		¹ H NMR
	Solvent	log <i>P</i>	Pressure mbar	$\overline{M}_{n,SEC}$ kg/mol	Đ	$\overline{M}_{n,NMR}$ kg/mol
1	Benzyl methyl ether	1.4	150	4.7	1.9	3.1
2	Anisole	2.1	150	13.8	2.3	6.7
3	Phenetole	2.5	150	10.9	2.2	4.9
4	Toluene	2.7	1000	4.0	1.7	1.7
5	<i>n</i> -dibutyl ether	2.9	400	2.6	1.6	1.2
6	Xylene	3.2	1000	2.1	1.4	1.0
7	Xylene	3.2	500	4.1	1.6	1.8
8	Xylene	3.2	400	4.8	1.9	1.7
9	Dibenzyl ether	3.5	150	4.3	2.0	2.1
10	Diphenyl ether	4.1	1000	2.2	1.4	1.0
11	Diphenyl ether	4.1	150	6.8	2.3	4.1
12	Diphenyl ether	4.1	20	12.2	2.1	5.1
13	Dodecane	6.1	1000	3.5	1.4	1.4
14	Dodecane	6.1	20	5.1	1.6	2.4

3. Results and discussion

3.1. Optimization of the CALB-catalyzed transesterification conditions

Succinic acid, a biobased and largely available building block for polyesters synthesis, is generally seen as a poor candidate for enzymatic polycondensation due to its low solubility in 1,4-BDO and in the most common solvents used in enzyme-catalyzed polyesters syntheses, leading to low polymerization efficiency [38]. To avoid phase separation during polymerization, diethyl succinate (DES) was used as an acyl donor monomer. Moreover, DES is more hydrophobic than succinic acid and thus has a higher octanol-water partition coefficient (log *P*) value. Previous studies showed that the efficiency of the enzymatic polycondensation/transesterification had a tendency to increase when the hydrophobicity of the system had log *P* optimum values between 1.9 and 4.5 [39,40]. Indeed, for lipase-catalyzed ester bond formation, systems should contain only traces of water for optimal enzymatic activity [41,42]. Furthermore, with DES, ethanol is a by-product of the reaction and can be easily removed from the reaction mixture unlike water. Thus, products should be miscible in the solvent to avoid a phase separation. Finally, a favorable interaction is necessary between the N435 acrylic carrier matrix and the solvent; likewise a high enzyme-solvent affinity is required in order to have the optimal 3D shape of the binding site to allow for the site to be accessible to reactive groups.

In order to select the best solvent (*i.e.*, leading to polyester of higher molar mass), different CALB-catalyzed syntheses of poly(1,4-butylene succinate) (PBS) from 1,4-BDO and DES in various solvents and at different pressures were performed, and summarized in Table 1. The molar mass of synthesized PBS was determined by SEC ($\overline{M}_{n,SEC}$) and ¹H NMR ($\overline{M}_{n,NMR}$) according to the method developed in SI.2 using the relative intensities of PBS and end-groups characteristic signals. Table 1 clearly shows that regardless of the solvent used, lowering the pressure led to increase polyesters molar masses. $\overline{M}_{n,NMR}$ values were smaller than $\overline{M}_{n,SEC}$ values, as the SEC calibration is based on PS standards. Only three solvents led to the synthesis of relatively high molar mass PBS (*i.e.*, > 10 kg/mol). $\overline{M}_{n,SEC}$ of 10.9, 12.2 and 13.8 kg/mol were observed for syntheses in phenetole, diphenyl ether and anisole, respectively. All other tested solvents were deemed inadequate for this reaction as the synthesized polyesters had low molar masses (*i.e.*, $\overline{M}_{n,SEC}$ < 5 kg/mol). PBS precipitated quickly (after 24–32 h) in *n*-dibutyl ether and xylene. The N435 acrylic carrier matrix showed poor affinity to dodecane. This led to the reduction of the extent of reaction as this solvent is too hydrophobic (*i.e.*, log *P* = 6.1); therefore, ultimately leading to a modification of the local water content which affected the enzyme 3D structure.

Anisole was determined as the most adequate solvent for the CALB-catalyzed synthesis of PBS. Moreover, this system was performed under a lower vacuum when compared to the reaction carried out in diphenyl ether (*i.e.*, 150 mbar instead of 20 mbar), as anisole has a lower boiling point. Nevertheless, as seen in Table 2, preliminary results of the CALB-catalyzed synthesis of poly(2,3-butylene succinate) (PB'S) from 2,3-BDO and DES had inferior molar mass in anisole compared to in diphenyl ether (presented

Table 2

Enzymatic synthesis of PB'S at 90 °C catalyzed with 10 wt.% of N435 in 150 wt.% of solvent.

Entry	Experimental conditions			SEC		¹ H NMR
	Solvent	log <i>P</i>	Pressure mbar	$\overline{M}_{n,SEC}$ kg/mol	Đ	$\overline{M}_{n,NMR}$ kg/mol
1	Anisole	2.1	150	3.0	1.3	2.1
2	Phenetole	2.5	150	3.7	1.4	3.2
3	Toluene	2.7	1000	0.9	1.5	0.7
4	Diphenyl ether	4.1	20	4.3	1.5	3.0
5	Dodecane	6.1	20	3.6	1.3	2.3

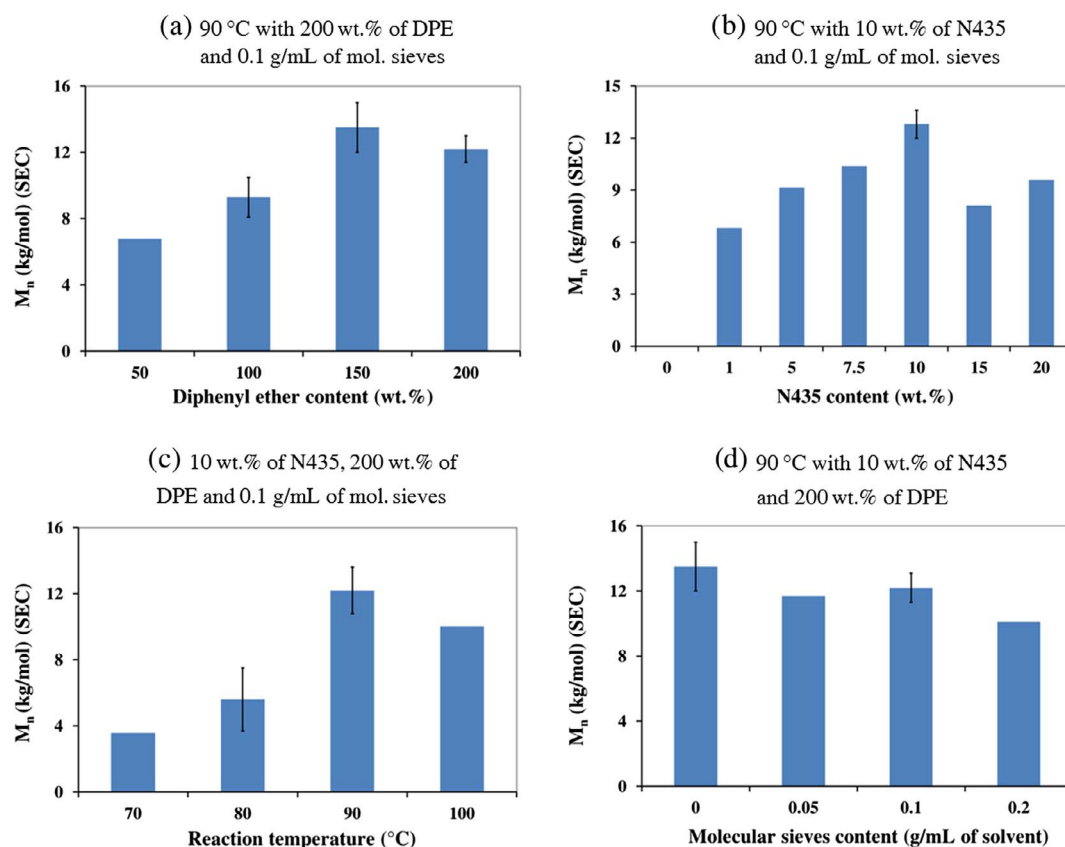


Fig. 1. Optimization of (a) the N435 content, (b) the diphenyl ether content, (c) the reaction temperature and (d) the molecular sieves content for the CALB-catalyzed PBS synthesis in diphenyl ether (DPE).

below), thus the latter was chosen as the most adequate solvent for PBB'S synthesis. This results is in agreement with the literature as it has been previously reported that diphenyl ether is the preferred solvent to achieve high molar mass for lipase-catalyzed polyesters synthesis [12,21,38].

