

Selecting Polymers for Two-Phase Partitioning Bioreactors (TPPBs): Consideration of Thermodynamic Affinity, Crystallinity, and Glass Transition Temperature

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*Two-phase partitioning bioreactor technology involves the use of a secondary immiscible phase to lower the concentration of cytotoxic solutes in the fermentation broth to subinhibitory levels. Although polymeric absorbents have attracted recent interest due to their low cost and biocompatibility, material selection requires the consideration of properties beyond those of small molecule absorbents (i.e., immiscible organic solvents). These include a polymer's (1) thermodynamic affinity for the target compound, (2) degree of crystallinity (w_c), and (3) glass transition temperature (T_g). We have examined the capability of three thermodynamic models to predict the partition coefficient (PC) for *n*-butyric acid, a fermentation product, in 15 polymers. Whereas PC predictions for amorphous materials had an average absolute deviation (AAD) of $\geq 16\%$, predictions for semicrystalline polymers were less accurate (AAD $\geq 30\%$). Prediction errors were associated with uncertainties in determining the degree of crystallinity within a polymer and the effect of absorbed water on *n*-butyric acid partitioning. Further complications were found to arise for semicrystalline polymers, wherein strongly interacting solutes increased the polymer's absorptive capacity by actually dissolving the crystalline fraction. Finally, we determined that diffusion limitations may occur for polymers operating near their T_g , and that the T_g can be reduced by plasticization by water and/or solute. This study has demonstrated the impact of basic material properties that affects the performance of polymers as sequestering phases in TPPBs, and reflects the additional complexity of polymers that must be taken into account in material selection.*

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Introduction

Two-phase partitioning bioreactor (TPPB) technology can improve the efficiency and product titers of bioconversion processes by reducing substrate and/or product concentrations below cytotoxic levels.^{1–3} TPPBs have led to process improvements in systems ranging from alcohol/organic acid fermentation^{4–10} to the biodegradation of more hydrophobic xenobiotics.^{11–15} This is accomplished by solute absorption into a non-aqueous phase (NAP) that provides a combination of high solute affinity and good biocompatibility. High molecular weight elastomers and semicrystalline thermoplastics have recently been studied as alternatives to immiscible organic solvents and oligomeric liquids such as polypropylene glycol and silicone oil.² These materials offer advantages such as ease of handling, low cost, simple recovery from bioreactor media, and improved biocompatibility.¹⁶ We have also previously described limitations of solid polymers, namely, their lower inherent diffusivities¹⁷ and potential for negative hydrodynamic effects.¹⁸ We have now concluded that differences in the absorptive^{19–21} properties of rubbery,

glassy, and crystalline domains within solid polymers require a retooling of previous liquid NAP selection approaches.²²

Selecting a polymer for a TPPB process requires consideration of many of the same factors important in other separation processes such as pervaporation, perstraction, and vapor permeation,^{23–25} as well as food packaging applications^{26,27} and protective clothing design.²⁸ These aspects include (1) polymer-solute thermodynamic affinity, (2) degree of crystallinity (w_c), and (3) polymer glass transition temperature (T_g).^{29,30} In the case of TPPBs, these considerations will determine the *rate* and *extent* to which a toxic solute will be absorbed/sequestered in the polymer phase, thereby reducing its inhibitory effects. A common quantitative measure to describe the absorption capacity of a polymer is the partition coefficient (PC), defined as the equilibrium concentration of a solute in the polymer phase divided by that in the aqueous phase. As such, estimating the PC provided by a polymer for the target solute is central to TPPB development.^{29,31}

Our recent study³⁰ of polymer-solute thermodynamics found that UNIFAC-vdW-FV and Flory-Huggins-based models were able to predict the equilibrium absorption (PC) of solutes into a single amorphous material, poly(*n*-butyl acrylate). One purpose of this study was to examine the ability of the same thermodynamic models to describe the sequestration of *n*-butyric acid (recently

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produced in a TPPB system³²) into a broad range of amorphous materials that were screened from a standard polymer solubility database.³³ The bioproduction of *n*-butyric acid is an attractive alternative to current petrochemical processes to supply food, fragrance, pharmaceutical, and chemical industries.^{34–36} However, the accumulation of *n*-butyric acid results in low product titers and lost productivity,^{9,37} and therefore a TPPB system may have practical utility.³⁸ This work extends previous binary (solute + polymer) PC prediction methods³⁰ by incorporating experimental water sorption data, enabling ternary polymer phase activity predictions (water + *n*-butyric acid + polymer). In the biological production of *n*-butyric acid, as in all fermentation processes, water is an inevitable component, making this approach a necessary step toward more realistically representing an actual biological process.

