Spectroscopic and Thermodynamic Study of Biopolymer Adsorption Phenomena in Heterogeneous Solid–Liquid Systems

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ABSTRACT: Molecular selective adsorption processes at the solid surface of biopolymers in mixed solvent systems are poorly understood due to manifold interactions. However, the ability to achieve adsorptive fractionation of liquid mixtures is posited to relate to the role of specific solid–liquid interactions at the adsorbent interface. The hydration of solid biopolymers (amylose, amylopectin, cellulose) in binary aqueous systems is partly governed by the relative solvent binding affinities with the biopolymer surface sites, in accordance with the role of textural and surface chemical properties. While molecular models that account for the surface area and solvent effects provide reliable estimates of hydration energy and binding affinity parameters, spectroscopic and thermal methods offer a facile alternative experimental approach to account for detailed aspects of solvation phenomena at biopolymer interfaces that involve solid–liquid adsorption. In this report, thermal and spectroscopic methods were used to understand the interaction of starch- and cellulose-based materials in water–ethanol (W–E) binary mixtures. Batch adsorption studies in binary W–E mixtures reveal the selective solvent uptake properties by the biomaterials, in agreement with their solvent swelling in pure water or ethanol. The nature, stability of the bound water, and the thermodynamic properties of the biopolymers in variable hydration states were probed via differential scanning calorimetry and Raman spectroscopy. The trends in biopolymer–solvent interactions are corroborated by dye adsorption and scanning electron microscopy, indicating that biopolymer adsorption properties in W–E mixtures strongly depend on the surface area, pore structure, and accessibility of the polar surface groups of the biopolymer systems, in agreement with the solvent-selective uptake results reported herein.

INTRODUCTION

Hydration phenomena in heterogeneous systems at solid–liquid interfaces play a key role in physical and biophysical processes in condensed media. In general, hydration phenomena influence the thermodynamics of supramolecular complex formation and self-assembly in fields such as biocatalysis, protein folding, drug delivery systems, and macromolecule-based chemical separations. Biopolymer adsorbent technology that utilize starch and cellulose (CE) has gained increasing attention due to its relative abundance, low-cost, and low energy inputs with minimal infrastructure requirements for adsorptive-based fractionation. In a previous study, we reported the unique adsorption-based fractionation of water (W) from binary mixtures of water and ethanol (W–E) using biopolymer sorbents and their modified forms. Elsewhere, we reported on the unique adsorption of water vapor using starch biopolymer adsorbents. In condensed media, the adsorptive fractionation of solvents and organic mixtures using biopolymers such as starch and cellulose encompasses variable solute–solvent (s–l), solute–solvent (s–l), and solvent–solvent (l–l) interactions. In a recent isothermal study of starch–water vapor systems, the importance of biopolymer–solvent (s–l) interactions was shown by markedly different thermodynamic properties of starch-based adsorbents. By comparison, the heterogeneous fractionation of water from W–E binary solutions using starch- and cellulose-based adsorbents was reported that extends beyond the pioneering work of Ladisch and Dyck. However, there is a limited molecular-level understanding of the factors that govern hydration and solvent-selective fractionation in solid-solution (s–l) sorption processes, especially for native and modified (cross-linked) biopolymer systems. This knowledge gap limits the rational design and utilization of biopolymers for applications such as advanced drug delivery and industrial-scale chemical separations.

Numerous reports have applied theoretical and experimental techniques to investigate solvent–solvent interactions and biopolymer hydration. It is also noteworthy that the subject of hydration from the viewpoint of solution chemistry and chemical physics is an expansive and complex field of research. In particular, the field has elicited a heated debate and stimulated investigations of solvent–solvent interactions, which is beyond the scope of the current work. In the case of s–l interactions, the present work is focused on biopolymer solvation processes in the context of s–l interactions.
adsorption processes. While the adsorption phenomena of this type involve an ensemble of interactions, the relative affinities of solvent with the active surface sites of the solid-phase biopolymer is the focus herein. Although, a molecular-level understanding of biopolymer adsorption processes is limited in heterogeneous s−l systems, solvent swelling of native and modified biopolymers in mixed solvents indicates that variable solvent-selective adsorption occurs. Greater solvent-selective adsorption occurs for biopolymers that were incrementally cross-linked with epichlorohydrin, where the effect was assigned to changes in the hydrophile−lipophile balance (HLB) of the biopolymer. Thus, it is posited that the role of surface accessible functional groups play a key role in solvent−solute interactions, where changes to the Gibbs surface energy of the system depend on the textural properties of the adsorbent system. Models that incorporate surface area and solvent effects provide reliable estimates of hydration energy and binding affinity that may account for hydration phenomena in structurally well-defined macromolecular systems. By contrast, a complementary approach that employs experimental methods such as spectroscopy (Fourier-transform infrared, NMR, and Raman) and thermal analysis (differential scanning calorimetry, DSC) techniques offer a facile alternative for the study of biopolymer solvation in materials with complex tertiary structures. Collectively, these techniques enable a detailed study of interactions in biopolymer−solvent (s−l) systems with consideration of the biopolymer morphology, thermal, swelling, and rheological properties that complement a large body of research in this field.

