Shear structuring as a new method to make anisotropic structures from soy–gluten blends

Katarzyna J. Grabowska, Stavroula Tekidou, Remko M. Boom, Atze-Jan van der Goot *

Food Process Engineering Laboratory, Wageningen University, Bornse weilanden 9, 6708 WG Wageningen, The Netherlands

Abstract

The concept of shear-induced structuring was applied to concentrated blends of soy protein isolate (SPI) and wheat gluten (WG) to create novel semi-solid food textures. Concurrent simple shear deformation and heating (95 °C) of the protein blends generated original structures consisting of fibers or layers. The ratio of SPI to vital WG and the final concentration determined the morphology of the structure. It is hypothesized that the spatial distribution of the SPI-rich phase and the WG-rich phase in a blend was altered by the shear flow. When both phases became aligned horizontally in the shear cell, a fibrous structure was formed; when they became aligned vertically in the shear cell, a layered structure was formed. The structures obtained were analyzed visually and using texture analysis and scanning electron microscopy.

© 2014 Elsevier Ltd. All rights reserved.

1. Introduction

A sustainable alternative to intensive meat production is the development of plant protein products with a fibrous and meat-like texture (Boland et al., 2013). Formation of such anisotropic structure from a concentrated protein blend involves reorganization of the initial structure followed by fixation of the newly formed morphology. Traditional techniques that are used to structure protein (spinning, extrusion, and mixing) already succeed in formation of highly anisotropic morphologies but they have certain limitations. Spinning requires pure protein fraction and very low concentration while extrusion needs high dry matter concentration (above 60 wt.%) (Noguchi, 1989; van Hoof, Seegers, Corsten, & Neirynck, 2008). Besides, spinning causes large wastewater streams from the coagulation and washing bath, which contain large amounts of salts and organic matter. Extrusion puts limitations on product shape, although it delivers fully fibrillized protein (Liang, Huff, & Hsieh, 2002). A mixing process, as described in a patent by van Hoof et al. (2008), requires additional polysaccharide-like alginates to generate small fibrous elements.

In this article, we present a novel technique to create anisotropic, plant-based food structures. This shear-induced process has already been shown to fibrillize calcium caseinate on multiple length scales (Manski, van der Goot, & Boom, 2007). The structure formation depended on the specific properties of calcium caseinate: formation of micelles and mild attractions between those micelles (Grabowska, van der Goot, & Boom, 2012; Sprakel, Spruijt, Stuart, Michels, & van der Gucht, 2009). Nevertheless, the encouraging results obtained with calcium caseinate led us to the research aim to create an anisotropic plant-based structure. For this purpose, we used blends consisting of a commercial SPI and WG with different concentrations (20–40 wt.%) and ratios of soy protein isolate (SPI) and wheat gluten (WG) (1:4, 2:3, 3:2, 4:1). The structures obtained were analyzed visually and using texture analysis and scanning electron microscopy (SEM).

2. Materials and methods

2.1. Materials

Commercial SPI and vital WG were used to produce the structures. The SPI (Supro 500E IP) is a powder containing at least 90 wt.% (N × 6.25) proteins, maximum 5 wt.% ash, and maximum 6 wt.% moisture as stated in the manufacturer’s specifications (Solae, St Louis, MO). The water-holding capacity (WHC) was assessed using the centrifugation method described by Chou and Morr (1976), and was found to be 8.9 g water per 1 g dry SPI powder.

Vital WG, produced by Roquette (Lestrem, France), contains at least 83 wt.% (N × 6.25) protein based on the dry matter and 8% moisture, according to the manufacturer's specifications. The WHC of WG was 1.9 g water per 1 g powder as tested with the centrifugation method described by Chou and Morr (1976).

Food-grade sodium chloride (NaCl), dimethyl sulfoxide (DMSO), glutaraldehyde, and ethanol (96% purity) used in the sample preparation for
SEM analysis were obtained from Sigma-Aldrich (Zwijndrecht, The Netherlands).