In addition, practical limitations related to semicrystalline polymers, whose crystalline domains are generally nonabsorptive,^{29,39,40} were experimentally quantified in this work. In particular, the challenges in determining and applying measurements of the degree of crystallinity (w_c) in PC predictions were examined through studies of *n*-butyric acid sorption into seven semicrystalline polymers. In a related aspect, the ability of water and/or solute to affect the polymer's melting temperature (T_m) and degree of crystallinity were quantified with respect to the material's overall ability to sequester the toxic target molecule.

A third aspect when selecting a polymeric absorptive phase is the material's glass transition temperature (T_g). The difference between a bioreactor's operating temperature and the polymer's T_g can provide a general indication of a material's diffusivity for a target solute molecule.⁴¹ In the production of biomolecules in TPPBs, solute uptake must be fast enough to prevent the accumulation of toxic metabolites, making materials with low T_g s particularly attractive.²⁹ This work therefore also included a study of water and *n*-octanol absorption into poly(vinyl acetate) (PVAc), a polymer selected for its T_g (45.7°C) being near to typical fermentation temperatures. The abundance of $\log K_{o/w}$ values in literature combined with *n*-octanol's low polarity, strong affinity for PVAc, and large molecular size (compared to water) made it an ideal model compound. Partitioning experiments were conducted at two temperatures, above and below the material's as-received T_g (30 and 60°C), to quantify the ability of water and *n*-octanol to reduce T_g by plasticization and by extension, their impact on diffusivity.

In all, this article describes both the theory and implementation of three important criteria that need to be considered when selecting a polymer for toxic solute removal in TPPBs. These results enable a more efficient and focused approach to selecting effective absorptive polymers, while expanding upon previous studies by considering semicrystalline materials and polymers that absorb water. These considerations are of particular importance for more hydrophilic biomolecules, such as organic acids and alcohols, due to their affinity for more polar polymers. Finally, in addition to polymer screening based exclusively on the extent of solute absorption (PC), the rate of absorption is considered with respect to a material's T_g and its proximity to a bioreactor's operating temperature.

Materials and Methods

Materials and material preparation

All chemicals (purity $\geq 99\%$) were purchased from either Sigma-Aldrich (Canada) or Fisher Scientific (Canada). All

polymers were purchased from Scientific Polymer Products (Ontario, NY).

For *n*-butyric acid partitioning tests, polymers were used as-received with the exception of poly(methyl acrylate), poly(ethyl acrylate), poly(*n*-propyl acrylate), and poly(*n*-butyl acrylate), which were isolated from toluene solution by solvent evaporation at room temperature for 48 h followed by 48 h at 60°C.

Prior to T_g depression studies, poly(vinyl acetate) (PVAc) beads with a nominal molecular weight of 350 kg/mol and bead diameter of ~ 0.2 –1.0 mm were gently heated in an aluminum pan and formed into a fused mass using a spatula. The fused polymer was cooled to room temperature and ground using a rotary mill. Two stainless-steel wire mesh sieves with clear openings of 0.980 and 1.524 mm were utilized to normalize the PVAc particle size, and ensure a consistent diffusional path length between studies.

Differential scanning calorimetry

Glass Transition Temperature (T_g). Determination of glass transition temperature was performed using a DSC Q 100 by TA Instruments. Shortly after the partition tests were initiated (< 12 h), the PVAc particles conjoined to form a flattened fused mass. DSC samples were prepared by cutting the polymer in quarters and removing the sample from the center of the fused mass. Samples were scanned between -40 and 80°C at a heating rate of $10^\circ\text{C}/\text{min}$ under a nitrogen purge. The midpoint T_g was determined according to ASTM Test Method D7426 – 08 using TA Universal Analysis software.

Melting Temperature (T_m) and Crystalline Weight Fraction (w_c). Melting temperature (T_m) and crystalline weight fraction (w_c) were determined with a DSC Q 100 by TA Instruments operating between -85°C and 85°C at a heating rate of $10^\circ\text{C}/\text{min}$ under a nitrogen purge. For poly(ethylene succinate) ($T_m = 102^\circ\text{C}$), the upper temperature limit was 300°C . The melting endotherm recorded upon the sample's first heating was integrated using TA Universal Analysis software and divided by the standard enthalpy of fusion (ΔH_f°) to calculate the weight fraction of the crystalline domain (w_c).

