Molecular insight into the wetting behavior and amphiphilic character of cellulose nanocrystals

David C. Malaspina

ICMAB-CSIC, Campus UAB Bellaterra, Barcelona, Spain.

Jordi Faraudo

ICMAB-CSIC, Campus UAB Bellaterra, Barcelona, Spain.

Abstract

The study of nanocellulose is a field of growing interest due to its many applications and its use in the development of biocompatible and eco-friendly materials. In spite of the vast number of studies in the field, many questions about the role of the molecular structure in the properties of cellulose are still subject of debate. One of these fundamental questions is the possible amphiphilic nature of cellulose and the relative role of hydrogen bonding and hydrophobic effect on the interactions of cellulose. In this work we present an extensive molecular dynamics simulation study of this question by analyzing the wetting of cellulose with water and organic solvent, its interaction with hydrophilic and hydrophobic ions and its interaction with a protein (human epidermal growth factor, hEGF). We consider two characteristic cellulose crystal planes of I β cellulose with very different roughness, different hydrogen bonding capability and different exposure of cellulose hydrophobic groups (the (010) plane which has exposed -OH groups and the (100) plane with buried -OH groups). Our results show that both surfaces are simultaneously hydrophilic and lipophilic, with both surfaces having very similar contact angles. In spite of the global similarity of wetting of both surfaces, the molecular details of wetting are very different and substantial local wetting heterogeneities (which strongly depend on the surface) appear for both solvents. We also observe a weak interaction of both surfaces with hydrophobic and hydrophilic solutes. These weak interactions are attributed to the simultaneous lipophilic and hydrophilic character of both (100) and (010) cellulose surfaces. Interestingly, we found a substantial interaction of both cellulose planes with polar and apolar residues of the hEGF protein.

*Corresponding author

Email address: jfaraudo@icmab.es (Jordi Faraudo)

1. Introduction

Cellulose is one of the most abundant polymers on Earth and it has been used as a source for materials in the form of flax, hemp, and cotton fibers since prehistoric times [1]. Nowadays, its nanoscale form, nanocellulose (i.e., cellulose with at least one dimension in the nano-scale) and nanocellulose composite based materials are emerging as interesting candidates for the development of eco-friendly advanced materials to substitute petroleum-based materials [2, 3, 4]. This growing interest in the development of new cellulose and nanocellulose based materials is accompanied by a revamped interest in the understanding of the relation between the properties of the material, its chemical structure and its hierarchical supramolecular organization [5, 6] which are not well understood in such a complex material. Interestingly, fundamental physico-chemical questions such as the role of hydrogen bonding and hydrophobic interactions (which are common in many subfields of colloid and interface science) are believed to play key roles in properties of cellulose and nanocellulose which are not fully understood [7].

Chemically (see Figure 1), cellulose is a polysaccharide formed by linearly connected repeated units of anhydrocellobiose (i.e. a disaccharide of two glucose molecules linked together through an oxygen covalently bonded to C1 of one glucose ring and C4 of the adjoining ring, the so-called β (1-4) glycosidic bond). Cellulose materials are made by these polysaccharide chains organized in highly hierarchical structures [2, 5, 6].

Cellulose chains assemble in nano-fibrils (sheet-like elongated structures with typical diameters in the ~ 10 nm scale) which also assemble in fibers that are the common basic constituent of cellulosic materials. Fibrils are semi-crystalline, with regions of low order coexisting with crystalline domains. There are substantial variations in basic properties of the fibrils such as the degree of crystallinity, shape, length and diameter as well as crystal structure depending on its origin (bacterial cellulose or cellulose obtained from wood for example [6, 8]).

What are the basic molecular and supramolecular driving forces responsible for the hierarchical organization of cellulose? As shown in Figure 1, cellulose has an extensive network of inter-molecular and intra-molecular hydrogen bonds. The properties of cellulose have been traditionally explained in terms of these hydrogen bond chains. Cellulose has a hygroscopic character [9] and in water tends to form hydrogel structures, a behavior which has been related to the presence of many polar groups in cellulose and its ability to form H-bonds. Intramolecular H-bonds are believed to provide chain stiffness and intermolecular hydrogen bonds are usually invoked to justify the assembly of the linear polymers into fibrillar structures. However, a different perspective has been proposed in the literature (the so-called Lindman hypothesis) [7, 10, 11, 12], which considers that the relevance of hydrogen bonding in cellulose has been overemphasized and points out the importance of hydrophobic interactions in cellulose (which may give a sort of amphiphilic character to cellulose). This proposed role of hydrophobic interactions, which is a controversial issue in the field of cellulose [13], is based on molecular structure considerations and on a careful

Figure 1: Molecular structure of cellulose. (a) Anhydrocellobiose repeated unit of cellulose, (b) Top view of two cellulose chains with indication of possible intra and inter molecular hydrogen bonds and (c) lateral view of the two cellulose chains shown in (b).

analysis of cellulose dissolution experiments [10]. As seen in Figure 1, cellulose chains possess several hydrogen bond donors that give an hydrophilic character to the molecule, but at the same time pyranose rings in the molecule have a more hydrophobic character, so that cellulose molecules have an anisotropy in their interactions, producing a complex competition between hydrophobic and hydrophilic character. Remarkably, it is very difficult to dissolve cellulose in both polar and apolar solvents. The solvents available for cellulose dissolution are of highly different nature which include unusual options (from simple or multicomponent mixtures, aqueous and organic media, inorganic and organic salts, with peculiar experimental conditions such as high and low temperatures or high and low pH) [11, 12]. This complex pattern is a clear indication of the complexity of the interactions involving cellulose chains.

The fundamental question we want to address in this work is the connection between the molecular and supramolecular structure of cellulose and the role of molecular-level interactions (hydrophobic effect, hydrogen bonding, ...) in cellulose-solvent and cellulose-molecule interactions. More specifically, we want to address theoretically, by using atomistic simulations, the question of the possible amphiphilic, hydrophilic or hydrophobic character of cellulose starting from the chemical structure of cellulose and its crystal structure.

65 The analysis of simulation results obtained from well-defined cellulose crystal structures in situations as diverse as wetting from different solvents or the interaction with different molecules (with different solvation characteristics and different hydrogen bonding capability) will allow us to discuss in full detail the roles of hydrophobic and H-bonding capabilities of cellulose at different scales.

70 The paper is organized as follows. In section 2, we will briefly summarize basic aspects of atomistic modelling of cellulose. In section 3, we will consider MD simulations of wetting of cellulose crystals by different types of solvents (more specifically, we consider a droplet of water and a droplet of an organic solvent onto a cellulose crystal at ambient temperature). In section 4, we consider the

75 interaction of cellulose with suspensions in water containing solutes of different hydrophobic or hydrophilic character and of different size (from small molecules to a protein). We end-up the paper with conclusions and perspectives (Section 5).

Figure 2: Atomistic modeling of I β cellulose (molecular representation and coloring uses the CPK convention). a) Nanocrystal (from proposed maize fibril shape[14]) with ≈ 4 nm diameter and ≈ 12 nm length (18144 atoms) generated with the "cellulose-builder" toolkit [15]. b) Cross section of (a) indicating the different crystallographic planes. c) Detail of the (010) plane in (b) with indication of surface -OH groups that are exposed and available for interaction for other molecules and solvents. d) Detail of the (100) plane in (b) indicating the location of the surface -OH groups involved in intramolecular hydrogen bonding. Image made with VMD [16].

2. Atomistic modeling of Cellulose

80 Molecular Dynamics (MD) simulation [17] is a suitable tool for performing atomistic computer simulations of materials and their interactions with molecules and solvents under given thermodynamic conditions (temperature, pressure). Conceptually, the method is simple. It is based on the fact that at
85 ordinary conditions (not under extreme temperatures or pressures), the motion of atoms of any molecular system can be simulated by implementing numerically the Newton laws of motion in a computer. This allows the prediction of supramolecular properties starting from an atomistic description of the system of interest.

