International Journal of Applied Engineering Research ISSN 0973-4562 Volume 12, Number 21 (2017) pp. 10711-10718


# Effect of Fixative Types on The Natural Dye Efficacy of Rambutan (*Nephelium Lappaceum*) Peel Extract for the Cotton Fabric Application

**Vita Paramita*, Heny Kusumayanti, Wahyuningsih Rizka Amalia and Fransisca and Sri Nugraheni Setiawati**

*Department of Chemical Engineering, Diponegoro University,*
*Prof. Sudarto, SH Street, Tembalang, Semarang 50275, Indonesia.*
**Corresponding author*

*Orcid: 0000-0003-0384-4631*

**Abstract**

This present study evaluated the effect of various metal salts of fixative agent ($CaCO_3$, $Al_2(SO_4)_3$, and $FeSO_4$) on the efficacy of rambutan (*Nephelium lappaceum*) peel of anthocyanin and flavonoid, on the shades produced on cotton fabric and the optimization of dyeing process parameter (rambutan peel concentration and fixative concentration). Rambutan peel is abundant and renewable of natural dye sources without any commercial valuable and easily obtained as food household waste. The dyed cotton fabric was analyzed the fastened total anthocyanin content (TAC) and total flavonoid content (TFC) frrom rambutan peel extract on the cotton fabric regarding to the concentration of rambutan peel extract and fixative agent. The optimization of total anthocyanin and flavonoid content attached on the cotton fabric were also studied by using central composite design-response surface methodology. Mature color were obtained on the cotton fabric after storage for 6 moths. The concentration of rambutan peel gave the highest influenced parameters for TAC, while the fixative type performed the highest influenced parameter for TFC. The TAC provided a better accuracy of model than the TFC. The predicted optimum values of TAC obtained by using fixative of $FeSO_4$ were found at 29.09 % of rambutan peel's concentration and 31.06 % of fixative content with the coefficient of determination obtained was 0.9743 (97.43 %). This high $R^2$ value indicated that TAC significantly effect of shades on dyed fabric color.

**Keywords**: rambutan peel, natural dyes, fixative types, cotton fabric

## INTRODUCTION

Natural dyes gained high attention for the last ten decades due to their environment friendly characteristic, such as noncarcinogenic, non-toxic, biodegradable, renewable and produce soothing shades to fabric [1–5]. Plant dyes offered as potential sources due to their abundant availability in nature. Rambutan (*Nephelium lappaceum*) outer skins are one of the valuable potential source of natural dyes because it commonly discarded as food waste without any commercial value. It is a tropical fruit and native from Southeast Asia region. Rambutan peel is rich in anthocyanins, ellagitannins, ellagic acid, corilagin, geraniin, syringic acid and p-coumaric acid [6–7].

In order to stabilize the natural dyes in fabric, its application require the use of mordant solution [4–5]. Staining bonding occurs between the metal ions from mordant, which serve as electron acceptors, and the electron donors from the dye molecules and result a metal complex compound. This complex compound is insoluble in water and creates a dye retention on fabric. Most used mordants are including alum, lime, chrome, copper sulphate, ferrous sulphate, and so on [8]. Since chrome and copper are categorized as heavy metals, its considered as harmful for human skin and not environmentally friendly [9].

Cotton is the most recommended fabric for daily use in the tropical area, includes Indonesia, due to skin health purposes. This area have mostly humid with warm temperature and light breeze most of the year, therefore cotton fabric is applied. Clothes serve the closest environment to the skin, and therfore the characteristics of cloth materials are important for people who have sensitive skin stimulations [10]. Nature of cotton provides the inherent hydrophilic [11], therefore it can easily absorb the sweat and dried again. Many studied of rambutan peel as natural dyes published, however the efficacy of its content, anthocyanin and flavonoid, on the shades produced on cotton fabric and the optimization of dyeing process parameter (rambutan peel concentration and fixative concentration) regarding to the fixative types have not been studying.

