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DEVELOPMENT OF DIGITAL IMAGE COLORIMETRY (DIC) METHOD FOR DETERMINING THE FRESHNESS OF MULLET FISH (*Mugil cephalus*) USING A SMARTPHONE

Pengambangan Metode Digital Image Colorimetry (DIC) Untuk Penentuam Kesegaran Ikan Belanak (*Mugil cephalus*) Berbasis Smartphone

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ABSTRACT

Fish is a highly perishable fishery commodity that rapidly deteriorates after being caught; therefore, a rapid, accurate, and practical method for freshness detection is required. Conventional methods such as organoleptic tests are limited by their subjectivity, thus necessitating the development of alternative approaches based on digital technology. This study aimed to develop a smartphone-based Digital Image Colorimetry (DIC) method for detecting fish freshness using a natural dye extracted from Butterfly Pea Flower (Clitoria ternatea) and to compare its accuracy with the conventional organoleptic method. The sample used was Mullet Fish (Mugil cephalus), which was filleted (without bones) and divided into three portions, each weighing 5 grams. The fish portions were placed in transparent bottles equipped with indicator labels made from Butterfly Pea extract, and color changes were observed during storage at room temperature. The indicator labels were periodically photographed using a smartphone camera, and the captured images were analyzed using ImageJ software to obtain RGB values as color change parameters. Anthocyanins in the Butterfly Pea extract act as pH indicators sensitive to volatile compounds (ammonia) produced during spoilage. The results showed a gradual color change of the indicator label from purple to blue as storage time increased, with RGB value patterns consistent with the deterioration process. Based on organoleptic tests, the fish was categorized as spoiled after 12 hours of storage, which was consistent with the DIC results. Therefore, the Digital Image Colorimetry (DIC) method using Butterfly Pea extract was proven to be accurate, practical, and promising as a smartphone-based alternative for detecting fish freshness.

Keywords: Digital Image Colorimetry, Fish Freshness, Mugil cephalus, Smartphone

ABSTRAK

Ikan merupakan komoditas perikanan yang mudah mengalami penurunan mutu setelah ditangkap, sehingga diperlukan metode deteksi kesegaran yang cepat, akurat, dan praktis. Metode konvensional seperti uji organoleptik memiliki keterbatasan karena bersifat subjektif, sehingga perlu dikembangkan metode alternatif berbasis teknologi digital. Penelitian ini bertujuan untuk mengembangkan metode Digital Image Colorimetry (DIC) berbasis smartphone dalam mendeteksi tingkat kesegaran ikan menggunakan pewarna alami ekstrak Bunga Telang (*Clitoria ternatea*), serta membandingkan hasilnya dengan metode uji organoleptik konvensional. Sampel yang digunakan adalah ikan belanak (*Mugil cephalus*) yang dipotong tanpa tulang menjadi tiga bagian dengan masing-masing berat 5 gram. Potongan ikan ditempatkan dalam botol transparan yang telah dilengkapi label indikator berbasis ekstrak Bunga Telang, kemudian diamati perubahan warnanya selama penyimpanan pada suhu. Antosianin pada ekstrak Bunga Telang berfungsi sebagai indikator pH yang peka terhadap senyawa volatil (amonia) yang terbentuk seiring penurunan mutu ikan. Hasil pengamatan menunjukkan bahwa terjadi perubahan warna label indikator dari ungu menjadi biru seiring bertambahnya waktu penyimpanan, dengan pola perubahan nilai RGB yang konsisten terhadap proses pembusukan. Berdasarkan hasil uji organoleptik, ikan mulai dikategorikan busuk pada jam ke-12 penyimpanan, yang selaras dengan hasil analisis DIC. Dengan demikian, metode Digital Image Colorimetry (DIC) berbasis smartphone terbukti cukup akurat, praktis, dan potensial digunakan sebagai alternatif metode konvensional dalam mendeteksi kesegaran ikan secara digital.

Kata Kunci: Kesegaran Ikan, Kolorimetri Citra Digital, *Mugil cephalus*, Smartphone

INTRODUCTIONS

Fish is a major source of animal protein widely consumed by people in both urban and rural areas. As a marine product, fish contains long-chain fatty acids such as omega-3 (DHA) and omega-6, which play an important role in supporting growth and maintaining body health. These components are rarely found in terrestrial animal or plant-based products (Dewi *et al.,* 2018). In addition to its high nutritional value and relatively affordable price, fish is also an essential food source for humans. However, due to its perishable nature, fish requires special handling to maintain its quality.

Fresh or wet fish refers to fish that has not undergone any preservation process using additives, except for cooling with ice. Fish is considered optimally fresh when its characteristics still resemble those of live fish in terms of appearance, aroma, taste, and texture. Improper handling may cause a decline in its quality or freshness. Handling fresh fish includes all processes carried out from the moment the fish is caught until it reaches the consumer, involving various stakeholders such as fishermen, traders, processors, distributors, and retailers (Nurqaderianie *et al.,* 2016).