2.2. Methods

2.2.1. WHC

The WHC was tested using the centrifugation method described by Chou and Morr (1976). A 10 wt.% SPI dispersion in demineralized water was prepared using a magnetic stirrer and vortexed for 1–2 min. Afterwards, the proteins were kept at ambient temperature for 30 min. The dispersion was then centrifuged (Allegra X22R, Beckman Coulter) at 2200 rpm and 20 °C for 30 min. The supernatant and the pellet were separated and weighed (Sartorius AG MA30, Göttingen, Germany). The total dry matter content in the pellet was measured with an infrared moisture analyzer (Sartorius AG, MA30). The WHC was expressed in grams of water per gram of dry matter (g water/g DM).

2.2.2. Differential scanning calorimetry

Thermographs of SPI, WG, and an SPI-WG blend were recorded with a differential scanning calorimeter (DSC) (Perkin Elmer Diamond; PerkinElmer Co., Norwalk, CT). The instrument was calibrated using indium and gallium. The temperature of the sample was increased from 20 °C to 200 °C using a heating rate of 10 °C/min. An empty aluminum pan was used as a reference. Measurements were taken in duplicate and the results were analyzed using Pyris software (PerkinElmer Co., Norwalk, CT).

2.2.3. Structure formation process

2.2.3.1. Preparation of the protein blend. Protein blends of 20–40 wt.% concentration were prepared in demineralized water with 1 wt.% of NaCl added. First, the SPI was pre-moisturized at a room temperature for 30 min by thorough mixing of the SPI in the NaCl solution. The pre-moisturized SPI was combined and mixed with WG before the structuring process. The weight ratio between the SPI and WG was varied as presented in Table 1.

2.2.3.2. Structuring process. The protein blends were subjected to simple shear at a temperature of 95 °C. The design of the cone-cylinder shear device used (referred to as the shearing device) was based on previously reported equipment (Habeych, Dekkers, van der Goot, & Boom, 2008; Manski, van der Zalm, van der Goot, & Boom, 2007; Peighambaroud, Hamer, Boom, & van der Goot, 2008; van der Zalm, van der Goot, & Boom, 2009), but upgraded for use at higher temperatures (above 100 °C) by the addition of an improved closing system that prevents water evaporation. Shear was applied by rotating the bottom cone of the device. The temperature was controlled by circulating oil (Julabo Oil Bath, PrestoPlus LBH5). The blends were exposed to a constant shear of 30 rpm for 15 min in the preheated (95 °C) shearing cell. After shearing, the sample was cooled under quiescent conditions to 4 °C within 30 min before performing further analysis. The composition of all blends is shown in Table 1.

2.2.4. Large deformation tests

The mechanical properties of the samples were analyzed using a texture analyzer (Instron Testing System, type 5564, load cell 2000 N). The samples were cut in two ways: parallel to the shear flow and perpendicular to the shear flow applied during the structuring process. These are referred to as parallel cross-section and perpendicular cross-section, respectively. The breaking behavior of the cross-sections was tested in triplicate. A pre-cut, bone-shaped mold was used to make samples (length 25.0 mm, width 4.4 mm, thickness 1–3 mm). The thickness was verified for every sample individually and included in further calculations. The surfaces of the grips were roughened with abrasive tape to prevent slipping of the sample. The measurement was conducted at room temperature with a crosshead speed of 1 mm/s. Blue Hill software (Instron) was used to calculate the tensile fracture stress \( \sigma \) (kPa) and the tensile fracture strain \( \varepsilon \) (mm/mm) at the break point. The tensile stress \( \sigma \) (kPa) and the tensile strain \( \varepsilon \) (mm/mm) were used to estimate the stress and strain-based anisotropy index (AI) according to the following equations:

\[
AI_\parallel = \frac{\tau_\parallel}{\tau_\perp}
\]

where \( \tau_\parallel \) and \( \tau_\perp \) represent the tensile stress parallel and perpendicular to the shear flow, respectively, and

\[
AI_{\circ} = \frac{\sigma_\parallel}{\sigma_\perp}
\]

where \( \sigma_\parallel \) and \( \sigma_\perp \) are the tensile strains parallel and the perpendicular to the shear flow, respectively.