Herein, we report a multi-instrumental approach for the study of the adsorption properties of cellulose (CE) and several starch biopolymers derived from corn and maize that contain variable amylose (AM) and amylopectin (AP) contents in the presence of binary solvent mixtures. The starch materials herein include high AP maize starch (98% AP, 2% AM), soluble corn starch (50% AM/AP), and high AM starch (98% AM, 2% AP). These biopolymers are denoted as Table 1. Selected Physicochemical Properties of Biopolymers and Solvents (NR = Not Recorded; ND = No Data)

| Sample ID | Polysaccharide | Cellulose | Maize Starch | Soluble Starch | High Amylose Starch |
|-----------|----------------|-----------|--------------|----------------|-------------------|
| AM50      | CE             | AP        | AM           |
| AM        |                |           |              |

- AP content (%): 98, 50, 50, 2
- Molar mass (g/mol): 2.7 × 10^4–9.0 × 10^5, 5.30 × 10^7, 342.30, average > 1.5 × 10^5
- Average > 1.5 × 10^5

- Surface area (cm^2/g): 0.96, 1.52, NR, 0.56
- Pore size (Å): 2.5 × 10^−3, 1.8 × 10^−5, NR, 6.0 × 10^−4
- Pore volume (Å^3): 2.5 × 10^−3, 1.8 × 10^−5, NR, 6.0 × 10^−4
- Water solubility (g/L): insoluble, 10%, soluble, negligible
- Dielectric constant (ε): 1.2, 3.6, ND, ND
- Solvents: water (W), ethanol (E)
- Molar volume (V_m; mL/mol): 18.0, 58.0, 24.5
- Dielectric constant (ε): 18.0, 58.0, 24.5
- Enthalpy of vaporization (ΔH_vap; °C): 2257, 841

“Based on nitrogen adsorption isotherm results."
The objectives of the current study are manifold: (1) to characterize the textural and surface properties of the biopolymers, (2) to study the fractionation properties of water in W–E binary mixtures, and (3) to understand the molecular-level details of biopolymer—solvent adsorptive processes. Scanning Electron Microscopy (SEM) and related imaging methods provide information on the biopolymer morphology and textural properties. A dye-based adsorption method with phenolphthalein (phth) was employed, where the degree of phth decolorization in the presence of polysaccharides provides an assessment of the biopolymer hydroxyl (−OH) group accessibility. The biopolymer—solvent-selective uptake properties were investigated using batch adsorption and solvent swelling studies in neat water (W) and ethanol (E), where trends in W–E uptake is understood on the basis of differences in adsorptive affinity due to the variation in the textural and surface properties of the biopolymer materials.

The unique solvent selectivity/uptake of such adsorbents in W, E, and mixed W–E solvents with variable composition provides an avenue to establish a molecular-level understanding of the biopolymer—solvent adsorption process. Accompanying processes such as l–l and s–l interactions can be inferred from DSC and Raman spectroscopy results. The present study contributes to an understanding of the role of hydration phenomena at solid biopolymer interfaces and provides further insight into the molecular-level process of solvent fractionation in W–E mixtures via a complementary experimental approach. The role of biopolymer solvation is a topic of continued interest due to the utilization and design of improved biopolymer materials for green chemistry and sustainable industrial processing. The present research is expected to catalyze further developments in these areas that employ platform biomaterials such as starch and cellulose because of their natural abundance, along with their unique structure and interfacial properties. Biomaterials will play an ever-increasing role in the future, especially in diverse applications as functional coatings for energy storage and exchange, biocatalyst supports, advanced drug delivery, and solid-phase extraction (SPE) materials for food processing and environmental remediation.

**RESULTS**

As indicated above, spectral and thermal characterization of solid-phase starch and cellulose polymers in solvent systems are described below, along with the corresponding solvent swelling and adsorption properties in condensed media.