The optimization of the reaction conditions in diphenyl ether was then investigated. The influence of (i) the N435 content, (ii) the solvent amount, (iii) the reaction temperature and (iv) the molecular sieve content on the CALB-catalyzed PBS synthesis in diphenyl ether will be discussed and main results are plotted in Fig. 1. First, the control reaction (blank reaction without N435) did not allow for the synthesis of polyester, thus proving that the acylation between monomers observed for the other reactions was actually due to CALB (Fig. 1b). \overline{M}_n of synthesized PBS increased with increasing N435 content from 1 to 10 wt.% (i.e., a CALB content between 0.1 and 1 wt.%), in agreement with previous data [21], and then decreased for higher N435 amounts. The same tendency was observed for the solvent content with an optimal diphenyl ether amount at 150 wt.% (Fig. 1a), in agreement with Jiang et al. [12]. Furthermore, \overline{M}_n increased with the reaction temperature from 70 to 90 °C and then decreased due to a loss of activity resulting from CALB denaturation at high temperatures. The important \overline{M}_n gap occurring between 80 and 90 °C was mainly due to the PBS which has a high melting temperature (i.e., 113 °C) and precipitated after 32–48 h at 80 °C in diphenyl ether (Fig. 1c). Finally, \overline{M}_n decreased with the molecular sieve content (Fig. 1d). In conclusion, the optimal conditions for PBS synthesis were performed with 150 wt.% of diphenyl ether at 90 °C with 10 wt.% of N435 (i.e., 1 wt.% of CALB) and without molecular sieves.

Different CALB-catalyzed syntheses of PB'S in various reactive systems were performed with the optimized reaction conditions as previously determined, and summarized in Table 2. Results showed that, contrary to PBS, diphenyl ether permitted the highest molar mass ($\overline{M}_{n,SEC}$ of 4.3 kg/mol) for the synthesis of PB'S. Results with anisole were poor. Dodecane and phenetole permitted the synthesis of PB'S with similar molar masses just a bit lower than the one obtained with diphenyl ether. Interestingly, 2,3-BDO was able to react with DES in such enzymatic system to obtain oligomers with noticeable molar masses. Indeed, 2,3-BDO monomer is a very short diol containing only secondary OH groups and both parameters are known to highly decrease the reactivity with CALB [21,43–46]. Even if some papers showed that CALB was able to react with secondary OH functions [28–30], this study is the first to report the enzymatic synthesis of polyesters with 2,3-BDO monomer in significant extent.

3.2. Enzymatic synthesis of PBB'S copolyesters

Different PBB'S copolyesters were synthesized from 1,4-BDO, 2,3-BDO and DES at different 1,4-BDO/2,3-BDO feed molar ratios

according Scheme 1 using the optimized conditions previously determined, i.e. at 90 °C, in 150 wt.% diphenyl ether, under a 20 mbar vacuum and with 10 wt.% of N435 (i.e., 1 wt.% of CALB). In order to avoid any monomer loss, the vacuum was decreased stepwise inside the system allowing for the synthesis of less volatile oligomers before significantly decreasing the pressure. Average and detailed results are given in this paper and in Supporting Information SI.6, respectively.

The final architectures of the synthesized copolyesters were verified by ^1H , ^{13}C NMR and FTIR (Fig. SI.2). ^1H NMR results of PBB'S copolyesters and their corresponding homopolyesters are presented in Fig. 1a and SI.1. In ^1H NMR, the presence of esters functions was verified by the signal at $\delta = 4.12$ and 4.98 ppm assigned to $\text{COO}-\text{CH}_2-\text{CH}_2-$ and $\text{COO}-\text{CH}(\text{CH}_3)-$ protons from 1,4-BDO and 2,3-BDO repeating units, respectively. ^1H chemical shifts at $\delta = 1.15$, 1.70 and 2.62 ppm were ascribed to $\text{COO}-\text{CH}(\text{CH}_3)-$ protons from 2,3-BDO repeating units, $\text{COO}-\text{CH}_2-\text{CH}_2-$ protons from 1,4-BDO repeating units and $\text{CO}-\text{CH}_2-$ protons from succinate repeating units, respectively. Finally, the presence of 1,4-BDO, 2,3-BDO and ester (DES) end-groups were observed at $\delta = 3.67$, 3.74 and 1.25 ppm, respectively. The ester end-group proportion is calculated according to Eq. (SI.6) (more details in SI.2).

Copolyesters have mostly ester end-groups (i.e., 80–90 mol.%), whereas the feed ratio between OH and ester function was 1/1. A first hypothesis could be that a small amount of diol was lost during the synthesis, even if a particular attention was paid to decrease stepwise the vacuum to prevent the evaporation of monomers. However, Nakaoki et al. observed that the enzyme-catalyzed polymerization of adipic acid and 1,6-hexanediol preferentially yielded hydroxyl terminated products [47], in agreement with the mechanism proposed by Binns et al. [48], at the early stage. Thus, when increasing the reaction time, the product is more likely to be carboxyl terminated by the transesterification reaction [47]. Furthermore, the 1,4-BDO/2,3-BDO molar composition of synthesized copolyesters was determined from ^1H NMR spectra using relative intensities of methine and methylene protons in α of ester functions in 1,4-BDO and 2,3BDO, respectively. The 2,3-BDO content was calculated according to Eq. (2),

$$X_{2,3\text{-BDO}} = \frac{I_{4.98}}{I_{4.98} + \frac{I_{4.12} - I_{3.67} - 2/3 \times I_{1.25}}{2}} \quad (2)$$

Calculations showed that the molar content of 1,4-BDO was slightly higher than the feed content for all copolyesters containing both diols. This discrepancy can be due to the difference of reactivity between both diols. However, even by increasing the reaction time before decreasing the pressure, no increase of the 2,3-BDO content was observed. Previously, our group synthesized copolyesters from 1,4-BDO and 2,3-BDO via an organometallic two-stage melt transesterification route, and the difference between the feed and the final 2,3-BDO content was higher (i.e., 33–45 mol.% of 2,3-BDO in final copolyester from a 50 mol.% feed composition according to the method used) than by the enzymatic process [11]. Thus, less monomer loss was observed when compared to the present study. One can suppose that the difference of the experimental conditions (i.e., equimolar amounts of monomer for the enzymatic process and a 25 mol.% excess of diol for the organometallic process), the difference of reaction type (i.e., esterification in bulk vs. transesterification in solution) and the favorable reactivity of 2,3-BDO with CALB were the main reasons for the higher 2,3-BDO content by the enzymatic process. Indeed, during the organometallic process, a 25 mol.% excess of diol was used and thus a feed 0.67/0.67/1 (1,4-BDO/2,3-BDO/adipic acid) ratio was introduced and since 1,4-BDO was approximately 10 times more reactive than 2,3-BDO with adipic acid, some 2,3-BDO were not able to react and were removed from the reaction mixture [11]. Likewise, the esterification in bulk with an organometallic catalyst quickly reached an equilibrium prior to attaining 100% conversion, reducing the incorporation of less reactive species such as 2,3-BDO, causing significant raw material losses [11].

^{13}C NMR was performed to obtain more detailed information about the PBB'S architecture. Corresponding results are presented in Fig. 2b and in SI.3. In addition, Distorsionless Enhancement Polarization Transfer (DEPT) 135 ^{13}C - and Heteronuclear Single Quantum correlation (HSQC) 2D NMR spectra of PBB'S copolyesters are presented in Figs. SI.5 and SI.7, respectively. ^{13}C chemical shifts at $\delta = 25.3$ and 64.3 ppm were ascribed to $\text{COO}-\text{CH}_2-\text{CH}_2-$ and $\text{COO}-\text{CH}_2-\text{CH}_2-$ carbons from 1,4-BDO repeating units, whereas ones at $\delta = 16.1$ and 71.8 ppm were ascribed to $\text{COO}-\text{CH}(\text{CH}_3)-$, and $\text{COO}-\text{CH}(\text{CH}_3)-$ carbons from 2,3-BDO repeating units, respectively. Finally, ^{13}C chemical shifts at $\delta = 29.1\text{--}29.2$ (multiple peaks), 171.7 (two peaks) and 172.4 (two peaks) ppm were ascribed to $\text{CO}-\text{CH}_2-$, $\text{CO}-\text{CH}_2-$ next to a 2,3-butylene repetitive unit and $\text{CO}-\text{CH}_2-$ next to a 1,4-butylene repetitive unit, respectively. The sensitivity of ^{13}C NMR to small differences in the chemical environment enabled us to identify the different triads BSB, BSB', and B'SB' (with S, B and B' standing for succinate, 1,4-BDO and 2,3-BDO, respectively) presented in Fig. 3a. Succinate

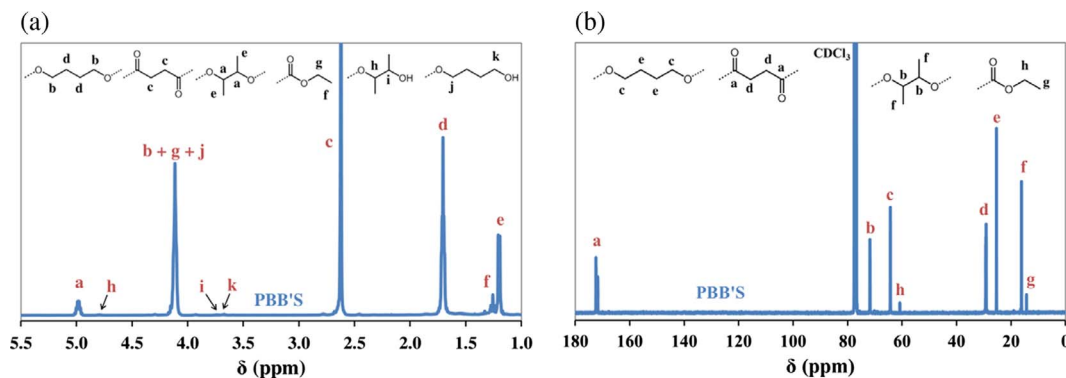


Fig. 2. (a) ^1H and (b) ^{13}C NMR of PBB'S copolyesters in CDCl_3 .

