Settling of Bentonite Particles in Gelatin Solutions for Stickwater Treatment

Rashid M. Shamsuddin\textsuperscript{a,b,*}, Casparus J.R. Verbeek\textsuperscript{a} and Mark C. Lay\textsuperscript{a}

\textsuperscript{*}School of Engineering, Faculty of Science and Engineering, University of Waikato, Private Bag 3105, Hamilton 3240, New Zealand
\textsuperscript{a}Chemical Engineering Department, Faculty of Engineering, Universiti Teknologi Petronas, 32610 Bandar Seri Iskandar, Malaysia

Abstract

The settling of calcium and sodium bentonite coated with adsorbed protein from gelatin solutions was investigated across pH 3 to 9. The particle settling or sedimentation was achieved at acidic environment regardless of the amount of protein adsorbed by the particles. The settling is attributed to the formation of “house of cards” structure by bentonite platelets and it is strongly influenced by solution pH. The collapse of this formation resulted in floc formation or particle suspension where no settling can be observed. Few possible settling mechanisms were proposed based on the charge balance and interactions between bentonite platelets and protein molecules in the system. The findings are intended for treatment of gelatin-rich stickwater (meat rendering waste water) using bentonite as low-cost adsorbent. Integrating adsorption and settling into one unit operation has multiple benefits; reducing BOD by removing the proteins and bentonite is directly modified with organics and can be recovered via gravity settling for composite additive application.

Keywords: Settling; sedimentation; bentonite; gelatin; stickwater; house of card

1. Introduction

Bentonite platelets consist of silica surfaces (permanently negatively charged) and alumina edges (pH-dependent charge). The negative charge accounts for 95 \% of the total surface area and is balanced out by cationic adsorption [1, 2]. In solution, the dissociation of alumina and silica elements as well as the hydroxyl (OH) groups at the edge determines the overall charge. Aluminium has point of zero charge (PZC) at pH 7 and on the clay lattice, contributing toward the overall negative charge. At low pH, aluminium and silica are positively charged and OH\textsuperscript{2-} is formed on the surface. This amphoteric character makes bentonite a versatile adsorbent material.

Bentonite is commonly used in adsorption as well as being added into composites to improve mechanical properties. Current research regarding bentonite is limited to adsorption or composite development only. As an adsorbent, it has shown great success in various protein recoveries [1, 3-5], wine clarification [6, 7], heavy metals [8, 9] and dye pigments removals [10, 11]. As a bio-composite filler, bentonite has to be modified with organics to make it compatible with the matrix materials [12]. Modification with amine, bovine serum albumin (BSA), gelatin, alkylammonium and starch could be expensive. Stickwater on the other hand is a waste water from the meat rendering industry containing mostly degraded gelatin.

* Corresponding author. Tel.:+60-5-368-7590; fax:+60-5-365-6176.
E-mail address: mrashid.shamsuddin@petronas.com.my
In recent work, we have examined protein recovery from gelatin [13] and stickwater [14] solutions using raw and modified bentonite. At the highest, about 60% of the protein in stickwater was adsorbed onto the clay which corresponds to an organic content of 30-35 dry wt%. Surface adsorption as well as intercalation were observed in gelatin and stickwater adsorption. Through this approach, not only stickwater is partially treated, but the bentonite particles are organically and economically modified to increase its compatibility as filler in biomatrix polymers such as in bloodmeal-based plastic called Novatein thermoplastic protein (NTP). Previously, we found that stickwater-modified sodium bentonite improved the tensile strength of NTP composites by 23 % at only 0.5 parts per hundred bloodmeal (pphBM) dosage [14], compared to octadecylamine-modified sodium bentonite that gave 20 % increase at 2 pphBM [15].

For solid-liquid separation, gravity settling or sedimentation is widely studied as separation process due to its simplicity. The study of sedimentation at solid-liquid interface has always been a challenge in science especially when the system becomes more complicated. Unlike the settling of solid in gas, there are various possible interactions between solid and liquid phases that complex the sedimentation [16, 17]. Sedimentation can potentially be integrated into adsorption process as a 2-in-1 waste water treatment- adsorbing contaminants as well as recovering the spent adsorbent. However, the research fields of sedimentation and adsorption have remained separated.