The method requires the formulation of an atomistic model of the system
90 with an atomistically detailed structure and a method for the calculation of all interactions between atoms of the system, which in principle can be done at different levels of theory, ranging from quantum mechanical electronic structure methods to analytical force fields (which can be of semi-empirical origin or fitted from electronic structure calculations [18]).

95 Atomistic simulations of a full fibril containing both crystalline and amorphous regions are not possible, since in order to be realistic they will require the consideration of system sizes of the order of microns [6] which are beyond the possibilities of atomistic simulations. Therefore, atomistic MD simulations focused on fully crystalline fragments of fibrils (elementary nanofibrils or nanocrystals)
100 and also in their interactions with solvents and other molecules. Typical dimensions of the crystals in the simulations are in the order of ≈ 10 nm [19, 20]. Simulations of larger structures are possible by considering MD simulations of coarse-grained (not atomistic) models [21, 22].

The first ingredient needed in MD simulations is the generation of atomic
105 coordinates of the initial structures to be employed in the simulations. In the case of cellulose, this can be done using for example the "cellulose-builder" computational toolkit developed in Ref [15] which generates atomic coordinates of nanofibrils of any length with any of the known crystal structures of cellulose ($I\alpha$, $I\beta$, II, and III_I polymorphs [23]). This tool can be combined with other
110 packages for decoration of the cellulose fibrils with hemicellulose and lignin for simulations in the case of plant cellulose. An example of a nanofibril with the $I\beta$ crystal structure generated using this tool is shown in Figure 2a.

In the interactions of cellulose with solvents and molecules, the properties of the cellulose surface play an essential role. As shown in Figure 2b, different sur-
115 faces planes with different H-bond capability can be obtained from crystalline cellulose $I\beta$ [24]. Plane (010) $I\beta$ display the highest accessibility for hydrophilic groups (-OH) and highest surface roughness (figure 2c) while plane (100) $I\beta$ display the lowest accessibility for hydrophilic groups and lowest surface roughness (figure 2d). Plane (110) has an intermediate character between planes (010) and
120 (100). Crystalline $I\alpha$ cellulose has similar crystal planes in terms of hydrophilic groups accessibility and surface roughness [24]. Therefore, simulation studies of cellulose interactions with molecules and solvents should take into account this structural diversity in the character of cellulose surfaces.

Concerning the modelization of the atomic interactions, there are several
125 works discussing the suitability of molecular force fields for describing cellulose.
For example, in Ref[25] MD simulations using Gromos force field were employed
to study the thermal expansion and the Young modulus of $I\beta$ cellulose obtain-
ing a good agreement with experimental data. However, later simulations with
longer simulation times employing also the Gromos force field indicated limita-
130 tions in reproducing the known crystal structures of cellulose [26]. In the same
study, better results were obtained by using CHARMM C35 or GLYCAM06
force fields. A more recent, exhaustive comparison [19] between different well-
known molecular force fields (CHARMM C35, GLYCAM06 and CSFF) shows
that these three force fields correctly predict the (experimentally known) crystal
135 structures of several carbohydrates. Therefore, using any of these force fields for
the modelization of atomic interactions, it is possible to perform atomistic MD
simulations of cellulose crystals and their interactions with different solvents
and with other molecules at the desired temperatures and pressures.

In fact, this MD force field methodology described above has been employed
140 in many theoretical studies of cellulose. The self-assembly of cellulose chains in
water and the effect of different factors (chain length, ionization state) has been
studied in Ref [27] using CSFF forcefield for cellulose and TIP3P model for water
and in Ref [28] using the Glycam forcefield for cellulose and the TIP4P/2005
model for water. The interaction of $I\beta$ crystalline cellulose with water has
145 been studied using MD simulations with different force fields. CHARMM force
field for cellulose and TIP3P water model was employed in Refs [29, 30], the
PCFF force field for cellulose and SPC model for water was employed in Refs
[31, 24] and Glycam06 forcefield for cellulose and TIP3P model for water was
employed in Ref [32]. The interaction of cellulose crystals with peptides has
150 also been considered in Ref [30], modeling the cellulose and the protein with
the CHARMM forcefield and the water with the TIP3P model. Concerning
simulations in other solvents, in Ref [33] the authors performed MD simulations
of a cellulose nanocrystal in a water/octane emulsion using CHARMM force
field for cellulose and octane and the TIP3P model for water. The transfer of a
155 cellulose chain from the surface of a cellulose crystal surface towards bulk liquid
(water or organic solvent) was simulated in Ref [34], using the GROMOS96 45A4
force field for cellulose, a simple Lennard-Jones liquid model for the organic
solvent and the SPC model for water. There are also MD simulation studies
of the interaction of cellulose with molecules and materials in water solvent.
160 For example, Chen et al. [35] considered the interaction of urea in water with
cellulose and Alqus et al. [36] considered the interaction of cellulose covered
with graphene in water.

Without entering into the details of the results of all these diverse computa-
tional studies, we can say that they confirm the view that in cellulose crystals
165 both hydrogen bonding capability and hydrophobic interactions play a substan-
tial role. Heterogeneity is also important, in the sense that these studies show a
significant local dependence of hydrogen bonding and hydrophobic interactions.

We will further test this view here, by considering MD simulations of a $I\beta$
cellulose nanocrystal (Figure 2c, d) in several different situations, employing the

Table 1: Summary of wetting simulations (solvent droplet over a surface). We indicate the name for each system, cellulose crystal plane, number of solvent molecules, total number of atoms, simulation time, and simulation box size.

Name	Solvent	Cellulose I β crystal plane	Solvent molec.	Total atoms	Sim. time	Sim. box (nm $\times$ nm $\times$ nm)
S1	water	(010)	884	28,736	20 ns	$9.4\times 12\times 10.5$
S2	water	(100)	884	33,776	20 ns	$12\times 9.8\times 10.3$
S3	tetradecane	(010)	83	28,852	20 ns	$9.3\times 16\times 10.5$
S4	tetradecane	(100)	83	33,892	20 ns	$14\times 9.8\times 10.3$

170 same model for the cellulose crystal to study the interaction of its (010) and
(100) facets with different solvents (water and organic solvent) and different
molecules (with different solvation characteristics and different hydrogen bond-
ing capability). The analysis of MD results obtained from well-defined cellulose
crystal structures in these diverse situations will allow us to discuss in full de-
175 tail the roles of hydrophobic and H-bonding capabilities of cellulose at different
scales.

In the simulations reported in the following sections, we always employ the
CHARMM36 force field for modeling cellulose [37] since this general force field
allows also the simulation of different solvents [38] and also proteins and ions in
180 solution [39], making possible the study of the interaction of the interaction of
cellulose crystals with them. For water, we will employ the TIP4P/2005 model
[40] which is the water model that (up to now) best capture hydrogen-bonding
features of liquid water at all temperatures and pressures [41] and also has the
best performance in reproducing water properties (see [42] for a detailed com-
185 parison between the performance of different water models). Further technical
details are given in the Appendix.

3. Wetting of Cellulose: water and tetradecane

In this section, we consider MD simulations of wetting of (010) and (100)
surfaces of I β cellulose by water and by an apolar solvent (tetradecane). We
190 consider these two surfaces since, as discussed in the previous section, they
have very different hydrogen bonding capabilities, being the (010) plane the one
with the highest accessibility for hydrophilic groups (-OH) and highest surface
roughness (see figure 2c) while the plane (100) has the lowest accessibility for
hydrophilic groups and lowest surface roughness (see figure 2d).

195 The details of these four simulations (two surfaces and two solvents) are
summarized in Table 1. In the simulations, we initially place a solvent (water or
tetradecane) nanodroplet of about 3 nm size near the surface ((010) or (100)).
The results were analyzed once the droplet reaches an equilibrium configuration
over the surface. The results for both surfaces are shown in Figure 3 for water
200 and in Figure 4 for tetradecane.