The results showed a large standard deviation, especially when analyzing materials containing with a higher concentration that gave fibrous materials (30–40 wt.%). This is related to the characteristic fibrous nature of the materials tested.

2.2.5. SEM

To visualize the morphology of the structure, a dried sample was prepared according to the method described by Muller, van Aelst, Humbel, van der Krift, and Boekhout (2000) and Nijsse and van Aelst (1999). The structure was cut into a small trapezium (the longest edge 8 mm) and stored in glacial acetic acid in demineralized water (3% v/v) overnight at 4 °C. The next day, the sample was immersed in a series of solutions of dimethyl sulfoxide (DMSO) (15%, 30%, and 50%, v/v) within 60 min. Slow freezing of the sample was executed in cold gaseous nitrogen. The samples were then immersed in liquid nitrogen and freeze fractured with a razor blade. The freeze fracturing was done parallel and perpendicular to the shear flow exerted on the sample. After fracturing, the sample was immersed in a series of DMSO solutions within 60 min (50%, 30%, and 15% v/v). The sample was then washed twice in demineralized water within 40 min. The sample was dehydrated within 80 min in a graded series of ethanol solutions (10%, 30%, 50%, 70%, v/v). The sample was then kept overnight in a 70% v/v ethanol solution. The next day, the sample was submersed in 90% v/v and 100% v/v ethanol before critical point drying with carbon dioxide (CPD 020, Balzers, Liechtenstein). The sample was then glued onto a sample holder using conductive carbon cement, and sputter coated with platinum. The fractured surface was analyzed with the Magellan SEM.

3. Results

The application of simple shear flow (denoted as shearing) to SPI, WG and SPI-WG blends resulted in a variety of structures (Table 2). Heating was sufficient to form a solid solid structure, whereas the combination of heating and shearing was required to form an anisotropic

<table>
<thead>
<tr>
<th>Table 1</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Concentration</th>
<th>40 wt.%</th>
<th>35 wt.%</th>
<th>30 wt.%</th>
<th>25 wt.%</th>
<th>20 wt.%</th>
</tr>
</thead>
<tbody>
<tr>
<td>40 wt.%</td>
<td>24 + 16</td>
<td>16 + 24</td>
<td>8 + 32</td>
<td>0 + 40</td>
<td></td>
</tr>
<tr>
<td>35 wt.%</td>
<td>21 + 14</td>
<td>14 + 21</td>
<td>7 + 28</td>
<td>0 + 35</td>
<td></td>
</tr>
<tr>
<td>30 wt.%</td>
<td>24 + 6</td>
<td>18 + 12</td>
<td>6 + 24</td>
<td>0 + 30</td>
<td></td>
</tr>
<tr>
<td>25 wt.%</td>
<td>20 + 5</td>
<td>15 + 10</td>
<td>10 + 15</td>
<td>5 + 20*</td>
<td></td>
</tr>
<tr>
<td>20 wt.%</td>
<td>16 + 4</td>
<td>16 + 4</td>
<td>16 + 4</td>
<td>16 + 4</td>
<td></td>
</tr>
<tr>
<td>5:0</td>
<td>3:2</td>
<td>2:3</td>
<td>1:4</td>
<td>0:5</td>
<td></td>
</tr>
</tbody>
</table>
structure. We found that the SPI alone did not give anisotropy, whereas WG gave a pronounced fibrous structure, in agreement with earlier findings (Hansen, Johnston, & Ferrel, 1975; Wieser, 2007). Without shearing, WG gave randomly oriented small fibers. The combination of SPI and WG gave an aligned structure containing horizontally oriented fibers that were thicker and longer than the randomly oriented fibers found in the non-sheared WG sample.

