

Proceeding Paper

Assessment of the Physicochemical and Textural Properties of Food Hydrogels Obtained Using Pea Protein and Gellan Gum [†]

Adonis Hilal ^{*}, Anna Florowska, Tomasz Florowski and Małgorzata Wroniak

Department of Food Technology and Assessment, Institute of Food Science, Warsaw University of Life Sciences-SGGW, 02-787 Warsaw, Poland; email1@email.com (A.F.); email2@email.com (T.F.); email3@email.com (M.W.)

^{*} Correspondence: adonis_hilal@sggw.edu.pl

[†] Presented at the 4th International Electronic Conference on Foods, 15–30 October 2023; Available online: <https://foods2023.sciforum.net/>.

Abstract: The aim of this research was to evaluate the physicochemical and textural properties of food hydrogels produced using pea protein and gellan gum. The obtained samples were analyzed in terms of their volumetric gelling index, microrheology, texture, physical stability, and color parameters. Most of the obtained samples formed a gel structure. In the case of microrheology parameters, by combining PP and GG, the solid-liquid balance was shifted towards more solid-like properties, which was also observed while analyzing the elasticity index (EI) and the textural properties of the obtained hydrogels. Additionally, a color difference between the obtained samples was noticed. Depending on the properties that the final food product must exhibit, a binary protein-poly-saccharide hydrogel can be used as a matrix to contribute to that product's physicochemical and textural properties.

Keywords: delivery systems; functional food; plant-based food; structure; biopolymer; gels; food texture

1. Introduction

Hydrogels have emerged as an essential component in food applications, offering many benefits when developing new functional food systems. These food hydrogels' significant potential properties are related to their distinctive three-dimensional structure, composed of hydrophilic polymers capable of absorbing and retaining large amounts of water [1,2]. They provide several valuable features when developing new food systems, including moisture retention, structure stability enhancement, and textural attribute enhancement (including emulating the mouthfeel and texture of fats while significantly reducing caloric content when used as fat mimetics) [3–5]. Furthermore, food hydrogels can play an integral role in entrapping vitamins, minerals, flavors, and other functional ingredients within their structure, providing a pathway for regulating release kinetics and protecting these compounds from environmental degradation [6]. Hydrogels as a delivery system ensure that the nutritional value and bioactivity of the substances entrapped in the gel matrix are preserved, contributing to the health benefits of the final product when consumed [7,8].

Binary food hydrogels comprise two different biopolymers, each with distinct properties that set them apart from hydrogels made with only one type of biopolymer. The polymer composition of binary food hydrogels allows for the customization of the gel's properties to meet the needs of various food products [9]. Each polymer adds unique properties to the hydrogel network, allowing for precise control over parameters such as gel strength, flexibility, texture, stability (resistance to environmental stresses), and release kinetics [10,11].

Citation: Hilal, A.; Florowska, A.; Florowski, T.; Wroniak, M. Assessment of the Physicochemical and Textural Properties of Food Hydrogels Obtained Using Pea Protein and Gellan Gum. *2023*, *26*, x. <https://doi.org/10.3390/xxxxx>

Academic Editor(s): Name

Published: date



Copyright: © 2023 by the authors. Submitted for possible open access publication under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

While both pea protein [12,13] and gellan gum [14,15] have been studied individually for their applications in food systems, combining them to create a hydrogel has the potential to offer unique properties and functionalities. A pea protein-gellan gum hydrogel could be used as a plant-based, protein-rich, and structurally stable matrix in producing various food products, including meat substitutes, dairy-free products, and plant-based desserts [10,11,16]. Conducting studies on binary protein-polysaccharide food hydrogel is critical because of their enhanced properties, tailored functionality, and controlled release capabilities. As a result, the food industry can drive innovation, differentiation, and advancements by incorporating these hydrogels in developing new food systems, resulting in more functional, appealing, and health-promoting food products.

2. Materials and Methods

2.1. Material

Pea protein (PP, NUTRALYS® F85F, protein content 88%, ash 10%) was obtained from Roquette Freres, (Lestrem, France). Gellan gum (GG, high acyl Type 900, particle size: min. 95% mesh through 80 mesh) was obtained from C.E. Roeper GmbH, (Hamburg, Germany).

2.2. Samples Preparation

The samples were obtained in 8 combinations (Table 1)—pea protein (PP) concentration 0, 10, and 12.5% and gellan gum (GG) concentration 0, 0.5 and 0.75%. Pea protein was dispersed in distilled water for 60 min. Afterwards, the protein dispersion was heated (to a temperature of 80 °C) while continuously mixing for 30 min. Gellan gum was then added to the dispersion and the dispersion was stirred for 10 min. Then the obtained dispersions were stored for 24 h at a temperature of 8 °C.