The most important result of our simulations is that both surfaces show
wetting behavior that corresponds simultaneously to hydrophilic and lipophilic

Figure 3: Cellulose wetting behavior with water. a) Cellulose plane (010) wet by a nanodroplet of water, observed from the side view. b) Detail of the hydrogen bonds formed. c) Cellulose plane (100) wet by a nanodroplet of water, observed from the side view. d) Detail of the hydrogen bonds formed. e) 2-D density profile of water molecules on plane (010), side view. Resulting contact angles are indicated in the figure. f) 2-D density profile of water molecules on plane (100), side view. Resulting contact angles are indicated in the figure.

behavior.

In the case of wetting by water simulations (S1 and S2 in table 1), we observe the formation of a well-defined water droplet for both surfaces (see snapshots in Figure 3a and Figure c). The contact angles in the simulations are calculated from the average 2-D water density profiles of the droplet. We obtain a contact

angle of $\approx 16^\circ$ for the (010) plane and $\approx 23^\circ$ for the (100) plane, as seen in Figures 3e and 3f.

210 It is interesting to compare our results with previous simulation and experimental results, which showed mixed conclusions. Early simulations [31] with the older SPC water model and the PCFF force field for cellulose predicted a high contact angle (95°) for plane (100) and a small contact angle (46°) for plane (110) (this plane has an intermediate character between plane (100) and
215 plane (010), see Figure 2). More recent MD results [30] using also CHARMM forcefield but with the TIP3P water model (instead of the TIP4P/2005 model employed here) predicted a contact angle of 25° for water with cellulose plane (100), very similar to our results shown in Figure 3f (no results were reported for other planes in this case).

220 Experimental results show a wide variability in reported contact angles [43, 44, 45], due to many practical issues (specific experimental technique or protocol employed, type of sample, ...). Advancing and receding contact angles for water/air of 19° and 12° were observed for bacterial cellulose in Ref. [43]. In Ref [44], time-dependent contact angles were observed for crystalline cellulose
225 which started at 45° and were observed to decrease up to 32° . Time dependent contact angles were also observed for cellulose fibers [45], starting at 25° and decreasing up to 10° . Besides the variability associated with these results, we can say that experimental contact angles are consistent with the hydrophilic character obtained in our simulations.

230 In the case of wetting by tetradecane (S3 and S4 in table 1), we observe that the organic solvent spreads completely over both surfaces so the observed contact angles are zero (see Figure 4). Up to the best of our knowledge, there are no previous wetting simulations of cellulose by organic solvents. Also, we have not found experiments on wetting of crystalline nanocellulose by organic
235 solvents. There are of course reports on wetting of wood by different liquids, which may be related to the wetting properties of cellulose. Interestingly several organic liquids completely wet wood with contact angles of zero [46].

There are also recent studies that highlight the amphiphilic behavior of cellulose. For example, Kalashnikova et al [47] studied the stabilization of oil/water
240 emulsion by cellulose crystals. They found that crystals with low surface charge groups favor the stability of emulsions suggesting that the amphiphilic nature of cellulose is the main driving force for the stabilization. MD simulations [33] of a cellulose crystal in oil/water emulsion also support this interpretation.

Our simulations presented here suggest that the proposed amphiphilic behavior (which suggests the presence of hydrophilic and hydrophobic regions) can
245 be better described as a simultaneous hydrophilic and lipophilic behavior. All surfaces of cellulose crystal present this dual hydrophilic and lipophilic behavior. Noting that the contact angles for the organic solvent are smaller than the contact angles for water, we can also say that cellulose has larger affinity for
250 organic solvents than for water (is more lipophilic than hydrophilic).

Although the wetting behavior is the same for both (010) and (100) surfaces and both solvents (in the sense that both behave as hydrophilic and lipophilic), there are substantial differences in the molecular organization of the solvents at

Figure 4: Cellulose wetting behavior with organic solvent. Cellulose plane (010) wet by a nanodroplet of tetradecane, observed from: a) side view and b) top view. Cellulose plane (100) wet by a nanodroplet of tetradecane, observed from: c) side view and d) top view.

the interface with each of these surfaces. In the case of water at the (010) surface
 255 (Figure 3 a, b), we observe the penetration of water in the interstices between
 cellulose molecules, due to the high roughness of the surface. We also observe the
 formation of hydrogen bonds between water molecules and the exposed surface
 -OH groups of cellulose. In the case of the (100) plane (Figure 3 c, d), we observe
 260 the formation of water structures at the interface, with characteristic empty gaps
 on top of the apolar rings of cellulose. This peculiar ordering of water at plane
 (100) I β has been also reported in several previous MD simulations [29, 32].

We also observe the formation of hydrogen bonds between water and the
 -OH groups of the interface, groups which initially were not accessible to water.
 Comparing the initial crystal structure shown in Figure 2d with the structure
 265 obtained in presence of a water droplet (Figure 3b), we see that cellulose
 molecule-molecule H-bonds have been replaced by cellulose-water H-bonds.

We recall that, in spite of these molecular-level differences in the organiza-
 tion of water molecules, the contact angles in both cases are very similar (see
 Figure 3c). It is also clear that the wetting behavior of water at both surfaces
 270 has heterogeneities. This may reflect an heterogeneous character of the interac-
 tions at the interface which may impact the interaction with ions with different
 degrees of hydration or molecules with hydrophobic and hydrophilic patches as

Table 2: Summary of simulations with a molecule in water interacting with a cellulose surface. In all cases the molecules are negatively charged and the system is globally neutral, being the counterions Na^+ .

Name	Molecule	Cellulose I β crystal plane	Water molec.	Total atoms	Sim. time	Sim. box (nm $\times$ nm $\times$ nm)
S5	CO_3^{2-}	(010)	2,260	15,094	190 ns	$4.6 \times 4.1 \times 5.9$
S6	CO_3^{2-}	(100)	2,289	15,210	190 ns	$4.9 \times 4.1 \times 5.7$
S7	Ph_4B^-	(010)	2,431	15,818	170 ns	$4.6 \times 4.1 \times 6.2$
S8	Ph_4B^-	(100)	2,859	17,530	170 ns	$4.9 \times 4.1 \times 6.5$
S9a	EGF	(010)	24,665	124,665	83 ns	$9.5 \times 9.7 \times 10.3$
S9b	EGF	(010)	24,198	122,797	120 ns	$9.5 \times 10.4 \times 9.2$
S10a	EGF	(100)	25,318	132,317	78 ns	$10 \times 9.8 \times 10.3$
S10b	EGF	(100)	24,761	130,089	140 ns	$9.9 \times 10.4 \times 9.2$

proteins. This aspect will be investigated in the following section.

The arrangement of tetradecane in both surfaces is also different, as seen in Figure 4. In the case of the (010) surface, tetradecane molecules follow the stripes defined by the surface roughness, while in the case of the (100) surface, tetradecane molecules tend to assembly in clusters with local order.

4. Interaction of Cellulose with molecules in water

The results obtained in the previous section emphasize the presence of heterogeneities in the wetting of cellulose by water (see Figure 3). A way to probe the effect of these molecular-scale heterogeneities is to investigate the interaction of cellulose with molecules. We will consider the case of small molecules with sizes similar to those observed in the heterogeneities and the case of a protein, which have patches with hydrophobic and hydrophilic regions. These simulations are summarized in Table 2.

We will start by considering the interaction of molecular ions with cellulose. It is known that the interaction of ions with surfaces and interfaces is dominated by its hydration shell (which determines its hydrophobic or hydrophilic character) and the hydrophobic or hydrophilic character of the surface [48, 49, 50], giving the so-called Hoffmeister series (which classify ions with the same charge and different hydration).

We will consider here two extreme cases, carbonate ions (CO_3^{2-}) and tetraphenyl borate (Ph_4B^-) ions. Carbonate ions are strongly hydrophilic [48]. They have an interaction with water molecules stronger than the water-water interaction and they tend to retain their hydration shell [51]. Due to this strong interaction they tend to be repelled from the air/water interface [48] and attracted to hydrophilic surfaces. On the other hand, Ph_4B^- is a highly hydrophobic organic ion. It has a strong tendency to accumulate to lipophilic/hydrophobic surfaces, onto hydrophobic regions of polymers and at hydrophobic patches of proteins, as shown in several experimental and simulation studies [49, 50, 52, 53]. In

both cases, we include in the simulations Na^+ as a counterion which is a weakly hydrophilic ion [50].