Identifying the level of fish freshness is a crucial stage in fish processing that must be performed quickly and accurately, especially when handling large quantities of fish. The degree of freshness can be determined through visual color changes, particularly in the fish's eyes. Several conventional methods commonly used to assess fish freshness include chemical or biochemical analysis, microbiological evaluation, and sensory testing (Saputra *et al.,* 2022). Freshness assessment is a critical aspect of the fisheries industry because it directly affects product quality, flavor, and market value for both producers and consumers.

One of the commonly used methods for evaluating fish freshness is spectrophotometry, a scientific technique that analyzes the chemical and physical properties of fish by measuring the light spectrum absorbed by a sample. Spectrophotometry provides high accuracy in detecting chemical changes such as Total Volatile Basic Nitrogen (TVB-N) and

Trimethylamine Oxide (TMAO), which are widely recognized as indicators of fish freshness. However, despite its accuracy, the use of spectrophotometers presents several limitations, including high operational costs (Rp 300,000–500,000 per sample), lengthy processing time from sample preparation to data acquisition (6–24 hours), and its destructive nature (Wojnowski *et al.,* 2017). In addition, the price of a spectrophotometer can range from tens to hundreds of millions of rupiah, and its operation is not always practical for field applications. Therefore, it is important to develop an accurate and efficient method for detecting fish freshness.

The advancement of smartphone technology equipped with high-resolution cameras (>12 MP) has opened new opportunities for developing more practical and affordable fish freshness analysis methods. Modern smartphones are equipped with sensitive color sensors, autofocus capability, LED flash, and are supported by powerful processors and on-device machine learning features (Masawat *et al.,* 2015). With smartphone penetration reaching 89% in Indonesia (GSMA, 2023) and relatively affordable prices, this technology provides an ideal platform for developing innovative analytical methods. Smartphones offer great potential in chemical analysis due to their accessibility and practicality. In this regard, smartphones can be used as diagnostic tools through colorimetric methods (Firdaus *et al.,* 2025).

The Digital Image Colorimetry (DIC) method is a technique that utilizes digital image processing to measure the color or color components of an object or sample. Its basic principle involves analyzing the color values from digital images to obtain quantitative color data. Digital image analysis uses RGB (Red, Green, Blue) data, which represent reflected light intensity values ranging from 0 to 255 units. The reflected light from an object is captured by the charge-coupled device (CCD) of a digital camera (Dinata *et al.,* 2019). The freshness of fish is strongly influenced by several factors, including storage conditions, temperature, and time, which lead to chemical and physical changes that can be detected through color variation.

Based on the above discussion, the development of a smartphone-based Digital Image Colorimetry (DIC) method for determining fish freshness has significant potential to provide a practical solution for monitoring fish quality throughout the supply chain. This study focuses on developing an integrated system that encompasses image acquisition, data processing, and result interpretation, which can be widely implemented in the fisheries industry. The findings of this research are expected to make a substantial contribution to improving the efficiency and effectiveness of fish freshness determination in Indonesia.

The objectives and benefits of this research are to develop a Digital Image Colorimetry (DIC) method for detecting fish freshness using natural dyes extracted from Butterfly Pea Flower (Clitoria ternatea), and to evaluate the accuracy of the freshness values obtained through the DIC method compared to those obtained using the conventional organoleptic assessment.

METHODS

This research was conducted from April to August 2025 at the Fisheries Laboratory, Department of Marine Science, Faculty of Agriculture, Bengkulu University. The study employed an experimental method with quantitative data analysis. To measure the freshness level of fish, data collection was carried out using the Digital Image Colorimetry (DIC) method. This method was applied to analyze color changes in fish through digital images captured using a smartphone camera. The data obtained from this method were then statistically analyzed to determine the correlation between color parameters and the freshness level of the fish.

Tools and Materials

The materials used in this research included fresh fish samples, butterfly pea (*Clitoria ternatea*) flower extract, absolute ethanol, Natrium Hydroxide, hydrochloric acid (HCl), and distilled water. The equipment utilized consisted of a smartphone with a minimum 12 MP camera, a portable light box with LED illumination, transparent glass bottles, a pH meter, dark bottles, a mortar, a glass filter, Whatman filter papers No. 42 and No. 1, a dropper pipette, a refrigerator, an analytical balance, a measuring cylinder, and data processing software.

Sampel Testing Using the DIC Method

1. Preparation of Natural Dye Extract from Butterfly Pea Flowers

The natural dye used in this study contains anthocyanins, pigments that are sensitive to pH changes. The extract was obtained from butterfly pea (*Clitoria ternatea*) flowers, which are rich in anthocyanins. The extraction was performed using a soaking method with a buffer solution to maintain stability.