### 3.1. Structure formation of SPI

A minimum concentration of SPI is necessary for the formation of a (soft solid) gel (Hermansson, 1978; Renkema, 2001; Wang & Damodaran, 1991). That is why different concentrations of SPI were investigated (12.5–40 wt.%). Below 25 wt.%, an isotropic material was formed. The blends with a concentration greater than 25 wt.% generated anisotropic structures, which consisted of densely packed vertical layers. Above 35 wt.%, incomplete hydration was obtained leading to a non-coherent material that broke when shearing. Processing blends with an even higher concentration (40–45 wt.%), was impossible due to the lack of plasticity caused by high deficiency of water. Those experiments, therefore, suggest that SPI was not able to form a fibrous material, which is in line with studies describing the use of extrusion (Cheftel, 1986; Cheftel, Kitagawa, & Quequiner, 1992). They did not obtain an anisotropic structure, nor did they report on the presence of a layered material as found after shear-induced structuring.

Samples containing 25%, 30%, and 35% SPI were analyzed for their mechanical properties (Fig. 1). There is no indication of anisotropy, because the tensile stress and strain were the same for the parallel and the perpendicular samples. The measurements became less reproducible at the highest concentration (30 wt.%), which indicates a larger inhomogeneity of the matrix. This is consistent with the finding that the 30 wt.% material crumbled and no longer formed a coherent matrix.

#### 3.1.1. Microstructure

The morphology of a 30 wt.% SPI structure visualized with SEM is presented in Fig. 2. The parallel cross-section shows oblong flattened pores (Fig. 2b), and the perpendicular cross-section showed round pores (Fig. 2a). The pores, most likely created through water evaporation during sample preparation, seemed to be affected by the shear flow, although this sign of anisotropy is not reflected in the mechanical properties as described earlier (Fig. 1).

On a smaller length scale, the sheared SPI shows coexistence of two different protein networks: one made of packed globular domains (cauliflower morphology; Fig. 3a) and a second made of string-like domains (spider web morphology; Fig. 3b). The presence of both morphologies is typical for gel formation of globular proteins (Doi, 1993; Hermansson, 1994; KinSELLA, 1976; THANH & Shibasaki, 1978). In addition, pieces of cell wall remaining after the refining process can be seen (Fig. 3a, lower right quadrant).

### 3.2. Structure formation with WG

To make a soft solid material from WG, a higher concentration of WG was necessary compared with SPI. A concentration below 30 wt.% led to the formation of an aqueous phase, due to the low WHC of WG (1.9 g water/1 g WG). WG consists of diverse proteins of which the glutenins...
and gliadins are the most important. The elastomeric proteins of WG, particularly the high molecular weight glutenin subunits, are able to aggregate (Laszity, 1984; Song & Parkinson, 2012) on hydrating and mixing (Day, Augustin, Batey, & Wrigley, 2006). It was observed that WG forms fibers spontaneously on mixing with water (Zaidel, Chin, Rahman, & Karim, 2008). We obtained a truly fibrous structure from a 30 wt.% WG blend when WG was sheared with 30 rpm at 95 °C for 30 min. The fibers were organized parallel to the shear flow applied.

The formation of a fibrous structure seems to lower the water-holding properties because a certain amount of water was expelled from the WG on processing. Shearing the WG blends with a higher concentration (35 and 40 wt.%) resulted in a layered structure (a so-called lamellar structure). Some fibers could be distinguished, although it cannot be excluded that those fibers were obtained through tearing of a layer during visual observations. From the results, it was concluded that WG can be used to form fibrous materials but only in a very narrow range of concentration.