Hence, our interest now is to determine how is the interaction of these ions with $\text{I}\beta$ cellulose, which according to our wetting simulations reported in the previous section, is both lipophilic and hydrophilic.

The results are shown in Figure 5 for carbonate and in Figure 6 for tetraphenyl borate. Both ions show a weak interaction with the (010) and the (100) plane of $\text{I}\beta$ cellulose.

Let us discuss first the results for the case of the carbonate ion and the (010) surface. As shown in Figure 5a, there is a depletion of this ion from the interface. The configuration of maximum approach of the ion at the interface is shown in Figure 5b. In this case, the maximum approach to the surface is made in a way that the two Na^+ counterions are adsorbed at the exposed -OH groups and the carbonate ion is in contact with these two adsorbed ions, making two ionic bridges with the surface. In the case of the (100) plane, we observe a similar result. As shown by the density profile (Figure 5c), there is a depletion of carbonate near the surface. The configuration of maximum approach is also illustrated in the snapshot of Figure 5d. The carbonate is not in direct contact with the surface, but rather there is a Na^+ counterion making a bridge between carbonate and the surface. This Na^+ counterions in contact with the surface, is in contact with one of the surface -OH groups (recall here that these -OH groups are not exposed to the interface in the crystal structure but modify their orientation in contact with water, see Figures 2d and 3d).

In the case of Ph_4B^- , we observe a weak attraction towards both surfaces, as indicated by an increased density profile near the surface Figure 6. The configuration of maximum approach between the Ph_4B^- ion and the surfaces is illustrated in the snapshots shown in Figure 6. In the case of the (010) surface, the orientation of the ion is influenced by the rugosity of the surface. This surface has "bumps" and also "holes" in which cellulose rings are exposed to water. The Ph_4B^- ion at the (010) surface is oriented in a way that corresponds to one of the phenyl rings inserted into one of these "holes" defined at the surface (see Figure 6b). In the case of the (100) surface, the configuration of maximum approach between the ion and the surface corresponds to one of the rings in contact with the surface (Figure 6d).

Therefore, the simultaneous hydrophilic and lipophilic behavior of the cellulose surface (with a smaller contact angle for the organic solvent), translates into a weak repulsion of hydrophilic ions and a weak attraction of hydrophobic ions.

Now, we consider the interaction of both cellulose surfaces with a protein (simulations S9a, S9b, S10a and S10b in Table 2). We considered the case of the human Epidermal Growth Factor protein (hEGF), which could be of interest in potential uses of cellulose for tissue repair. The protein structure employed in the simulations was obtained from solution NMR at neutral pH, deposited in the Protein Data Bank under PDB code 2KV4 [54] (see Appendix for further details). At neutral pH, the protein has a charge of -5e, according to the protonation states obtained using PropKa [55]. In the simulations, we

Figure 5: Cellulose interaction with carbonate ion. a) Atomic number density profile of carbonate (including all atoms) and Na^+ counter ions as a function of the distance to surface of cellulose plane (010). b) Snapshot of carbonate interacting with plane (010). Na^+ counter ions are shown in vdW representation. c) Atomic number density profile of carbonate (including all atoms) and Na^+ counter ions as a function of the distance to surface of cellulose plane (100). d) Snapshot of carbonate interacting with plane (100). Na^+ counter ions are shown in vdW representation. All density profiles for carbonate ion are calculated including all the atoms of the molecule. We include the atom density for cellulose to appreciate the proximity with the surface. We only included O and C atoms in this density calculation for cellulose and cellulose density was divided by a factor of 400 in order to appear in a similar scale.

consider 5 Na^+ counterions to neutralize the system.

The results (Figure 7) show that in both planes (010) and (100) the protein
 350 a some point in the simulation prefers to be at the cellulose surface rather than
 to be in solution. In our simulations, hEGF protein was placed at 0.7 nm of
 distance of both cellulose planes as starting configuration. We considered two
 different, opposite orientations *a* and *b*, indicated as a suffix in simulation names
 of Table 2. In orientation *a* the protein is oriented with its long axis parallel to
 the surface and with the residues ASN1 and LEU52 placed in front of the surface.
 355 In orientation *b* the protein is rotated 180° on the longitudinal direction such
 as ASN1 and LEU52 are opposite to the surface. During a short equilibration
 run of a few ns, the protein moved to contact with the surface in all simulations
 (S9a, S9b, S10a and S10b), exploring the surface and changing its orientation
 depending on the exposed crystal plane. Protein adsorption shows substantial

Figure 6: Cellulose interaction with tetraphenyl borate ion (Ph_4B^-). a) Atom density profile of Ph_4B^- as function to the distance to surface of cellulose plane (010). b) Snapshot of Ph_4B^- interacting with plane (010). c) Atom density profile of Ph_4B^- as function to the distance to surface of cellulose plane (100). d) Snapshot of Ph_4B^- interacting with plane (100). All density profiles for Ph_4B^- ion are calculated including all the atoms of the molecule. We include the atom density for cellulose to appreciate the proximity with the surface. We only included O and C atoms in cellulose density calculation and cellulose density was divided by a factor of 40 in order to appear in a similar scale.

360 differences between cellulose surface (010) and (100), as we will discuss next. At this point, we note that in ref [29] the authors identify an extended hydration layer that can preclude the approach of proteins (enzymes) to cellulose but we do not observe any evidence for this effect in our simulations.

Let us first discuss the results for simulation S9a and S9b (Figure 7a) corresponding to adsorption at the cellulose cellulose surface (010). In simulation S9a, hEGF protein binds at the very beginning of simulation (during the equilibration run) and remains adsorbed during all the production run, as seen in Figure 7a (black line). In simulation S9b (which considers an initial configuration of the protein rotated 180° with respect to S9a) the hEGF protein explores the surface before adsorbing, changing its relative orientation towards the surface. Finally, it adsorbs at the cellulose surface with the same orientation as obtained in simulation S9a and it remains adsorbed during 40 ns. The protein adsorption to cellulose plane (010) in both simulations S9a and S9b takes place through the same regions of the hEGF protein, which are two coils determined by the

Figure 7: a) Plot of minimum distance between hEGF protein and cellulose plane (010) vs simulation time for simulations S9a and S9b (see Table 2). Inset: Snapshot of simulation S9b with hEGF protein adsorbed at cellulose surface (010) at simulation time of 80 ns. The residues within 3.7 Å of the surface are indicated in the inset. b) Plot of minimum distance between hEGF protein and cellulose plane (100) vs simulation time for simulations S10a and S10b (see Table 2). Inset: Snapshot of simulation S10b with hEGF protein adsorbed at cellulose surface (100) at simulation time of 113 ns. The residues within 3.7 Å of the surface are indicated in the inset. The structure of the hEGF protein appears in cartoon representation and the protein surface was also indicated as obtained from the surface algorithm implemented in VMD [16].

375 aminoacids ASN1, SER2, ASP3, SER4, GLU5 and ILE23, GLU24, ALA25 and
 LEU26. At a given instant of time, we typically observe four of these aminoacids
 in contact with the cellulose surface, as shown in the inset of Figure 7a) for a
 particular time. We recall here that this list of aminoacids involved in protein
 adsorption at the cellulose surface include both polar aminoacids (that interact
 380 by hydrogen bonding) and nonpolar aminoacids (that interact by hydrophobic
 contacts). This is consistent with the results of the previous section that show
 a simultaneous hydrophilic and hydrophobic behaviour of this cellulose surface.
 The binding free energy associated with the interaction of these regions was es-
 timated by Adaptive Bias Force method (ABF), as described in the Appendix.
 385 We obtain a value of 30 Kcal/mol per each of the two above mentioned coil
 regions, which corresponds to an average of 15 Kcal/mol per adsorbed residue.