Procedure for preparing the natural dye extract:

a. Prepare 20 grams of cleaned and dried butterfly pea flowers.
b. Crush the flowers using a mortar, and add 100 mL of ethanol solvent and distilled water in a 7:3 ratio.
c. After crushing, add 1 M hydrochloric acid (HCl) until the pH reaches 2.
d. Transfer the extract into a dark bottle and store it in a refrigerator for 24 hours.
e. After 24 hours, filter the extract using Whatman filter paper No. 42.
f. The resulting supernatant was separated and stored in a dark bottle for use as the butterfly pea flower extract dye.

2. Preparation of Colorimetric Labels

Cellulose paper was used as the substrate for absorbing the natural dye. Whatman filter paper No. 1 was cut into strips measuring 6 cm × 1 cm. Each strip was immersed in the butterfly pea extract solution (pH 2) for 30 seconds and then air-dried at room temperature. Once dried, the colorimetric labels were ready to be used for detecting color changes caused by fish spoilage.

3. Testing with Fish Samples

Fresh fish fillet samples weighing 5 grams were placed in sealed containers together with the colorimetric labels. The containers were stored at room temperature, and the color changes on the labels were monitored every hour over a 24-hour period. In addition, organoleptic tests were conducted according to the Indonesian National Standard (SNI 01-2729.1-2006) on the remaining portions of the fish stored at room temperature. These sensory evaluations were performed at the same hourly intervals (0–24 hours).

4. Color Change Analysis

The color changes on the labels were documented using digital photography. The captured images were analyzed using ImageJ software to quantitatively measure changes in color intensity. The results of the color analysis were then compared with organoleptic test results to determine the correlation between label color variation and fish freshness level.

5. Result Evaluation

The observed color changes on the labels served as the basis for determining the freshness scale of the fish. An increase in temperature generally accelerated the color change, indicating that storage temperature influences the rate of fish spoilage (Ayu *et al.,* 2022).

RESULT

Sampel Characteristic

In this study, the fish samples used were mullet (Mugilidae), obtained through angling activities in the estuary of the Jenggalu River, Bengkulu City. The fish were still alive when

caught, ensuring that the initial quality was fresh. The total weight of the fish samples was approximately 100 grams, with an average total length of about 22 cm. After collection, the abdominal flesh of the fish was filleted and divided into three portions, each weighing 5 grams. The initial testing (0 hours) was conducted immediately in the field to obtain a baseline of freshness changes from the moment the fish were first caught.

Mullet is an economically important fish species commonly found in coastal waters, estuaries, and brackish areas. Morphologically, mullet has a relatively large and broad body, two dorsal fins, a slightly flat and large head, and scales with a dark coloration on the back and silvery color on the belly (Sukma & Hartono, 2024). Its adaptability to fluctuating salinity in estuarine environments makes it a readily available fishery resource for coastal communities (Okfan & Muskananfola, 2015). In addition, mullet has high nutritional value, particularly in protein and fatty acid content (Hafiludin *et al.,* 2012), making freshness an essential quality parameter to be maintained post-harvest.

The selection of mullet as the research sample was based on its availability in the study area and its biological characteristics, which are suitable for fish quality assessment. The condition of the fish being alive at the time of capture provided a reliable baseline for observing gradual freshness degradation. Through the application of the Digital Image Colorimetry (DIC) method, it is expected that freshness changes caused by the formation of volatile compounds such as ammonia can be detected through color variations in the butterfly pea flower–based indicator. Therefore, this study not only describes the post-harvest freshness dynamics of mullet but also evaluates the potential of smartphone-based DIC as a simple technology for field detection of fish freshness.

Figure 1. Mullet Fish
(Source: Personal Documentation)

The classification of mullet (Mugil cephalus) according to Kurniawan (2022) is as follows:Kingdom : Animalia
Phylum : Chordata
Classis : Actinopterygii
Ordo : Mugiliformes
Familia : Mugilidae
Genus : *Mugil*
Species : *Mugil cephalus*

Extraction of Butterfly Pea Flowers

The Butterfly Pea Flower (Clitoria ternatea) extract used in this study was obtained through an extraction process using Absolute Ethanol and distilled water (Aquadest) as solvents. A total of 20 grams of fresh Butterfly Pea Flowers were used, taking only the purple-

colored petals, as this part contains the highest concentration of anthocyanins compared to other floral structures. The selected flowers were then washed with clean water to remove dirt and debris, followed by drying to reduce moisture content so that it would not interfere with the extraction process.

The Butterfly Pea Flowers were ground using a mortar until they became fine to increase the contact surface area between the material and the solvent, thus allowing optimal extraction of anthocyanin compounds. The extraction process was carried out by adding Absolute Ethanol and Aquadest in a 7:3 ratio, specifically 25 mL of Absolute Ethanol and 10.7 mL of Aquadest for 20 grams of material. Absolute Ethanol was used due to its semi-polar nature, enabling effective dissolution of anthocyanin pigments, while the addition of Aquadest enhanced the dissolution of polar compounds and helped maintain color stability (Sirwutubun *et al.,* 2016).