First heating scans were chosen to represent w_c and T_m , as this represents polymer samples as-received, which were subjected to partitioning tests with no further handling. For polymers for which the DSC melting endotherm spanned the operating temperature ($T = 30^\circ\text{C}$), only the crystalline fraction that existed $> 30^\circ\text{C}$ was considered. To determine the impact of water or an *n*-butyric acid solution (20 g/L) on polymer crystallinity and melting temperature, polymer samples were soaked at 30°C in an Innova 4400 incubator shaker at 180 rpm for 7 days and tested alongside dry polymer samples as-received from the supplier. Poly(tetramethylene glycol) (PTMG) (2.9 kg/mol) samples were also tested at 10 and 30 g/L *n*-butyric acid to further investigate the atypical behavior of this system.

Water Uptake. Water absorption experiments were conducted on polymer samples equilibrated for a minimum of 48 h in Type I ultrapure water at 30°C . Water-soaked samples (8–15 mg) were pat dry to remove surface water and immediately heated in a TA Instruments Q500 thermogravimetric analyzer from ~ 25 to 200°C ($10^\circ\text{C}/\text{min}$). The samples were held at 200°C until the rate of water loss dropped below 0.05 wt%/min. An 11.4% average standard deviation on water absorption values was estimated from duplicate runs performed for five polymers (amorphous and semicrystalline).

Experimental Partition Coefficients (PC_{expt}). Partition coefficient tests were performed in triplicate using three polymer masses, ranging from 0.25 to 1.5 g of dry mass. Each target molecule was prepared in separate aqueous solutions (0.4 g/L *n*-octanol or 20 g/L *n*-butyric acid) using Type I ultrapure water. In addition to the polymer mass, 10–20 mL aliquots of aqueous solution were added to each scintillation vial, sealed tightly with a foil lined cap, and allowed to equilibrate in an Innova 4400 incubator shaker at 30 or 60°C at 180 rpm.

Aqueous *n*-butyric acid concentrations before and after equilibrating for 7 days were measured by HPLC using a Varian Hi-Plex H column (300 × 7.7 mm) at 60°C with a 10 mM H₂SO₄ mobile phase at 0.7 mL/min, and a UV–Vis detector (Varian Prostar, PS325) at 220 nm. *n*-Octanol concentrations were measured using a Varian 450-GC gas chromatography unit equipped with a CP-8410 AutoInjector, VF-5 ms 30 m capillary column and FID detector. In T_g depression trials, control solutions (without polymer) were taken at every other time point. Aqueous *n*-butyric acid concentrations had a standard deviation of 1.4% ($n = 3$) while *n*-octanol concentrations had a standard deviation of 6.7% ($n = 3$).

A mass balance was performed to determine the solute concentration in the polymer. Swelling of the polymer by water and the simultaneous decrease in aqueous phase volume was accounted for using experimental water uptake data. Experimental PC values were calculated using aqueous and polymer phase weight fractions, w_i^{aq} and w_i^{poly} , in Eq. 1. Standard deviation ($n = 3$) values were calculated from triplicate samples to establish a mean value for the equilibrium PC.

$$PC = \frac{w_i^{\text{poly}}}{w_i^{\text{aq}}} \quad (1)$$

In the case of T_g depression trials, where systems are not in a state of equilibrium, the ratio of solute concentrations in the polymer and aqueous phase ($n = 1$) as a function of time, $PC^*(t)$, can be defined as follows:

$$PC^*(t) = \frac{w_i^{\text{poly}}(t)}{w_i^{\text{aq}}(t)} \quad (2)$$

Partition coefficient predictions (PC_{pred})

Partition coefficients (PC) can be predicted for systems only at thermodynamic equilibrium, for which the PC has been defined as³⁰

$$PC = \frac{w_i^{\text{poly}}}{w_i^{\text{aq}}} = \frac{\Omega_i^{\text{aq}}}{\Omega_i^{\text{poly}}} \quad (3)$$

In this study, the non-random two liquid (NRTL) model⁴² was selected to estimate aqueous phase activity coefficients (Ω_i^{aq}) at experimental conditions using parameters provided by the Dortmund Data Bank. As described in our previous work,³⁰ polymer phase activity coefficients (Ω_i^{poly}) were predicted using the Flory–Huggins solution theory⁴³ in conjunction with Hildebrand solubility parameters (FH-Hildebrand),⁴⁴ Hansen solubility parameters (FH-HSP),⁴⁵ or using UNIFAC–vdW–FV.⁴⁶ Water absorption was accounted for in the UNIFAC–vdW–FV model by setting the polymer phase water weight fraction to the value determined from experimental data. The adjustable parameters in the FH-HSP model was set to $\alpha = 1$ and for FH-Hildebrand $\beta = 0.34$.^{30,44}