Table 1. The explanation of the samples coding.

Samples Code	Pea Protein (PP) [%]	Gellan Gum (GG) [%]
C1	10	0
C2	12.5	0
C3	0	0.5
C4	0	0.75
H1	10	0.5
H2	10	0.75
H3	12.5	0.5
H4	12.5	0.75

2.3. Methods

2.3.1. Volumetric Gelling Index

This parameter expresses the ability of dispersions to form a gel structure. When no gel structure forms, VGI equals zero, and when the sample is completely gelled, VGI equals 100%. The following equation is used to calculate VGI [17].

$$VGI = \frac{V_G}{V_T} \cdot 100 \quad (1)$$

where V_G —volume of the formulated gel, V_T —total volume of the sample. The reported values represent the averages of three replicates.

2.3.2. Microrheological Properties

The hydrogels' microrheological properties were investigated using the Rheolaser Master device (Formulation, L'Union, France). The device uses the dynamic MS-DWS (Multi Speckle Diffusing Wave Spectroscopy) technique—wavelength of 650 nm. The

detector captures the interfering backscattered waves, and the measurement results are recorded using the Rheotest software. The raw data was used to calculate the following microrheological parameters: solid-liquid balance (SLB) [nm^{-2}], elasticity index (EI) [nm^{-2}], and macroscopic viscosity index (MVI) [nm^{-2}]. The values reported are the averages of three replicates.

2.3.3. Textural Properties

A texture analyzer (TA.XT Plus, Stable Micro Mixtures, Surrey, UK) was used for the texture analysis. The texture analyzer was outfitted with a 0.5 cm diameter cylindrical flat probe (P/0.5R) to measure the hydrogels' strength [N]. The sample penetration depth was set to 8 mm, the measurement speed to 1.0 mm/s, and the temperature to 20 °C. To measure the spreadability [N·s] of the obtained hydrogels, the texture analyzer was equipped with the TTC Spreadability Rig. Exponent version 6.1.4.0 (Stable Micro Mixtures, Surrey, UK) equipment software was used to process the collected data. The reported values are the averages of three replicates.

2.3.4. Physical Stability

The physical stability of the obtained hydrogels was evaluated using the LUMiSizer 6120-75 (L.U.M. GmbH, Berlin, Germany). The hydrogels were centrifuged while the entire sample's cell was illuminated with near-infrared (NIR) light—STEP technology (Space and Time Extinction Profiles). The dispersion volume was 1.8 mL, the used wavelength was equal to 870 nm, light factor 1, 1500 rpm, experiment period 15 h 10 min, interval time 210 s, and temperature 20 °C. The instability index was calculated after obtaining the destabilization behavior (fingerprint) from the recorded data. The reported values are the averages of three replicates.

2.3.5. Color Parameters

The color components in the CIE $L^* a^* b^*$ at the surface of the obtained hydrogels were measured using a Minolta CR-200 colorimeter (Minolta, Japan; light source D65; measuring head hole: 8 mm). Using the obtained $L^* a^* b^*$ parameter, the total color difference (ΔE), whiteness (WI), and yellowness index (YI) indices were calculated. To determine the color differences between the obtained hydrogels, ΔE was computed. The total color difference E was calculated using the equation below [18].

$$\Delta E = \sqrt{(L_{s1}^* - L_{s2}^*)^2 + (a_{s1}^* - a_{s2}^*)^2 + (b_{s1}^* - b_{s2}^*)^2} \quad (2)$$

where L_{s1}^* ; a_{s1}^* ; b_{s1}^* and L_{s2}^* ; a_{s2}^* ; b_{s2}^* refer to the color parameters of the compared hydrogels. The whiteness (WI) and yellowness (YI) index were calculated using the following equations [19]:

$$WI = 100 - \sqrt{(100 - L^*)^2 + a^{*2} + b^{*2}} \quad (3)$$

$$YI = 142.86 \cdot \frac{b^*}{L^*} \quad (4)$$

where L^* , a^* , and b^* refer to the color parameters of each analyzed hydrogel. The reported values represent the averages of three replicates.