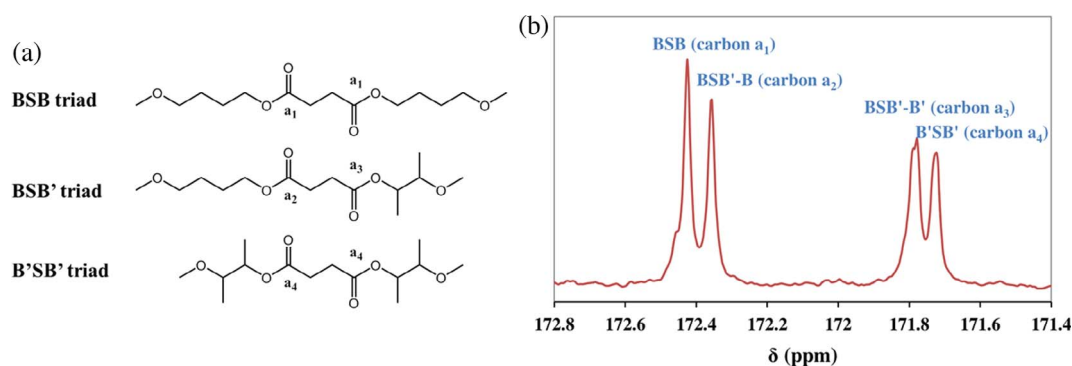


Fig. 3. ¹³C NMR spectra of PPB'S copolyester in CDCl₃ centered on carbonyl peaks and chemical structures corresponding to used abbreviations in the figure.

moieties are present in all three triads at distinctly different resonance absorptions close to chemical shifts of PBS (PB₁₀₀B'₀S) or PB'S (PB₀B'₁₀₀S). Such results, with the splitting of the two signals (carbon atoms a₁, a₂, a₃ and a₄ in Fig. 3b), enable the calculation of the average sequence length of BS and B'S units (L_{BS} and L_{B'S}, respectively) and the degree of randomness (R) using Eqs. (SI.9)–(SI.11) (more details in SI.5).

The corresponding data are listed in Table 3. According to the 1,4-BDO/2,3-BDO compositions, average block lengths of segments (L_{BS} and L_{B'S}) varied between 1.2 and 4.7. PPB'S copolyesters containing both diols all showed a randomness degree of approximately 1.0 meaning a random distribution sequence behavior between 1,4-butyl and 2,3-butyl segments along the polyester chain. Such a random distribution was expected since CALB is known to catalyze ester bond formation and also intermolecular transesterification leading to random structures [49–53].

The molar masses and dispersities of synthesized copolyesters were analyzed by SEC in chloroform and are presented in Table 3. $\bar{M}_{n,SEC}$ of copolyesters after 72 h varied with the 2,3-BDO content from 13.5 ± 1.5 to 4.3 ± 1.1 kg/mol for PBS and PB'S, respectively. Moreover, the evolution of the molar mass as a function of the reaction time of various PPB'S copolyesters is presented in Fig. SI.8. For all compositions, the molar mass increased continuously for 72 h of reaction time, even if the increase was not significant between 48 and 72 h. This confirmed the initial choice to perform the syntheses over 72 h. However, the molar mass increase slowed down in the presence of 2,3-BDO. Likewise, \bar{D} decreased with the 2,3-BDO content from 2.0 to 1.4. For PBS, a more accurate molar mass value using the Mark-Houwink-Sakurada (MHS) equation and parameters (K and a) from Garin et al. [54] or Charlier et al. [55] could be calculated from overestimated \bar{M}_n values determined by SEC due to the calibration based on PS standards using Eq. (SI.12). The MHS-corrected \bar{M}_n of PBS was approximately 6.0–6.5 kg/mol ($DP_n \sim 35$ –38). The copolyester chain length decrease was also observed for molar mass obtained by ¹H NMR (i.e., from 4.6 to 2.6 kg/mol). $\bar{M}_{n,NMR}$ was determined according to relative intensities of characteristics signals using Eq. (3),

$$M_{n,NMR} = \frac{\left(\frac{I_{4.98}}{2} + \frac{I_{4.12} - I_{3.67} - 2I_{31.25}}{4}\right) \times 88.10 + \frac{I_{2.62}}{4} \times 84.07 + \left(I_{3.74} + \frac{I_{3.67}}{2}\right) \times 89.11 + \frac{I_{1.25}}{3} \times 45.06}{0.5 \times \left(I_{3.74} + \frac{I_{3.67}}{2} + \frac{I_{1.25}}{3}\right)} \quad (3)$$

where 88.10, 84.07, 89.11 and 45.06 are the molar mass (in g/mol) of the diol unit, succinate unit, hydroxyl end-groups unit and ester end-groups unit, respectively (more details in SI.2). $\bar{M}_{n,NMR}$ of PBS (i.e., 4.6 kg/mol) was lower than the one determined by the MHS equation. This difference could be due to the fact that, for high molar mass polyesters, ¹H NMR signals of the end-groups were less prominent and were difficult to differentiate from the baseline. Therefore, their intensities might be slightly overestimated leading to the underestimation of the calculated $\bar{M}_{n,NMR}$. The decrease of the molar mass could be explained by the use of 2,3-BDO which is less reactive than 1,4-BDO. Nevertheless, to the best of your knowledge, this study is the first one to report the lipase-catalyzed polyesters from high 2,3-BDO content with molar mass higher than 1.0 kg/mol.

Since the structure of 2R,3R-(–)-butanediol contains two asymmetric carbons, synthesized copolyesters containing this monomer

Table 3

Enzymatic synthesis of PPB'S copolyesters of various compositions at 90 °C catalyzed by 10 wt.% of N435 in 150 wt.% of diphenyl ether.

		¹ H NMR		SEC			¹ H NMR				¹³ C NMR				
Sample	Feed composition 1,4-BDO/2,3-BDO mol.%	Exp. composition 1,4-BDO/2,3-BDO mol.%	Yield %	$\overline{M}_{n,SEC}$	\overline{M}_w	Đ	$\overline{M}_{n,NMR}$	1° OH	2° OH	Ester	L _{BS}	L _{B'S}	R	[α]	
				kg/mol	kg/mol		kg/mol	%	%	%				°	
PBS	100/0	100/0	87	13.5	26.2	2.0	4.6	15	0	85	-	-	-	0	
PB ₇₉ B' ₂₁ S	75/25	79/21	82	7.9	15.5	2.0	3.4	7	5	88	4.7	1.2	1.02	5.6	
PB ₅₃ B' ₄₇ S	50/50	53/47	65	7.0	12.7	1.8	2.9	7	10	83	2.2	1.7	1.03	11.1	
PB ₂₈ B' ₇₂ S	25/75	28/72	69	5.4	8.8	1.6	2.6	2	16	82	1.6	2.7	1.00	18.6	
PB'S	0/100	0/100	64	4.3	5.8	1.4	3.0	0	12	88	-	-	-	25.0	

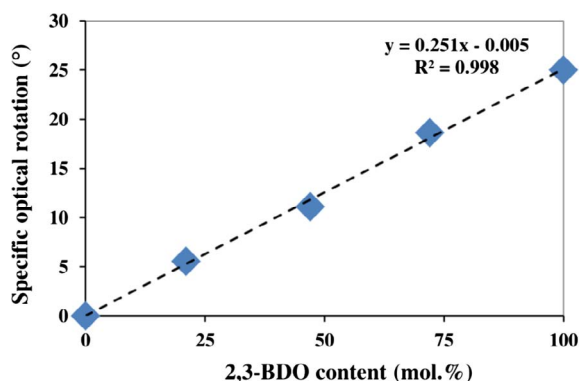


Fig. 4. Variation of the specific optical rotation of PBB'S copolyesters in function of the 2,3-BDO content.

possess also asymmetric carbons. In order to study the optical properties of copolyesters with 2,3-BDO, their specific optical rotation ($[\alpha]$) were determined in chloroform at 20 °C. Results are summarized in Table 3 and detailed in Table SI.3. Since PBS did not contain any asymmetric carbons, this copolyester showed, as expected, $[\alpha]$ value of 0. However, PB₇₉B'₂₁S, PB₅₃B'₄₇S, PB₂₈B'₇₂S and PB'S exhibited $[\alpha]$ values of 5.9, 11.1, 18.6 and 25.0°, respectively. $[\alpha]$ of PBB'S copolyesters increased proportionately with increasing 2,3-butylene content (Fig. 4). Using a line equation ($y = 0.251x - 0.005$), this analytical method could be used as a complement to ¹H NMR technique to determine the molar composition of synthesized PBB'S copolyesters, as it has already been used in a previous study on another copolyester architecture [11].