The pH of the extract was then adjusted to pH 2 by gradually adding Hydrochloric Acid (HCl). A pH of 2 was selected because it provides the most optimal condition for maintaining the stability and color intensity of anthocyanins. Under acidic conditions, anthocyanins exist in the form of flavylium cations, which is the most stable form and produces an intense reddish-purple coloration (Mahmudatussa'adah *et al.,* 2014). This structure has high light absorbance, resulting in a stronger and more stable color that is less susceptible to degradation by oxidation or light exposure.

Preliminary tests showed that extracts at pH 2 produced the highest color absorption on the indicator label paper. This is due to the strong interaction between the positively charged anthocyanin molecules in acidic conditions and the negatively charged hydroxyl groups in the cellulose fibers of the paper, producing a more intense, stable, and long-lasting color. In contrast, at higher pH values, anthocyanins tend to shift into quinonoidal or carbinol base forms, causing the color to fade toward blue or even become colorless (Ayun and Ajeng, 2022).

The stability of the Butterfly Pea Flower extract was maintained by storing the solution in a tightly sealed dark bottle placed in a light-protected environment, such as inside a refrigerator. Storage in a dark bottle serves to prevent photodegradation and oxidation of anthocyanin pigments, as exposure to light and oxygen may break down the flavylium structure, resulting in color fading. Therefore, the extraction and storage process conducted at pH 2 was proven effective in producing a stable, intense purple-colored extract suitable for use as a natural pH-based indicator for detecting fish freshness.

After the extraction process, the extract solution was stored for 24 hours at room temperature in a tightly closed dark bottle (Figure 2). This stage allowed sufficient time for the complete and uniform dissolution of active compounds, particularly anthocyanins, into the solvent. Meanwhile, the 24-hour soaking period facilitated optimal diffusion between the solvent and the plant material, resulting in a higher pigment concentration. Once the soaking period was completed, the extract was filtered using Whatman Paper No. 42 to separate the flower residue from the clear anthocyanin-containing filtrate. The use of this filter paper is essential to obtain a clean, homogeneous solution free of solid particles to avoid interference in the indicator label coloring process. After filtration, the extract was ready for use in the production of the indicator label paper.

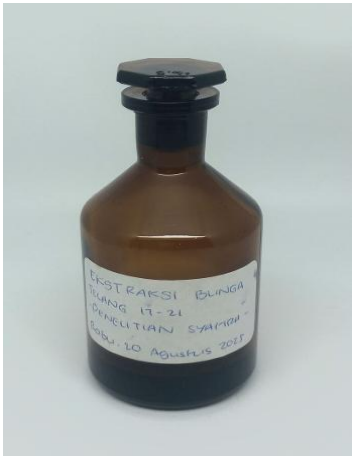


Figure 2. Butterfly Pea Flower (Clitoria ternatea) Extract in a Dark Bottle
(Source: Personal Documentation)

The Butterfly Pea Flower (Clitoria ternatea) extract used in this study functioned as a natural indicator solution for the indicator label paper. Butterfly Pea Flowers are known to be rich in anthocyanin pigments, a class of flavonoid compounds with polyphenolic structures containing chromophore groups capable of absorbing light at specific wavelengths (Purwaniati *et al.,* 2020). Anthocyanins are highly sensitive to changes in environmental pH, causing their color to shift depending on the acidity or alkalinity of the medium (Yessica, 2023). Under acidic conditions (low pH), anthocyanins typically appear reddish-purple; under neutral conditions they shift to blue, and under alkaline conditions (high pH) they turn greenish (Angriani, 2019). This property makes Butterfly Pea Flower extract highly suitable as a fish-freshness indicator, as protein degradation in fish during storage produces basic volatile compounds such as ammonia, which can increase the pH.

The color change of the Butterfly Pea Flower extract on the indicator paper occurs due to the interaction between anthocyanin molecules and hydrogen ions (H^+) in the surrounding environment (Safitri and Findari, 2024). During spoilage, microbial deamination of amino acids produces ammonia and volatile amines (components of TVB-N). Increases in these ammonia/amine levels alter the acid–base equilibrium (NH_3/NH_4^+) within the tissue, causing the system to become more alkaline (increasing pH). The rise in pH triggers structural transformations of anthocyanins (flavylium → quinonoidal/anion → pseudobase → chalcone), resulting in observable color changes that can be used as pH or freshness indicators (Zhuang *et al.,* 2021). This mechanism explains why the indicator paper dipped in Butterfly Pea Flower extract undergoes color shifts as ammonia levels in fish flesh increase.

The extraction of Butterfly Pea Flowers is commonly carried out using polar solvents such as ethanol and distilled water, as anthocyanins dissolve well in polar media (Unawahi *et al.,* 2022). In this study, the produced extract was applied to Whatman Paper No. 1 to obtain indicator paper with an initial reddish-purple color. When the paper is exposed to ammonia vapor generated from fish protein degradation, gradual color changes can be observed visually or quantified through RGB values using the Digital Image Colorimetry (DIC) method. Thus, the use of Butterfly Pea Flower extract not only takes advantage of a natural, safe, and inexpensive material, but also supports the development of fish freshness detection technology based on color-indicator reactions.