2.3.6. Statistical Analysis

Statistica 13.1 (StatSoft, Krakow, Poland) was used to analyze the experiment data. Analysis of variance (ANOVA) was used to determine the effect of PP and GG concentration on the observed results of the experiments. Tukey's test at $\alpha = 0.05$ was used to determine the significance of the differences. Additionally, the results were also assessed using principal component analysis (PCA) and hierarchical cluster analysis (HCA).

3. Results and Discussion

Based on the conducted research, it was found that the volumetric gelling index (VGI) of most of the obtained samples was equal to 100% (Table 1). SLB represents the ratio of the modulus of elasticity and the modulus of viscosity loss (G'/G''). C3 and C4 samples exhibited mostly solid-like properties ($SLB < 5$), while the rest of the samples exhibited more liquid-like properties ($SLB > 5$). Additionally, it was observed that by combining PP and GG, the value of SLB shifted towards 0.5. This could be due to the effect GG had on the protein structure [9]. Furthermore, the macroscopic viscosity index (MVI) showcased a change in the apparent viscosity at zero shear of sample H3, when comparing to C2 and H4.

The sample C4 (produced using only GG, Table 2) had the highest strength and spreadability values, which also was showcased by [GG] having the only significant effect of those parameters. The combination of PP and GG didn't affect the value of strength, nor the value of spreadability. However, the addition of psyllium husk (2%) to a pea protein hydrogel (protein concentration 12.5%) in previous research significantly affected the values of both strength and spreadability [20].

Table 2. The volumetric gelling index and microrheological properties of the obtained samples.

Samples	VGI [%]	SLB [nm^{-2}]	EI $\times 10^{-3}$ [nm^{-2}]	MVI $\times 10^{-4}$ [nm^{-2}]	Strength [N]	Spreadability [N·s]
C1	82 ^a \pm 1.5	0.89 ^e \pm 0.02	0.7 ^a \pm 0.01	0.08 ^a \pm 0.0	0.06 ^a \pm 0.00	0.12 ^a \pm 0.01
C2	89 ^b \pm 1.5	0.65 ^{cd} \pm 0.01	1.3 ^a \pm 0.01	16.4 ^{ab} \pm 3.1	0.06 ^a \pm 0.00	0.17 ^a \pm 0.01
C3	100 ^c \pm 0.0	0.47 ^{ab} \pm 0.04	1.5 ^a \pm 3.8	30.2 ^b \pm 8.1	0.73 ^b \pm 0.03	2.54 ^d \pm 0.04
C4	100 ^c \pm 0.0	0.43 ^a \pm 0.04	2.1 ^a \pm 4.4	39.4 ^b \pm 3.5	1.14 ^c \pm 0.01	3.89 ^e \pm 0.09
H1	100 ^c \pm 0.0	0.68 ^d \pm 0.04	1.7 ^a \pm 0.2	0.58 ^a \pm 0.2	0.07 ^a \pm 0.00	0.78 ^b \pm 0.11
H2	100 ^c \pm 0.0	0.60 ^{cd} \pm 0.06	2.6 ^a \pm 0.1	4.27 ^a \pm 0.8	0.07 ^a \pm 0.00	1.85 ^c \pm 0.19
H3	100 ^c \pm 0.0	0.67 ^d \pm 0.03	16 ^b \pm 0.07	1.54 ^a \pm 0.4	0.08 ^a \pm 0.00	0.76 ^b \pm 0.01
H4	100 ^c \pm 0.0	0.55 ^{bc} \pm 0.03	20 ^b \pm 0.2	18.5 ^{ab} \pm 2.6	0.10 ^a \pm 0.00	1.97 ^c \pm 0.05
Statistic ANOVA, η^2 [-]						
[PP]	0.682	0.692	ns	0.297	ns	ns
[GG]	ns	0.587	ns	0.275	0.949	0.983
[PP]·[GG]	0.811	0.716	ns	ns	0.969	ns

All the values are mean with standard deviation ($n = 3$). According to Tukey's test the values followed by the same letter (a–e) do not differ significantly ($q > 0.05$). η^2 —coefficient indicating the extent of the effect of factors [PP]—pea protein concentration, [GG] gellan gum concentration, and [PP]·[GG]. ns—non-significant.

In terms of physical stability (Table 3), all the obtained samples exhibited high stability (instability index values < 0.2) [1]. The addition of GG to PP significantly reduced the values of the instability index. Moreover, samples containing only GG showed the lowest instability index values.

Table 3. The textural properties, physical stability, and color parameters of the obtained samples.