The FTIR analysis of different PBB'S copolyesters is presented in Fig. 5. First, FTIR spectra of copolyesters showed two sharp signals at approximately 1715 and 1160 cm⁻¹ assigned to the C=O and C–O–C stretching vibrations from ester confirming the presence of ester linkage in the polymer chemical structure. Moreover, no broad resonance around 3400 cm⁻¹ originating from OH functions were observed in all samples confirming the low amount of OH end-groups as previously stated by NMR. Other signals in spectra depended highly on the 1,4-BDO/2,3-BDO molar composition of copolyesters. PB'S exhibited a signal at 2985 cm⁻¹ assigned to the symmetrical C–H stretching vibrations of –CH₃ groups which decreased with the 1,4-BDO content until having a small shoulder for PBS due to the presence of ester end-groups. Likewise, signals at 2883, 1450 and 1377 cm⁻¹ assigned to C–H stretching vibrations of methine, C–H asymmetrical and symmetrical bending vibration of methyl groups, respectively, were characteristic of PB'S and their intensities decreased with increasing 1,4-BDO content. Conversely, signals at 2945, 2855 and 1330 cm⁻¹ assigned to symmetrical C–H stretching vibrations, asymmetrical C–H stretching vibration and symmetric deformation of –CH₂– groups, respectively, were characteristic of 1,4-BDO and their intensity decreased with increasing 2,3-BDO content. Finally, characteristic signals of PBS observed at 1046, 955, 805 and 653 cm⁻¹ assigned to the O–C–C vibration in 1,4-BDO, the C–O symmetric stretching, the methylene in succinate in-plane bending and the –COO– bending shifted to 1085 (O–C–C vibration in 2,3-BDO), 1020, 860 and 750 cm⁻¹ for PB'S, respectively [56,57].

The MALDI-TOF MS spectrum of PB'S with peaks interpretation is shown in Fig. 6, whereas PBS, PB₇₉B'₂₁S and PB₂₈B'₇₂S spectra are presented in Fig. SI.9. First, all copolyesters exhibited mass peak patterns in the m/z region between 500 and 2500 (or 3500 for PBS). This analysis confirmed the decrease of the molar mass with increasing 2,3-BDO content. Different patterns were observed for PBB'S copolyesters, as listed in Table SI.4. First, the different patterns were separated by an m/z value of 172, in agreement with the molar mass of the PBB'S repetitive unit. Then, all copolyesters exhibited mostly ester/ester or acid/ester end-groups and ester/

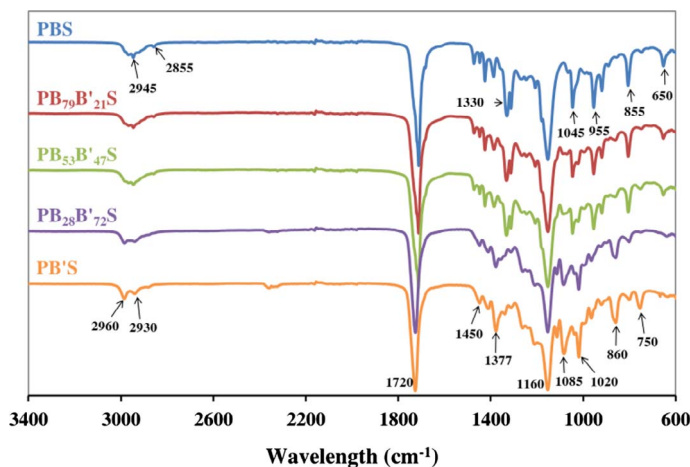


Fig. 5. FTIR spectra of PBB'S copolyesters.

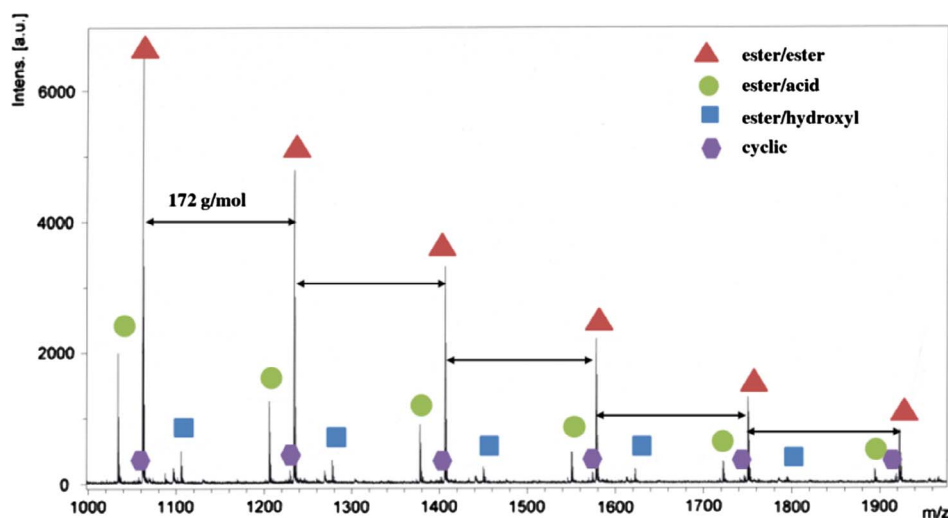


Fig. 6. MALDI-ToF MS spectrum of PB'S with peak assignment.

hydroxyl end-groups in low quantity, which was in agreement with NMR results and previous studies [36,58,59]. Acid end-groups were produced from the hydrolysis of ester end-groups during or after the reaction [60]. Moreover, in many samples, cyclic species were detected in tiny amount, in agreement with previous studies [36,59]. However, since cyclic species of PBB'S copolyesters have almost the same m/z signal than copolyesters with ester/ester end-groups, both signals were close to each other and these cyclic species were difficult to observe. Furthermore, PBS exhibited the presence of acid/hydroxyl end-groups and hydroxyl/hydroxyl end-groups, in tiny quantities. PBS seemed thus to have an increased hydrolysis sensitivity when compared to other copolyesters. This can be due to the local reduced ester function density of PBS, coming from longer 1,4-BDO units, which yields a polyester with higher chain mobility. Finally, PB₇₉B'₂₁S exhibited a low intensity signal assigned to ester/vinyl end-groups. One can suppose the formation of vinyl end-groups from the hydroxyl dehydration. However, no extra resonance indicative of the presence of vinyl groups was observed in NMR spectra of PB'S and PB₇₉B'₂₁S.

3.3. Influence of the reaction temperature on the PBB'S structure

The influences of the synthesis temperature on the PBB'S structure and on the presence of 2,3-BDO units inside the corresponding copolyesters were also investigated. Several copolyesters, with a 50/50 (1,4-BDO/2,3-BDO) ratio feed, were synthesized at different reaction temperatures (*i.e.*, 50, 60, 70, 80 and 90 °C). Results are summarized in Table 4. The temperature rise did not have a significant effect on the incorporation of 2,3-BDO units in the copolyesters (*i.e.*, 44–47 mol.%), even if a tiny increase trend seemed to be observed with the increase of the temperature. One can suppose that, by decreasing the temperature, the esterification rate of CALB decreased and thus less 2,3-BDO units reacted with DES to form oligomers (before the high vacuum step leading to 2,3-BDO loss by evaporation). All the copolyesters showed a random structure. Molar masses seemed to display an overall increase trend with increasing temperature, whereas \bar{D} stayed globally constant (*i.e.*, 1.6–1.8). For example, $\bar{M}_{n,SEC}$ rose from 4.1 to 6.2 kg/mol between 50 and 90 °C, respectively. However, this effect was less obvious between 60 and 80 °C. The temperature did not affect the sequence distribution along the chain, whereas the end-groups proportion study showed no significant effect with mostly ester end-groups (75–87%). Moreover, it was noteworthy to observe that the proportion of 2,3-BDO end-groups was higher than 1,4-BDO end-groups for all samples confirming the lower reactivity of the secondary OH groups with acyl donor groups by CALB compared to primary OH groups.