In the case of adsorption onto the cellulose surface (100) (simulation S10a
 and S10b in Table 2), the protein remains at the cellulose surface or close to the
 cellulose surface without binding to the cellulose surface during a substantial
 390 simulation time, as seen in Figure 7b. After exploring the surface, in simulation
 S10b we observe the adsorption of the protein onto the surface and the protein
 remains adsorbed until the end of the simulation. The contact of the protein
 with the cellulose plane is mostly due to the apolar LEU52, TRP50, LEU47
 and TYR44 residues, as shown in 7b). We also observe some configurations in
 395 which in addition to these residues, the protein also has one or two residues of the
 ASN1, SER2, ASP3, SER4, GLU5 tail adsorbed onto the cellulose surface. Our
 results show that the interaction of the hEGF protein with the (100) cellulose
 surface is mainly hydrophobic. These results are also consistent with previous
 results [30] which considered an extensive analysis of the adsorption of different
 400 aminoacids, dipeptides and tripeptides on plane (100) I β . In Ref [30] it was
 found that aminoacids with aromatic side chains are more prone to interact
 with plane (100) that have most of the hydrogen bond donors buried. We have
 estimated the free energy of adsorption of the hEGF protein to plane (100) (see
 Appendix for details), obtaining ≈ 150 Kcal/mol. This large value of free energy
 405 for interaction with plane (100) includes not only the interaction with LEU52,
 TRP50, LEU47 and TYR44 apolar residues but also the interaction of this
 cellulose plane with the tail formed by the residues ASN1, SER2, ASP3, SER4
 and GLU5 which tries to approach to the surface as we detach the hydrophobic
 residues during the ABF biased simulation.

410 Therefore, the particular surface of cellulose involved in the interaction plays
 an important role in protein adsorption, with the hEGF protein being able
 to bind to the cellulose surface by different regions depending on the exposed
 crystallographic plane.

5. Conclusion and Discussion

415 In this study we have analyzed interactions of different molecules with cel-
 lulose crystal planes (100) and (010) of cellulose I β . These cellulose plane have
 very different chemical structure since plane (100) have most of the hydrogen
 bond donor groups buried in the surface, while plane (010) have most of this

groups exposed. Other crystallographic planes will have an intermediate character between (100) and (010) so they will show an intermediate character in their interactions with solvents and molecules [24]. The conclusions obtained from our MD simulations of I β cellulose nanocrystals can be summarized as follows:

- The obtained contact angles for water and tetradecane onto (010) and (100) surfaces of I β cellulose indicate that both surfaces are simultaneously hydrophilic and lipophilic. This result is remarkable considering the very different hydrogen bonding capabilities of the two surfaces and their different exposure of hydrophobic groups at the surface.
- Wetting of the cellulose surface is strongly heterogeneous at the molecular scale in the case of both water and organic solvents. This complex surface behavior suggests the possibility of new and interesting applications for the use of nanocellulose materials which will deserve further modeling studies.
- Both hydrophilic and hydrophobic ions have a weak interaction with nanocellulose surfaces (although hydrophobic ions seem to have a somewhat stronger interaction with cellulose than hydrophilic ones), a result which can be attributed to the simultaneous hydrophilic and lipophilic behavior of cellulose surfaces.

The soluble hEGF protein binds to both the (010) and (100) cellulose surface through different regions of the protein which contain both polar and apolar residues. In the case of the (010) surface, the aminoacids involved in adsorption are mostly polar aminoacids whereas in the case of adsorption at the (100) surface most of the involved aminoacids are apolar. This result suggests that the simultaneous hydrophilic and hydrophobic character of cellulose induces a substantial interaction of cellulose with proteins (which contain both polar and apolar aminoacids available for interaction with substrates).

Finally, it is important to note that our conclusions are based on the study of a untreated crystal cellulose surface based on crystallographic data. But real nanocellulose based materials are expected to be even more complex. For example, measurements by different groups systematically indicate that cellulose is negatively charged under neutral pH, with an iso-electric point of 3.7 [43], a result completely unexpected from the chemical structure of an unmodified cellulose crystal. This is a clear indication of the presence of acid groups at the surface of cellulose. Measurements on pure cellobiose show that the molecule is neutral below a pH of about 10.5, in agreement with theoretical calculations from molecular structure [28]. It seems plausible that these acid groups present at cellulose surface are due to surface modifications on the cellulose nano-fibrils produced during its bio-synthesis. Some of the most common synthesis methods of cellulose nanoparticles are believed to induce the presence of surface groups such as sulfate ester groups (sulfuric acid hydrolysis), hydroxyl

groups (hydrochloric acid hydrolysis), acetyl groups (acetic acid hydrolysis), carboxylic acid groups (carboxymethylation route) and others [56]. Thus, modeling and simulation of the interactions of real cellulose materials with solvents and molecules requires to include these chemical surface groups in the model, which
 465 means that models should take into account the synthesis route of the cellulose. The appropriate way to modify present models (based on crystal structure) to include these surface chemical groups and its dependence on the synthesis route in the models is far from obvious, but it is clear that this should be the next step in theoretical studies involving the modeling of cellulose.

470 Acknowledgments

We acknowledge financial support from the Spanish Government through the MAT2015-64442-R grant and the Severo Ochoa Grant SEV-2015-0496 for Research Centres of Excellence awarded to ICMAB. D.C. Malaspina is supported by the European Union’s Horizon 2020 research and innovation programme
 475 under Marie Skłodowska-Curie grant agreement No 6655919. The simulations reported here were performed using the Edison Supercomputing facility of the National Energy Research Scientific Computing Center (NERSC), a U.S. Department of Energy Office of Science User Facility operated under Contract No. DE-AC02-05CH11231. Useful discussions with Prof. A. Roig and Dr A.
 480 Laromaine from the Nanoparticles and Nanocomposites Group at ICMAB are acknowledged.

Appendix: Simulation Methods

Molecular dynamics (MD) simulations were performed using NAMD 2.12 software [57]. All simulation snapshots were made using Visual Molecular Dynamics (VMD) software [16]. Cellulose crystal planes were build using the Cel-
 485 lulose builder toolkit [15] and described using the carbohydrate section of the CHARMM36 force field [37]. In all simulations, water was described employing the TIP4P/2005 model [40, 42] which best capture hydrogen-bonding features of liquid water at all pressures [41]. Tetraphenyl borate, tetradecane, carbonate
 490 and methane were described by CHARMM36 general force field (CGenFF) [38]. Human epidermal growth factor protein (hEGF) structure was obtained from the Protein Data Bank (PDB code:2KV4) [54]. The force field employed for the protein was the CHARMM36 force field protein section [58]. The composition, simulation box size, simulation time and other details for all simulated systems
 495 are described in Tables 1 and 2. In all simulations we employed periodic boundary conditions in all directions. The protocol followed in all simulations includes an initial minimization, equilibration and production runs. In all simulations we control temperature with a Langevin thermostat at 298 K using a damping coefficient of 1 ps^{-1} . Simulations of wetting by a solvent droplet (S1-S4 in Table
 500 1) were made in the NVT ensemble. Simulations of interaction of cellulose with molecules (s5-S10 in Table 2) were made in the NPT ensemble. The barostat