The utility of these models is dependent on the accuracy of the inputs entered into the model, namely the polymer's density, molecular weight, solubility parameters (FH-Hildebrand and FH-HSP), and functional groups (UNIFAC–vdW–FV). Amorphous polymer densities (where available) were sourced from Brandrup et al.⁴⁷ Where amorphous phase density values were not available, the group contribution method outlined by van Krevelen and te Nijenhuis⁴⁸ was used. Nominal molecular weights were provided by the supplier. Solubility parameters were sourced from the database provided in the HSPiP v.4.0.04 computer program.³³

Results and Discussion

Thermodynamic affinity

Polymer candidates for the sequestration of *n*-butyric acid were screened using the FH-HSP method applied to a database of 650 polymers,³³ of which eight amorphous polymers were tested for their experimental partition coefficients (PC_{expt}). The data listed in Table 1 show values ranging from 1.4 to 2.6, with more hydrophilic polymers generally yielding higher PCs. Note that all three activity coefficient models—FH-Hildebrand, FH-HSP, and UNIFAC–vdW–FV—predicted *n*-butyric acid's activity coefficient satisfactorily in each case. When combined with aqueous phase activity coefficient estimates generated with the NRTL model, the average absolute deviation (AAD%) between predicted PC estimates and experimental PC_{expt} data was 16.5% for FH-Hildebrand, 31.4% for FH-HSP, and 53.5% for UNIFAC–vdW–FV. Moreover, all three models correctly identified poly(*n*-hydroxybutyl methacrylate), poly(hydroxyethyl methacrylate), and poly(*n*-hydroxypropyl methacrylate) as the top-three performing polymers in terms of butyric acid absorption (PC).

Several polymers that exhibit a strong affinity for *n*-butyric acid also absorb water (Table 1). In theory, consideration of water absorption in PC predictions provides a more rigorous analysis of the polymer phase solute activity. In this respect, UNIFAC–vdW–FV is advantageous over Flory–Huggins models as it can predict activity in ternary (polymer + *n*-butyric acid + water) and higher phase systems. However, although remaining qualitatively correct, refining the UNIFAC–vdW–FV model by including water absorption data resulted in a distinct reduction in predictive accuracy (AAD = 343%), particularly for polymers with high water uptake (Table 1). Decreased prediction accuracy at higher water concentrations may be indicative of UNIFAC–vdW–FV's inability to account for the formation of polymer-bound and unbound states of water.^{49,50}

Degree of crystallinity

Experimental PC values for semicrystalline polymers can be expressed as a function of the amorphous partition coefficient (PC_{am}) and the crystalline phase fraction (w_c). Equation 4 is based on the well-documented observation that intact crystallites in semicrystalline materials are nonabsorptive.^{29,39,40}

$$PC_{\text{expt}} = PC_{\text{am}}(1 - w_c) \quad (4)$$

We have shown that PC_{am} can be estimated with a reasonably good degree of accuracy using activity coefficient models (PC_{pred}) (see Table 1). However, the degree of crystallinity (w_c) is dependent on a material's thermal history and, as such, it cannot be predicted accurately. It should also be noted that

Table 1. Experimental (PC_{expt}) and Predicted (PC_{pred}) Partition Coefficient Values for *n*-Butyric Acid with Various Amorphous Polymers