**Characterization of Biopolymers.** Starch is a semicrystalline biopolymer linked by α(1 → 4) and α(1 → 6) glycosidic bonds that contain linear (amylose; AM) and branched (amylopectin; AP) oligomers (cf. Figure 1a,b), respectively. Cellulose (cf. Figure 1c) contains β(1 → 4) glycosidic bonds with greater rigidity over starch, and variable crystallinity due to the extensive intra- and inter-molecular H-bonding between the neighboring glucopyranose monomer units and cellulose chains. This unique H-bonding of cellulose leads to the supramolecular self-assembly of its polymer strands to aggregate into micro- and macro-fibril structures. The structural variability of starch and cellulose affects the textural (pore size distribution and surface area) properties and surface accessible functional groups of these biopolymers.

**Textural Properties.** Textural properties (porosity and surface area) play a key role in determining the relative accessibility of the active sorption sites of the biopolymer materials and their subsequent s–l interactions can be inferred from DSC and Raman spectroscopy results. The present study contributes to an understanding of the role of hydration phenomena at solid biopolymer interfaces and provides further insight into the molecular-level process of solvent fractionation in W–E mixtures via a complementary experimental approach. The role of biopolymer solvation is a topic of continued interest due to the utilization and design of improved biopolymer materials for green chemistry and sustainable industrial processing. The present research is expected to catalyze further developments in these areas that employ platform biomaterials such as starch and cellulose because of their natural abundance, along with their unique structure and interfacial properties. Biomaterials will play an ever-increasing role in the future, especially in diverse applications as functional coatings for energy storage and exchange, biocatalyst supports, advanced drug delivery, and solid-phase extraction (SPE) materials for food processing and environmental remediation.
the biopolymer, as noted by the textural features in Figure 2d. While the SEM results provide useful structural insight into the morphology of biopolymers, there is a need to account for the role of solvent effects due to the amphiphilic nature of such biopolymers. The role of solvent on the biopolymer structure is supported by the unique rheological behavior of starch at variable concentrations.32

**Surface Chemical Properties.** The surface accessible functional groups and dipolar character of biopolymers are important aspects that govern their structure and hydration properties. The surface accessibility of the −OH groups of the biopolymers was estimated using a dye-based method with phenolphthalein (phth) as a probe,38 based on the decolorization effect of phth upon complex formation with polysaccharides at an alkaline pH39 (cf. Figure S1). The extent to its unbound form.37 Figure 3 illustrates the decolorization where the formation of an optically transparent biopolymer of dye decolorization in the presence of biopolymers generally phth complex is the result of a pKa shift of bound phth relative to its unbound form.37 Figure 3 illustrates the decolorization results obtained from the adsorption profile of phth in the saturation region of the isotherm at a fixed biopolymer dosage (ca. 30 mg). The relative dye decolorization for the biopolymers are listed in descending order: AM > AM50 > AP ≫ CE, according to the relative absorbance change (ΔAbs) observed in Figure 3. The results indicate greater −OH accessible sites for AM, and the least accessibility for CE. It is worth mentioning that ca. 30% of the −OH groups for CE are accessible based on the relative peak intensity of CE with respect to AM, in agreement with an independent study.12

**Swelling and Uptake Properties.** Variable solvent swelling relates to the solvation of biopolymers that ultimately influence their properties in applications such as controlled-release delivery systems40,41 and adsorptive fractionation.42 The thermodynamics of the solvent uptake by biopolymer adsorbents can be partly related to surface interactions that vary according to the textural and surface chemical properties of the materials. Solvent swelling encompasses solvent−solvent (1−1) and biopolymer−solvent (s−l) interactions since it is a sorption (adsorption + absorption) process. By contrast, isotherm parameters (Qm, n) in Table 2 relate to adsorption phenomena that may be ascribed mainly to biopolymer−solvent (s−l) interactions. The solvent swelling [S_{w,l} (%)] for biopolymers imbibed in neat water and ethanol (Table 2) provide evidence that selective solvent uptake occurs, especially when absorption processes prevail. Similar to the dye decolorization results, the solvent swelling generally increases with the amyllose content, whereas CE shows unique swelling in water, as follows: AM (250) > CE (170) > AM50 (160) > AP (153). The trends in solvent swelling are in agreement with the results for monolayer water uptake capacity for biopolymers (Qm) in binary W−E mixtures (cf. Table 2) where CE (1.6) > AM (0.61) > AM50 (0.20) ≈ AP (0.21).6 The offset in the Qm values in Table 2 for water and ethanol uptake with the biopolymers is accounted for by the differences in the dielectric constant and molar volumes of each solvent listed in Table 1 (vide infra).