employed in NPT simulations was a Langevin piston kept at 1 atm with a piston period of 100 fs and piston decay of 50 fs. The pressure was applied in the direction of the water slab (z direction) using the anisotropic barostat option of NAMD. In these simulations, the dimensions of the simulation box in x and y directions was maintained constant. In all simulations Newton's equation of motion were integrated every 2 fs and electrostatic interactions updated every 4 fs. All bonds between heavy atoms and hydrogen atoms were maintained rigid using rigid bonds. Short range electrostatic and Lennard-Jones potential was computed with a cutoff of 1.2 nm and a switching distance of 1.0 nm. For long range electrostatic we used Particle Mesh Ewald (PME) algorithm using a grid spacing of 1.0 nm. In all simulations we added Na^+ ions to obtain electro-neutrality of the system. For systems S9 and S10 of Table 2, hEGF protein was placed initially at 0.7 nm from the cellulose surface during the equilibration run, but during the production run the protein is initially near the surface. For the calculation of density profiles in Figure 5 and in Figure 5 we implement our own Fortran code and for the 2-D water density of Figure 3 we implemented our own code in Python. The value of contact angle was approximated by superposition of different trial angles over the density profile of Figure 3. The minimum distance in Figure 7 was calculated using gmx mindist tool from GROMACS[59]. For simulations S9b and S10b we have performed free-energy calculations using the Adaptive Biasing Force (ABF) technique as implemented in NAMD [60]. In these biased MD simulations we obtain the potential of mean force (PMF) for the interaction of hEGF from cellulose. We selected as a reaction coordinate the z -axis projection of the distance between the center of mass of the cellulose surface and the center of mass of a group of protein residues involved in the protein adsorption. The force constant employed in the ABF calculation was 10 kcal mol⁻¹ Å⁻¹ and the PMF was obtained with 0.1 Å resolution. In the case of the PMF calculation for the S9B simulation, we start from a configuration with an initially adsorbed protein over the cellulose surface (as obtained from simulation S9B). We performed the ABF simulation by considering as generalized coordinate the center of mass distance between cellulose and the aminoacids 23, 24, 25 and 26 of the protein aminoacid sequence, which correspond to one of the two coils of the protein initially adsorbed to cellulose. Once these aminoacids were detached from the surface, we performed a second ABF simulation in which the generalized coordinate was the center of mass distance between cellulose and the aminoacids 1, 2, 3 and 4, which corresponds to the other coil of the protein that adsorbs over cellulose. In this way, the protein was fully detached from the surface. In the case of the PMF calculation for the S10B simulation, we considered as generalized coordinate the center of mass distance between cellulose and the aminoacids 43, 44, 47, 50, 52, 15 and 8 of the protein aminoacid sequence, which correspond to the region of the protein initially adsorbed to cellulose. In this case, we find that the convergence of the ABF simulations to obtain a well defined PMF were difficult, so we decided to perform two different, complementary ABF simulations. In the first one, we started from a configuration with an initially adsorbed protein over the cellulose surface, as obtained from simulation S10B. In the second ABF simulation, we reversed the previous ABF simulation,

starting from an initially desorbed protein and ending with the protein adsorbed onto cellulose. The two simulations produced almost identical PMF values.

550 References

- [1] S. U. Wisseman, W. S. Williams, *Ancient Technologies and Archaeological Materials*, Taylor & Francis, 2013.
URL <https://books.google.es/books?id=BXkuAgAAQBAJ>
- [2] D. Klemm, E. D. Cranston, D. Fischer, M. Gama, S. A. Kedzior, D. Kralisch, F. Kramer, T. Kondo, T. Lindström, S. Nietzsche, K. Petzold-Welcke, F. Rauchfuß, *Nanocellulose as a natural source for groundbreaking applications in materials science: Today's state*, *Materials Today* 21 (7) (2018) 720–748. doi:10.1016/j.mattod.2018.02.001.
URL <http://dx.doi.org/10.1016/j.mattod.2018.02.001>
- [3] M. Zeng, A. Laromaine, W. Feng, P. A. Levkin, A. Roig, *Origami magnetic cellulose: controlled magnetic fraction and patterning of flexible bacterial cellulose*, *J. Mater. Chem. C* 2 (31) (2014) 6312–6318. doi:10.1039/c4tc00787e.
URL <http://dx.doi.org/10.1039/c4tc00787e>
- [4] A. Laromaine, T. Tronser, I. Pini, S. Parets, P. A. Levkin, A. Roig, *Free-standing three-dimensional hollow bacterial cellulose structures with controlled geometry via patterned superhydrophobic/hydrophilic surfaces*, *Soft Matter* 14 (19) (2018) 3955–3962. doi:10.1039/c8sm00112j.
URL <http://dx.doi.org/10.1039/c8sm00112j>
- [5] S. Ling, D. L. Kaplan, M. J. Buehler, *Nanofibrils in nature and materials engineering*, *Nature Reviews Materials* 3 (4) (2018) 18016+. doi:10.1038/natrevmats.2018.16.
URL <http://dx.doi.org/10.1038/natrevmats.2018.16>
- [6] R. J. Moon, A. Martini, J. Nairn, J. Simonsen, J. Youngblood, *Cellulose nanomaterials review: structure, properties and nanocomposites*, *Chemical Society Reviews* 40 (7) (2011) 3941–3994. doi:10.1039/c0cs00108b.
URL <http://dx.doi.org/10.1039/c0cs00108b>
- [7] B. Lindman, *From surfactant to cellulose and DNA self-assembly. A 50-year journey*, *Colloid and Polymer Science* 294 (11) (2016) 1687–1703. doi:10.1007/s00396-016-3927-2.
URL <http://dx.doi.org/10.1007/s00396-016-3927-2>
- [8] M. Martínez-Sanz, M. J. Gidley, E. P. Gilbert, *Hierarchical architecture of bacterial cellulose and composite plant cell wall polysaccharide hydrogels using small angle neutron scattering*, *Soft Matter* 12 (5) (2016) 1534–1549. doi:10.1039/c5sm02085a.
URL <http://dx.doi.org/10.1039/c5sm02085a>

- [9] T. Heinze, O. A. El Seoud, A. Koschella, Cellulose derivatives : synthesis, structure, and properties, Springer Series on Polymer and Composite Materials, 2018.
590 URL <http://www.worldcat.org/isbn/9783319731681>
- [10] B. Medronho, B. Lindman, Brief overview on cellulose dissolution/regeneration interactions and mechanisms, *Advances in Colloid and Interface Science* 222 (2015) 502–508. doi:10.1016/j.cis.2014.05.004.
URL <http://dx.doi.org/10.1016/j.cis.2014.05.004>
- 595 [11] L. Alves, B. F. Medronho, F. E. Antunes, A. Romano, M. G. Miguel, B. Lindman, On the role of hydrophobic interactions in cellulose dissolution and regeneration: Colloidal aggregates and molecular solutions, *Colloids and Surfaces A: Physicochemical and Engineering Aspects* 483 (2015) 257–263. doi:10.1016/j.colsurfa.2015.03.011.
600 URL <http://dx.doi.org/10.1016/j.colsurfa.2015.03.011>
- [12] L. Alves, B. Medronho, F. Antunes, D. Topgaard, B. Lindman, Dissolution state of cellulose in aqueous systems. 1. Alkaline solvents, *Cellulose* 23 (1) (2016) 247–258. doi:10.1007/s10570-015-0809-6.
URL <http://dx.doi.org/10.1007/s10570-015-0809-6>
- 605 [13] W. Glasser, R. Atalla, J. Blackwell, R. Malcolm Brown, W. Burchard, A. French, D. Klemm, Y. Nishiyama, About the structure of cellulose: debating the Lindman hypothesis, *Cellulose* 19 (3) (2012) 589–598. doi:10.1007/s10570-012-9691-7.
URL <http://dx.doi.org/10.1007/s10570-012-9691-7>
- 610 [14] S.-Y. Ding, M. E. Himmel, The Maize Primary Cell Wall Microfibril: A New Model Derived from Direct Visualization, *Journal of Agricultural and Food Chemistry* 54 (3) (2006) 597–606. doi:10.1021/jf051851z.
URL <http://dx.doi.org/10.1021/jf051851z>
- [15] T. C. F. Gomes, M. S. Skaf, Cellulose-Builder: A toolkit for building crystalline structures of cellulose, *Journal of Computational Chemistry* 33 (14) (2012) 1338–1346. doi:10.1002/jcc.22959.
615 URL <http://dx.doi.org/10.1002/jcc.22959>
- [16] W. Humphrey, A. Dalke, K. Schulten, VMD: Visual molecular dynamics, *Journal of Molecular Graphics* 14 (1) (1996) 33–38. doi:10.1016/0263-7855(96)00018-5.
620 URL [http://dx.doi.org/10.1016/0263-7855\(96\)00018-5](http://dx.doi.org/10.1016/0263-7855(96)00018-5)
- [17] D. Frenkel, B. Smit, *Understanding Molecular Simulation*, 2nd Edition, Academic Press, Inc., Orlando, FL, USA, 2001.
- [18] J. H. Jensen, *Molecular modeling basics*, CRC Press, 2010.
625 URL <http://www.worldcat.org/isbn/9781420075267>