## MATERIALS AND METHODS

### *Materials*

Rambutan outer skin (*Nephelium lappaceum*) was obtained from the local rambutan farmer in Semarang area. Mordant solution (70 wt% of glucose), $CaCO_3$, $Al_2(SO_4)_3$ and $FeSO_4$ were purchased from Bratachem (Semarang, Indonesia). Cotton fabric of "primisima" was purchased from Pekalongan district. Quercetin standard was purchased from Sigma-Aldrich, Inc. (Singapore).

***Cotton fabric dyeing by using rambutan peel extract***

Rambutan outer skin was extracted by using warm water at 40 $^0$C at varied concentration of 15, 20, and 25 %, then cooled into room temperature. The prepared cotton fabric of "primisima" was immersed by using the cooled rambutan peel's extract for ten minutes, following by the immersion on the mordant solution for 10 minutes. These stages were repeated up to three times and wind-blown until there were no dripping anymore. After immersion processes, the wet but non-dripping fabrics were fixating by using fixative agent ($CaCO_3$, $Al_2(SO_4)_3$ or $FeSO_4$) on the fixative concentration of 5, 25, and 45 %. After the fixation process, the cotton fabric was ready for analysing the content of flavonoids and anthocyanins which stained in the cotton fabric.

***Determination of total anthocyanin content (TAC)***

Total anthocyanin content (TAC) was determined according to the pH spectroscopic differential method [12] by using UV-Visible *recording spectrophotometer* (UV-160A, Shimadzu, Japan). 3 ml of extract was diluted in 5 ml of two different buffers; respectively, 0.025 M of potassium chloride at pH = 1.0 and 0.4 M of sodium acetate at pH = 4.5. After 30 min incubation at room temperature, absorbance (A) was measured at wavelengths of 510 and 700 nm. After incubation at room temperature for 30 min, the absorbance (A) was measured at wavelengths of 510 and 700 nm. The results were calculated by using the equations as [13]:

$$A_{sp} = (A_{510} - A_{700})_{pH_{1,0}} - (A_{510} - A_{700})_{pH_{4,5}} \quad (1)$$

The calculation of total anthocyanin content (TAC) follows the equation:

$$\text{TAC} = \frac{(A_{sp} \times M \times DF \times 1000)}{(\varepsilon \times \lambda \times m)} \quad (2)$$

where, DF is the dilution factor, ε is the molar absorption coefficient, 29.600 $M^{-1}cm^{-1}$[14], M is the molecular weight, 448.8 g/mol, λ is the long quadrilateral optical path, 1 cm, and m is the sample weight (g) [15]. The TAC are expressed as mg anthocyanin per 2 $cm^2$ of the fabric cotton test of sample area.

***Determination of total flavonoid content (TFC)***

Total flavonoid content (TFC) was expressed as mg quercetin equivalent (QE) [16]. One milliliter of extract or standard quercetin solution (20, 40, 60, 80 and 100 µg/ml) was added to a 10 ml measuring flask containing 4 ml of distilled water. Subsequently, 0.3 ml of 5% $NaNO_2$ was added, five minutes later, 0.3 ml of 10% $AlCl_3$ was then added. After the next five minutes, 2 ml of 1 M NaOH was added and the volume was made up to 10 ml with distilled water. The solution was mixed and the absorbance of the blank and the samples was measured at 510 nm by using UV-Visible *recording spectrophotometer* (UV-160A, Shimadzu, Japan).