Table 4

Influence of the reaction temperature on the enzymatic PBB'S copolyester synthesis from a 50/50 (1,4-BDO/2,3-BDO) ratio in 150 wt.% of diphenyl ether with 10 wt.% of N435.

Temperature °C	Exp. composition 1,4-BDO/2,3-BDO mol.%	SEC			¹ H NMR			¹³ C NMR	
		$\bar{M}_{n,SEC}$ kg/mol	\bar{M}_w kg/mol	\bar{D}	$\bar{M}_{n,NMR}$ g/mol	Primary OH %	Secondary OH %	ester %	R
50	56 /44	4.1	6.5	1.6	2.1	5	20	75	0.99
60	54/46	5.2	9.0	1.7	2.6	8	11	81	1.01
70	55/45	4.7	8.4	1.8	2.6	4	10	86	1.00
80	54/46	5.4	9.5	1.8	2.9	4	9	87	0.99
90	53/47	6.2	10.9	1.7	3.0	7	10	83	0.99

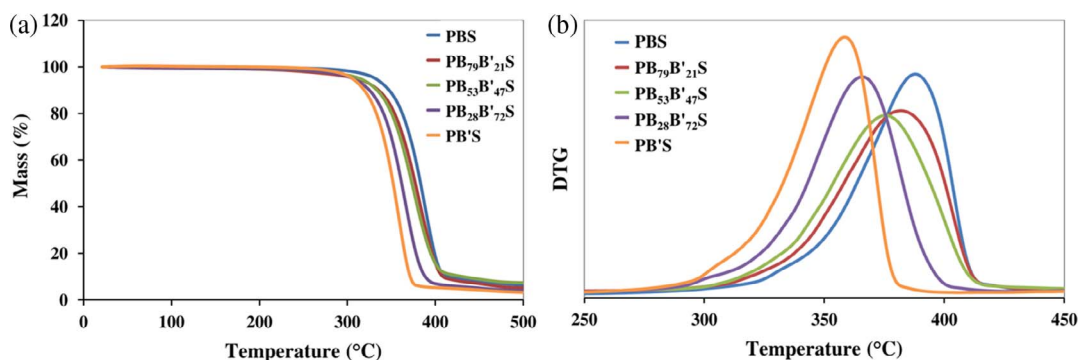


Fig. 7. (a) Mass loss and (b) DTG curves of PBB'S copolyesters under helium.

3.4. Thermal stability of PBB'S copolyesters

The TGA mass loss traces of PBB'S copolyesters and their derivatives curves (DTG) are shown in Fig. 7, and results are summarized in Table SI.5. Under helium, all copolyesters degraded in three main steps, which involved competitive mechanisms. First, all samples were thermally stable until 250 °C and a small (< 3%) mass loss was observed at 250–300 °C due to the possible presence of residual solvent, the degradation of low molar mass chains along with the cyclization at the chain-ends and backbiting reactions [61,62]. Furthermore, a major degradation occurred between 300 and 425 °C, with a substantial mass loss of approximately 85–90% corresponding to the thermal degradation, mostly by β - and α -hydrogen bond scissions [11,63]. These reactions are responsible for the decomposition of polyesters into small compounds such as diacids, vinyl compounds, aldehydes and anhydrides [61]. After this decomposition step, mass residues of 3–7% still remained under non-oxidative atmosphere (helium). This multi-steps degradation was in agreement with previous reports [11,61,64].

The 50% mass loss degradation temperature ($T_{d,50\%}$) and the maximal degradation temperature ($T_{deg,max}$) were approximately 350–385 and 360–390 °C, respectively, and both decreased with the 2,3-BDO content. The thermal degradation profile of “2,3-BDO rich” copolyesters was, thus, sharper compared to “1,4-BDO rich” copolyesters. To resume, the thermal degradation profile of PBB'S depended highly on the 1,4-BDO/2,3-BDO molar ratio. This result was in contradiction with a previous recent study performed on PBB'A in which no influence of the 1,4-BDO/2,3-BDO composition on the thermal degradation of poly(1,4-butylen adipate-co-2,3-butylen adipate) was observed [11]. One may suppose that the decrease of the molar mass linked to the 2,3-BDO units incorporation was responsible for the decrease of $T_{deg,max}$.

3.5. Crystalline structure and thermal properties of PBB'S copolyesters

The WAXS analyses of PBB'S copolyesters are presented in Fig. 8. Results showed that PB'S and PB₂₈B'₇₂S were amorphous at 25 °C. Contrary to the two aforementioned samples, PBS is a semi-crystalline polyester and its crystal structure has been already reported [65]. The main PBS diffraction peaks appeared at $2\theta = 19.7^\circ$, 22.0° and 22.7° . PB₇₉B'₂₁S and PB₅₃B'₄₇S copolyesters exhibited WAXS patterns similar to PBS, indicating that the crystal structure in these copolyesters has the same characteristics as the PBS lattice, but with lower intensities. The co-monomeric unit (2,3-BDO in this case) incorporated in minor amounts was found to be partially integrated or excluded from the PBS crystal lattice. This phenomenon decreased the crystal strength and lamellae sizes due to the presence of “2,3-BDO-based defects” in the copolyesters chains. Thus, the degree of crystallinity decreased with increasing 2,3-BDO content.

In complement to WAXS analyses, the crystallinity and thermal properties of copolyesters were studied by DSC. Prior to this, it has

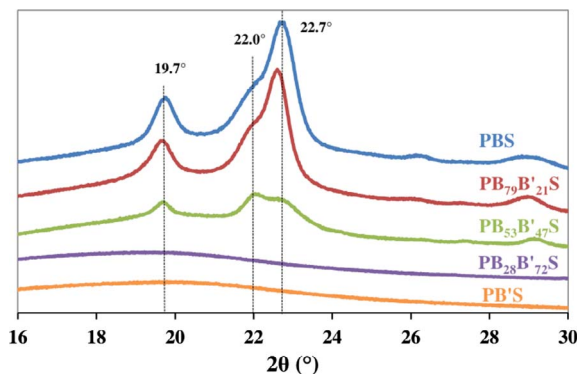


Fig. 8. WAXS patterns of PBB'S copolyesters.

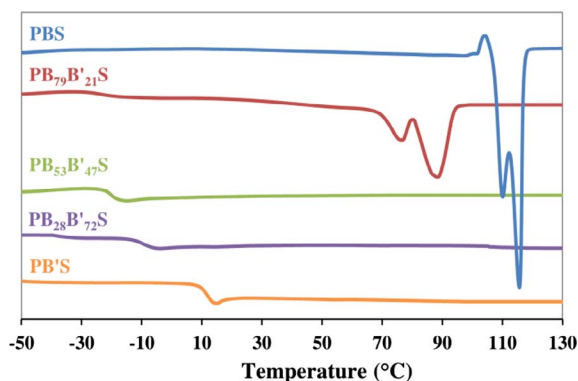


Fig. 9. Second heating run curves of PBB'S copolyesters at 10 °C/min.

been verified from TGA results that no significant degradation occurred in the samples amongst the DSC analysis temperature range. Second heating run curves are presented in Fig. 9. The variation of thermal properties is discussed as a function of the ester function density (D_{ester}) calculated according to Eq. SI.13. The degree of crystallinity (X_c) of copolyesters was calculated by Eq. (1), taking into account the melting enthalpy value of 100% crystalline phase of PBS ($\Delta H_{m,\text{PBS}}^\circ = 210 \text{ J/g}$) determined by Papageorgiou et al. [66], and WAXS results, which showed that samples exhibited only PBS crystals.

PBS, which has the lowest D_{ester} due to 1,4-butylene segments, is a semi-crystalline polyester with a sharp crystallization temperature (T_c) at 88 °C, a high melting temperature (T_m) of 115 °C and $X_c = 37\%$. During the second heating run, a tiny cold-crystallization peak preceded a melting phenomenon decomposed on two peaks which was attributed to a fusion-recrystallization phenomenon which is rather common for aliphatic polyesters [11,67]. $\text{PB}_{79}\text{B}'_{21}\text{S}$, similarly to PBS, is a semi-crystalline polyester, but with lower T_c , T_m and X_c of 48 °C, 88 °C and 23%, respectively. By increasing the 2,3-BDO content, PBB'S copolyesters exhibited an amorphous behavior. More specifically, $\text{PB}_{53}\text{B}'_{47}\text{S}$ exhibited a small endotherm at 45 °C with $X_c = 15\%$ during the first heating run but it did not crystallize during the cooling or the second heating run due to the high 2,3-BDO content. *A fortiori*, $\text{PB}_{28}\text{B}'_{72}\text{S}$ and PB'S were completely amorphous, in agreement with WAXS results.