Samples	Instability Index	Color Parameters				
		L*	a*	b*	WI	YI
C1	0.18 ^e \pm 0.00	73.5 ^{bc} \pm 1.55	-4.4 ^c \pm 0.16	17.9 ^c \pm 0.44	67.7 ^{cd} \pm 1.02	34.9 ^{cd} \pm 0.21
C2	0.03 ^c \pm 0.00	74.5 ^c \pm 0.39	-3.7 ^d \pm 0.05	19.3 ^d \pm 0.37	67.8 ^{cd} \pm 0.29	37.0 ^{de} \pm 0.65
C3	0.01 ^a \pm 0.00	16.5 ^a \pm 0.23	-0.19 ^e \pm 0.01	-0.90 ^a \pm 0.12	16.5 ^a \pm 0.23	-8.0 ^a \pm 1.15
C4	0.00 ^a \pm 0.00	17.7 ^a \pm 0.48	-0.07 ^e \pm 0.02	-0.10 ^a \pm 0.13	17.6 ^a \pm 0.48	-7.9 ^a \pm 1.27
H1	0.09 ^d \pm 0.00	72.5 ^{bc} \pm 0.04	-5.6 ^a \pm 0.25	14.3 ^b \pm 0.42	68.5 ^d \pm 0.24	28.2 ^b \pm 0.84
H2	0.02 ^b \pm 0.00	72.8 ^{bc} \pm 0.53	-4.8 ^b \pm 0.11	17.3 ^c \pm 0.71	67.4 ^{cd} \pm 0.23	34.0 ^c \pm 1.19
H3	0.02 ^b \pm 0.00	72.0 ^b \pm 0.94	-4.2 ^c \pm 0.24	18.0 ^c \pm 0.65	66.4 ^{bc} \pm 0.64	35.7 ^{cd} \pm 1.10
H4	0.02 ^b \pm 0.00	71.9 ^b \pm 0.41	-4.1 ^c \pm 0.05	19.3 ^d \pm 0.25	65.7 ^c \pm 0.22	38.4 ^e \pm 0.31

Statistic ANOVA, η^2 [-]						
[PP]	0.997	ns	0.924	0.891	0.644	0.891
[GG]	0.977	ns	0.581	0.756	ns	0.757
[PP]:[GG]	0.996	ns	0.665	0.753	0.686	0.762

All the values are mean with standard deviation ($n = 3$). According to Tukey’s test the values followed by the same letter (a–e) do not differ significantly ($q > 0.05$). η^2 —coefficient indicating the extent of the effect of factors [PP]—pea protein concentration, [GG] gellan gum concentration, and [PP]:[GG]. ns—non-significant.

With regards to the color parameters (Tables 3 and 4), GG and PP didn’t have a significant effect on L^* . Moreover, PP had the highest effect on the values of a^* and b^* parameters. The effect of pea protein on the color parameters was also observed in case of the whiteness and yellowness indices—this is because GG produce transparent gel structures, while pea protein gels are less transparent [1]. When comparing C1 to H1 and H1 to H3 a clear color difference was noticed ($3.5 < \Delta E < 5$). Furthermore, only an experienced observer was able to notice the difference in color between C1, H3 and H2, between H2 and H3, between H3 and H4, between C4 and C3, and between C2 and C1.

Table 4. The color difference parameter (ΔE) between the obtained samples (values are mean; $n = 3$).

Samples	H4	H3	H2	H1	C4	C3	C2	C1
C1	2.13	1.57	1.06	3.98	59.14	60.18	1.81	0.00
C2	2.58	2.88	2.85	5.73	60.44	61.47	0.00	
C3	59.12	58.70	59.31	58.23	1.13	0.00		
C4	58.09	57.67	58.27	57.18	0.00			
H1	5.28	3.98	3.15	0.00				
H2	2.28	1.22	0.00					
H3	1.36	0.00						
H4	0.00							

The values presented in that table are mean ($n = 3$). Depending on the ΔE values the color difference between the analyzed hydrogels can be estimated as not noticeable for the observer ($0 < \Delta E < 1$), only experienced observer can notice the color difference between the hydrogels ($1 < \Delta E < 2$), unexperienced observer also can notice the color difference ($2 < \Delta E < 3.5$), clear color difference in noticed ($3.5 < \Delta E < 5$), and observer can notice different colors ($5 < \Delta E$).

The PCA and HCA of the obtained result are presented in Figure 1. The principal component analysis (PCA) indicates the relation between the investigated parameters. Two major factors were identified: factor 1 describing 78.81% and factor 14.25% of the variance (93.06% in total). As shown in PCA and HCA, the analyzed samples differed significantly and could be divided into four groups.