Organoleptic Test Results

The organoleptic test is a conventional method widely used to assess fish freshness, including Mugil cephalus. This assessment is based on sensory observation of the fish's physical and sensory characteristics, such as skin and eye color, odor, flesh texture, and body

elasticity (Nisah *et al.,* 2021). In this study, the organoleptic test was employed as a comparative approach to the Digital Image Colorimetry (DIC) method for evaluating the freshness level of Mugil cephalus both visually and sensorially.

The organoleptic panel consisted of several observers who evaluated the samples using a standardized quality scale, such as a 1–9 scoring system or a fresh–spoiled category (Wittriansyah *et al.,* 2019). The main parameters observed included eye clarity, gill color, characteristic fish odor, and flesh firmness. A decrease in organoleptic quality is generally indicated by cloudy eyes, pale or darkened gills, a strong fishy odor, and soft or easily broken flesh texture (Mardiah *et al.,* 2022). These sensory evaluations provide a subjective yet practical representation of fish freshness levels.

In this study, organoleptic testing was conducted by eight semi-trained panelists who assessed the freshness of Mugil cephalus based on key sensory attributes—color, odor, texture, and general appearance. Each panelist assigned a score ranging from 1 to 9, where a score of 9 indicated a very fresh fish, and a score of 1 represented an unfit-for-consumption sample. The average values obtained from the eight panelists were then calculated to provide an overall picture of organoleptic quality changes during 24 hours of storage (Table 3).

Table 3. Organoleptic Test Results

Time (Hours)	Average Value	DS
0	9,00	0,00
2	7,88	0,64
4	7,13	0,35
6	6,63	0,52
8	6,00	0,53
10	5,00	0,53
12	4,75	0,71
14	4,25	0,71
16	3,38	0,52
18	2,50	0,53
20	1,88	0,35
22	1,00	0,00
24	1,00	0,00

Based on the organoleptic assessment conducted by eight panelists on mullet fish, an average score within the range of 1–9 was obtained, where a score of 9 indicates a very fresh condition and a score of 1 indicates a condition unfit for consumption. The assessment results showed a gradual decline in organoleptic scores as storage time increased over a 24-hour period.

At 0 hours, the fish received an average score of 9, indicating a very fresh condition characterized by clear eyes, bright red gills, a natural fresh odor, and firm, elastic flesh texture. After 2–4 hours, the organoleptic score began to decrease to 7. Although the fish was still acceptable for consumption, the panelists detected slight changes, such as a more noticeable fishy odor and a slight reduction in the brightness of the flesh color.

By 6–10 hours, the organoleptic score dropped to the range of 6–5, indicating that the fish was beginning to show signs of declining freshness. The panelists observed reduced elasticity of the flesh, a stronger fishy smell, and duller gills. At 12–16 hours, the score further

declined to 4–3, suggesting that the fish was at the limit of freshness or nearly unacceptable for consumption. The odor became stronger and unpleasant, accompanied by a softer flesh texture.

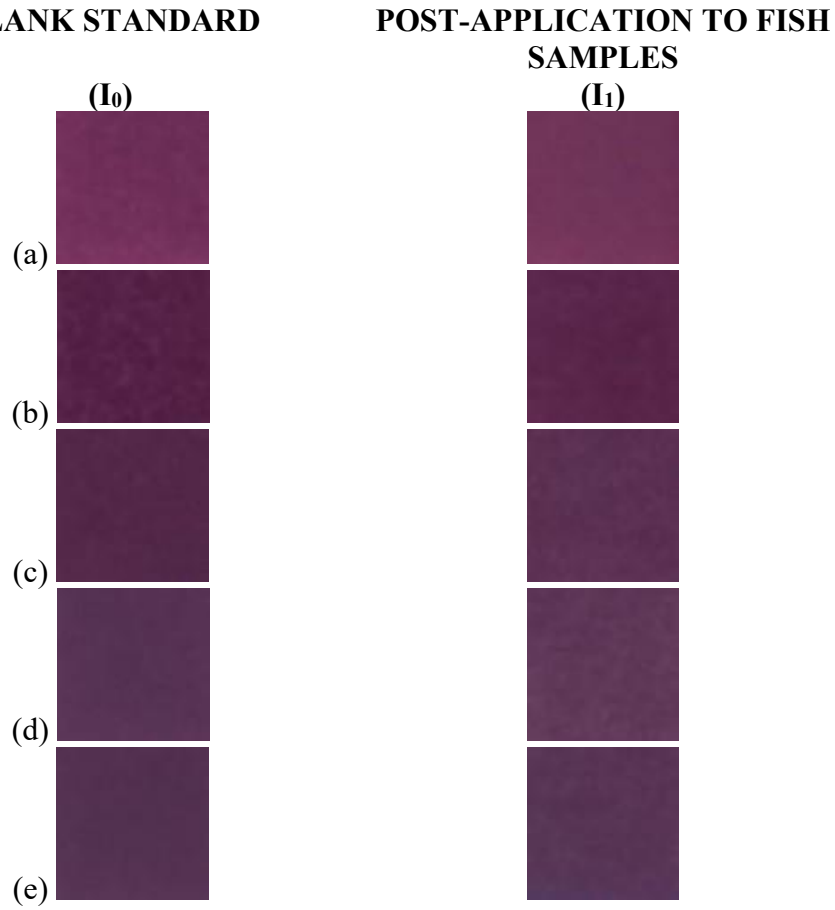
At longer storage durations, specifically 18–24 hours, the organoleptic score declined further to 2–1, indicating that the fish had entered an advanced stage of spoilage. The panelists reported a pronounced rotten smell, very soft flesh texture, and increasingly pale body coloration. Thus, it can be concluded that the mullet fish examined experienced a significant decline in organoleptic quality within 24 hours, and based on the panelists' evaluations, the fish was no longer suitable for consumption after 18 hours of storage.