Fig. 4 shows the mechanical properties of the structure formed with WG. The tensile stress of the parallel sample was three times larger than the tensile stress of the perpendicular sample, which confirms the anisotropy of the material. The tensile strain measurement did not show pronounced anisotropy. The increase in strength along the fiber direction could be due to stretching of the fibers (accompanied by strain hardening), whereas the weaker interactions between the bundles of the fibers give a lower perpendicular strength. With increasing protein concentration, the differences in parallel and perpendicular strength diminished, indicating that the bonds responsible for the material strength are more randomly distributed at higher concentrations. It is somewhat surprising that the overall (average) strength of the sample is not strongly dependent on the concentration. At higher concentration, the material becomes inhomogeneous and is not completely hydrated. The disappearance of the anisotropy in the case of WG was associated with a change in structure from fibers to layers (Fig. 4).

3.3. Structure formation in SPI–WG blends

Shearing SPI–WG blends gave a variety of anisotropic structures, ranging from layers to fibers (Fig. 5). The morphology depended on the SPI/WG ratio and the dry matter concentration.

DSC analysis of an SPI-WG blend suggests that SPI and WG form separate domains. Fig. 6 shows the changes in heat flow for each sample. WG undergoes a transition at about 70 °C, which is not disturbed by the addition of SPI. The SPI used did not show any specific first-order transitions on heating and therefore is expected to be fully denatured during the isolation process (among others due to thermal drying). The existence of the separate domains may be important for the generation of the fibrous morphology.

As stated, the structure obtained depended on the total dry matter content and the SPI/WG ratio. The 20% SPI–WG blends did not absorb all the water, especially at higher WG content. Most blends with a concentration of 20 wt.% (except the 4:1 ratio) resulted in a fragile and porous structure. Although fibers were present in these structures, they were weak and randomly distributed, similar to the fibers in the non-
sheared WG structures (Table 2). Most likely, these fibers were obtained spontaneously at the first contact with water during mixing. The samples produced with 20 wt.% protein content broke during handling before the texture analysis could be completed, except the sample with the highest proportion of SPI (4:1). In that material, similar values for the tensile parameters for both orientations were found, indicating lack of anisotropy (Figs. 7 and 8). Increasing the concentration to 25% gave structures containing horizontally distributed fibers, and thus anisotropy, for all SPI/WG ratios. The SPI and WG phases could be distinguished based on differences in their surface porosity as shown by SEM. They were aligned alternately resulting in a lamellar morphology. A higher WG content led to a weaker structure, but with a more fibrous character.

The mechanical test of the structures with a ratio of 4:1 and 3:2 confirmed their anisotropic character (Figs. 8 and 9). The sample with an SPI/WG ratio of 2:3 did not show anisotropy in the mechanical test, confirming the visual observation that no fibers were present. The sample with the highest WG content was too weak for mechanical analysis.

A further increase in the concentration to 30% led to more distinct alignment and a compact structure. Strong and well-pronounced horizontal fibers were formed from blends with an SPI/WG ratio of 4:1 and 3:2. The blend converted into layers (lamellar structure) at the highest WG concentration (Fig. 5). When tearing such a layered structure, fibers became visible.

The increase in the SPI–WG concentration to 30% resulted in a number of interesting observations. As expected, the samples were stronger due to the higher concentration (Figs. 7 and 8). The materials with ratios of 4:1, 3:2 and 1:4 were anisotropic in the expected manner given that the material was stronger parallel to the shear flow. The highest tensile stress and strain were recorded for the 2:3 sample. This sample consisted of well-pronounced vertical layers and therefore no difference in the mechanical properties of the specimens cut along and across the layer was expected. A further increase in WG content (1:4) reduced the strength but resulted in a fibrous material again.

We further analyzed a fibrous sample (4:1) using SEM microscopy. Two phases were visible, especially when cut along the shear flow direction (Fig. 9). A compact gel with large pores was surrounded by a second phase consisting of highly structured fibrous domains (Fig. 9). The latter is probably WG, because it is well known for its network-forming properties and in this sample it is present at a lower concentration. A perpendicular cross-section of the same sample shows very dissimilar morphology, which is in line with the anisotropy (Fig. 10): spherical
air bubbles and domains surrounded by another phase with different porosity.