**Solvent−Biopolymer Interactions in Mixed Solvents.** The solvation properties of biopolymers (AP, AM50, AM, and CE) were further examined using DSC and Raman spectroscopy, in conjunction with the textural and surface chemical properties described above. It should be noted that the preparation of solvated biopolymer (solid−liquid) samples enable a study of biopolymer−solvent interactions. One exception is for the case of solvent swelling measurements since this method involves sorption, as noted above. The use of adsorption isotherm models and complementary methods (Raman spectroscopy and DSC) in neat W or E, and mixed W−E systems affords further insight into the primary role of biopolymer−solvent and secondary role of solvent−solvent interactions.

**Differential Scanning Calorimetry.** DSC provides unique complementary information on the state of biopolymer solvation and s−l interactions by monitoring the trends in endothermic profiles for vaporization (ΔH_{vap}). DSC has been widely used to study the thermal properties of starch materials, following the seminal work of Steven and Elton.35 The use of DSC for the study of the hydration properties of functional materials was recently reviewed,22,46 where starch and its modified forms display notably complex thermal behavior.

| Table 2. Swelling and Isotherm Uptake Properties of Biopolymers in Water−Ethanol (W−E) Mixtures |  |
|---|---|---|---|---|
| materials | AP | AM50 | AM | CE |
| S_E (%) | 2.1 ± 0.5 | 8.0 ± 2 | 14 ± 6 | 2.0 ± 0.1 |
| S_W (%) | 153 ± 10 | 160 ± 21 | 250 ± 12 | 170 ± 10 |
| Q_{m,E} (g/g) | 0.032 ± 0.001 | 0.051 ± 0.008 | 0.0084 ± 0.0025 | 1.03 ± 0.13 |
| n_E | 1.68 ± 0.102 | 0.895 ± 0.156 | 3.08 ± 1.43 | 3.18 ± 2.13 |
| Q_{m,W} (g/g) | 0.21 ± 0.03 | 0.20 ± 0.08 | 0.61 ± 0.11 | 1.6 ± 0.2 |
| n_W | 3.86 ± 1.23 | 2.44 ± 1.76 | 2.02 ± 0.666 | 1.15 ± 0.55 |

*S_E/S_W* swelling of ethanol (E) and water (W). *Q_{m,E} and Q_{m,W} are the adsorption capacity of E and W for binary W−E mixtures of variable composition. Data from ref 26.
In spite of the structural complexity of bound water at such biopolymer surfaces, DSC can be used to probe the thermodynamic state of the bound solvent in biopolymers. It follows that the adsorbent–solvent (s−l) interactions from calorimetry can be evaluated to afford molecular-level interpretations of such processes as noted for hydrated mineral oxides. In particular, the unique thermal properties of starch (amylose and amyllopectin) and cellulose in their solvated state in water–ethanol mixtures are expected to differ due to the structural variation among these biopolymers. For instance, the nature of biopolymer–solvent (s−l) interactions can be inferred based on thermal events related to adsorbed solvent, since free, weakly/strongly bound, and desorbed water reveals unique DSC profiles with variable heat flow, intensity, and temperature shifts.

In Figure 4a–d, the DSC thermograms reveal dehydration and gelatinization endotherms for biopolymer (s−l) systems with a variable W−E content over the 30−140 °C range. The relative stability of the bound water is greatest for CE (Figure 4d) and least for AM (c), as follows: CE > AP ≥ AM50 > AM. This stability profile was inferred from the full-width at half-maximum (FWHM) results of the Raman data (cf. Table S1) and the DSC dehydration transition temperatures in Figure 4. Starch materials with a higher amylose content (AM and

Figure 4. Differential scanning calorimetry results for various biopolymer materials; (a) AP, (b) AM50, (c) AM, and (d) CE, with a variable water−ethanol content.

Figure 5. Raman spectra of the biopolymer materials in W−E solvent systems containing D2O (10% w/w), where the relative water−ethanol (%W & E) content is shown.
AM50; Figure 4b,c) have greater affinity for water, as evidenced by a wide thermal transition range (40−140 °C) for the observed endotherms. By contrast, a narrow thermal range (ca. 80−120 °C; Figure 4a,d) occurs for high amylopectin (AP) starch and cellulose CE, in accordance with their known hydrophobic character. Furthermore, the $\Delta H_{ap}$ varies among the biomaterials in neat W versus neat E, suggesting that solvation effects depend on the nature of solvent (vide infra). The foregoing provides support that variable solute−solvent (s−l) binding affinity occurs among the various biopolymer systems.