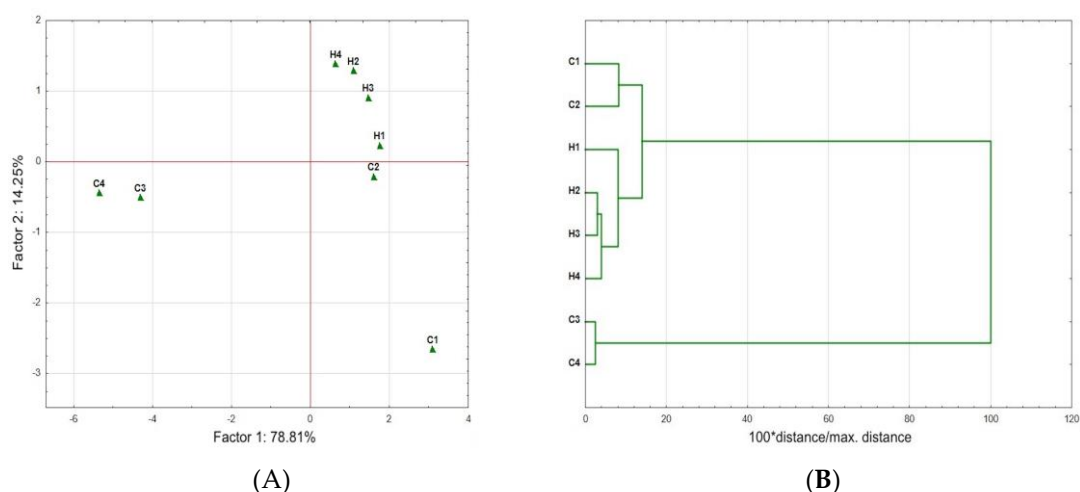


Figure 1. Principal component analysis PCA (A) and hierarchical cluster analysis HCA (B) of the analyzed samples.

4. Conclusions

By varying the concentrations of pea protein and gellan gum, the physicochemical and textural properties of the resulting binary hydrogels can be controlled. In terms of the analyzed properties, the most optimal variant was the one containing 12.5% pea protein and 0.75% gellan gum. Depending on the properties that the final food product must exhibit, a binary protein-polysaccharide hydrogel can be used as a matrix to contribute to that product's physicochemical and textural properties.

Author Contributions: Conceptualization, A.H.; methodology, A.H.; data curation, A.H.; writing—original draft preparation, A.H.; writing—review and editing, A.F., T.F. and M.W. All authors have read and agreed to the published version of the manuscript.

Funding: Research equipment (Rheolaser Master) was purchased as part of “the Food and Nutrition Centre—modernization of the WULS campus to create a Food and Nutrition Research and Development Centre (CZiZ)” co-financed by the European Union from the European Regional Development Fund under the Regional Operational Program of the Mazowieckie Voivodeship for 2014–2020 (project no. RPMA.01.01.00-14-8276/17).

Institutional Review Board Statement:

Informed Consent Statement:

Data Availability Statement:

Conflicts of Interest: The authors declare no conflict of interest.

References

- Hilal, A.; Florowska, A.; Florowski, T.; Wroniak, M. A Comparative Evaluation of the Structural and Biomechanical Properties of Food-Grade Biopolymers as Potential Hydrogel Building Blocks. *Biomedicines* **2022**, *10*, 2106. <https://doi.org/10.3390/biomedicines10092106>.
- Ho, T.C.; Chang, C.C.; Chan, H.P.; Chung, T.W.; Shu, C.W.; Chuang, K.P.; Duh, T.H.; Yang, M.H.; Tyan, Y.C. Hydrogels: Properties and Applications in Biomedicine. *Molecules* **2022**, *27*, 2902.
- Zha, F.; Rao, J.; Chen, B. Plant-Based Food Hydrogels: Constitutive Characteristics, Formation, and Modulation. *Curr. Opin. Colloid. Interface Sci.* **2021**, *56*, 101505. <https://doi.org/10.1016/j.cocis.2021.101505>.
- Cheng, W.; Wu, X.; Zhang, Y.; Wu, D.; Meng, L.; Chen, Y.; Tang, X. Recent Applications of Hydrogels in Food Safety Sensing: Role of Hydrogels. *Trends Food Sci. Technol.* **2022**, *129*, 244–257.
- Yang, D. Recent Advances in Hydrogels. *Chem. Mater.* **2022**, *34*, 1987–1989.
- Ćorković, I.; Pichler, A.; Buljeta, I.; Šimunović, J.; Kopjar, M. Carboxymethylcellulose Hydrogels: Effect of Its Different Amount on Preservation of Tart Cherry Anthocyanins and Polyphenols. *Curr. Plant Biol.* **2021**, *28*, 100222. <https://doi.org/10.1016/j.cpb.2021.100222>.

