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Article

Development of Bio-Based Color Sensor from Myofibrillar Protein–Based Edible Film Incorporated with Sappan Wood (*Caesalpinia sappan* L.) Extract for Smart Food Packaging

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Abstract: The use of intelligent sensor-based packaging in food products provide quick evaluation of the quality of the food. This study was done to develop bio-based color sensor from surimi-based color sensor incorporated with sappan wood extract (SWE) for smart food packaging. The SWE with different concentration (0.15%, 0.25%, and 0.35%) was incorporated to the myofibrillar protein-based edible film. The sappan wood-surimi edible film (SSEF) was subjected to physical properties analysis and the color changes at different pH values and soaking time at different condition were evaluated. Based on the results, different concentration of the SWE significantly ($p < 0.05$) affected the physical properties of the film. With the increasing of pH values, the darkness, redness, and blueness of the film was increased. Based on the evaluation of the SSEF with different soaking condition, the color changes of the film in acidic condition were more stable than in neutral and alkaline condition. The results from this study showed that SSEF have the possibilities to be used as a smart food packaging possessing the capabilities to act as color sensor due to its sensitivity to the changes in pH condition of the product.

Keywords: natural dye; pH sensor; smart packaging; surimi-based; tilapia protein

1. Introduction

The development of new packaging solutions is a result of how packaging technology develops in response to consumer expectations for food products that are fresh, safe, and of an exceptional quality [1,2]. Bioactive substances, such as antimicrobials [3] and antioxidants [4], are present in active food packaging materials to increase shelf life, preserve the quality, and stabilize food-packaged commodities [5]. The goal of intelligent food packaging solutions is to monitor the environment or the conditions of the food, identify any physical, chemical, or biological changes, and take appropriate action. The outcome is intended to provide a quick evaluation of food quality [6–8].

According to the variables that can be controlled, intelligent sensor-based packaging materials can be divided into time-temperature, gas, and freshness indicators to track improper temperature changes along the supply chain, variations in the gas composition in the headspace of the food package, particularly in modified atmosphere packaging, and freshness decay, through alterations in the amount of metabolites indicators of microbial growth and, consequently, altering the freshness of food [2,9]. The most popular freshness indicators are made consisting of a solid support and a dye that reacts to pH changes by changing color and giving a visual response to the environment inside

the package [2,6]. Despite the fact that several synthetic dyes have been investigated as pH indicators in numerous research [10–12], leaching of the dye and consumer knowledge of the negative effects induced by chemically produced dyes raises concerns due to their toxicity and bioaccumulation [2,6,13]. Due to their biodegradability, lack of toxicity, lack of carcinogenicity, and ecologically benign manufacture, natural dyes derived from a variety of sources seem to be a potential option [14,15]. Anthocyanins from barberry (*Berberis vulgaris* L.), black carrots (*Daucus carota* L.), saffron (*Crocus sativus* L.), red cabbage (*Brassica oleraceae*), and the red naphthoquinone pigment shikonin from the root of gromwell are a few examples of natural pigments used in colorimetric indicator systems [7,8,16,17].

Sappan wood (*Caesalpinia sappan* L.) is naturally present across Asia, including China, Japan, and Thailand. This wood plant is now grown in a number of other parts of the world, including Africa, Europe, North America, and South America, due to its many beneficial uses. The wood has the potential to be used as a natural red dye as opposed to a synthetic dye since it is cheap and lacks a distinctive flavor [18]. Brazilein, a white phenolic compound with two aromatic rings, one pyrone, and one five-membered ring, is the primary active component of sappan. However, the hydroxyl group in the brazilein structure is easily oxidized and can convert into a carbonyl group, leading to a structural change and the creation of brazilein, a colorful molecule. Being a polyphenolic substance, brazilein is expected to change color as a result of changes in pH that impact the hydroxyl group in its molecule [19,20]. In addition to brazilein, *C. sappan* is also thought to be a possible source of anthocyanins, natural substances that might be candidates to replace synthetic dyes because of their attractive, vivid colors (orange, red, and purple), which exhibit rapid color fading when exposed to light, oxygen, hot temperatures, pH, salt stress, and enzymes [21].

The incorporation of biopolymers in active packaging has attracted the attention of numerous researchers. These biopolymers include the units created by a covalent peptide link, proteins [22]. Many crucial sources of protein can be obtained in various plant or animal sources. Researchers began to extract polypeptides from a wide range of vegetable and animal products or by-products due to the abundance of resources inside these fundamental products [23,24]. Surimi, a by-product prepared from minced, deboned fish meat, contains concentrated myofibrillar fish proteins [25]. The attributes of myofibrillar protein-based films are slightly superior to those of known protein films, and myofibrillar protein exhibited outstanding film-forming capacities in both acidic and alkaline environments [26–28].