The variable s−l interactions may be accounted for by differences in the textural and surface chemical properties of the materials as described in the sections below.

Raman Spectroscopy. The nature of biopolymer hydration in W−E binary mixtures was probed using Raman spectroscopy, because the spectral intensity of various Raman signatures are known to be sensitive to changes in the microenvironment. This effect results due to the changes in polarization and solute−solvent interactions of macromolecules in s−l systems.49,50 In particular, the relatively low scattering cross-section of water aids in the study of hydrated biopolymers using Raman spectroscopy. Figure 5 illustrates the Raman spectral results of various solid−phase biopolymers imbibed with variable levels of solvent (W or E) over the spectral region between 2200 and 3200 cm$^{-1}$. The bound water fraction contains D$_2$O (10% w/w), where the O−H2O mixtures yields a band assigned to the HOD uncoupled scattering cross-section of H$_2$O is low, the use of 10% D$_2$O/C-H band at 2900 cm$^{-1}$ strongly C-H stretching bands at 2800−2900 cm$^{-1}$ shows subtle red/blue Raman shifts (15 cm$^{-1}$) for water. The spectral features (intensity, FWHM, and shift) of this band are summarized in Table S1 in the Supporting Information (SI). The intensity of this band relates to its variable HLB character, arising due to its unique hydration in W versus neat E, consistent with the high swelling values listed in Table 2.

The Raman spectra in Figure 5 are dominated by vibrational bands at ~2900 cm$^{-1}$, where polysaccharides typically display strong C−H stretching bands at ~2800−3000 cm$^{-1}$ with broad O−H bands at ~3100−3600 cm$^{-1}$.15,51 In the case of the Raman spectra of W−E mixtures, the solvent O−H band appears ca. 3000−3400 cm$^{-1}$ and the observed C−H stretching bands (ca. 2900 cm$^{-1}$) relate to combined vibrational contributions from both starch and ethanol that vary with the composition of the mixed (W−E) solvent. The C−H band at 2900 cm$^{-1}$ shows subtle red/blue Raman shifts up to ~15 cm$^{-1}$, which indicate that the microenvironment effects are evident at variable W−E content. While the Raman scattering cross-section of H$_2$O is low, the use of 10% D$_2$O/H$_2$O mixtures yields a band assigned to the HOD uncoupled oscillator (~2500 cm$^{-1}$) for water. The spectral features (intensity, FWHM, and shift) of this band are summarized in Table S1 in the Supporting Information (SI). The intensity of the HOD band for the biopolymers increase as the water content increases from 25 to 75%. In general, the observed Raman shifts (~15 cm$^{-1}$) do not follow a well-defined trend among the biopolymer systems. However, the starch materials with a greater amylose content (i.e., AM and AM50) have somewhat greater FWHM values with larger Raman shifts (cf. Figure 5 and Table S1 in SI) compared to AP and CE.