The porosity of the sample is a dominant feature. Fig. 10 shows spherical domains that have been partly fused together; in between the domains, there is a smaller porous phase. It is possible that the SPI-rich phases surrounded by WG fibers on a macro scale are experienced as thick fibers. That would explain why both thin (most likely WG) and thick (possibly strings of SPI domains) fibers were observed.

A further increase in the dry matter content to 35 wt.% or even 40 wt.% led to samples in which not all the material was fully hydrated, especially at high SPI content. At these concentrations, mostly layered structures were formed, although the 35 wt.% sample made with a ratio of 3:2 had strong anisotropic properties (Figs. 7 and 8). The materials were weaker than the samples at lower concentrations because of incomplete hydration. At 40% only, the sample with the highest WG content resulted in a fully hydrated and stronger gel with a certain anisotropy. The SPI–WG sample with a 2:3 ratio showed unexpected high anisotropy of 3.5 based on the tensile stress measurements.

As stated, most samples formed layers at higher concentrations. The layered morphology was confirmed for the sample with a 1:4 ratio by SEM imaging. The sample was prepared using two different techniques, brittle fracture above liquid nitrogen (Fig. 11a) and a smooth cut with a razor (Fig. 11b), in order to capture different aspects of the microstructure. The layered character of the sample was clearly visible by fracturing above the nitrogen, whereas the smoothly cut sample showed gaps between both phases and adhesion between the phases, most likely due to local deformation on cutting (Fig. 11b). The aligned domains in Fig. 11a are the edges of the layers. A single layer had a thickness of 5–8 μm. The same sample cut smoothly showed the porosity of the WG phase (main fraction) and the more globular domains of SPI proteins (minor fraction). This indicates that the layers consisting of WG are separated by SPI protein domains; thus WG must be the continuous phase and SPI is the dispersed phase (Fig. 11b).

4. Discussion

The use of well-defined deformation has a great potential as an alternative structuring method to produce new anisotropic materials consisting of SPI and WG. Almost all SPI–WG samples were anisotropic after processing, which was visible by eye on a macro scale, although the mechanical test did not always confirm the anisotropy.
We believe that the mechanism of the structure formation is based on phase separation between SPI and WG under mild shear flow, similarly to the previous study (Wolf, Scirocco, Frith, & Norton, 2000). It was shown that shear stress affects the shape of particles formed in a phase-separated liquid–liquid biopolymer mixture processed in a cone-plate rheometer and fixated through gelation of one or both phases. Changes in shear stress generated different extents of anisotropy.

Here, on shearing, the domains are deformed and broken up, and ultimately align in the flow direction for most of the samples. Without shear, the two phases are deformed less and are oriented randomly.
leading to macroscopically isotropic structures. These findings are in line with studies by Tolstoguzov (1993), who pointed out that incompatible biopolymers should be mixed to form anisotropy by extrusion. A similar relation was found in a study on shear-induced structuring of starch–zein blends (Habeych, van der Goot, & Boom, 2009; Habeych et al., 2008). In order to deform the material by the shear forces, a certain viscosity of the biopolymer is required. High viscosity allows the transfer of the stresses onto the individual phases in the material, which leads to a minimum concentration necessary to obtain an anisotropic structure.

A prerequisite for preserving the anisotropic structure is the simultaneous occurrence of deformation and solidification. Here, we used the thermal gelation by heating above 50–60 °C (Damodaran & Kinsella, 1982; Kinsella, 1979; Morales & Kokini, 1999; Singh & MacRitchie, 2004) as a solidification step.

To further understand the structure formation process, the WHC of both proteins must be considered. The WHC of SPI is much higher than the WHC of WG. Hermansson (1972) observed that the swelling of SPI particles in a dispersion resulted in an increase in the dispersion viscosity, which was confirmed by Anón, Sorgentini, and Wagner (2001). We observed that the rate of water absorption is much higher in the case of WG. Therefore, mixing water and a dry powder blend of SPI and WG resulted in inhomogeneous water distribution in the blend; much of the water is initially in the WG phase. Blends with more WG required higher concentrations than the blends with a higher proportion of SPI, which is probably related to the lower overall blend viscosity.