To the best of our knowledge, no study has yet been conducted on the changes on physical properties and color of myofibrillar protein-based edible film incorporated with sappan wood extract at varied pH and soaking time in different condition. Thus, the aim of the present research is to investigate the physical properties of the myofibrillar protein-based edible film added with different concentration of sappan wood extract. Additionally, the color of the film at different pH and soaking condition also evaluated.

2. Materials and Methods

2.1. Raw materials

The surimi was obtained from the processing of Nile tilapia (*Oreochromis niloticus*) were bought from a supermarket (local market, West Java, Indonesia), with the process was done following the method described by Shiku et al. (2004) with modification. The HCl (PubChem CID 313), and NaOH (PubChem CID 14798) were procured from LOBA CHEMIE PVT. LTD., India. All chemicals were analytical grade.

2.2. Preparation of sappan wood surimi edible film

The preparation of sappan wood-surimi edible film (SSEF) was done by following the method described by [29] with slight modification. Frozen surimi was thawed for 20 min and the thawed surimi (10% w/w) was mixed with distilled water (150 mL) and 1M HCl until the pH was 3. The mixture was homogenized using a homogenizer (PRO250 Homogenizer, Thomas 1204B63, Thomas

Scientific, USA) for 30 min at 55°C with the addition of 50% glycerol from the weight of the surimi (w/w) and sieved using 150 mesh nylon sieve. The sappan wood extract (SWE) was added into the film mixture with different concentration (0.15%, 0.25%, and 0.35% w/v) as natural dye. The SSEF mixture was homogenized, poured into a glass plate (20 x 20 cm), and dried in a hot air oven (Binder, Binder GmbH, Germany) at 50°C for 48 h. After the drying process, the SSEF film was packed in a polyethylene bag and put in a desiccator for further use.

2.3. Physical properties analysis

2.3.1. Thickness

The thickness of the SSFEs was measured by using a digital micrometer (Mitutoyo, Tokyo, Japan). The measurement was done from different areas of the films. The thickness measurement was done in triplicate

2.3.2. Transparency

The film transparency (%) was measured using a UV spectrophotometer (model UV-160, Shimadzu, Kyoto, Japan) at 600 nm, according to the method of [30]. The transparency value of the film was calculated by the following equation:

$$\text{Transparency value} = (-\log T_{600})/x \quad (1)$$

where T_{600} is the fractional transmittance at 600 nm and x is the thickness of the film (mm).

2.3.3. Mechanical properties

The tensile strength (TS) and elongation at break of the SSEFs were measured according to the standard protocols [31]. Films were cut into 1 cm x 10 cm strips and kept in a desiccator containing NaBr solution with RH of 57% for 72 h prior to the test. The measurement was done using a texture analyzer (SMT5, Santam, Tehran, Iran) equipped with 100 N load cell, 10 cm distance between grips, and the crosshead speed of 10 mm/min. The measurement of the color values was conducted in quintuplet.

2.4. Color values at different pH and soaking time at different condition

The color (L^* , a^* and b^*) values of the SSFEs at different pH (1 to 14) using a buffer solution at respective pH and soaking time (0 - 20 min, observed periodically every 2 min) at different condition (acid, neutral, and alkaline) were measured using a Colorimeter (ColorFlex, Hunter Lab Inc., USA), with the L^* represents the dark-light spectrum, the a^* represents red intensity, and the b^* represents yellow intensity [32]. The measurement of the color values was conducted in triplicate. The total color difference (ΔE) was calculated as follows:

$$\Delta E = \sqrt{((\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2)} \quad (1)$$

where ΔL^* , Δa^* , and Δb^* were the differences between color parameters of control sample and the color parameters obtained at different pH and soaking time at different condition.

2.5. Statistical analysis

All obtained results were analyzed and reported as mean \pm SD (standard deviation). The statistical analysis was conducted using SPSS software version 17 (SPSS Inc., Chicago, IL, USA) using Duncan's Multiple Range Test (DMRT) with the significance level determined at 95% confidence limit.

3. Results

3.1. Physical properties

The results on the physical properties of the SSEFs are shown in Table 1. The thickness of the SSEFs was in the range of 0.17 - 0.22 mm. Based on the results, the transparency of the SSEFs was in the range of 0.84 - 2.16. The determination of the TS and the elongation at break of the SSEF revealed that the values of these properties were in the range of 1.70 - 10.15 MPa and 12.68 - 15.70%, respectively. The addition of SWE at different concentration in the formulation of SSEF significantly ($p < 0.05$) affected the properties of the resulted film.

Table 1. Physical properties of SSEF with different SWE concentration.

Treatment (%)	Thickness (mm)	Transparency	Tensile strength (mPa)	Elongation at break (%)
0.15	0.17 ± 0.01 ^c	2.16 ± 0.13 ^a	10.15 ± 1.1 ^a	12.68 ± 1.17 ^b
0.25	0.19 ± 0.01 ^b	1.26 ± 0.03 ^b	8.48 ± 1.0 ^b	14.80 ± 0.96 ^a
0.35	0.22 ± 0.01 ^a	0.84 ± 0.04 ^c	7.70 ± 0.7 ^b	15.70 ± 1.26 ^a















Remarks: Data were presented as mean ± SD. Different superscript letter means significant difference ($p < 0.05$) between treatment.