DISCUSSION

The results from the complementary thermal (DSC) and Raman data above reveal interesting molecular-level details regarding solute−solvent (s−l) interactions in W−E binary mixtures. In particular, the DSC trends for the thermal stability of the bound ethanol are listed as: CE > AP > AM50 > AM. By comparison, a reversed trend of the biopolymer affinity to water is given as follows: AM > AM50 > AP > CE. The above trends can be related to the biopolymer textural and surface chemical properties consonant with interactions anticipated with hydrophilic and apolar domains of the respective biopolymers. The microporous character and apolar domains of CE and AP is supported by the SEM, DSC, and dye decolorization results above. The unique tertiary structure for each biopolymer provides an account of the characteristic water adsorption and swelling properties for these biopolymers. For example, water can be enclathrated within the amorphous and hydrophobic domains of AP and CE to a greater extent than AM, since the latter likely undergoes s−l hydrogen bonding interactions with accessible −OH groups of the biopolymer, in accordance with the key role of H-bonding interactions for AM and AM50 in the solid “dry” state. The trends in water swelling (AM > CE > AM50 > AP) and monolayer water adsorption (CE > AM > AM50 > AP) properties among the biopolymers differ in accordance with textural and surface chemical properties. The trends in solvent swelling suggest that the factors vary due to the differing role of surface adsorption versus absorption energetics among the biopolymers. The forces may include capillary condensation (absorption or enclathration) within the pores of the biopolymer, even at apolar sites, as well as dipolar interactions with surface functional −OH groups (adsorption). The broader DSC endotherm transitions for high amylose starches (AM and AM50) relate to their greater propensity to adsorb water, consistent with the high swelling values listed in Table 2. The affinity of AM and AM50 for water parallels other reports that describe the reactivity of starch granules with enzymatic reagents,36,37,54 in accordance with greater surface accessibility of active sites at the quaternary structural level of starch. AM and AM50 have greater surface accessible −OH groups for H-bonding due to their linear morphology (cf. Figure 1a and Scheme 1a). In contrast, AP and CE are characterized by branched or double helical (Scheme 1b) and fibril structures (Scheme 1c), respectively. The surface accessible −OH functional groups of CE was estimated at 30% (cf. Figure 3), where its greater water uptake relative to AP (cf. Table 2) relates to its variable HLB character, arising due to its unique microenvironment. This effect results due to the changes in polarization and solute−solvent−solvent non-covalent interactions of macromolecules in s−l systems.49,50 In particular, the relatively low scattering cross-section of water aids in the study of hydrated biopolymers using Raman spectroscopy. Figure 5 illustrates the Raman spectral results of various solid−phase biopolymers imbibed with variable levels of solvent (W or E) over the spectral region between 2200 and 3200 cm$^{-1}$. The bound water fraction contains D$_2$O (10% w/w), where the line-width/shape of the uncoupled oscillator HOD Raman bands is variable and relates to the microenvironment effects due to contributions assigned to bound (l−l or s−l) and free water.95,50

The Raman spectra in Figure 5 are dominated by vibrational bands at ~2900 cm$^{-1}$, where polysaccharides typically display strong C−H stretching bands at ~2800−3000 cm$^{-1}$ with broad O−H bands at ~3100−3600 cm$^{-1}$. In the case of the Raman spectra of W−E mixtures, the solvent O−H band appears ca. 3000−3400 cm$^{-1}$ and the observed C−H stretching bands (ca. 2900 cm$^{-1}$) relate to combined vibrational contributions from both starch and ethanol that vary with the composition of the mixed (W−E) solvent. The C−H band at 2900 cm$^{-1}$ shows subtle red/blue Raman shifts up to ~15 cm$^{-1}$, which indicate that the microenvironment effects are evident at variable W−E content. While the Raman scattering cross-section of H$_2$O is low, the use of 10% D$_2$O/H$_2$O mixtures yields a band assigned to the HOD uncoupled oscillator (~2500 cm$^{-1}$) for water. The spectral features (intensity, FWHM, and shift) of this band are summarized in Table S1 in the Supporting Information (SI). The intensity of the HOD band for the biopolymers increase as the water content increases from 25 to 75%. In general, the observed Raman shifts (~15 cm$^{-1}$) do not follow a well-defined trend among the biopolymer systems. However, the starch materials with a greater amylose content (i.e., AM and AM50) have somewhat greater FWHM values with larger Raman shifts (cf. Figure 5 and Table S1 in SI) compared to AP and CE.
quaternary fibril structure. The properties of CE are contrasted with starch biopolymers that possess greater hydrophilic character and conformational lability. The narrow DSC endotherms at higher temperatures (for AP and CE) are in agreement with the greater stability of the bound water, in accordance with the microporous structure and apolar domains of these biopolymers. It is worth noting that the DSC endotherm profiles of starch shift to higher temperature as the AP content increases, AP > AMS0 > AM, in agreement with the unique thermal properties of linear versus branched starches. Starch materials that contain a greater AP content display higher dehydration temperatures and enthalpic changes due to greater chain entanglement and s-s adhesive interactions over AM starches. The lower melting (gelatinization/dehydration endotherm) temperature of AM starch is supported by its reduced crystallinity. In general, higher enthalpy values in Figure 4 correlate with its greater water content (75% water).

By contrast, the endotherm events for the biopolymer–solvent systems (s-l) in W-E mixtures ca. 80 °C relate to ethanol vaporization (ΔH_vap), in agreement with its normal boiling point (cf. Table 1). It is worthwhile to note from the DSC results that secondary l-l interactions can be indirectly inferred from the trends for the dehydration endotherms of the biomaterials in neat W or E, and at 50–50 W-E content (wt %). The variable trends for the biopolymers suggest that hydrophobic hydration may act synergistically for hydrophilic (AM and AM50) biopolymers or competitively for hydrophobic (AP and CE) biopolymers upon imbibing in mixed and neat solvents.