Intra or inter-chains interactions between ester groups were more noticeable in PB'S than in PBS due to the increase of D_{ester} with the 2,3-BDO content from 25 to 33% for PBS and PB'S, respectively (Table SI.6). This led to the decrease of the chain mobility responsible for a global increase of the T_g from -37 to 12 °C for PBS and PB'S, respectively. Moreover, the presence of the two methyl side groups in 2,3-butylene segments seemed to prevent the crystallization. For instance, poly(ethylene succinate) is a semi-crystalline polyester [66], whereas PB'S is amorphous. The ability of 2,3-BDO to restrain crystallization has been employed previously to obtain amorphous copolyesters [11,20].

4. Conclusion

An efficient approach for the green synthesis of fully biobased random poly(1,4-butylene succinate-*ran*-2,3-butylene succinate) (PBB'S) copolyesters by CALB-catalyzed enzymatic transesterification process from 1,4-butanediol (1,4-BDO), 2,3-butanediol (2,3-BDO) and diethyl succinate at 90 °C in diphenyl ether under vacuum was successfully demonstrated.

After the determination of the optimal CALB-catalyzed transesterification conditions on PBS synthesis, the procedure was applied for the PBB'S copolyesters elaboration. First, random copolyesters of relatively high molar masses and high 2,3-BDO contents were obtained. These copolyesters exhibited 2,3-BDO contents slightly lower than feed ones. \overline{M}_w of copolyesters decreased from 26.2 to 5.8 kg/mol with the increase in the 2,3-BDO content from 0 to 100 mol.%, respectively. These results highlighted the lower reactivity of 2,3-BDO compared to 1,4-BDO, and confirmed that CALB prefers diols with longer chain lengths and primary OH functions. Moreover, we have shown with copolyesters with 50/50 (1,4-BDO/2,3-BDO) feed ratio that the reaction temperature did not have a significant influence on the 2,3-BDO final content (44–47 mol.%), contrary to the \overline{M}_w that increased from 6.5 to 10.9 kg/mol, between 50 to 90 °C, respectively. CALB activity towards secondary alcohols seemed, thus, to increase with the temperature over the studied range. In addition, copolyesters containing 2,3-BDO exhibited a small positive specific optical rotation, which was found to be proportional to the 2,3-BDO content. This can be used as an additional technique to evaluate the molar composition. Furthermore, from the thermal behavior of PBB'S, it has been noticed that the chain mobility of the copolyester decreased as the 2,3-BDO content increased leading to a global increase of the T_g from -37 to 12 °C and a significant decreased of the degree of crystallinity. Amorphous copolyester are obtained for 2,3-BDO content higher than 47 mol.%. Finally, all copolyesters showed an excellent thermal stability with an onset decomposition temperature above 250 °C and the thermal degradation profile of PBB'S copolyesters was mainly driven by the 1,4-BDO/2,3-BDO composition, resulting in an acceleration of the degradation rate after the onset in copolyesters containing 2,3-BDO.

Conclusively, the possibility to use 2,3-BDO as a potentially biobased monomer and a product from biomass valorization, makes the PBB'S copolyester, with tunable thermal properties and excellent thermal stability, a promising sustainable material for some engineering applications such as adhesives, or for some biomedical applications. Moreover, the opportunity to use this monomer *via* an enzymatic process will increase the number of macromolecular engineering tools at our disposition and, thus, bring added value to

the enzyme-catalyzed polymer technology.

Acknowledgements

This work has received funding from the European's Union 7th Framework Program for research, technological development and demonstration under Grant Agreement No. 311815 (SYNPOL Project). In addition, the authors are grateful to Ms. Khantutta-Kim Tremblay-Parrado for her help in proofreading this article.

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at <http://dx.doi.org/10.1016/j.eurpolymj.2017.04.045>.

References

- [1] S. Laurichesse, L. Avérous, Chemical modification of lignins: towards biobased polymers, *Prog. Polym. Sci.* 39 (2014) 1266–1290, <http://dx.doi.org/10.1016/j.progpolymsci.2013.11.004>.
- [2] C. Vilela, A.F. Sousa, A.C. Fonseca, A.C. Serra, J.F.J. Coelho, C.S.R. Freire, A.J.D. Silvestre, The quest for sustainable polyesters – insights into the future, *Polym. Chem.* 5 (2014) 3119–3141, <http://dx.doi.org/10.1039/C3PY01213A>.
- [3] A. Gandini, The irruption of polymers from renewable resources on the scene of macromolecular science and technology, *Green Chem.* 13 (2011) 1061–1083, <http://dx.doi.org/10.1039/C0GC00789G>.
- [4] J.J. Bozell, G.R. Petersen, Technology development for the production of biobased products from biorefinery carbohydrates—the US Department of Energy's "Top 10" revisited, *Green Chem.* 12 (2010) 539, <http://dx.doi.org/10.1039/b922014c>.
- [5] T.A. Werpy, J.E. Holladay, J.F. White, Top Value Added Chemicals from Biomass: I. Results of Screening for Potential Candidates from Sugars and Synthesis Gas, Pacific Northwest National Laboratory (PNNL), Richland, WA (US), 2004 <https://www.osti.gov/scitech/biblio/926125-top-value-added-chemicals-from-biomass-screening-potential-candidates-from-sugars-synthesis-gas> (accessed March 23, 2016).
- [6] J. Becker, A. Lange, J. Fabarius, C. Wittmann, Top value platform chemicals: bio-based production of organic acids, *Curr. Opin. Biotechnol.* 36 (2015) 168–175, <http://dx.doi.org/10.1016/j.copbio.2015.08.022>.
- [7] S. Choi, C.W. Song, J.H. Shin, S.Y. Lee, Biorefineries for the production of top building block chemicals and their derivatives, *Metab. Eng.* 28 (2015) 223–239, <http://dx.doi.org/10.1016/j.mbs.2014.12.007>.