Polymer	MW (kg/mol)	Water Uptake (wt%)		PC_{expt}	FH-Hildebrand		FH-HSP		UNIFAC-vdW-FV		UNIFAC-vdW-FV (with H ₂ O)	
		Uptake	UNIFAC-vdW-FV		PC_{pred}	Rank	PC_{pred}	Rank	PC_{pred}	Rank	PC_{pred}	Rank
Poly(<i>n</i> -hydroxybutyl methacrylate)	100	31.0 ± 3.5	12.8	2.6 ± 0.2	2.5	1	3.1	2	4.7	1	14.2	1
Poly(hydroxyethyl methacrylate)	20	38.5 ± 4.4	22.8	2.3 ± 0.4	2.2	3	3.1	3	4.5	3	13.4	3
Poly(<i>n</i> -hydroxypropyl methacrylate)	400	32.0 ± 3.6	17.2	2.3 ± 0.4	2.5	2	3.2	1	4.7	2	13.5	2
Poly(ethyl acrylate)	100	4.9 ± 0.6	2.3	1.8 ± 0.3	2.0	6	2.1	5	1.9	5	5.4	6
Poly(<i>n</i> -propyl acrylate)	140	2.1 ± 0.2	0.9	1.7 ± 0.4	1.8	8	1.8	8	1.3	8	2.5	7
Poly(<i>n</i> -butyl acrylate)	100	1.0 ± 0.1	0.9	1.5 ± 0.3	1.9	7	1.9	7	1.5	7	2.1	8
Poly(methyl acrylate)	40	14.5 ± 1.6	3.1	1.4 ± 0.5	2.1	5	2.1	6	1.5	6	9.7	4
Poly(vinyl propionate)	30	4.4 ± 0.5	2.7	1.4 ± 0.3	2.2	4	2.3	4	2.2	4	5.6	5

Partition coefficient values were obtained at 30°C using an initial aqueous concentration of 20 g/L. Experimental errors shown are ±1 standard deviation.

w_c values reported in the literature are generally measured for an as-received dry polymer, and may not reflect the material once exposed to an aqueous fermentation medium. Table 2 provides melting point (T_m) and w_c data for a range of semicrystalline materials in their as-received states, as well as after soaking in water and a 2 wt% butyric acid solution. In most cases, neither water nor solute changed these properties. However, poly(tetramethylene glycol) (PTMG) proved to be highly sensitive to the presence of *n*-butyric acid, as w_c declined from 0.55 for the dry polymer to 0.38 for the acid-soaked sample. This loss of crystallinity within PTMG is illustrated in Figure 1, which shows the transition from a semicrystalline solid to viscoelastic polymer melt upon immersion in an aqueous *n*-butyric acid solution. The polymer melt's increasingly liquid-like behavior and increased tackiness as w_c is reduced could cause operational challenges in a TPPB, including more difficult handling, separation, and adsorption to internal bio-reactor components.¹⁶

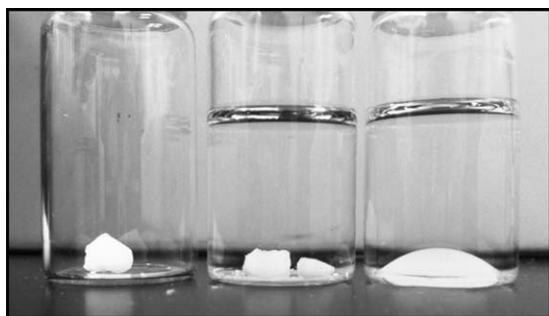
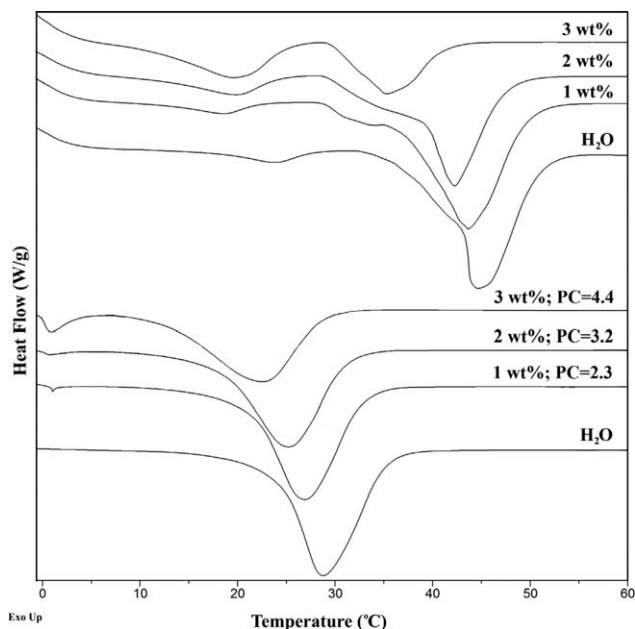
Nevertheless, the loss of mechanical integrity as crystallinity is lowered is counteracted by the resultant increase in the amount of absorptive amorphous material. To demonstrate this, the unusual behavior of the PTMG + *n*-butyric acid system was further investigated by measuring PC, T_m , and w_c for polymer exposed to a range of acid concentrations (Figure 2; Table 3). Although soaking PTMG in 1 wt% *n*-butyric acid had no measurable impact on crystallinity, a 3 wt% solution reduced w_c to just 0.12. In addition, increasing the *n*-butyric acid concentration from 1 to 3 wt% increased PC_{expt} from 2.3 ± 0.2 to 4.4 ± 0.2 (Table 3), indicating an increase in the polymer's capacity to remove the acid. The observed relationship between PC and crystallinity is consistent with Eq. 4, in that the correction of PC_{expt} to the degree of crystallinity (using w_c found at each concentration: 1, 2, and 3 wt%) yields the same amorphous phase partition coefficient ($PC_{\text{am}} = 5.1 \pm 0.1$).