In the case of the Raman spectral results, the trends in the shifts of the uncoupled OD vibrations and the FWHM are inferred to relate to the nature of intermolecular H-bonding of adsorbed solvent for the biopolymer–solvent (s-l) systems. Similar to the DSC results, the OD bands for AM and AM50 starch systems display a broader appearance with red shifts relative to AP and CE.

The Raman results for the HOD bands with variable FWHM and intensity indicate a greater level of adsorbed water in the biopolymer–solvent ensemble that adopts weakly-versus strongly-bound water for AM and CE, respectively. Relative to bulk water, AM and AM50 are characterized by microenvironments with weakly-bound water due to surface binding (s-l). By contrast, AP and CE reveal attenuated OD bands with a reduced FWHM. The Raman results provide evidence of highly ordered water for AP and CE due to the propensity of greater H-bonding within the micropore and fibril domains, due to the properties that are similar to bulk water (l-l). This is in agreement with the narrow DSC gelatinization/dehydration endotherms in Figure 4. The micropore networks in these biopolymers are supported by their unique morphology as revealed by the SEM results (cf. Figure 2). Thus, variable H-bonding occurs for AP and CE due to adsorbed versus absorbed water, as described above. The Raman spectra of hydration and bulk water typically consist of three spectral components at ~2400, 2500, and 2600, where the 2600 cm⁻¹ band describes nonhydrogen bonded water (OD). The broader band widths for AM and AM50 may relate to the weakly-bound water in the nonporous biopolymer domains, in agreement with the greater swelling and gelation properties of the linear starches.

The ongoing discussion of biopolymer–solvent (s-l) interactions reveals that variable solvent swelling, surface uptake, and affinity occurs in W-E systems due to several factors: (1) the surface accessibility of the biopolymer functional groups, (2) the overall biopolymer HLB, and (3) biopolymer textural properties based on the values shown in Table 1 and the SEM results (Figure 2). High amylose starches (AM and AM50) adopt a linear morphology that have more surface accessible –OH groups for effective H-bonding. By contrast, high amylopectin (AP) and cellulose have variable HLB due to reduced –OH accessibility, along with pore domains that accounts for bound water within micropore sites. In addition to the above factors, solvent physicochemical properties for W and E may account for the variable uptake by the biopolymer systems: (i) solvent dielectric constant (ε_W ≈ 80; ε_E ≈ 24), (ii) solvent molar volume: V_m(W) = 18 cm³/mol and V_m(E) = 58 cm³/mol, and (iii) the structuredness of the hydrogen bonding network of the solvent (three-dimensional (3-D) for water versus two-dimensional (2-D) for ethanol).

On the basis of the foregoing considerations and those listed in Table 1, water is more adaptive at forming stable hydrogen bonded clusters and hydration shells based on its smaller size, large dielectric constant, where the 3-D network of water over the 2-D ethanol solvent parallels the large cohesive energy density and enclathration properties of water. By contrast, the H-bond network of ethanol has greater 2-D character with a propensity to form W-E or E-E clusters due to favorable l-l interactions. According to the hydrophobic hydration of ethanol in water–ethanol mixtures, water envelops ethanol in a clathrate-like configuration. The variable trends for hydrophobic hydration of the biopolymers described herein arise due to differences in the biopolymer–solvent (s-l) affinities in W-E systems, in conjunction with the solvent physicochemical properties. The comparative hydration properties of starch systems and cellulose in pure water studied herein are conceptually illustrated in Scheme 2.

Scheme 2. Conceptualized View of Hydrated Biopolymers: (a) Amylose, (b) Amylopectin, and (c) Cellulose in Pure Water“

“*The degree of swelling of the respective biopolymers in water is denoted by the yellow spheres on the right.
CONCLUSIONS

The adsorption properties of the various starch- and cellulose-based biopolymers (AP, AM50, AM, and CE) were studied using DSC, Raman spectroscopy, SEM, and solvent swelling in neat/mixed water-ethanol (W–E) solvents. The swelling of the biopolymers in neat solvents was greater for high amylose (AM) and soluble starch (AM50) due to the flexible nature of the biopolymer backbone and greater surface accessibility of the hydroxyl groups. The flexible starch biopolymers (AM and AM50) have limited SA and pore structure due to efficient packing in the solid state. In contrast, the rigid fibril structure of cellulose (CE) contributes permanent porosity with less solvent swelling, in agreement with the SEM results.