- [8] N.R. Barton, A.P. Burgard, M.J. Burk, J.S. Crater, R.E. Osterhout, P. Pharkya, B.A. Steer, J. Sun, J.D. Trawick, S.J.V. Dien, T.H. Yang, H. Yim, An integrated biotechnology platform for developing sustainable chemical processes, *J. Ind. Microbiol. Biotechnol.* 42 (2014) 349–360, <http://dx.doi.org/10.1007/s10295-014-1541-1>.
- [9] M. Reulier, L. Avérous, Elaboration, morphology and properties of renewable thermoplastics blends, based on polyamide and polyurethane synthesized from dimer fatty acids, *Eur. Polym. J.* 67 (2015) 418–427, <http://dx.doi.org/10.1016/j.eurpolymj.2014.11.036>.
- [10] C. Bueno-Ferrer, E. Hablot, M. del C. Garrigós, S. Bocchini, L. Avérous, A. Jiménez, Relationship between morphology, properties and degradation parameters of novative biobased thermoplastic polyurethanes obtained from dimer fatty acids, *Polym. Degrad. Stab.* 97 (2012) 1964–1969, <http://dx.doi.org/10.1016/j.polydegradstab.2012.03.002>.
- [11] T. Debuissy, E. Pollet, L. Avérous, Synthesis of potentially biobased copolyesters based on adipic acid and butanediols: kinetic study between 1,4- and 2,3-butanediol and their influence on crystallization and thermal properties, *Polymer* 99 (2016) 204–213, <http://dx.doi.org/10.1016/j.polymer.2016.07.022>.
- [12] Y. Jiang, A.J.J. Wootman, G.O.R.A. van Ekenstein, K. Loos, Enzyme-catalyzed synthesis of unsaturated aliphatic polyesters based on green monomers from renewable resources, *Biomolecules* 3 (2013) 461–480, <http://dx.doi.org/10.3390/biom3030461>.
- [13] M. Köpke, C. Mihalcea, F. Liew, J.H. Tizard, M.S. Ali, J.J. Conolly, B. Al-Sinawi, S.D. Simpson, 2,3-Butanediol Production by acetogenic bacteria, an alternative route to chemical synthesis, using industrial waste gas, *Appl. Environ. Microbiol.* 77 (2011) 5467–5475, <http://dx.doi.org/10.1128/AEM.00355-11>.
- [14] X.-X. Wang, H.-Y. Hu, D.-H. Liu, Y.-Q. Song, The implementation of high fermentative 2,3-butanediol production from xylose by simultaneous additions of yeast extract, Na₂EDTA, and acetic acid, *New Biotechnol.* 33 (2016) 16–22, <http://dx.doi.org/10.1016/j.nbt.2015.07.004>.
- [15] R.W. Watson, N.H. Grace, J.L. Barnwell, Production and properties of 2,3-butanediol. XXXVI. Linear polyesters of 2,3-butanediol, *Can. J. Res.* 28b (1950) 652–659, <http://dx.doi.org/10.1139/cjr50b-078>.
- [16] Ralph W. Watson, 2,3-butanediyl phthalate resins, US2502686, 1950. < <http://www.google.com/patents/US2502686> > .
- [17] B.A.J. Noordover, V.G. van Staalduinen, R. Duchateau, C.E. Koning, van Benthem, M. Mak, A. Heise, A.E. Frissen, J. van Haveren, Co- and terpolyesters based on isosorbide and succinic acid for coating applications: synthesis and characterization, *Biomacromol* 7 (2006) 3406–3416, <http://dx.doi.org/10.1021/bm060713v>.
- [18] E. Gubbels, L. Jasinska-Walc, C.E. Koning, Synthesis and characterization of novel renewable polyesters based on 2,5-furandicarboxylic acid and 2,3-butanediol, *J. Polym. Sci. Part Polym. Chem.* 51 (2013) 890–898, <http://dx.doi.org/10.1002/pola.26446>.
- [19] E. Gubbels, L. Jasinska-Walc, B.A.J. Noordover, C.E. Koning, Linear and branched polyester resins based on dimethyl-2,5-furandicarboxylate for coating applications, *Eur. Polym. J.* 49 (2013) 3188–3198, <http://dx.doi.org/10.1016/j.eurpolymj.2013.06.019>.
- [20] X. Hu, X. Shen, M. Huang, C. Liu, Y. Geng, R. Wang, R. Xu, H. Qiao, L. Zhang, Biodegradable unsaturated polyesters containing 2,3-butanediol for engineering applications: synthesis, characterization and performances, *Polymer* 84 (2016) 343–354, <http://dx.doi.org/10.1016/j.polymer.2016.01.007>.
- [21] A. Mahapatro, B. Kalra, A. Kumar, R.A. Gross, Lipase-catalyzed polycondensations: effect of substrates and solvent on chain formation, dispersity, and end-group structure, *Biomacromolecules* 4 (2003) 544–551, <http://dx.doi.org/10.1021/bm0257208>.
- [22] M.C. Tanzi, P. Verderio, M.G. Lampugnani, M. Resnati, E. Dejana, E. Sturani, Cytotoxicity of some catalysts commonly used in the synthesis of copolymers for biomedical use, *J. Mater. Sci. – Mater. Med.* 5 (1994) 393–396, <http://dx.doi.org/10.1007/BF00058971>.
- [23] R.D. Schmid, R. Verger, Lipases: interfacial enzymes with attractive applications, *Angew. Chem. Int. Ed.* 37 (1998) 1608–1633, [http://dx.doi.org/10.1002/\(SICI\)1521-3773\(19980703\)37:12<1608::AID-ANIE1608>3.0.CO;2-V](http://dx.doi.org/10.1002/(SICI)1521-3773(19980703)37:12<1608::AID-ANIE1608>3.0.CO;2-V).
- [24] S. Kobayashi, Recent developments in lipase-catalyzed synthesis of polyesters, *Macromol. Rapid Commun.* 30 (2009) 237–266, <http://dx.doi.org/10.1002/marc.200800690>.
- [25] L. Gustini, C. Lavilla, W.W.T.J. Janssen, A. Martínez de Ilarduya, S. Muñoz-Guerra, C.E. Koning, Green and selective polycondensation methods toward linear sorbitol-based polyesters: enzymatic versus organic and metal-based catalysis, *Chemsuschem* 9 (2016) 2250–2260, <http://dx.doi.org/10.1002/cssc.201600626>.
- [26] Y. Jiang, K. Loos, Enzymatic synthesis of biobased polyesters and polyamides, *Polymers* 8 (2016) 243, <http://dx.doi.org/10.3390/polym8070243>.
- [27] L. Gustini, B.A.J. Noordover, C. Gehrels, C. Dietz, C.E. Koning, Enzymatic synthesis and preliminary evaluation as coating of sorbitol-based, hydroxy-functional polyesters with controlled molecular weights, *Eur. Polym. J.* 67 (2015) 459–475, <http://dx.doi.org/10.1016/j.eurpolymj.2014.12.025>.
- [28] J. Ottosson, K. Hult, Influence of acyl chain length on the enantioselectivity of Candida antarctica lipase B and its thermodynamic components in kinetic resolution of sec-alcohols, *J. Mol. Catal. B Enzym.* 11 (2001) 1025–1028, [http://dx.doi.org/10.1016/S1381-1177\(00\)00088-6](http://dx.doi.org/10.1016/S1381-1177(00)00088-6).
- [29] S. Raza, L. Fransson, K. Hult, Enantioselectivity in Candida antarctica lipase B: a molecular dynamics study, *Protein Sci.* 10 (2001) 329–338, <http://dx.doi.org/10.1110/ps.33901>.