3.2. Color values of SSEF at different condition

3.2.1. Color values at different pH

The sensitivity of the SSEF with different SWE concentration to pH changes was evaluated through immersion in buffer solutions with pH values ranging from 1 to 14 (Table 2). Based on the color measurement of SSFE with different SWE concentration at same pH value, the L* of the SSEF with 0.15% SWE exhibited significantly ($p < 0.05$) higher value (65.86 - 74.97) than the film with 0.25% (64.75 - 73.56) and 0.35% (44.35 - 60.96) SWE. Conversely, the a* and b* value of the SSEF containing the highest concentration of SWE showed significantly ($p < 0.05$) higher value (16.72 - 58.25 and 25.14 - 43.58, respectively) than the SSEF with 0.25% (7.68 - 50.22 and 15.65 - 29.87, respectively) and 0.15% (5.43 - 48.87 and 11.75 - 22.15, respectively) extract (Table 1). Consequently, the total color difference (ΔE) of the SSEF with the addition of 0.15%, 0.25%, and 0.35% SWE were increased significantly (2.30 - 44.20, 8.37 - 43.90, and 3.77 - 45.86, respectively; $p < 0.05$)

Table 2. Apparent color and colorimetric parameters (L*, a*, and b*) of SSEF with different SWE concentration at different pH values (1 to 14).

Treatment	pH													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
0.15 %														
	74.97							72.45					69.22	65.86
	±	74.83	74.28	73.96	73.55	73.29	72.96	±	72.03	71.85	71.25	70.49	±	±
	0.02 ^A	±	±	±	±	±	±	0.02 ^H	±	±	±	±	0.03 ^M	0.02 ^N
	a	0.02 ^{Ba}	0.03 ^{Ca}	0.02 ^{Da}	0.02 ^{Ea}	0.02 ^{Fa}	0.01 ^{Ga}	a	0.02 ^{Ia}	0.02 ^{Ja}	0.01 ^{Ka}	0.03 ^{La}	a	a
a*	5.43	6.75												
	±	±	8.29	9.45		10.39	10.67	11.33	12.24	16.33	19.51	34.25	47.28	48.87
	0.01 ^N	0.04 ^M	±	±	9.86 ±	±	±	±	±	±	±	±	±	±
	c	c	0.02 ^{Lc}	0.07 ^{Kc}	0.02 ^{Jc}	0.01 ^{Ic}	0.01 ^{Hc}	0.01 ^{Gc}	0.01 ^{Fc}	0.01 ^{Ec}	0.04 ^{Dc}	0.02 ^{Cc}	0.03 ^{Bc}	0.02 ^{Ac}