From a material selection standpoint, lower crystallinity maximizes the absorptive amorphous phase and improves solute uptake. Table 4 further highlights the deleterious effect of crystallinity on PC, shown as the difference between PC_{am} (100% amorphous material) and PC_{expt} (semicrystalline material). Table 4 also demonstrates the challenge in predicting the performance of semicrystalline polymers, indicated by the generally poor agreement between predicted partition coefficients (PC_{pred}) and experimental measurements (PC_{am}). The amorphous phase activity coefficients are consistently overpredicted when water was not considered, with AAD% values of FH-Hildebrand (49%), FH-HSP (51%), and UNIFAC-vdW-FV (51%). In contrast, incorporating equilibrium water swelling data into UNIFAC-vdW-FV(H₂O) improved PC predictions to an AAD of 30%. In conjunction with PC predictions for amorphous polymers (Table 1), the results in Table 4 demonstrate the system-dependence of model accuracy and the benefits of employing a broad range of predictive tools for effective polymer identification.

Finally, uncertainty in w_c values undoubtedly contribute to errors in PC prediction, given the difficulty in integrating broad DSC melting endotherms, as well as variations in the morphology and quality of polymer crystallites from those used to generate literature values for the heat of fusion. We conclude that the selection of semicrystalline materials for TPPB applications is more qualitative than the selection of amorphous polymers, and more of an exercise in identifying

Table 2. Effect of H₂O and 2 wt% *n*-Butyric Acid Solution on the Crystalline Weight Fraction (w_c) and Melting Temperature (T_m) for Various Semicrystalline Polymers

Polymer	T_m (°C)			w_c		
	As-Received	Water Soaked	Acid Soaked (2 wt%)	As-Received	Water Soaked	Acid Soaked (2 wt%)
Poly(tetramethylene glycol)	47	45	42	0.55	0.50	0.38
Poly(ϵ -caprolactone)	69	67	64	0.73	0.73	0.70
Poly(butylene adipate)	61	59	60	0.62	0.62	0.62
Poly(trimethylene adipate)	40	45	44	0.40	0.44	0.40
Poly(ethylene adipate)	54	53	50	0.58	0.61	0.61
Poly(trimethylene succinate)	52	47	47	0.27	0.30	0.32
Poly(ethylene succinate)	102	101	97	0.42	0.41	0.44

**Figure 1.** PTMG (2.9 kg/mol) as-received (left) and after soaking in pure water (center) and in a *n*-butyric acid solution (2 wt%) (right).**Figure 2.** First (top) and second (bottom) DSC heating scans of poly(tetramethylene glycol) (2.9 kg/mol) for samples after equilibration with pure water and aqueous *n*-butyric acid solutions with initial concentrations of 1, 2, and 3 wt%. Melting temperature (T_m) values were derived from first heating scans.

promising candidates (by rank) than in predicting PC values accurately.

Glass transition temperature (T_g)

For a polymeric NAP to be effective, it must absorb/desorb the solute fast enough to maintain the desired aqueous phase concentration during TPPB operation. This gener-

Table 3. PTMG's Crystalline Weight Fraction (w_c), Melting Temperature (T_m), and Experimental (Bulk Polymer) Partition Coefficient (PC_{expt}) and Corrected Amorphous Phase Partition Coefficient (PC_{am}) After Equilibration with *n*-Butyric Acid at Varying Initial Aqueous Concentrations

	w_c	T_m (°C)	PC_{expt}	PC_{am}
Dry (as-received)	0.55	47	–	–
Water soaked (100 wt%)	0.50	45	–	–
<i>n</i> -Butyric acid soaked (1 wt%)	0.56	44	2.3 ± 0.2	5.2 ± 0.5
<i>n</i> -Butyric acid soaked (2 wt%)	0.38	42	3.2 ± 0.2	5.2 ± 0.3
<i>n</i> -Butyric acid soaked (3 wt%)	0.12	35	4.4 ± 0.2	5.0 ± 0.2