The surface accessible biopolymer –OH groups were estimated by a dye decolorization method with phenolphthalein. Starch materials possess greater accessibility over cellulose due to the unique structural differences between each biopolymer. CE and AP show similar water adsorption properties in accordance with their hydrophobic character and biopolymer steric effects, as supported by dye adsorption, DSC, and Raman results. High amylose (AM and AM50) and high amylepectin (AP) starches differ in their hydrated state and HLB profile due to variable branching, swelling, and hydrophobic effects. The unique biopolymer–solvent interaction of CE in W–E relates to its quaternary fibril structure and pore domains that account for water uptake due to sorption (adsorption + adsorption) processes. Solvation of the biopolymer (s–l interactions) is influenced by the physicochemical properties of the system as follows: (1) the propensity of the biopolymer to undergo swelling, (2) functional group accessibility of the biopolymer, and (3) the nature (size, polarity, etc.) and composition of the solvent system. The complementary methods reported herein provide further insight into the factors governing biopolymer structure-function that relate to adsorption phenomena in s–l systems. In turn, we envisage that the structure of biopolymer sorbents can be modified in a controlled manner to yield materials with tailored properties for applications in food production, carrier devices, and as solid-phase extraction (SPE) materials for environmental remediation.

EXPERIMENTAL METHODS

Materials and Chemicals. Cellulose (CE) and starches from various sources (corn or maize) containing variable amylose and amylepectin contents were chosen as the biopolymer materials (cf. Table 1). All of the biopolymers and ethanol (100% w/w) were purchased from Sigma-Aldrich (Oakville, ON, Canada) and were used as received. Deionized Millipore water was used for the preparation of all aqueous solutions. The selected physicochemical properties of the polysaccharides and the solvent components are listed in Table 1.

Solvent Swelling Tests. The swelling properties of the polysaccharides were measured using 20 mg of material equilibrated in 7 mL of Millipore water or neat ethanol for 48 h. The degree of swelling in water (Sw) or ethanol (S_e) for the biopolymers is determined using eq 1 as shown below, where W_s is the wet sample and W_d is the dry sample after oven drying at 60 °C.

\[ S_w = \left( \frac{W_s - W_d}{W_d} \right) \times 100\% \]  

Phenolphthalein Decolorization Studies. The surface accessibility of the biopolymer –OH groups was estimated by the decolorization of phenolphthalein (phth) in aqueous solution using a method described elsewhere. A 7 mL solution containing phth in NaHCO₃ aqueous buffer at pH 10.5 was added to vials with variable sample masses (1–10 mg) that were allowed to equilibrate with shaking for 24 h at 295 K. Phase separation was achieved by centrifugation (Precision Micro-Semi Micro Centricone, Precision Scientific Co.) of the solution mixture at 1550 rpm and the measurement of absorbance was performed using a double beam spectrophotometer (Varian CARY 100) at 295 ± 0.5 K and \( \lambda_{max} \) of 552 nm.

Differential Scanning Calorimetry (DSC). The DSC thermograms were acquired from starch or cellulose samples that were equilibrated with known amounts of water and ethanol solvents. A biopolymer (20 mg) was added to 4 dram glass vials containing 10 mL of solvent with a variable water–ethanol (W–E) weight (%) content. The sample vials were incubated by shaking (Poly Science, Dual Action Shaker) for 24 h at 160 rpm at ambient pH and temperature to achieve equilibrium. Thereafter, the samples were separated from the solutions using a vacuum filter to obtain solvated solid (biopolymer–solvent) samples for analysis by DSC. The samples were added to DSC pans and sealed with a hole punched in the sample lid to allow for outgassing of vapor during the heating cycle over the temperature range of 30–150 °C.

Raman Spectroscopy. The samples for Raman spectroscopy were prepared as above (cf. DSC section), where the water content was isotopically mixed with 10% (w/w) D₂O/H₂O. The hydrated biopolymer samples were isolated by centrifuging (Precision Micro-Semi Micro Centricone, Precision Scientific Co.) at 1800 rpm for 1 h to obtain solvated solid (biopolymer–solvent) samples for further analysis.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acsomega.8b01663.

The surface accessibility of the biopolymer –OH groups were estimated using a dye-based method with phenolphthalein (phth), along with Raman spectral parameters (intensity, FWHM, and shift) of the biopolymers and are reported in 10% (w/w) D₂O/H₂O mixtures (PDF)

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Notes

The authors declare no competing financial interest.

ACKNOWLEDGMENTS

The Government of Saskatchewan (Ministry of Agriculture) through the Agriculture Development Fund (Project#: 20160266) is acknowledged for supporting this research.
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