0.25 %	b *	22.15	21.85	21.22	20.26	19.89	19.04	18.74	18.18	17.84	16.72	15.68	13.39	12.46	11.75
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.02 ^{Ac}	0.03 ^{Bc}	0.03 ^{Cc}	0.01 ^{Dc}	0.03 ^{Ec}	0.01 ^{Fc}	0.01 ^{Gc}	0.01 ^H	0.02 ^{Ic}	0.04 ^{Jc}	0.02 ^{Kc}	0.01 ^{Lc}	0.01 ^M	0.03 ^{Nc}
	Δ E	2.30	1.93	2.33	3.40				5.88	6.89	11.04	14.42	29.13	42.14	44.20
		±	±	±	±	3.81 ±	4.62 ±	5.01 ±	±	±	±	±	±	±	±
		0.02 ^{Lc}	0.01 ^M	0.01 ^{Lc}	0.07 ^K	0.03 ^{Ib}	0.01 ^{Ic}	0.01 ^{Hc}	0.01 ^{Gc}	0.02 ^{Fc}	0.02 ^{Ec}	0.04 ^{Dc}	0.01 ^{Cc}	0.02 ^{Bb}	0.01 ^A
			c		b										b
	L *	73.56	73.04	72.81	72.22	71.74	71.55	71.11	70.86	70.53	70.18	69.57	67.85	64.92	64.75
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.01 ^A	0.05 ^{Bb}	0.02 ^{Cb}	0.02 ^D	0.01 ^{Eb}	0.02 ^{Fb}	0.01 ^{Gb}	0.02 ^H	0.01 ^{Ib}	0.02 ^{Ib}	0.02 ^{Kb}	0.01 ^{Lb}	0.01 ^M	0.02 ^N
	a*	7.68	8.12	9.55	10.89	11.58	11.95	12.32	12.77	13.98	18.32	21.75	38.63	49.78	50.22
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.01 ^N	0.01 ^M	0.01 ^{Lb}	0.02 ^K	0.02 ^{Ib}	0.01 ^{Ib}	0.01 ^{Hb}	0.01 ^G	0.01 ^{Fb}	0.01 ^{Eb}	0.02 ^{Db}	0.02 ^{Cb}	0.01 ^{Bb}	0.01 ^A
	b *	29.87	29.34	28.75	27.55	26.98	25.54	24.66	22.78	21.97	21.01	19.56	17.45	15.92	15.65
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.01 ^A	0.04 ^{Bb}	0.03 ^{Cb}	0.02 ^D	0.01 ^{Eb}	0.03 ^{Fb}	0.01 ^{Gb}	0.01 ^H	0.01 ^{Ib}	0.01 ^{Ib}	0.01 ^{Kb}	0.02 ^{Lb}	0.01 ^M	0.02 ^N
	Δ E	8.37	8.18	8.10	8.61	8.89 ±	10.14	10.84	12.61	13.54	15.91	18.86	32.78	43.38	43.90
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.01 ^{La}	0.03 ^M	0.03 ^N	0.03 ^K	0.01 ^{Ja}	0.02 ^{Ia}	0.01 ^{Ha}	0.02 ^{Ga}	0.02 ^{Fa}	0.01 ^{Ea}	0.02 ^{Db}	0.01 ^{Ca}	0.01 ^{Ba}	0.01 ^{Ac}
			a	a	ba										
0.35 %	L *	60.96	59.45	58.97	58.24	57.55	57.21	56.75	55.65	54.17	53.67	51.11	48.25	44.86	44.35
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.02 ^{Ac}	0.05 ^{Bc}	0.02 ^{Cc}	0.02 ^{Dc}	0.02 ^{Ec}	0.02 ^{Fc}	0.02 ^{Gc}	0.01 ^H	0.01 ^{Ic}	0.02 ^{Ic}	0.04 ^{Kc}	0.02 ^{Lc}	0.02 ^M	0.02 ^{Nc}
	a*	16.72	17.56	18.75	19.53	19.94	20.89	21.77	22.14	23.34	27.76	34.28	45.73	55.25	58.25
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.01 ^N	0.04 ^M	0.03 ^{La}	0.01 ^{Ka}	0.02 ^{Ja}	0.02 ^{Ia}	0.02 ^{Ha}	0.01 ^{Ga}	0.03 ^{Fa}	0.02 ^{Ea}	0.01 ^{Da}	0.01 ^{Ca}	0.02 ^{Ba}	0.01 ^A
	b *	43.58	43.25	42.63	42.21	41.75	40.44	39.37	38.21	37.96	35.72	33.55	30.12	26.85	25.14
		±	±	±	±	±	±	±	±	±	±	±	±	±	±
		0.01 ^A	0.03 ^{Ba}	0.04 ^{Ca}	0.02 ^{Da}	0.03 ^{Ea}	0.01 ^{Fa}	0.01 ^{Ga}	0.03 ^H	0.02 ^{Ia}	0.01 ^{Ja}	0.02 ^{Ka}	0.03 ^{La}	0.02 ^M	0.02 ^N























		3.77	3.03	2.83	3.26			8.28		9.65	13.61	20.31	31.94	42.42	45.86
Δ		±	±	±	±	3.98 ±	5.36 ±	6.69 ±	±	±	±	±	±	±	±
E		0.01 ^{Lb}	0.05 ^N	0.05 ^M	0.02 ^K	0.04 ^{Lb}	0.01 ^{Lb}	0.01 ^{Hb}	0.02 ^G	0.01 ^{Fb}	0.01 ^{Fb}	0.02 ^{Da}	0.02 ^{Cb}	0.02 ^{Bb}	0.02 ^A
			b	b	b			b							a












Remarks: Data were presented as mean ± SD. Different uppercase superscript letter means significant difference (*p* < 0.05) between pH. Different lowercase letter means significant difference (*p* < 0.05) between concentration.

3.2.2. Color values at different soaking time in different condition

The SSEF with different SWE concentration also subjected to different soaking time at various condition (Tables 3–5). Based on the color measurement of the SSEF at different soaking time in acidic condition (Table 3), the L* of the SSEF with 0.15% SWE exhibited significantly (*p* < 0.05) higher value (73.09 - 73.41) than the film with 0.25% (67.56 - 68.74) and 0.35% (59.75 - 60.62) SWE. Conversely, the a* and b* value of the SSEF containing the highest concentration of SWE showed significantly (*p* < 0.05) higher value (15.31 - 19.96 and 39.42 - 45.07, respectively) than the SSEF with 0.25% (10.22 - 10.81 and 28.95 - 34.79, respectively) and 0.15% (5.65 - 6.29 and 19.61 - 21.15, respectively) extract. The total color difference (ΔE) of the SSEF with the addition of 0.15%, 0.25%, and 0.35% SWE were increased significantly (0.29 - 1.69, 0.26 - 5.99, and 1.49 - 7.37, respectively).

Table 3. Apparent color and colorimetric parameters (L*, a*, and b*) of the SSEF with different SWE concentration and soaking time at acidic condition.