ally requires a T_g that is significantly lower than the TPPB operating temperature, since this difference correlates strongly with polymer segment mobility, solute diffusivity,⁴¹ and absorption rate.²⁹ Poly(vinyl acetate) (PVAc) has a reported T_g of 46°C and, as such, is expected to exist in a nonabsorptive glassy state when used at 30°C, and an absorptive rubbery state when immersed in an aqueous *n*-octanol solution at 60°C. However, the data plotted in Figure 3 show that PVAc exhibits good thermodynamic affinity for *n*-octanol at both test temperatures, with instantaneous partition coefficients ($PC^*(t)$) evolving to high equilibrium PC values, albeit at significantly different rates.

Figure 3 also reports the T_g of the polymer phase as a function of time, revealing a rapid decline in glass transition temperature from 46°C in a dry sample to about 15°C upon immersion in an *n*-octanol solution. This T_g reduction accounts for the absorption observed at the 30°C test condition, but it is the speed of this decline at both 30 and 60°C that is particularly interesting. Whereas *n*-octanol absorption rates were highly temperature dependent, polymer plasticization was rapid in both experiments. The data listed in Table 5 confirm that plasticization results from a rapid uptake of a small amount of water, which in turn facilitates *n*-octanol absorption by improving polymer chain mobility. The high diffusivity of water has been noted for several polymers of relevance to aqueous–organic separations,^{29,49,51–55} and the observed influence of penetrant size and shape on *n*-octanol uptake by PVAc is consistent with reports on several other polymer–solute systems.^{56–58}

In the context of TPPB design, plasticization by water and/or solute can significantly improve absorption/desorption dynamics, allowing polymers with T_g s slightly above the bioprocess temperature to function effectively. In all cases, a large difference between the operating temperature and the *in situ* polymer phase T_g is advantageous, such that $T \gg T_g$. In the PVAc/*n*-octanol system, operating 45°C above the

Table 4. Experimental (PC_{expt}), Corrected Amorphous Phase (PC_{am}), and Predicted (PC_{pred}) Partition Coefficient Values for *n*-Butyric Acid with Various Semicrystalline Polymers

Polymer	MW (kg/mol)	Water Uptake		PC_{expt}	PC_{am}	FH-Hildebrand		FH-HSP		UNIFAC-vdW-FV		UNIFAC-vdW-FV (with H ₂ O)	
		Uptake (wt%)	UNIFAC-vdW-FV (wt%)			PC_{pred}	Rank	PC_{pred}	Rank	PC_{pred}	Rank	PC_{pred}	Rank
Poly(tetramethylene glycol)	2.9	4.9 ± 0.6	9.6	3.2 ± 0.2	5.2 ± 0.3	1.7	7	1.6	7	2.9	1	6.7	1
Poly(ϵ -caprolactone)	55	0.9 ± 0.1	2.0	1.5 ± 0.2	5.0 ± 0.7	1.9	6	1.9	5	1.9	2	2.4	7
Poly(butylene adipate)	12	1.9 ± 0.2	3.5	1.8 ± 0.2	4.7 ± 0.5	2.0	5	1.9	6	1.8	3	2.9	5
Poly(trimethylene adipate)	3.8	2.5 ± 0.3	2.1	2.2 ± 0.4	3.7 ± 0.7	2.1	2	2.0	4	1.8	4	3.4	3
Poly(ethylene adipate)	10	3.4 ± 0.4	2.2	1.2 ± 0.2	3.0 ± 0.5	2.1	3	2.1	3	1.6	5	3.6	2
Poly(trimethylene succinate)	9.5	3.2 ± 0.4	2.9	1.8 ± 0.4	2.6 ± 0.6	2.2	1	2.2	1	1.6	6	3.4	4
Poly(ethylene succinate)	10	2.4 ± 0.3	3.7	1.0 ± 0.2	1.7 ± 0.4	2.1	4	2.2	2	1.4	7	2.5	6

Partition coefficient values were obtained at 30°C using an initial aqueous concentration of 20 g/L. Experimental errors shown are ±1 standard deviation.