From the Standard Deviation (SD) values, it can be observed that at 0 hours, the SD was 0, indicating that all panelists gave identical scores for the sample. However, at subsequent time intervals, SD values ranged from 0.35 to 0.71, indicating variations among panelists' assessments, although the differences were not substantial. For instance, at 12 hours, the average organoleptic score was 5 with an SD of 0.71, meaning that some panelists considered the fish still moderately acceptable, while others perceived a more noticeable decline in quality.

These findings align with the theory that fresh fish deteriorates rapidly after death due to enzymatic autolysis, bacterial activity, and lipid oxidation, which lead to the formation of volatile compounds such as ammonia and trimethylamine (Huss, 1995). These compounds contribute to changes in odor, flavor, texture, and color, thereby influencing the overall organoleptic scores.

Digital Image Colorimetry (DIC) Analysis Results

The following are the observed color changes on the indicator label analyzed using the Digital Image Colorimetry (DIC) method. The data show variations in the RGB color intensity values that occurred during the fish storage period (Figure 3).



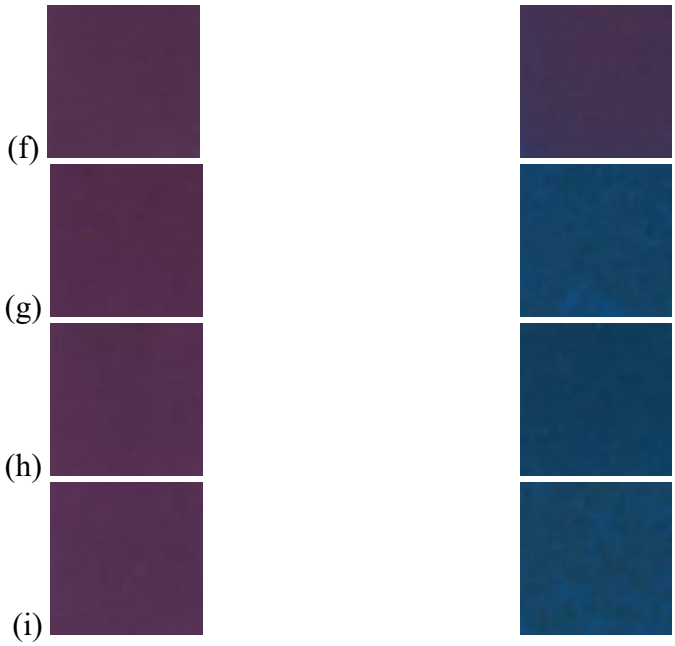


Figure 3. Color Changes on the Indicator Label

Based on the visual observations of the indicator label paper (Figure 2), the color changes became more apparent as the fish storage time increased. From 0 to 12 hours, the indicator paper displayed a predominantly reddish-purple color. This color indicates that the fish was still in a fresh condition, where the levels of volatile compounds such as ammonia (NH_3) were still low. At this stage, the pH remained relatively close to neutral, allowing the anthocyanins in the Butterfly Pea Flower to exist in a structural equilibrium between the flavylium cation form (red) and the basic form (blue), resulting in the characteristic purple hue. According to Herfayati et al. (2020), the red coloration is formed by flavylium cations, in which methoxy groups in the anthocyanin structure are more dominant than hydroxyl groups.

From 15 to 24 hours, the indicator paper showed a stronger shift toward bluish-purple to dark blue. This shift occurred due to the increasing concentration of basic volatile compounds produced from the microbial decomposition of proteins, which caused the pH of the system to rise. Under more alkaline conditions, anthocyanins undergo structural transformation from the flavylium cation form to the more stable quinonoidal base form, resulting in a more pronounced blue coloration. According to Herfayati et al. (2020), under alkaline conditions the extract turns bluish due to the formation of the pseudobase (carbinol) structure.