Presently, we attempted to find the ideal condition at which bentonite could result in settling in gelatin solution (model stickwater) after protein adsorption process. Two commonly available bentonite; calcium (CaBt) and sodium (NaBt) types were investigated with gelatin solutions under pH 3, 5.2 (PZC of gelatin), 7 and 9 for overnight settling. Sedimentation was assessed by turbidity measurement for clay concentration in solution and cryogenic scanning electron microscopy (cryo-SEM) for morphological study in semi-liquid environment. Finally, possible particle settling mechanisms in gelatin solution are discussed.

2. Experimental

2.1 Materials and equipment

The list of materials and equipment used is presented in Table 1.

<table>
<thead>
<tr>
<th>Materials</th>
<th>Supplier</th>
<th>Grade</th>
<th>Additional notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adsorption Calcium and sodium bentonite</td>
<td>Transform Minerals</td>
<td>Industrial</td>
<td>Volcanic origin</td>
</tr>
<tr>
<td>Gelatin</td>
<td>Davis Gelatin</td>
<td>Food</td>
<td></td>
</tr>
<tr>
<td>Chemicals Na₂HPO₄(s), NaH₂PO₄(s), NaCl(s), HCl(aq) and NaOH(s)</td>
<td>Ajax Finechem</td>
<td>Analytical</td>
<td>Phosphate buffer ingredients and pH adjusters</td>
</tr>
<tr>
<td>Equipment</td>
<td>Manufacturer and model</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Centrifuge</td>
<td>Eppendorf- Minispin Plas</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fine balance</td>
<td>Sartorius- CP225D</td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH meter</td>
<td>Eutech Instruments- CyberScan 100</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SEM</td>
<td>Hitachi- S-4700 with cryogenic attachment</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Settling column</td>
<td>Waikato Glass Blower</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Turbidity meter</td>
<td>Hach- 2100P</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water bath</td>
<td>Grant- GD120</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water pump</td>
<td>Little Giant Pump Co.- 2E-38NY</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

2.2 Adsorption and Settling

The adsorption of protein from gelatin solution using calcium and sodium bentonite has been reported in detail elsewhere [13] and was repeated at higher volume for settling study. Gelatin solutions at 20 mg/mL (2 %) were prepared by dissolving gelatin in 1200 ml of 0.02 M phosphate buffer solution (PBS). Calcium and sodium bentonite were added at 0.03 g/ml concentration or 36 g in 1200 ml of solution. The resulting mixtures were mixed using magnetic stirrers for one hour to reach equilibrium. Equilibrium solution was transferred into a settling column (Fig. 1A) and placed in a holding tank with heated water circulation (Fig. 1B). A water bath was used to keep circulating water in the holding tank at 37°C. Samples of 5 ml gelatin solution were withdrawn at 10, 30, 60, 90, 120 minutes and overnight at five outlet points using separate syringes (Fig. 1A). The samples were analyzed for turbidity readings which were then converted into clay concentration using the standard curves of “clay turbidity vs. clay dosages” prepared earlier.
2.3 Cryogenic Scanning Electron Microscopy

Equilibrium solutions of gelatin adsorption with calcium and sodium bentonite at pH 3 were centrifuged at minimum speed and time to increase the pellet viscosity. This enabled the samples to be transferred onto the standard SEM sample holders. The samples were then instantly frozen using liquid nitrogen and transferred into the SEM vacuum chamber. They were sputter-coated with platinum and the SEM was operated at 5 to 20 kV. Liquid nitrogen was continuously supplied to the cryogenic system.

3. Results and Discussion

3.1 Particle Settling in Gelatin Solution

Experimental results on settling experiments are shown in Fig. 2. Settling patterns suggest that the process is strongly influenced by solution pH. Calcium bentonite gave good settling at pH 3, minimal settling at pH 5.2 and did not settle at pH 7 and 9. No settling was observed for NaBt at all conditions. Conditions with settling shows the clay concentrations in solutions decreased over time across all outlets while there was little or no change in clay concentration for unsettled conditions. The former resulted in pellet formation at the bottom of the tank while floc or particle suspension was observed for unsettled conditions.
Fig. 2: Bentonite settling in gelatin at pH A) 3, B) 5.2, C) 7 and D) 9. Datapoints are mean values of three replicates. The lines are included to show the settling trends.