Treatment		Time (min)										
		0	2	4	6	8	10	12	14	16	18	20
0.15 %	L*											
		73.09	73.09	73.16	73.24	73.28	73.38	73.39	73.39	73.41	73.41	73.41
		±	±	±	±	±	±	±	±	±	±	±
	a*	0.01 ^{Ea}	0.02 ^{Ea}	0.03 ^{Da}	0.02 ^{Ca}	0.01 ^{Ba}	0.01 ^{Aa}	0.01 ^{Aa}	0.02 ^{Aa}	0.02 ^{Aa}	0.02 ^{Aa}	0.01 ^{Aa}
		6.29 ±	6.27 ±	6.13 ±	5.98 ±	5.86 ±	5.77 ±	73.39	73.39	5.66 ±	5.65 ±	5.65 ±
		0.02 ^{Ac}	0.01 ^{Ac}	0.02 ^{Bc}	0.01 ^{Cc}	0.03 ^{Dc}	0.02 ^{Ec}	±	±	0.01 ^{Gc}	0.03 ^{Gc}	0.01 ^{Gc}
	b*	21.15	20.86	20.52	20.33	19.82	19.66	19.65	19.63	19.63	19.61	19.61
		±	±	±	±	±	±	±	±	±	±	±
		0.01 ^{Ac}	0.02 ^{Bc}	0.01 ^{Cc}	0.02 ^{Dc}	0.01 ^{Ec}	0.02 ^{Fc}	0.02 ^{Fc}	0.02 ^{FG} _c	0.01 ^{FGc}	0.03 ^{Gc}	0.01 ^{Gc}
	ΔE	0	0.29 ±	0.65 ±	0.89 ±	1.41 ±	1.60 ±	1.63 ±	1.68 ±	1.68 ±	1.69 ±	1.69 ±
			0.02 ^{Ec}	0.02 ^{Dc}	0.02 ^{Cc}	0.02 ^{Bc}	0.02 ^{Ac}	0.02 ^{Ac}	0.02 ^{Ac}	0.01 ^{Ac}	0.02 ^{Ac}	0.01 ^{Ac}
0.25 %	L*											
		67.56	67.56	67.68	66.23	65.43	65.28	68.65	68.74	68.74	68.74	68.74
		±	±	±	±	±	±	±	±	±	±	±
		0.02 ^{Gb}	0.03 ^{Gb}	0.01 ^{Fb}	0.02 ^{Db}	0.01 ^{Eb}	0.01 ^{Fb}	0.01 ^{Bb}	0.03 ^{Ab}	0.01 ^{Ab}	0.02 ^{Ab}	0.03 ^{Ab}

		10.81	10.74	10.52	10.49	10.31	10.27	10.23	10.23	10.22	10.22	10.22
	a	±	±	±	±	±	±	±	±	±	±	±
	*	0.02 ^{Ab}	0.02 ^{Bb}	0.01 ^{Cb}	0.02 ^{Db}	0.02 ^{Eb}	0.01 ^{Fb}	0.02 ^{Gb}	0.02 ^{Gb}	0.01 ^{Gb}	0.02 ^{Gb}	0.01 ^{Gb}
		34.79	34.54	33.48	32.27	31.73	31.39	30.87	30.49	30.26	29.52	28.95
	b	±	±	±	±	±	±	±	±	±	±	±
	*	0.02 ^{Ab}	0.01 ^{Bb}	0.01 ^{Cb}	0.02 ^{Db}	0.01 ^{Eb}	0.01 ^{Fb}	0.01 ^{Gb}	0.01 ^{Hb}	0.01 ^{Ib}	0.01 ^{Jb}	0.01 ^{Kb}
	Δ	0	0.26 ±	1.35 ±	2.56 ±	3.13 ±	3.53 ±	4.11 ±	4.50 ±	4.72 ±	5.43 ±	5.99 ±
	E		0.01 ^{Ib}	0.01 ^{Ib}	0.02 ^{Hb}	0.01 ^{Gb}	0.01 ^{Fb}	0.01 ^{Eb}	0.02 ^{Db}	0.01 ^{Cb}	0.01 ^{Bb}	0.03 ^{Ab}
												