The color changes observed in this table align with the organoleptic test results, which showed a decline in freshness scores, and are further supported by the Digital Image Colorimetry (RGB) analysis. The Red values tended to decrease, while the Blue values increased during the later storage period, which is consistent with the visual transition from purple to blue. Thus, the anthocyanin-based indicator label from Butterfly Pea Flower extract can represent fish freshness in a simple manner, where a purple color indicates the fish is still fresh, while a blue color indicates that the fish has lost its freshness.

DISCUSSION

The graph of Red intensity changes (Figure 4) illustrates the variation in the Red component on the indicator paper during the storage of Mullets at room temperature. The Red value represents the intensity of the red color captured in the digital images obtained during observation. Changes in this component indicate the extent to which anthocyanin pigments undergo color shifts due to the influence of volatile compounds produced during the fish

spoilage process. A decrease in Red intensity generally indicates a reduction in reddish hues on the indicator, corresponding to the decline in fish freshness.

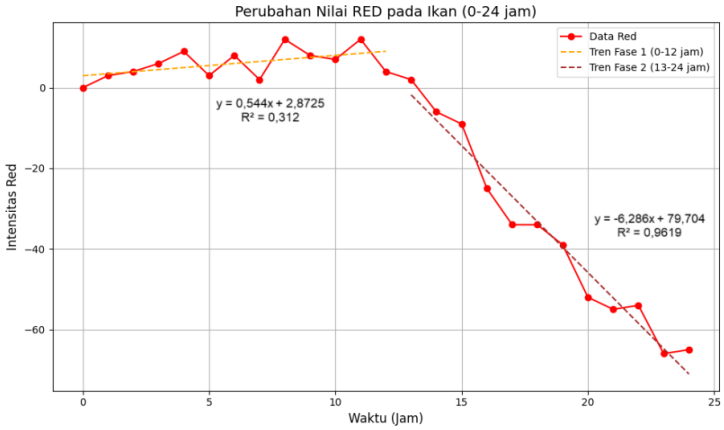


Figure 4. Red Value Graph

In the early phase (0–12 hours), the Red intensity values (Figure 4) showed a slight increase with the linear regression equation y = 0.544x + 2.8725 (R^2 = 0.312). This indicates that the red color remained relatively stable and even increased slightly during the initial storage period. However, from 12 to 24 hours, the Red intensity decreased sharply, following the regression equation y = –6.286x + 79.704 (R^2 = 0.9619). This significant decline reflects color changes associated with fish quality deterioration due to the accumulation of volatile compounds such as ammonia (NH_3) and trimethylamine (TMA), which increase surface pH (Byrne *et al.,* 2002).

Changes in the RGB color components are closely related to the complementary color behavior in the additive color system. In the RGB model, an increase in the intensity of one component will reduce the perception of its complementary color. For instance, a decrease in Red accompanied by an increase in Blue indicates a shift from a reddish-purple hue toward a bluish-purple tone. This phenomenon aligns with the visual observations of the anthocyanin-based Butterfly Pea Flower indicator paper, which exhibited color changes during fish storage (Ameri *et al.,* 2024). The initial purple color (a balanced combination of Red and Blue) shifted to blue as volatile compounds such as ammonia increased and pH rose. Thus, the digitally detected RGB value changes reflect the color reactions of anthocyanins in response to chemical alterations in the fish environment, further reinforcing the relationship between visual parameters and fish freshness levels (Vidana *et al.,* 2021).

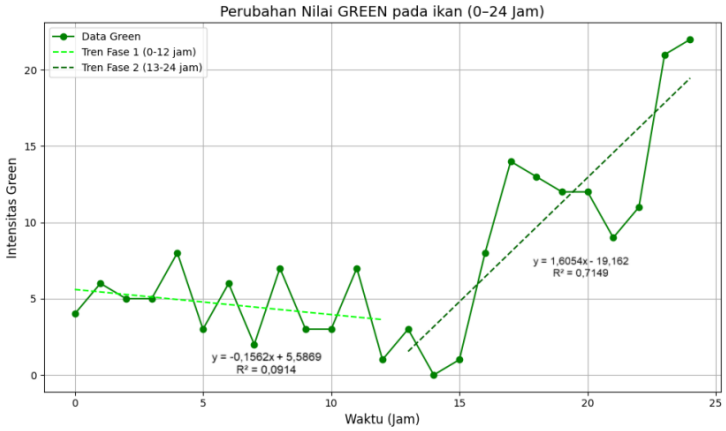


Figure 5. Green Value Graph

The Green intensity values (Figure 5) were relatively fluctuating during the 0–12 hour period, showing a decreasing trend (y = -0.1562x + 5.5869; R^2 = 0.0914), during which the indicator paper remained purple. This change was relatively small because the purple coloration is formed from a combination of red and blue, with the Green component still contributing to the overall color composition. During the 13–24 hour period, the Green values increased (y = 1.6054x - 19.162; R^2 = 0.7149), corresponding to the shift of the indicator color toward blue. This phenomenon occurs because visually perceived blue still contains a Green component in the RGB system; therefore, although green is not visible to the naked eye, the increase in Green intensity can still be detected digitally. Thus, the color shift of the indicator from purple to blue is also reflected in the rising Green intensity, indicating dynamic changes in pigment composition due to increasing pH and the formation of volatile basic compounds during fish storage. According to Fadhli *et al.,* (2022), spoilage processes in fish flesh generate volatile basic compounds that react with smart labels, causing noticeable color changes on the indicator.

