The Secret of Chia Gel Revealed - Nanoscale 3D Network Retards and Prolongs Food Release

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ABSTRACT

Chia gels have been associated with numerous health benefits over centuries, like curbing diabetes and obesity by retarding digestion rate and quenching craving for more food. However, the exact mode of gelation has always been quite ambiguous; the usual explanation often taken as the mucilage's swelling in or coagulation of water. Gel making is our passion and expertise. Our working hypothesis is that chia seed mucilage forms extended fibrous structures in water to ultimately form a 3D network at nanoscale. We are interested to probe the mechanism of gelation in many ways including microscopic techniques. Our preliminary studies entailed gelation tests, which revealed the polar nature of the mucilage. Subsequently, we systematically performed microscopic studies using optical microscope and scanning electron microscope. The optical micrographs revealed fiber bundles at microscale level while the SEM micrograph revealed a 3D network of nanoscale fibers with diameters around 50 nm, confirming our hypothesis. This breakthrough would surely revolutionize how we envision gelation process of Chia and other gel forming seeds and materials.

Keywords: chia gel, chia gel 3D network, chia nano fibers, chia digestion, chia slow food release

1. INTRODUCTION

Chia (Salvia hispanica) seed was native to the valleys of Mexico and northern Guatemala. Its uses included offering to the gods and painting of the face and body. Ancient people like the Mayans consumed it to prolong duration of fullness during war and other tasks. Chia is regarded as "super food" and contains nutritional and health benefits like antioxidants, omega-3 and -6, fibers, vitamins, and minerals.[1-4]

Unlike most other seeds, it has the unique property of gelling water. According to Munoz [5], its mucilage has water retaining and gelling properties with applications such as food thickening and emulsion stabilization. The mucilage, comprising mainly polysaccharides on the seed surface, swells and extends in water, trapping food particles (like sugars) and, thus, retarding digestion in the body.[5-6]

We aimed to understand the fundamental science underlying how chia seeds form 3D network that leads to gel formation. Despite the current notion that gel formation is merely due to the swelling and coagulation in water by the seed's mucilage, our hypothesis is that the mucilage of chia contains nanoscale fibers that extend in water to form 3D network, which entraps and "solidifies" water to form a gel.

We began by deducing the effect of various parameters like solvent polarity on gelation in order to discern conditions that influence network formation by the mucilage. Optical microscopy zoomed in and elucidated how the fibers grew around the seed in water. Finally, scanning electron microscopy was used to further zoom in and study the nature of fiber configuration and size in the network at nanoscale. This knowledge will be vital for future advancements in materials design and medicine, especially to provide the general public with convincing information to best adhere to effective health styles and to prevent conditions like diabetes and obesity.[7-9]

2. MATERIALS

Chia seeds were obtained from Costco, NJ. Unless otherwise stated, all the solvents including hexane, ethanol and distilled water were obtained from Sigma-Aldrich. Canola oil was obtained from Costco, NJ. Congo red dye and Methylene blue dye obtained from Sigma-Aldrich.

3. METHODS

3.1 Effects of Different Parameters on Gelling Efficiency

NOTE: Gelation was confirmed by observing no flow of the water/seed mixture due to gravity upon inverting vial upside down; swelling was confirmed when the seeds swelled instead of remaining the same or shrinking.

(A) EFFECT OF SEED CONCENTRATION: Different concentrations (4, 5, 6, 8, 10, 15 & 20% g/mL) of chia seed in deionized water were prepared. Each mixture was allowed time to gel and the minimum gelation concentration (MGC) capable of gelling water determined.

(B) EFFECT OF SOLVENT POLARITY: The MGC was used to prepare mixtures of chia in 100, 50, and 0 % water in water/ethanol mixture respectively.

(C) EFFECT OF MODE OF ADDITION OF COMPONENTS OF THE 50:50 SOLVENT MIXTURE: The MGC was used to prepare mixtures by changing the order of adding components in 50:50 water/ethanol.

(D) EFFECT OF HYDROPHOBIC SOLVENTS: Each of hexane and canola oil was added to chia based on MGC.

3.2 Imaging Fiber Growth

Prior to each imaging, the fibers were grown by gelling or swelling in water. For SEM, the water was completely removed while the fibers were extended by lyophilization.

(A) OPTICAL MICROSCOPY: The extruding fibers of chia seeds in water were viewed using the Nickon TiE optical microscope. The samples were prepared by adding Congo red or methylene blue dye to the seeds in water in 6x4 multiwell plate wells. The plate was loaded onto the stage, and the images focused at different magnifications and satisfactory images captured by an attached camera.

(B) SCANNING ELECTRON MICROSCOPY (SEM): The lyophilized, dry fibers were used for SEM imaging at statndard conditions using a scanning electron microscope (ZEISS Supra 55 VP). The SEM was operated at various magnifications to capture satisfactory SEM images.

4. RESULTS AND DISCUSSION

4.1 Results of Effects of Different Parameters

The results of the effects of various parameters on gelation are shown in Tables 1 - 4.

Table 1. Effect of chia seed concentration on gelation

Concentration (% g/mL)	4	5	6	8	10	15	20
Gelation Result	N	N	N	N	N	G	G

G = gel formed; N = gel not formed.