	L	59.75	59.74	59.95	60.23	60.33	60.54	60.69	60.64	60.63	60.62	60.62
	*	±	±	±	±	±	±	±	±	±	±	±
		0.01 ^{Gc}	0.02 ^{Gc}	0.01 ^{Fc}	0.01 ^{Ec}	0.02 ^{Dc}	0.02 ^{Cc}	0.02 ^{Ac}	0.02 ^{Bc}	0.02 ^{Bc}	0.03 ^{Bc}	0.01 ^{Bc}
		19.96	18.72	17.21	16.96	16.58	15.42	15.37	15.34	15.31	15.31	15.31
	a	±	±	±	±	±	±	±	±	±	±	±
	*	0.02 ^{Aa}	0.01 ^{Ba}	0.02 ^{Ca}	0.03 ^{Da}	0.01 ^{Ea}	0.02 ^{Fa}	0.01 ^{Ga}	0.01 ^{Ha}	0.02 ^{Ha}	0.02 ^{Ha}	0.01 ^{Ha}
		45.07	44.25	42.23	42.21	40.05	39.55	39.51	39.47	39.44	39.42	39.42
	b	±	±	±	±	±	±	±	±	±	±	±
	*	0.02 ^{Aa}	0.02 ^{Ba}	0.02 ^{Ca}	0.02 ^{Ca}	0.02 ^{Da}	0.02 ^{Ea}	0.02 ^{Fa}	0.01 ^{Ga}	0.02 ^{GH} a	0.01 ^{Ha}	0.01 ^{Ha}
	Δ	0	1.49 ±	3.96 ±	4.17 ±	6.08 ±	7.19 ±	7.27 ±	7.31 ±	7.35 ±	7.37 ±	7.37 ±
	E		0.02 ^{Fa}	0.01 ^{Ea}	0.02 ^{Da}	0.02 ^{Ca}	0.02 ^{Ba}	0.02 ^{Aa}	0.01 ^{Aa}	0.01 ^{Aa}	0.01 ^{Aa}	0.01 ^{Aa}

Remarks: Data were presented as mean ± SD. Different uppercase superscript letter means significant difference (*p* < 0.05) between soaking time. Different lowercase letter means significant difference (*p* < 0.05) between concentration.

In neutral condition (Table 4), the L* of the SSEF with 0.15% SWE also exhibited significantly (*p* < 0.05) higher value (71.61 - 73.06) than the film with 0.25% (65.26 - 67.56) and 0.35% (56.81 - 59.76) SWE. Conversely, the a* and b* value of the SSEF containing the highest concentration of SWE showed significantly (*p* < 0.05) higher value (19.96 - 21.08 and 38.04 - 45.07, respectively) than the SSEF with 0.25% (10.81 - 11.69 and 30.21 - 34.79, respectively) and 0.15% (6.29 - 7.59 and 18.91 - 21.15, respectively) extract. The total color difference (ΔE) of the SSEF with the addition of 0.15%, 0.25%, and 0.35% SWE were increased significantly (0.54 - 2.98, 0.46 - 5.20, and 1.09 - 7.70, respectively).

Table 4. Apparent color and colorimetric parameters (L*, a*, and b*) of the SSEF with different SWE concentration and soaking time at neutral condition.











Treatment	Time (min)										
	0	2	4	6	8	10	12	14	16	18	20
	0	2	4	6	8	10	12	14	16	18	20
											





















	0.01 ^G	0.03 ^G			0.01 ^C		0.03 ^A		0.02 ^A		
	a	a			a		a		a		
	45.07	44.12	41.86	41.35	39.54	38.25	38.11	38.11	38.05	38.04	38.04
	±	±	±	±	±	±	±	±	±	±	±
b	0.01 ^A	0.03 ^{Ba}	0.01 ^C	0.01 ^C	0.02 ^{Da}	0.02 ^{Ea}	0.02 ^{Ea}	0.03 ^{Ea}	0.01 ^{Ea}	0.02 ^{Ea}	0.02 ^{Ea}
*	a		a	a							
		1.09	3.34	4.22		7.46		7.63		7.70	
		±	±	±	6.06 ±	±	7.61 ±	±	7.68 ±	±	7.70 ±
Δ	0	±	0.01 ^D	0.01 ^C	0.01 ^{Ba}	0.01 ^A	0.01 ^{Aa}	0.02 ^A	0.11 ^{Aa}	0.02 ^A	0.02 ^{Aa}
E		0.03 ^{Ea}	a	a	a		a	a		a	

Remarks: Data were presented as mean ± SD. Different uppercase superscript letter means significant difference (*p* < 0.05) between soaking time. Different lowercase letter means significant difference (*p* < 0.05) between concentration.

The L* of the SSEF soaked in alkaline condition with 0.15% SWE also exhibited significantly (*p* < 0.05) higher value (60.73 - 73.09) than the film with 0.25% (54.02 - 67.55) and 0.35% (51.40 - 59.75) SWE. In contrast, the a* and b* value of the SSEF with the highest SWE concentration showed significantly (*p* < 0.05) higher value (19.96 - 42.21 and 32.71 - 45.07, respectively) than the SSEF with 0.25% (10.81 - 32.50 and 26.83 - 34.79, respectively) and 0.15% (6.29 - 24.79 and 13.85 - 21.15, respectively) extract. The total color difference (ΔE) of the SSEF with the addition of 0.15%, 0.25%, and 0.35% SWE were increased significantly (5.16 - 23.42, 2.71 - 26.78, and 4.60 - 26.79, respectively).

Table 5. Apparent color and colorimetric parameters (L*, a*, and b*) of the SSEF with different SWE concentration and soaking time at alkaline condition.