The Green value in the RGB color system plays an important role in maintaining overall color balance, particularly within secondary color ranges such as cyan, yellow, and purple. Based on additive color theory, green has a complementary color—magenta (a combination of red and blue)—which is located on the opposite side of the color wheel (Pridmore, 2021). This relationship confirms that the balance among Red, Green, and Blue in the additive system is interconnected and can be explained through complementary color interactions. This analysis further supports the interpretation that variations in digitally recorded RGB values result from the chemical transformation of anthocyanins due to increasing volatile compounds and pH changes in fish that have begun to experience freshness degradation (Kungsuwan *et al.,* 2014).

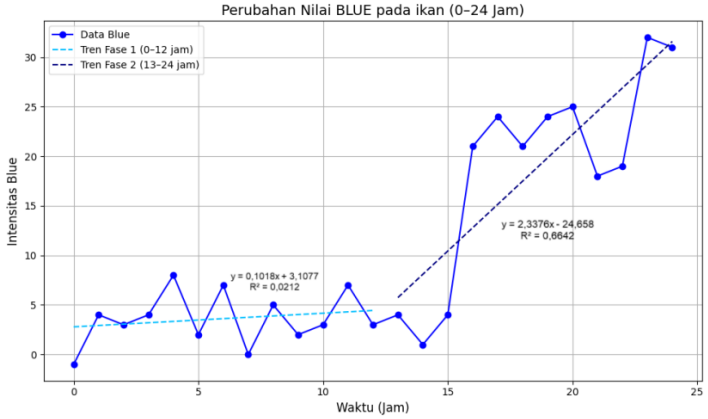


Figure 6. Blue Value Graph

The Blue intensity values (Figure 6) also showed a pattern similar to the Green component. During the early storage period (0–12 hours), the intensity remained relatively stable with a slight increase (y = 0.1018x + 3.1077; R^2 = 0.0212). However, after 12 hours, a significant increase occurred (y = 2.3376x - 24.658; R^2 = 0.6642), indicating a clear shift of the indicator color toward blue. This further strengthens the evidence that the rise in pH due to microbial activity and the formation of ammonia compounds directly influences the increase in blue intensity on the indicator label (Apriliani *et al.,* 2022).

In the Blue component graph presented in Figure 6, the increasing Blue values correspond with longer storage duration (or the altered condition of the indicator). The increased Blue values indicate that the blue aspect of the digital image is becoming more dominant. From the perspective of complementary color theory (in the RGB/light color space), blue has a complementary color located on the opposite side of the spectrum—typically orange,

or more precisely in the additive RGB model, blue is complemented by yellow-green/cyan depending on the model (Pridmore, 2021).

The variation observed in all three RGB components (Red, Green, and Blue) collectively reflects the complex color dynamics occurring on the anthocyanin-based indicator paper during fish storage. The decreasing Red values accompanied by the increasing Blue and Green values illustrate a chromatic shift from a reddish-purple hue toward bluish-green. This shift is closely associated with the halochromic properties of anthocyanins, which are highly sensitive to pH changes and volatile compounds produced during fish spoilage (Khezerlou *et al.,* 2023). In the context of complementary color theory, the dominance of blue and green indicates a reduced contribution of red—their complementary counterpart—in the additive RGB system, resulting in a progressively cooler appearance leaning toward the blue-green spectrum.

This phenomenon aligns with the chemical behavior of anthocyanins, whose molecular structures transform as pH increases, causing the color to shift from red or purple to blue-green. These findings demonstrate that RGB analysis not only records the visual changes but also represents the underlying chemical responses occurring during the decline in fish freshness.

CONCLUSION

Based on the findings of this study, it can be concluded that this research successfully developed a DIC method utilizing butterfly pea (Clitoria ternatea) extract as a natural dye for the indicator label. The anthocyanins in butterfly pea extract were proven to be highly sensitive to pH changes triggered by the increase of volatile compounds such as ammonia during fish storage, allowing the color changes on the indicator paper to serve as a reliable marker of fish freshness. The RGB values obtained through DIC analysis demonstrated a color-change pattern consistent with the organoleptic evaluation conducted by trained panelists. This finding confirms that the DIC method can serve as an accurate and practical alternative to conventional techniques for detecting the decline in fish quality during storage.

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