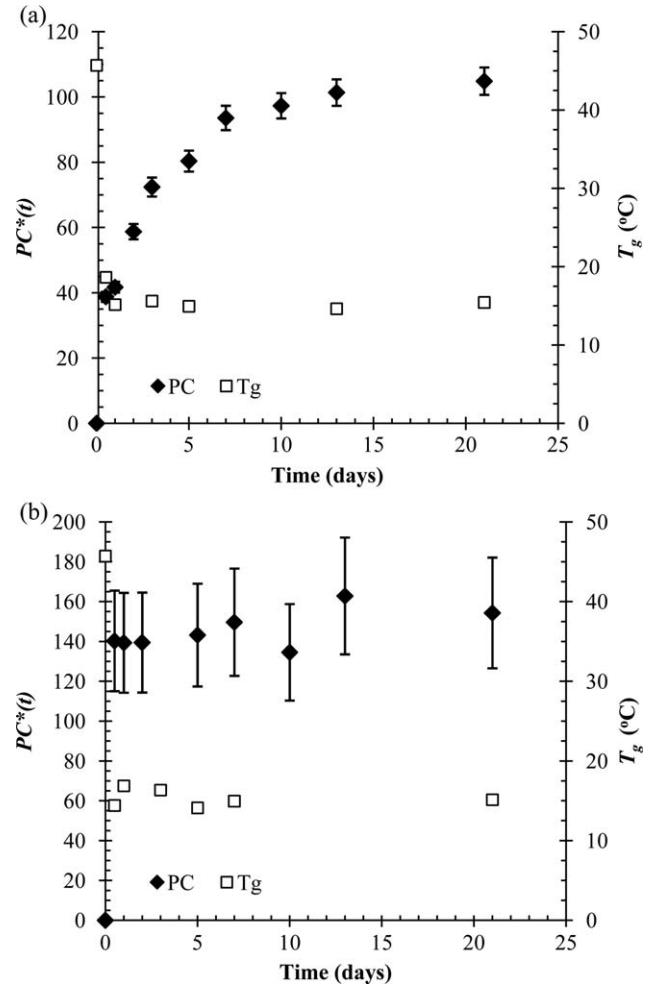


Figure 3. Instantaneous T_g and *n*-octanol partition coefficient for PVAc ($M_w \approx 350$ kg/mol) at (a) 30°C and (b) 60°C.

plasticized glass transition temperature ($T - T_g$) (Figure 3b) as opposed to $T - T_g = 15^\circ\text{C}$ (Figure 3a) resulted in much faster solute equilibration, as well as a higher overall partition coefficient. However, when increasing a bioreactor's operating temperature is not technically feasible, polymers with low T_g s should be preferentially selected for their likelihood to achieve more rapid solute uptake.

Conclusions

Thermodynamic affinity, degree of crystallinity (w_c), and glass transition temperature (T_g) determine the suitability of a polymer as an NAP for a given TPPB application. We have shown that the thermodynamic affinity of amorphous polymers can be predicted with a good degree of accuracy using Flory–Huggins-based models as well as UNIFAC-vdW-FV. However, challenges in quantifying crystallinity (w_c) under process operating conditions complicate similar analyses of semicrystalline materials, and this makes prediction of solute affinity using the above thermodynamic methods less satisfactory. This is particularly true in situations in which water and/or solute depress T_m below the bioreactor operating temperature, rendering a semicrystalline polymer amorphous. It should be noted that under such conditions, the polymer's amorphous phase fraction is increased, leading to higher overall PC values and a greater capacity to remove the toxic biomolecule.

Table 5. Plasticization of PVAc by Water and *n*-octanol

	T_g (°C)*		
	PVAc (15 kg/mol)	PVAc (100 kg/mol)	PVAc (350 kg/mol)
As-received	33	49	46
<i>n</i> -Octanol	37	51	52
Water	9	18	18
0.4 g <i>n</i> -Octanol/L	10	16	15

*3 weeks immersion at 30°C.

UNIFAC-vdW-FV predictions were also highly sensitive to water uptake; however, accuracy improvements were largely system dependent. In this work, it was found that consideration of water uptake improved PC predictions for semicrystalline materials but reduced predictive accuracy for amorphous polymers. Finally, interactions between the polymer and water and/or solute can greatly enhance absorption rates through polymer plasticization, and these effects should be considered where bioreactor operating temperatures are close to tabulated T_g values for dry, as-received polymer. However, in applications requiring very rapid absorption/desorption of target molecules, polymers with $T_g \ll T$ hold the most promise as they generally facilitate faster solute uptake. Overall, this work has contributed to establishing first-principles strategies for polymer selection in TPPB applications by evaluating the impact of several common polymer features and their effect on the extent and rate of solute uptake.

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