Treatment		Time (min)									
		0	2	4	6	8	10	12	14	16	20
0.15 %											
	L	73.09	72.45	70.26	65.36	63.21	60.86	60.74	60.74	60.73	60.73
	*	±	±	±	±	±	±	±	±	±	±
		0.03 ^A	0.03 ^{Ba}	0.02 ^{Ca}	0.02 ^D	0.02 ^{Ea}	0.02 ^{Fa}	0.02 ^G	0.02 ^{Ga}	0.02 ^G	0.01 ^G
		a			a			a		a	a
	a	6.29 ±	11.33	17.50	20.64	23.23	24.76	24.76	24.78	24.79	24.79
	*	0.02 ^{Fc}	0.01 ^{Ec}	0.02 ^{Dc}	0.01 ^{Cc}	0.03 ^{Bc}	0.02 ^A	0.01 ^A	0.02 ^{Ac}	0.01 ^A	0.03 ^A
							c	c		c	c
	b	21.15	20.26	18.74	16.39	14.53	13.95	13.86	13.86	13.85	13.85
	*	±	±	±	±	±	±	±	±	±	±
		0.01 ^{Ac}	0.03 ^{Bc}	0.01 ^{Cc}	0.01 ^{Dc}	0.02 ^{Ec}	0.02 ^{Fc}	0.02 ^G	0.02 ^{Gc}	0.02 ^G	0.02 ^G
								c	c	c	c
	Δ		5.16 ±	11.81	16.98	20.70	23.29	23.38	23.40	23.42	23.42
	E	0	0.01 ^{Fa}	0.02 ^{Ec}	±	±	±	±	±	±	±

		0.02 ^D				0.03 ^B	0.01 ^B	0.02 ^A	0.02 ^A	0.02 ^A	0.02 ^A
		b				b	b	a	a	a	a
0.25 %	L *										
		67.55	65.66	65.53	60.22	58.96	55.96	54.03	54.03	54.02	54.02
		±	±	±	±	±	±	±	±	±	±
		0.01 ^A b	0.03 ^{Bb}	0.02 ^{Cb}	0.01 ^D b	0.02 ^{Eb}	0.03 ^F b	0.02 ^G b	0.01 ^G b	0.01 ^G b	0.01 ^G b
	a *	10.81	12.44	20.02	25.64	27.22	32.54	32.51	32.50	32.50	32.50
		±	±	±	±	±	±	±	±	±	±
		0.02 ^G b	0.02 ^{Fb}	0.01 ^{Eb}	0.02 ^D b	0.01 ^{Cb}	0.01 ^A b	0.03 ^B b	0.02 ^{Bb}	0.02 ^B b	0.01 ^B b
		34.79	33.74	32.93	30.58	28.34	26.79	26.81	26.81	26.82	26.83
	b *	±	±	±	±	±	±	±	±	±	±
		0.02 ^A b	0.02 ^{Bb}	0.02 ^{Cb}	0.01 ^D b	0.02 ^{Eb}	0.03 ^F b	0.02 ^F b	0.02 ^F Gb	0.01 ^F Gb	0.01 ^F Gb
	Δ E	0	2.71 ±	10.22	17.07	19.62	25.90	26.79	26.78	26.78	26.78
			0.03 ^{Fc}	±	±	±	±	±	±	±	±
				0.01 ^{Eb}	0.02 ^D a	0.01 ^{Cc}	0.03 ^B b	0.02 ^A a	0.01 ^A a	0.02 ^A a	0.01 ^A a
0.35 %	L *										
		59.75	57.23	53.71	52.65	51.47	51.44	51.44	51.41	51.40	51.40
		±	±	±	±	±	±	±	±	±	±
		0.01 ^{Ac}	0.02 ^{Bc}	0.02 ^{Cc}	0.03 ^{Dc}	0.01 ^{Ec}	0.02 ^E Fc	0.02 ^E Fc	0.02 ^F Gc	0.02 ^G c	0.01 ^G c
	a *	19.96	22.56	26.11	28.96	39.32	42.39	42.25	42.25	42.23	42.21
		±	±	±	±	±	±	±	±	±	±
		0.01 ^H a	0.01 ^{Ga}	0.03 ^{Fa}	0.02 ^{Ea}	0.01 ^D a	0.02 ^A a	0.02 ^B a	0.03 ^{Ba}	0.03 ^B Ca	0.03 ^C a
		45.07	42.24	40.69	38.22	35.34	32.75	32.72	32.71	32.71	32.71
	b *	±	±	±	±	±	±	±	±	±	±
		0.01 ^A a	0.01 ^{Ba}	0.02 ^{Ca}	0.01 ^D a	0.01 ^{Ea}	0.01 ^{Fa}	0.01 ^G a	0.01 ^{Ga}	0.02 ^G a	0.02 ^G a
	Δ E	0	4.60 ±	9.67 ±	13.35	23.20	26.91	26.80	26.82	26.80	26.79
			0.01 ^{Eb}	0.01 ^D a	±	±	±	±	±	±	±
					0.03 ^{Cc}	0.01 ^{Ba}	0.02 ^A a	0.03 ^A a	0.02 ^A a	0.01 ^A a	0.02 ^A a

Remarks: Data were presented as mean ± SD. Different uppercase superscript letter means significant difference (*p* < 0.05) between soaking time. Different lowercase letter means significant difference (*p* < 0.05) between concentration.