7. Ćorković, I.; Pichler, A.; Šimunović, J.; Kopjar, M. Hydrogels: Characteristics and Application as Delivery Systems of Phenolic and Aroma Compounds. *Foods* **2021**, *10*, 1252.
8. Bordbar-Khiabani, A.; Gasik, M. Smart Hydrogels for Advanced Drug Delivery Systems. *Int. J. Mol. Sci.* **2022**, *23*, 3665.
9. Li, H.; Wang, T.; Hu, Y.; Wu, J.; Van der Meeren, P. Designing Delivery Systems for Functional Ingredients by Protein/Polysaccharide Interactions. *Trends Food Sci. Technol.* **2022**, *119*, 272–287.
10. Hilal, A.; Florowska, A.; Wroniak, M. Binary Hydrogels: Induction Methods and Recent Application Progress as Food Matrices for Bioactive Compounds Delivery—A Bibliometric Review. *Gels* **2023**, *9*, 68. <https://doi.org/10.3390/gels9010068>.
11. Yang, X.; Li, A.; Li, D.; Guo, Y.; Sun, L. Applications of Mixed Polysaccharide-Protein Systems in Fabricating Multi-Structures of Binary Food Gels—A Review. *Trends Food Sci. Technol.* **2021**, *109*, 197–210.
12. Klost, M.; Brzeski, C.; Drusch, S. Effect of Protein Aggregation on Rheological Properties of Pea Protein Gels. *Food Hydrocoll.* **2020**, *108*, 106036. <https://doi.org/10.1016/j.foodhyd.2020.106036>.
13. Zhu, P.; Huang, W.; Guo, X.; Chen, L. Strong and Elastic Pea Protein Hydrogels Formed through PH-Shifting Method. *Food Hydrocoll.* **2021**, *117*, 106705. <https://doi.org/10.1016/j.foodhyd.2021.106705>.
14. Das, M.; Giri, T.K. Hydrogels Based on Gellan Gum in Cell Delivery and Drug Delivery. *J. Drug Deliv. Sci. Technol.* **2020**, *56*, 101586. <https://doi.org/10.1016/j.jddst.2020.101586>.
15. Dey, M.; Ghosh, B.; Giri, T.K. Enhanced Intestinal Stability and PH Sensitive Release of Quercetin in GIT through Gellan Gum Hydrogels. *Colloids Surf. B Biointerfaces* **2020**, *196*, 111341. <https://doi.org/10.1016/j.colsurfb.2020.111341>.
16. Hilal, A. Hydrożele Białkowo-Polisacharydowe Jako Nośniki Substancji Bioaktywnych Na Przykładzie Kurkuminy. *Przemysł Spożywczy* **2022**, *1*, 15–18. <https://doi.org/10.15199/65.2022.12.2>.
17. Kim, Y.; Faqih, M.N.; Wang, S.S. Factors Affecting Gel Formation of Inulin. *Carbohydr. Polym.* **2001**, *46*, 135–145. [https://doi.org/10.1016/S0144-8617\(00\)00296-4](https://doi.org/10.1016/S0144-8617(00)00296-4).
18. Sobol, Z.; Jakubowski, T.; Nawara, P. Application of the CIE L*a*b* Method for the Evaluation of the Color of Fried Products from Potato Tubers Exposed to C Band Ultraviolet Light. *Sustainability* **2020**, *12*, 3487. <https://doi.org/10.3390/SU12083487>.
19. Tang, C.; Zhang, W.; Zou, Y.; Xing, L.; Zheng, H.; Xu, X.; Zhou, G. Influence of RosA-Protein Adducts Formation on Myofibrillar Protein Gelation Properties under Oxidative Stress. *Food Hydrocoll.* **2017**, *67*, 197–205. <https://doi.org/10.1016/j.foodhyd.2017.01.006>.
20. Hilal, A.; Florowska, A.; Wroniak, M. Evaluation of the Physicochemical and Textural Properties of Binary Protein-Polysaccharide Hydrogels. *Biol. Life Sci. Forum* **2022**, *18*, 41.

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.