- [19] H. Miyamoto, U. Schnupf, M. F. Crowley, J. W. Brady, Comparison of the simulations of cellulosic crystals with three carbohydrate force fields, *Carbohydrate Research* 422 (2016) 17–23. doi:10.1016/j.carres.2016.01.001.
 630 URL <http://dx.doi.org/10.1016/j.carres.2016.01.001>
- [20] S. Roig-Sanchez, E. Jungstedt, I. Anton-Sales, D. C. Malaspina, J. Faraudo, L. A. Berglund, A. Laromaine, A. Roig, Nanocellulose films with multiple functional nanoparticles in confined spatial distribution, *Nanoscale Horizons* doi:10.1039/c8nh00310f.
 635 URL <http://dx.doi.org/10.1039/c8nh00310f>
- [21] C. A. López, G. Bellesia, A. Redondo, P. Langan, S. P. S. Chundawat, B. E. Dale, S. J. Marrink, S. Gnanakaran, MARTINI Coarse-Grained Model for Crystalline Cellulose Microfibers, *J. Phys. Chem. B* 119 (2) (2015) 465–473. doi:10.1021/jp5105938.
 640 URL <http://dx.doi.org/10.1021/jp5105938>
- [22] A. Poma, M. Chwastyk, M. Cieplak, Coarse-grained model of the native cellulose I α and the transformation pathways to the I β allomorph, *Cellulose* 23 (3) (2016) 1573–1591. doi:10.1007/s10570-016-0903-4.
 URL <http://dx.doi.org/10.1007/s10570-016-0903-4>
- 645 [23] Y. Nishiyama, G. P. Johnson, A. D. French, V. T. Forsyth, P. Langan, Neutron Crystallography, Molecular Dynamics, and Quantum Mechanics Studies of the Nature of Hydrogen Bonding in Cellulose I , *Biomacromolecules* 9 (11) (2008) 3133–3140. doi:10.1021/bm800726v.
 URL <http://dx.doi.org/10.1021/bm800726v>
- 650 [24] K. Mazeau, On the external morphology of native cellulose microfibrils, *Carbohydrate Polymers* 84 (1) (2011) 524–532. doi:10.1016/j.carbpol.2010.12.016.
 URL <http://dx.doi.org/10.1016/j.carbpol.2010.12.016>
- 655 [25] M. Bergensträhle, L. A. Berglund, K. Mazeau, Thermal Response in Crystalline I Cellulose: A Molecular Dynamics Study, *J. Phys. Chem. B* 111 (30) (2007) 9138–9145. doi:10.1021/jp072258i.
 URL <http://dx.doi.org/10.1021/jp072258i>
- [26] J. F. Matthews, G. T. Beckham, M. Bergensträhle-Wohlert, J. W. Brady, M. E. Himmel, M. F. Crowley, Comparison of Cellulose I Simulations with Three Carbohydrate Force Fields, *Journal of Chemical Theory and Computation* 8 (2) (2012) 735–748. doi:10.1021/ct2007692.
 660 URL <http://dx.doi.org/10.1021/ct2007692>
- [27] M. Bergensträhle, J. Wohlert, M. E. Himmel, J. W. Brady, Simulation studies of the insolubility of cellulose, *Carbohydrate Research* 345 (14) (2010) 2060–2066. doi:10.1016/j.carres.2010.06.017.
 665 URL <http://dx.doi.org/10.1016/j.carres.2010.06.017>

- [28] E. Bialik, B. Stenqvist, Y. Fang, A. Östlund, I. Furó, B. Lindman, M. Lund, D. Bernin, Ionization of Cellobiose in Aqueous Alkali and the Mechanism of Cellulose Dissolution, *J. Phys. Chem. Lett.* 7 (24) (2016) 5044–5048. doi:10.1021/acs.jpcllett.6b02346. URL <http://dx.doi.org/10.1021/acs.jpcllett.6b02346>
- [29] J. F. Matthews, C. E. Skopec, P. E. Mason, P. Zuccato, R. W. Torget, J. Sugiyama, M. E. Himmel, J. W. Brady, Computer simulation studies of microcrystalline cellulose I, *Carbohydrate Research* 341 (1) (2006) 138–152. doi:10.1016/j.carres.2005.09.028. URL <http://dx.doi.org/10.1016/j.carres.2005.09.028>
- [30] G. Nawrocki, P.-A. Cazade, D. Thompson, M. Cieplak, Peptide Recognition Capabilities of Cellulose in Molecular Dynamics Simulations, *The Journal of Physical Chemistry C* 119 (43) (2015) 24404–24416. doi:10.1021/acs.jpcc.5b07118. URL <http://dx.doi.org/10.1021/acs.jpcc.5b07118>
- [31] K. Mazeau, A. Rivet, Wetting the (110) and (100) Surfaces of I Cellulose Studied by Molecular Dynamics, *Biomacromolecules* 9 (4) (2008) 1352–1354. doi:10.1021/bm7013872. URL <http://dx.doi.org/10.1021/bm7013872>
- [32] R. J. Maurer, A. F. Sax, V. Ribitsch, Molecular simulation of surface reorganization and wetting in crystalline cellulose I and II, *Cellulose* 20 (1) (2013) 25–42. doi:10.1007/s10570-012-9835-9. URL <http://dx.doi.org/10.1007/s10570-012-9835-9>
- [33] H. Miyamoto, D. M. Rein, K. Ueda, C. Yamane, Y. Cohen, Molecular dynamics simulation of cellulose-coated oil-in-water emulsions, *Cellulose* 24 (7) (2017) 2699–2711. doi:10.1007/s10570-017-1290-1. URL <http://dx.doi.org/10.1007/s10570-017-1290-1>
- [34] M. Bergensträhle, E. Thormann, N. Nordgren, L. A. Berglund, Force Pulling of Single Cellulose Chains at the Crystalline CelluloseLiquid Interface: A Molecular Dynamics Study, *Langmuir* 25 (8) (2009) 4635–4642. doi:10.1021/la803915c. URL <http://dx.doi.org/10.1021/la803915c>
- [35] P. Chen, Y. Nishiyama, J. Wohler, A. Lu, K. Mazeau, A. E. Ismail, Translational Entropy and Dispersion Energy Jointly Drive the Adsorption of Urea to Cellulose, *The Journal of Physical Chemistry B* 121 (10) (2017) 2244–2251. doi:10.1021/acs.jpcc.6b11914. URL <http://dx.doi.org/10.1021/acs.jpcc.6b11914>
- [36] R. Alqus, S. J. Eichhorn, R. A. Bryce, Molecular Dynamics of Cellulose Amphiphilicity at the GrapheneWater Interface, *Biomacromolecules* 16 (6) (2015) 1771–1783. doi:10.1021/acs.biomac.5b00307. URL <http://dx.doi.org/10.1021/acs.biomac.5b00307>