Because WG forms a more deformable phase than SPI, the WG phase is probably the continuous phase in most of the blends, thus making the SPI act as filler. The continuous WG phase forms fiber-like structures spontaneously (Kieffer, 2007; Wieser, 2007), which can be seen as fibers, or may act as a binder between strings of SPI domains, giving coarser fibers. The tensile analysis confirmed our hypothesis for the continuous phase. The tensile stress of the samples made of SPI–WG blends at a concentration of 30 wt.% (Fig. 7) is in same range of tensile stress values as the 30 wt.% WG samples oriented perpendicularly (Fig. 4a). The parallel WG cross-section is much stronger than the perpendicular cross-section, which might be caused by the strain hardening properties of WG fibers (Fig. 4). Similar strain hardening was shown by Peighambardoust, van der Goot, van Vliet, Hamer, and Boom (2006) after shearing of wheat dough. The WG and SPI phases had a weak interaction, which is indicated by the gaps between the phases observed in the SEM pictures (Fig. 11b). In the case of good compatibility, these gaps would not be visible (Habeych et al., 2008). This implies that the presence of the SPI phase, dispersed in the WG network, leads to a weakening of the overall structure. This is in line with a study on starch–zein blends, in which it was hypothesized that incompatibility between the starch and zein allowed the creation of anisotropic products but weakened, whereas the addition of a compatibilizer resulted in the formation of a more homogeneous and stronger material.

In the SPI–WG system, we observed differences in anisotropy. When the protein phases became aligned horizontally in the shear cell, a fibrous structure was formed, but when the phases were aligned vertically in the shear cell, a layered structure was formed. A layered structure is expected to have similar mechanical strength along the two dimensions of the lamellae, and thus one would not expect a difference in mechanical strength when the lamellae are oriented perpendicularly. The SPI–WG blends at ratios of 2:3 and 1:4, except for the blend with 40 wt.% protein overall did not show any mechanical anisotropy. It was previously shown in the literature that SPI inhibits WG aggregation (Ribotta, Arnulphi, León, & Anón, 2005). A high concentration of WG results in coalescence of protein fibers into a layer or film formation (Schofield, Bottomley, Timms, & Booth, 1983): reducing the WG concentration by introducing a second compound (e.g., SPI) to the blend inhibits the merging into films and allows fiber formation.

Overall, the creation of a fibrous structure requires a careful balance between the blend properties and the process conditions. Too much formation of fibers results in the creation of films (lamellae), whereas too little does not result in a consistent gel.

5. Conclusions

Anisotropic semi-solids can be prepared using blends of SPI and WG proteins at high moisture content (>60%), when the blend is subjected to well-defined shear flow at moderate temperatures. The natural functionality of plant proteins to gelate on heating was used to fixate the structure during the structuring process.

The structure formation mechanism is based on the existence of separate phases of two incompatible biopolymers: SPI and WG proteins. The two phases are deformed and aligned by the shear flow, leading to the formation of aligned zones and layered and, with a careful balance between the various phases, even fibrous material.

The morphologies found may be relevant as a basis for the development of plant-based meat analogs with an anisotropic texture.

Acknowledgments

This study was supported financially by ISPT (P10003) and the PEAS Foundation (The Netherlands). We are grateful to the following people: Erik Smiths (experimental help), Jacqueline Donkers (for taking the
SEM pictures), Herman de Beukelaer (assisting with the DSC analysis), Jos Sewalt (technical help), André Sanders, Hans de Rooij, Hans Meijer, Mees Schimmel, and Reinoud Hummelen (technical help). Barentz (Hoofddorp, The Netherlands) kindly donated raw materials.

References