Table 2. Effect of water/ethanol ratio on gelation

% of water in mixture	100	50	0
Gelation Result	G	N	N

G = gel formed; N = gel not formed.

Table 3. Effect of mode of adding 50:50 water/ethanol on gelation or swelling

Mode of addition	H_2O 1st	H_2O last	together
Gelation Result	SW	N	N

SW = seed swollen in solution; N = not swollen.

Table 4. Effect of hydrophobicity on gelation or swelling

Hydrophobic solvent	hexane	canola oil
Gelation Result	N	N

SW = seed swollen in solution; N = not swollen.

Based on the above results, the ideal conditions for chia seed gelation are a minimum of 15% g/mL seed concentration (Table 1) and a polar, aqueous solvent, preferably 100% water (Tables 2 to 4). This observation may be attributed to the gelling agent on the seed's surface being polar, thus, resulting in a more favorable interaction with water compared to with less polar solvents systems.

4.2 Results of Imaging Fiber Growth

Optical Microscopy

A photograph of chia seed gel and optical micrographs of grown chia seed fibers are shown in Figure 1. Chia seeds formed a gel which was verified by inverting the vial containing the gel upside down (Figure 1A); no flow despite the pull of gravity proved a gel was formed. The evenly spaced out seeds in the gel led us to believe that there must some sort of fibers responsible for separating the seeds apart as well as entrapping the water molecules.

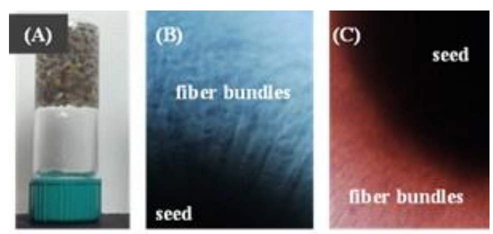

Figure 1. (A) Image of chia seed gel, inverted to verify a gel formation. (B & C) Optical micrograph of chia seed fiber bundles in water - dyed with Methylene blue (B) & Congo red (C)

To some extent, optical microscope gave some evidence that there were some kind of fibers covering the seed surface (Figure 1B & C). These figures show bundles of extended fibers of part of a chia seed soaked in water and dyed with Methylene Blue (Figure 1B) and Congo Red (Figure 1C). However, the difficulty to see distinct individual fibers still remained.

Electron Microscopy

Figure 2 shows SEM micrographs obtained using the ZEISS Supra 55 VP electron microscope. The SEM analyses resulted in an enhanced means of zooming in more to deduce the nature of the fibers covering the surface of the seeds. The samples were prepared by forming chia seed hydrogel, which was then lyophilized in order to carefully void the gel of water content while leaving the fibers in their extended state. The extremely dried, lyophilized samples were viewed under the SEM. The SEM imaging corroborated our hypothesis; observed was an extensive network of nanoscale fibers extended from proximate seeds to all directions (Figure 2). The magnification was ~1,000X. Furthermore, higher magnification (~30,000X) revealed nanoscale fibers with a diameters around 50 nm.

This discovery is a breakthrough towards understanding the fundamental science and the hypothesis that chia seed must form some sort of 3D network of fibers at nanoscale to induce gel formation in water.

This study has a major implication to science. Gelation has always been associated with the coagulation or solidification of a liquid solvent. The mechanism of gelation using small synthesized molecules, or even refined natural macromolecules, has been studied and somewhat quite understood to proceed via the interaction of nanoscale fibers to form a 3D network. [10-11] However, on the contrary, the mode of gelation using unrefined natural materials such as whole seeds like chia has been simply taken for granted as undergoing “coagulation”, “swelling”, etc. without a thorough investigation or understanding of the mechanism. Consequently, for the first time, our discovery that these whole seeds form such elegant 3D structures at nanoscale level will inevitably shed more light on the process of 3D network formation and gelation. This would surely provide the opportunity to study and hopefully mimic the efficiency of nature (biomimicry) in order to develop more efficient gels and nano softmaterials, as well as pave way to discovery more gel-forming seeds.

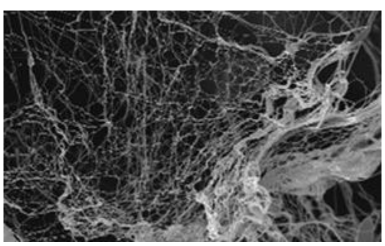

Figure 2. SEM micrograph of Chia seed fibers: 1000X magnification.

5. FUTURE DIRECTION

We plan to perform do more microscopic studies using transmission electron microscopy (TEM) to further elucidate the nature of the nanoscale network. We are also plan to study and confirm the effect of the nanoscale 3D network seed fibers on the hydrolysis of starch by salivary amylase (α-amylase).

6. CONCLUSION

Our studies have revealed that the gelation of chia seed is due to nanoscale fibers with diameters around 50 nm that formed a nanoscale 3D network. Based on the results, these fibers, covering seed surface, are most likely polar moieties that extends in polar water. These results would surely revolutionize how we envision gelation process of Chia and other gel forming seeds and materials, and potentially provide the opportunity to study and mimic chemistry in nature to develop more efficient gels and softmaterials.

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