4. Discussion

4.1. Physical properties

The strength of the material and the ability of the edible films to retain the integrity of the packed food are determined by their mechanical properties [33]. The increasing concentration of SWE increased the thickness of the edible film. According to reports, natural films made for food packaging range in thickness from 0.05 to 0.2 mm [34]. The results showed that the thickness of the films were within the acceptable range. Although the casting solutions have the same weight, this thickness variation can also be related to the varied film drying kinetics, which have an impact on the resulting thickness and structure, as has previously been noted in the literature [35–37]. Conversely, the increasing of the SWE concentration decreased the transparency of the film. The transparency of the film offers details about the size distribution of the particles in the matrix [38]. Based on the results, the highest TS value was obtained by the SSEF with the highest concentration of SWE.

4.2. Color values of SSEF at different condition

4.2.1. Color values at different pH

The increasing concentration of SWE at same pH condition decreased the L^* value of the film, while the a^* and b^* value was increased. Considering the pH value to the color of the SSEF, the increasing SWE concentration contributed to the increasing darkness, redness, and yellowness of the SSEF. Shown by the results on the color measurement, the values of the a^* , which indicates the redness of the sample, was increased by the increasing pH values. Moreover, the yellowness, indicated by the b^* value, was decreased with the increasing of the pH value. Brazilein showed a yellow color when the environment was acidic; the color changed to red as the pH rose to an alkaline condition. The lowering L^* value also reflects the increased darkness, which was caused by raising the pH to the alkaline region. The protonation and deprotonation of the hydroxyl (OH) group of polyphenolic compounds (such as anthocyanins and other flavonoids), which frequently occurred upon pH shift, resulted in alterations in the molecular structure of brazilein and caused changes in the color value [19].

4.2.2. Color values at different soaking time in different condition

The increasing of SWE concentration increased the a^* value of the film, while the L^* and b^* value was decreased. Prolonged soaking duration also shown to significantly ($p < 0.05$) affected the color values of the SSEF. The increased soaking duration in neutral condition increased the a^* value, while decreased the L^* and b^* value. Different with the neutral condition, the acidic and alkaline environment resulting in similar results. The L^* value was increased with the increasing of the SWE concentration, while the a^* and b^* value was decreased. Extended soaking time also shown to significantly ($p < 0.05$) affected the color values of the SSEF. The prolonged soaking duration in acidic and alkaline condition increased the a^* value, while decreased the L^* and b^* value.

As can be seen from the results, the gap of the color values between soaking period were getting wider with the increasing of the pH of the soaking condition, With the highest gap was observed at the SSEF soaked at alkaline condition. According to [19], brazilein exhibits stronger stability characteristics at lower pH values and reduced stability at higher pH values. These color variations are caused by the protonation and deprotonation of the hydroxyl (OH) group of brazilein. Similar results were seen for other polyphenolic pigments discovered by different researchers. According to studies by [39–42], the flavylium cation, the most common form of anthocyanins under an acidic condition (pH 3), had higher stability than the other forms, which exist at higher pH (e.g., quinoidal base, carbinol pseudobase and chalcone). In the case of phenolic compounds, decreased stability at higher pH is observed to be caused by the creation of quinone intermediate, which is quickly damaged through oxidation process [19]. Anthocyanin also play roles in the color change of the SSEF. Anthocyanins are sensitive to pH fluctuations since they start to lose their color at pH levels greater than 3.0. At pH levels below 3, anthocyanins mostly reside in the form of the extremely stable red

flavylium cation. The quick hydration of the flavylium cation causes the colored carbinol pseudobase to be produced when the pH rises from 4 to 5. At pH 6-7, a neutral quinonoidal base (purple to violet in color) results from the deprotonation of the flavylium cation; at pH 7-8, an anionic quinonoidal base is produced (blue color) [43–47].

5. Conclusions

It can be concluded that edible film made from surimi with the addition of SWE can be used as a bio-based color sensor that sensitive to pH changes. Based on the results, different concentration of the SWE significantly ($p < 0.05$) affected the physical properties of the film. The pH, soaking time, and soaking condition, also significantly ($p < 0.05$) affected the color values of the film. With the increasing of pH values, the L^* and b^* values were decreased, while the a^* value was increased. Based on the evaluation of the SSEF with different soaking condition at prolonged soaking duration, the color changes of the film in the acidic condition was more stable than in neutral and alkaline condition, while the acidic and alkaline environment resulting in similar results, which were increasing L^* value and decreasing a^* and b^* value, correlated to the stability of the bioactive compound exhibited in the extract of the sappan wood.

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Conflicts of Interest: The authors declare that there are no conflict of interest regarding the role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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