- [37] O. Guvench, S. S. Mallajosyula, E. P. Raman, E. Hatcher, K. Vanommeslaeghe, T. J. Foster, F. W. Jamison, A. D. MacKerell, CHARMM Additive All-Atom Force Field for Carbohydrate Derivatives and Its Utility in Polysaccharide and CarbohydrateProtein Modeling, *Journal of Chemical Theory and Computation* 7 (10) (2011) 3162–3180. doi:10.1021/ct200328p.
URL <http://dx.doi.org/10.1021/ct200328p>
- [38] K. Vanommeslaeghe, E. Hatcher, C. Acharya, S. Kundu, S. Zhong, J. Shim, E. Darian, O. Guvench, P. Lopes, I. Vorobyov, A. D. Mackerell, CHARMM general force field: A force field for drug-like molecules compatible with the CHARMM all-atom additive biological force fields, *Journal of Computational Chemistry* 31 (4) (2009) NA. doi:10.1002/jcc.21367.
URL <http://dx.doi.org/10.1002/jcc.21367>
- [39] A. D. MacKerell, D. Bashford, M. Bellott, R. L. Dunbrack, J. D. Evanseck, M. J. Field, S. Fischer, J. Gao, H. Guo, S. Ha, D. Joseph-McCarthy, L. Kuchnir, K. Kuczera, F. T. K. Lau, C. Mattos, S. Michnick, T. Ngo, D. T. Nguyen, B. Prodhom, W. E. Reiher, B. Roux, M. Schlenkrich, J. C. Smith, R. Stote, J. Straub, M. Watanabe, J. Wiórkiewicz-Kuczera, D. Yin, M. Karplus, All-Atom Empirical Potential for Molecular Modeling and Dynamics Studies of Proteins †, *The Journal of Physical Chemistry B* 102 (18) (1998) 3586–3616. doi:10.1021/jp973084f.
URL <http://dx.doi.org/10.1021/jp973084f>
- [40] J. L. F. Abascal, C. Vega, A general purpose model for the condensed phases of water: TIP4P/2005, *The Journal of Chemical Physics* 123 (23) (2005) 234505+. doi:10.1063/1.2121687.
URL <http://dx.doi.org/10.1063/1.2121687>
- [41] C. Calero, J. Martí, E. Guàrdia, 1H Nuclear Spin Relaxation of Liquid Water from Molecular Dynamics Simulations, *J. Phys. Chem. B* 119 (5) (2015) 1966–1973. doi:10.1021/jp510013q.
URL <http://dx.doi.org/10.1021/jp510013q>
- [42] C. Vega, J. L. F. Abascal, Simulating water with rigid non-polarizable models: a general perspective, *Physical Chemistry Chemical Physics* 13 (44) (2011) 19663–19688. doi:10.1039/c1cp22168j.
URL <http://dx.doi.org/10.1039/c1cp22168j>
- [43] K.-Y. Lee, F. Quero, J. Blaker, C. Hill, S. Eichhorn, A. Bismarck, Surface only modification of bacterial cellulose nanofibres with organic acids, *Cellulose* 18 (3) (2011) 595–605. doi:10.1007/s10570-011-9525-z.
URL <http://dx.doi.org/10.1007/s10570-011-9525-z>
- [44] G. Morandi, L. Heath, W. Thielemans, Cellulose Nanocrystals Grafted with Polystyrene Chains through Surface-Initiated Atom Transfer Radical Polymerization (SI-ATRP), *Langmuir* 25 (14) (2009) 8280–8286. doi:10.

1021/1a900452a.

750 URL <http://dx.doi.org/10.1021/1a900452a>

- [45] M. Abdelmouleh, S. Boufi, M. N. Belgacem, A. P. Duarte, A. Ben Salah, A. Gandini, Modification of cellulosic fibres with functionalised silanes: development of surface properties, *International Journal of Adhesion and Adhesives* 24 (1) (2004) 43–54. doi:10.1016/s0143-7496(03)00099-x.
755 URL [http://dx.doi.org/10.1016/s0143-7496\(03\)00099-x](http://dx.doi.org/10.1016/s0143-7496(03)00099-x)
- [46] G. I. Mantanis, R. A. Young, Wetting of wood, *Wood Science and Technology* 31 (5) (1997) 339–353. doi:10.1007/bf01159153.
URL <http://dx.doi.org/10.1007/bf01159153>
- [47] I. Kalashnikova, H. Bizot, B. Cathala, I. Capron, Modulation of Cellulose Nanocrystals Amphiphilic Properties to Stabilize Oil/Water Interface, *Biomacromolecules* 13 (1) (2012) 267–275. doi:10.1021/bm201599j.
760 URL <http://dx.doi.org/10.1021/bm201599j>
- [48] A. P. dos Santos, A. Diehl, Y. Levin, Surface Tensions, Surface Potentials, and the Hofmeister Series of Electrolyte Solutions, *Langmuir* 26 (13) (2010) 10778–10783. doi:10.1021/1a100604k.
765 URL <http://dx.doi.org/10.1021/1a100604k>
- [49] C. Calero, J. Faraudo, D. Bastos-González, Interaction of monovalent ions with hydrophobic and hydrophilic colloids: Charge inversion and ionic specificity, *Journal of the American Chemical Society* 133 (38) (2011) 15025–15035.
770
- [50] D. Bastos-González, L. Pérez-Fuentes, C. Drummond, J. Faraudo, Ions at interfaces: the central role of hydration and hydrophobicity, *Current Opinion in Colloid & Interface Science* 23 (2016) 19–28.
- [51] S. Yadav, A. Chandra, Structural and Dynamical Nature of Hydration Shells of the Carbonate Ion in Water: An Ab Initio Molecular Dynamics Study, *The Journal of Physical Chemistry B* 122 (4) (2018) 1495–1504. doi:10.1021/acs.jpcb.7b11636.
775 URL <http://dx.doi.org/10.1021/acs.jpcb.7b11636>
- [52] L. Pérez-Fuentes, C. Drummond, J. Faraudo, D. Bastos-González, Anions make the difference: insights from the interaction of big cations and anions with poly (N-isopropylacrylamide) chains and microgels, *Soft Matter* 11 (25) (2015) 5077–5086.
780
- [53] L. Pérez-Fuentes, C. Drummond, J. Faraudo, D. Bastos-González, Interaction of organic ions with proteins, *Soft matter* 13 (6) (2017) 1120–1131.
- [54] H.-W. Huang, S. K. Mohan, C. Yu, The NMR solution structure of human epidermal growth factor (hEGF) at physiological pH and its interactions with suramin, *Biochemical and Biophysical Research Communications* 402 (4) (2010) 705–710. doi:10.1016/j.bbrc.2010.10.089.
785 URL <http://dx.doi.org/10.1016/j.bbrc.2010.10.089>

- 790 [55] H. Li, A. D. Robertson, J. H. Jensen, Very fast empirical prediction and
rationalization of protein pKa values, *Proteins: Structure, Function, and
Bioinformatics* 61 (4) (2005) 704–721. doi:10.1002/prot.20660.
URL <http://dx.doi.org/10.1002/prot.20660>
- [56] A. Dufresne, *Nanocellulose: From Nature to High Performance Tailored
Materials*, De Gruyter textbook, De Gruyter, 2012.
795 URL <https://www.degruyter.com/viewbooktoc/product/129215>
- [57] J. C. Phillips, R. Braun, W. Wang, J. Gumbart, E. Tajkhorshid, E. Villa,
C. Chipot, R. D. Skeel, L. Kalé, K. Schulten, Scalable molecular dynamics
with NAMD, *Journal of Computational Chemistry* 26 (16) (2005) 1781–
800 1802. doi:10.1002/jcc.20289.
URL <http://dx.doi.org/10.1002/jcc.20289>
- [58] R. B. Best, X. Zhu, J. Shim, P. E. M. Lopes, J. Mittal, M. Feig, A. D. MacK-
erell, Optimization of the Additive CHARMM All-Atom Protein Force
Field Targeting Improved Sampling of the Backbone , and Side-Chain
805 1 and 2 Dihedral Angles, *Journal of Chemical Theory and Computation*
8 (9) (2012) 3257–3273. doi:10.1021/ct300400x.
URL <http://dx.doi.org/10.1021/ct300400x>
- [59] D. Van Der Spoel, E. Lindahl, B. Hess, G. Groenhof, A. E. Mark, H. J. C.
Berendsen, GROMACS: Fast, flexible, and free, *Journal of Computational
810 Chemistry* 26 (16) (2005) 1701–1718. doi:10.1002/jcc.20291.
URL <http://dx.doi.org/10.1002/jcc.20291>
- [60] J. Henin, G. Fiorin, C. Chipot, M. L. Klein, Exploring Multidimensional
Free Energy Landscapes Using Time-Dependent Biases on Collective Vari-
ables, *Journal of Chemical Theory and Computation* 6 (1) (2010) 35–47.
815 doi:10.1021/ct9004432.
URL <http://dx.doi.org/10.1021/ct9004432>