The settling behaviour of bentonite platelets at different pH is attributed to the “house of card” structure [7]. In solution, the bentonite layers swell or even exfoliate because of water uptake. At decreasing pH, the alumina edge is becoming more positively charged resulting in an “edge-face” interaction (between Al-edge and Si-surface), leading to a “house of cards” structure (Fig. 3). This structure is hypothesized for gelation of smectites at low pH and low electrolyte concentration. The formation is largely influenced by electrostatic forces and in an aqueous solution, the gel can be formed at as low as 2 wt% clay dispersion [18, 19].

As the pH increases, the edge charge shifts from positive to negative, causing the edge-face electrostatic interaction to weaken. The house of card formation eventually collapses into the original forms of platelet stacks at pH above the PZC of bentonite (Fig. 4) [18]. Thus, the tendency of the solid phase to stay in suspension increased at higher pH. At unsettling conditions, the platelets were covered with gelatin molecules and formed a stable suspension network of particles-gelatin as revealed by cryo-SEM (discussed in later section).
Apart from pH, the valency of the ions is important because the coagulating power of an ion increases rapidly with its valency for example Ca$^{2+}$ coagulates more than Na$^+$. The Ca$^{2+}$ can easily bridge two counterions from gelatin molecules compared to just one for Na$^+$. This is shown by polyacrylamide (PAM) adsorption using bentonite in water where PAM had a flocculation effect with CaBt and deflocculation effect with NaBt. Therefore, application of CaBt for settling process could be advantageous over NaBt due to bridging flocculation by Ca$^{2+}$ compared to Na$^+$.

3.2 Surface Morphology

As viewed under cryo-SEM (Fig. 5), the gel is a three-dimensional network of clay platelets and gelatin molecules. It is also appeared that NaBt particles exfoliated while CaBt particles swelled in solution. Gelatin molecules appear to have enveloped the platelets and acted as connecting bridges or crosslinks between particles. Calcium bentonite seemed to form more compact pellet with less moisture trapped in the particle-gelatin network while sodium bentonite showed stable suspensions with particles were connected by long gelatin chains. These observations are consistent with the settling results where NaBt did not settle due to particle exfoliation and high degree of particle crosslinking. Calcium bentonite on the other hand, agglomerated in gelatin solution into denser pellet and settled to the bottom at lower pH. This is attributed to both gelatin molecules and platelets were positively charged at pH 3, thus the particles had higher tendency for edge-face bonds compared to bonding between clay particles to gelatin. Therefore, more “house of cards” structures were formed at lower pH and settled due to weight.
4. Conclusion

Calcium bentonite gave good settling in gelatin adsorption at pH 3 and minimal settling at pH 5.2 while sodium bentonite did not result in any settling across pH 3 to 9. The ability of particle settling is mainly dependent on solution pH and the type of adsorbent rather than the amount of adsorbed protein. Bentonite settling is attributed to the formation of “house of card” structure in gelatin solution while floc formation resulted from platelets exfoliation. Microscopy analysis corroborated the above findings by depicting the morphological states of bentonite platelets in gelatin solution. At pH 3, NaBt particles were exfoliated while CaBt showed formations of platelet stacks or agglomerates which resulted in settling. For stickwater treatment, if maximum protein recovery is the priority, adsorption can be performed at pH 9 for CaBt and pH 5.2 for NaBt [13]. However, the use of CaBt at lower pH might be advantageous for subsequent recovery process due to its settling ability. Nonetheless, it has been shown that protein adsorption can occur at any pH conditions due to the amphoteric characteristics of gelatin and bentonite [13, 20, 22].

Acknowledgements

The authors would like to thank Transform Minerals for the bentonite and Ms. Helen Turner of Waikato University for the cryo-SEM assistance.

References