- [30] D. Rotticci, F. Hæffner, C. Orrenius, T. Norin, K. Hult, Molecular recognition of sec-alcohol enantiomers by *Candida antarctica* lipase B, *J. Mol. Catal. B Enzym.* 5 (1998) 267–272, [http://dx.doi.org/10.1016/S1381-1177\(98\)00047-2](http://dx.doi.org/10.1016/S1381-1177(98)00047-2).
- [31] S.W. Duchiron, E. Pollet, S. Givry, L. Avérous, Mixed systems to assist enzymatic ring opening polymerization of lactide stereoisomers, *RSC Adv.* 5 (2015) 84627–84635, <http://dx.doi.org/10.1039/C5RA18954C>.
- [32] H. Öztürk, E. Pollet, V. Phalip, Y. Güvenilir, L. Avérous, Nanoclays for lipase immobilization: biocatalyst characterization and activity in polyester synthesis, *Polymers* 8 (2016) 416, <http://dx.doi.org/10.3390/polym8120416>.
- [33] H. Öztürk Düşkünkörür, E. Pollet, V. Phalip, Y. Güvenilir, L. Avérous, Lipase catalyzed synthesis of polycaprolactone and clay-based nanohybrids, *Polymer* 55 (2014) 1648–1655, <http://dx.doi.org/10.1016/j.polymer.2014.02.016>.
- [34] H.Ö. Düşkünkörür, A. Bégué, E. Pollet, V. Phalip, Y. Güvenilir, L. Avérous, Enzymatic ring-opening (co)polymerization of lactide stereoisomers catalyzed by lipases. Toward the in situ synthesis of organic/inorganic nanohybrids, *J. Mol. Catal. B Enzym.* 115 (2015) 20–28, <http://dx.doi.org/10.1016/j.molcatb.2015.01.011>.
- [35] A. Khan, S.K. Sharma, A. Kumar, A.C. Watterson, J. Kumar, V.S. Parmar, Novozym 435-catalyzed syntheses of polyesters and polyamides of medicinal and industrial relevance, *Chemsuschem* 7 (2014) 379–390, <http://dx.doi.org/10.1002/cssc.201300343>.
- [36] Y. Jiang, A.J.J. Woortman, G.O.R.A. van Ekenstein, K. Loos, A biocatalytic approach towards sustainable furanic–aliphatic polyesters, *Polym. Chem.* 6 (2015) 5198–5211, <http://dx.doi.org/10.1039/C5PY00629E>.
- [37] K.R. Yoon, S.-P. Hong, B. Kong, I.S. Choi, Polycondensation of sebacic acid with primary and secondary hydroxyl groups containing diols catalyzed by *Candida antarctica* lipase B, *Synth. Commun.* 42 (2012) 3504–3512, <http://dx.doi.org/10.1080/00397911.2011.585267>.
- [38] H. Azim, A. Dekhterman, Z. Jiang, R.A. Gross, *Candida antarctica* lipase B-catalyzed synthesis of poly(butylene succinate): shorter chain building blocks also work, *Biomacromol* 7 (2006) 3093–3097, <http://dx.doi.org/10.1021/bm060574h>.
- [39] D. Yao, G. Li, T. Kuila, P. Li, N.H. Kim, S.-I. Kim, J.H. Lee, Lipase-catalyzed synthesis and characterization of biodegradable polyester containing L-malic acid unit in solvent system, *J. Appl. Polym. Sci.* 120 (2011) 1114–1120, <http://dx.doi.org/10.1002/app.33257>.
- [40] A. Kumar, R.A. Gross, *Candida antarctica* Lipase B catalyzed polycaprolactone synthesis: effects of organic media and temperature, *Biomacromol* 1 (2000) 133–138, <http://dx.doi.org/10.1021/bm990510p>.
- [41] M. Nordblad, P. Adlercreutz, Immobilisation procedure and reaction conditions for optimal performance of *Candida antarctica* lipase B in transesterification and hydrolysis, *Biocatal. Biotransfor.* 31 (2013) 237–245, <http://dx.doi.org/10.3109/10242422.2013.837240>.
- [42] P. Adlercreutz, Immobilisation and application of lipases in organic media, *Chem. Soc. Rev.* 42 (2013) 6406, <http://dx.doi.org/10.1039/c3cs35446f>.
- [43] H. Uyama, K. Inada, S. Kobayashi, Lipase-catalyzed synthesis of aliphatic polyesters by polycondensation of dicarboxylic acids and glycols in solvent-free system, *Polym. J.* 32 (2000) 440–443, <http://dx.doi.org/10.1295/polymj.32.440>.
- [44] Z.-L. Wang, K. Hiltunen, P. Orava, J. Seppälä, Y.-Y. Linko, Lipase-catalyzed polyester synthesis, *J. Macromol. Sci. Part A* 33 (1996) 599–612, <http://dx.doi.org/10.1080/10601329608010881>.
- [45] Y.-Y. Linko, M. Lämä, X. Wu, E. Uosukainen, J. Seppälä, P. Linko, Biodegradable products by lipase biocatalysis, *J. Biotechnol.* 66 (1998) 41–50, [http://dx.doi.org/10.1016/S0168-1656\(98\)00155-2](http://dx.doi.org/10.1016/S0168-1656(98)00155-2).
- [46] Y. Jiang, A.J.J. Woortman, G.O.R. Alberda van Ekenstein, K. Loos, A biocatalytic approach towards sustainable furanic–aliphatic polyesters, *Polym. Chem.* 6 (2015) 5198–5211, <http://dx.doi.org/10.1039/C5PY00629E>.
- [47] T. Nakaoki, M. Danno, K. Kurokawa, Enzyme-catalyzed polymerization mechanism of aliphatic polyester investigated by ¹H NMR and MALDI-TOF mass spectrum, *Polym. J.* 35 (2003) 791–797, <http://dx.doi.org/10.1295/polymj.35.791>.
- [48] F. Binns, P. Harfey, S.M. Roberts, A. Taylor, Studies of lipase-catalyzed polyesterification of an unactivated diacid/diol system, *J. Polym. Sci. Part Polym. Chem.* 36 (1998) 2069–2079, [http://dx.doi.org/10.1002/\(SICI\)1099-0518\(19980915\)36:12<2069::AID-POLA13>3.0.CO;2-4](http://dx.doi.org/10.1002/(SICI)1099-0518(19980915)36:12<2069::AID-POLA13>3.0.CO;2-4).
- [49] A. Kumar, R.A. Gross, *Candida antarctica* Lipase B-catalyzed transesterification: new synthetic routes to copolyesters, *J. Am. Chem. Soc.* 122 (2000) 11767–11770, <http://dx.doi.org/10.1021/ja002915j>.
- [50] A. Kumar, B. Kalra, A. Dekhterman, R.A. Gross, Efficient ring-opening polymerization and copolymerization of ε-caprolactone and ω-pentadecalactone catalyzed by *Candida antarctica* Lipase B, *Macromolecules* 33 (2000) 6303–6309, <http://dx.doi.org/10.1021/ma000344+>.
- [51] T. Takamoto, P. Kerep, H. Uyama, S. Kobayashi, Lipase-catalyzed transesterification of polyesters to ester copolymers, *Macromol. Biosci.* 1 (2001) 223–227, [http://dx.doi.org/10.1002/1616-5195\(20010801\)1:6<223::AID-MABI223>3.0.CO;2-B](http://dx.doi.org/10.1002/1616-5195(20010801)1:6<223::AID-MABI223>3.0.CO;2-B).
- [52] S. Namekawa, H. Uyama, S. Kobayashi, H.R. Kricheldorf, Lipase-catalyzed ring-opening polymerization and copolymerization of cyclic dicarbonates, *Macromol. Chem. Phys.* 201 (2000) 261–264, [http://dx.doi.org/10.1002/\(SICI\)1521-3935\(20000201\)201:2<261::AID-MACP261>3.0.CO;2-Q](http://dx.doi.org/10.1002/(SICI)1521-3935(20000201)201:2<261::AID-MACP261>3.0.CO;2-Q).
- [53] S. Namekawa, H. Uyama, S. Kobayashi, Enzymatic synthesis of polyesters from lactones, dicarboxylic acid divinyl esters, and glycols through combination of ring-opening polymerization and polycondensation, *Biomacromol* 1 (2000) 335–338, <http://dx.doi.org/10.1021/bm000030u>.
- [54] M. Garin, L. Tighzert, I. Vroman, S. Marinkovic, B. Estrine, The influence of molar mass on rheological and dilute solution properties of poly(butylene succinate), *J. Appl. Polym. Sci.* 131 (2014) 40887/1–40887/7, <http://dx.doi.org/10.1002/app.40887>.
- [55] Q. Charlier, E. Girard, F. Freymouth, M. Vandesteene, N. Jaquel, C. Ladavière, A. Rousseau, F. Fenouillot, Solution viscosity – molar mass relationships for poly(butylene succinate) and discussion on molar mass analysis, *Express Polym. Lett.* 9 (2015) 424–434, <http://dx.doi.org/10.3144/expresspolymlett.2015.41>.
- [56] R. Muthuraj, M. Misra, A.K. Mohanty, Injection molded sustainable biocomposites from poly(butylene succinate) bioplastic and perennial grass, *ACS Sustain. Chem. Eng.* 3 (2015) 2767–2776, <http://dx.doi.org/10.1021/acssuschemeng.5b00646>.
- [57] Y. Cai, J. Lv, J. Feng, Spectral characterization of four kinds of biodegradable plastics: poly (lactic acid), poly (butylenes adipate-co-terephthalate), poly (hydroxybutyrate-co-hydroxyvalerate) and poly (butylenes succinate) with fir and raman spectroscopy, *J. Polym. Environ.* 21 (2012) 108–114, <http://dx.doi.org/10.1007/s10924-012-0534-2>.
- [58] D.I. Habeych, P.B. Juhl, J. Pleiss, D. Vanegas, G. Eggink, C.G. Boeriu, Biocatalytic synthesis of polyesters from sugar-based building blocks using immobilized *Candida antarctica* Lipase B, *J. Mol. Catal. B Enzym.* 71 (2011) 1–9, <http://dx.doi.org/10.1016/j.molcatb.2011.02.015>.
- [59] Y. Jiang, A.J.J. Woortman, G.O.R. Alberda van Ekenstein, D.M. Petrović, K. Loos, Enzymatic synthesis of biobased polyesters using 2,5-bis(hydroxymethyl)furan as the building block, *Biomacromol* 15 (2014) 2482–2493, <http://dx.doi.org/10.1021/bm500340w>.
- [60] A.K. Chaudhary, E.J. Beckman, A.J. Russell, Biocatalytic polyester synthesis: analysis of the evolution of molecular weight and end group functionality, *Biotechnol. Bioeng.* 55 (1997) 227–239, [http://dx.doi.org/10.1002/\(SICI\)1097-0290\(19970705\)55:1<227::AID-BIT23>3.0.CO;2-H](http://dx.doi.org/10.1002/(SICI)1097-0290(19970705)55:1<227::AID-BIT23>3.0.CO;2-H).
- [61] O. Persenaire, M. Alexandre, P. Degée, P. Dubois, Mechanisms and kinetics of thermal degradation of poly(ε-caprolactone), *Biomacromol* 2 (2001) 288–294, <http://dx.doi.org/10.1021/bm0056310>.
- [62] K. Chrissafis, K.M. Paraskevopoulos, D.N. Bikiaris, Effect of molecular weight on thermal degradation mechanism of the biodegradable polyester poly(ethylene succinate), *Thermochim. Acta* 440 (2006) 166–175, <http://dx.doi.org/10.1016/j.tca.2005.11.002>.
- [63] T. Debuissy, E. Pollet, L. Avérous, Synthesis and characterization of biobased poly(butylene succinate-ran-butylene adipate). Analysis of the composition-dependent physicochemical properties, *Eur. Polym. J.* 87 (2017) 84–98, <http://dx.doi.org/10.1016/j.eurpolymj.2016.12.012>.
- [64] T. Debuissy, E. Pollet, L. Avérous, Titanium-catalyzed transesterification as a route to the synthesis of fully biobased poly(3-hydroxybutyrate-co-butylene dicarboxylate) copolyesters, from their homopolyesters, *Eur. Polym. J.* 90 (2017) 92–104, <http://dx.doi.org/10.1016/j.eurpolymj.2017.03.006>.
- [65] P. Pan, Y. Inoue, Polymorphism and isomorphism in biodegradable polyesters, *Prog. Polym. Sci.* 34 (2009) 605–640, <http://dx.doi.org/10.1016/j.progpolymsci.2009.01.003>.
- [66] G.Z. Papageorgiou, D.N. Bikiaris, Crystallization and melting behavior of three biodegradable poly(alkylene succinates). A comparative study, *Polymer* 46 (2005) 12081–12092, <http://dx.doi.org/10.1016/j.polymer.2005.10.073>.
- [67] T. Debuissy, E. Pollet, L. Avérous, Enzymatic synthesis of a bio-based copolyester from poly(butylene succinate) and poly((R)-3-hydroxybutyrate): study of reaction parameters on the transesterification rate, *Biomacromol* 17 (2016) 4054–4063, <http://dx.doi.org/10.1021/acs.biomac.6b01494>.