***Response Surface Methodology (RSM)***

In order to execute RSM, the design of experiments (DoE) applied was central composite designs (CCD) with design characteristic of face centered star points (α = 1) by applying two blocks-two factors, ten runs. Software Statistica 8.0 (StatSoft, Dell Software, Texas, United States) was used in this study. A second-order model can be constructed efficiently with this methodology [17].

$$Y = R_0 + \sum_{i=1}^{k} R_i X_i + \sum_{i=1}^{k} R_{ii} X_i^{\,2} + \sum_{i=1,i<j}^{k-1} \sum_{j=2}^{k} R_{ij} X_i X_j + \varepsilon \quad (3)$$

where Xi, Xj is input variables that affect the response Y; Ro, Ri, Rii and Rij (i = 1-k, j = 1-k) is a known parameter, and ε is a random error. A second order model is designed so that the variance of Y is constant for all points equidistant from the center of the design.

$$X_i = \left(\frac{X_i - X_0}{\Delta X_i}\right) \quad (4)$$

where Xi is the value of the code, Xo is the actual value at the center point and ΔXi is the value of the pace of change [18]. The dependent variable was TAC ($Y_1$) and TFC ($Y_2$), while the independent variables were concentration of rambutan peel (%, $X_1$) and fixative content (%, $X_2$) regarding to the fixative type ($CaCO_3$ and $FeSO_4$). The range of experimental levels values applied on RSM study was presented in Table 1.

**Table 1**. Experimental levels values of the independent variables

| Parameters | Variables | Coded levels | | |
|---|---|---|---|---|
| | | -1.000 | 0.000 | 1.000 |
| Concentration of rambutan peel (%) | $X_1$ | 15 | 20 | 25 |
| Fixative content (%) | $X_2$ | 5 | 25 | 45 |
| Type of enzyme (-) | | $CaCO_3$ | | $FeSO_4$ |

## RESULTS AND DISCUSSION

***Dyed cotton fabric***

The application of different fixative types produced variance shades on cotton dyed with rambutan pee extracts after 6 months storage which were shown in Table 2. Applying $CaCO_3$ and $Al_2(SO_4)_3$ as fixative compound resulted shades of brown, while applying $FeSO_4$ produced shades of grey. Shades of brown obtained from $Al_2(SO_4)_3$ shows lighter or

International Journal of Applied Engineering Research ISSN 0973-4562 Volume 12, Number 21 (2017) pp. 10711-10718


pale brown color than $CaCO_3$, which tends to yellowish brown color. Storage during 6 months affect the shades of the cotton fabric, significantly. Increasing the rambutan peel's concentration and decreasing the fixative concentration were resulting in a darker color. The colors were getting mature because the dye molecules are capable of forming metal complex with the positively charged metals. Dye of rambutan peels and metal cations have strong attraction towards negatively charged carboxyl groups of cotton [4].

Cellulose provides cotton absorbent because it contains a negative charge, which facilitate to attract the dipolar of dyed water molecules and absorb them. Moreover, cotton has hydrophilic properties [11] and the capillary action, therefore the cotton fibers can easily suck in water through the interior of the fiber. Once the water drawn in through the fibers, then it stored in the interior cell walls of the cotton and consequently evaporated. After the water dried out, the dye-metal complexed left on the cotton fabric. Cotton fabrics drying with anthocyanin pigments from Morus rubra fruits resulted a saturation red with acceptable fastnesses for potential commercial applications [19].

The anthocyanin from rambutan peel provided the natural dye efficacy for cellulosic fibers at high efficiency, probably due to the existence of acid content, i.e. ellagic acid, syringic acid and p-coumaric acid [6–7]. The anthocyanin content extracted from grape pomace were applied in textile dyeing operations using cotton fabric pre-mordanted with tannin as textile substrate and obtained red/violet shades. However, this biofixative of tannin used need for improvement to fulfil requirements for textile application [20]. The complex formation of the metal ion from fixative mordant and dye stuff possibly changes the color and shades of dyed fabric [4,21].

