Indigo Dye Derived from *Indigofera Tinctoria* as Natural Food Colorant

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**Abstract.** Recently, the uses of dyes are increasingly widespread especially in foods and beverages as food colors to attract the consumers. The dye that currently attracts is indigo. Indigo is a group of carbonyl compounds, one of the oldest known dyes in terms of natural blue dye derived from the *Indigofera tinctoria* plant. The use of indigo as a natural food dye intended to reduce the use of synthetic dyes are carcinogenic impact. The method used in this study includes the analysis of indigo using UV-Vis spectrophotometry and FTIR analysis. Based on the UV-Vis Spectrophotometer analysis results with the various concentrations of 0.002 mg/mL; 0.004 mg/mL; 0.006 mg/mL and 0.008 mg/mL were obtained maximum absorption peak at wavelength of 550-700 nm. The indigo dyes in various concentrations produce a stable pH at an average pH 9, therefore it can make the colors not easily fade (strong staining). Based on infrared spectrophotometer measurement were obtained absorption spectrum at 3100-3500 cm\(^{-1}\) as primary N-H and secondary amine, 1600 cm\(^{-1}\) as aromatic C=C, 1000-1350 cm\(^{-1}\) as aromatic C-N, 690-900 cm\(^{-1}\) as aromatic C-H.

1. **Introduction**

In recent years, there has been a revival of the use of dyes and colors of natural origin for coloring food, pharmaceutical, cosmetic and textile products. Among the natural dyes which are becoming widely recognized throughout the world, indigo which is one of the oldest known natural dyes [1], is a derivative of the colorless glucosides of the enol form of indoxyl, e.g. indican (indoxyl-D-glucoside). Indigo, after which this group of carbonyl dyes is named, is one of the oldest known natural dyes. It is a derivative of the colorless glucosides of the enol form of indoxyl, e.g.indican [2,3].

Extraction of indigo from the leaves of *Indigofera tinctoria Linn.* investigated chemical constituents including the chemical structure of major components in the water extract from both kinds of plant to develop the extraction method for preparation of ready-to-use natural dyes [4]. Coloring matter (Indigotin) from *Indigofera tinctoria* plant is usually present abundantly in flowers which give blue color mainly used to dye linen and hairs. Flavonoids, terpenoids, alkaloids, glycosides, Indigotine, Indirubin, rotenoids are the other related compounds abundantly present in the plant. These compounds were found to be
responsible for many pharmacological activities such as antihyperglycemic activity, antioxidant, anti-inflammatory activity, antibacterial, antihepatoprotective activity, antidiabetic activity and anticonvulsive agent [5].

Classification of scientifically conducted on alkaloid content: indicant glycosides, non-protein amino acids, nitro-propionic acid esters, cyanogenic glycosides, guanidine alkaloids, phenolic acids, phenolic glycosides flavonoids, iso flavonoids. Types of indigo which containing alkaloid compounds as above indicated as a dye [6]. The use of indigo as a dye should depend on the color. The dye stability will be influence by pH and their chromophore certain in dye.

2. Materials and Methods

2.1 Materials

The instruments were used for analysis are UV-Vis spectrophotometer Lambda 25 (Pelkin Elmer), and Fourier Transform Infrared (FTIR) Prestige 21 (Shimadzu). The Indigofera dyes extract was from Indigofera Tinctoria leaf as commercial product in Parakan, Temanggung, Central Java Indonesia.

2.2 Preparation

Indigo dyes of 0.002; 0.004; 0.006; and 0.008 mg, were dissolved in 100 mL of distilled water, respectively to obtain the mother liquor indigo. Then, the solutions were measured its pH. Influence of concentration on pH were studied. The homoogen solution of indigo dyes were analyzed by UV-Vis spectrophotometer to determine Indigo dye absorption spectra in various concentration samples. UV-Vis spectrophotometer analysis also aims to determine the wavelength of maximum absorption of the indigo. The chromophore groups of indigo was identify by FTIR.

3. Results and Discussion

Indigo dye (figure 1) usually has a contained of flavonoids, terpenoids, alkaloids, glycosides, indigotine, indirubin, rotenoids. These compounds were found to be responsible for many pharmacological activities such as antihyperglycemic activity, antioxidant, anti-inflammatory activity, antibacterial, antihepatoprotective activity, antidiabetic activity and anticonvulsive agent [7]. Because of their functions, indigo dyes can be use as a functional food dye. Based on indigo structural above, the different structure of indigo dye will be occur in acid and base conditions, which shown in figure 2.

![Figure 1. Structure of indigo [8-9]](image-url)
Figure 2. The indigo dyes structure in (a) acid condition and (b) base condition [4]

Figure 3. pH of indigo in various concentration (mg/mL)

The pH measurement in various concentration indigo dyes shown that the higher the indigo concentration, the higher the pH value. In the range pH of 8.8 – 9.2 is a stable of indigo dyes condition. Therefore, those conditions produce the color do not easily fade (strong staining). The absorption spectrum measurement of indigo dyes performed at a wavelength in range 200-800 nm. Indigo absorption spectrum was shown in figure 4.
Figure 4. The UV-Vis absorption of indigo dyes in various concentration of 0.02 mg/mL (---); 0.04 mg/mL (-----); 0.06 mg/mL (-----); and 0.08 mg/mL (-----)

The UV-Vis absorption of indigo dye by using UV-Vis spectrophotometer showed that the maximum absorption peak is at a wavelength in the visible range of 550-700 nm, where the absorbance maximum reached at a concentration of 0.008 mg/mL. In general, the concentration of dyes affected the color intensity. The strong spectrum in the whole visible range have gained the blue color of indigo dye. The blue color of indigo dye cause of the chromophore group in chemical structure. Electronic transitions of indigo chromophore group from both $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ were in visible range. The chromophore groups of dye was also have been identify by FTIR spectra. The FTIR absorption spectra of indigo dyes was shown in figure 5. Based on infrared spectrophotometer measurement were obtained absorption spectrum at 3100-3500 cm$^{-1}$ as primary N-H and secondary amine, 1600 cm$^{-1}$ as aromatic C=C, 1000-1350 cm$^{-1}$ as aromatic C-N, 690-900 cm$^{-1}$ as aromatic C-H (table 1).

| Wavenumber (cm$^{-1}$) | Functional Group                  |
|------------------------|-----------------------------------|
| 3100-3500              | N-H Primary and Secondary Amines  |
| 1600                   | C=C Aromatic                      |
| 1350-1000              | C-N Amines                        |
| 900-690                | C-H Aromatic                      |
4. Conclusion
Indigo dyes derived from Indigofera tinctoria plant was potentially as a natural blue dye for food colorant. The stable blue color of indigo dyes produce by strongly UV-Vis absorption in the broad range of 550-700 nm is base condition (8.8 – 9.2). The blue color on Indigo dye was generated from an electronic transition of the chromophore group in indigo dye structure as N-H primary and secondary amine, C=C aromatic, C=N amine, and C-H aromatic.

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