**Table 2:** Shades of dyed cotton fabric after 6 months of dyeing process

| Type of fixative (-) | $CaCO_3$ | | | $Al_2(SO4)_3$ | | | $FeSO_4$ | | |
|---|---|---|---|---|---|---|---|---|---|
| Concentration of rambutan peel (%) / Fixative content (%) | 15 | 20 | 25 | 15 | 20 | 25 | 15 | 20 | 25 |
| 5 |  |  |  |  |  |  |  |  |  |
| | | | | | | | | | |
| 25 |  |  |  |  |  |  |  |  |  |
| | | | | | | | | | |
| 45 |  |  |  |  |  |  |  |  |  |

***Efficacy of anthocyanin and flavonoid content from rambutan peel's extract***

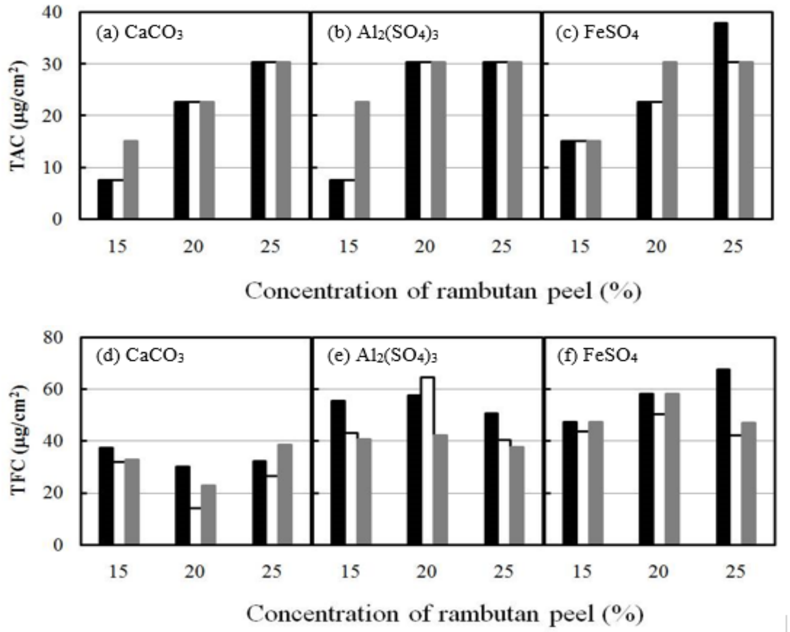


**Figure 1**: The total anthocyanin (TAC; a, b, c) and flavonoid (TFC; d, e, f) content left on the cotton fabric post mordanting by applying $CaCO_3$ (a, d), $Al_2(SO_4)_3$ (b, e) and $FeSO_4$ (c, f) as fixative agent. Fixative concentration: ■, 5 %; □, 25 %, ■, 45 %.

Figure 1a,b,c provide the effect of concentration of rambutan peel on the TAC regarding to the fixative types and concentrations. TAC were regardless on the $Al_2(SO_4)_3$ concentration when applying for 20 and 25 % of rambutan peel concentration, which were the same values at 30.32 µg/cm2. Decreasing the rambutan peel concentration at 15 % decreased the TAC (7.58 µg/cm2), mainly for the cotton treated by 5 and 25 % of $Al_2(SO_4)_3$ concentration. These low value of TAC were also obtained on the $CaCO_3$ treatment. The highest value of TAC obtained in the dyed cotton was found at 25 % of rambutan peel concentration with 5 % of $FeSO_4$ concentration. TAC tends to increase by increasing the concentration of rambutan peel and fixative content. Color variation from anthocyanins can be obtained by varying the pH or metal chelation agents [22]. This result is also in agreement with the data obtained in this work.

Figure 1d,e,f provide the effect of concentration of rambutan peel on the TFC regarding to the fixative types and concentrations. The observed result shows that the TFC detected at 25 % of CaCO3 concentration and 20 % of rambutan peel concentration was the lowest value (µg/cm2). The highest TFC (67.59 µg/cm2) was determined on the 25 % of rambutan peel concentration with 5 % of FeSO4 concentration as fixative agent. The application of $Al_2(SO_4)_3$ gave middle value of TFC detected on cotton fabric comparing to the $CaCO_3$ and $FeSO_4$ application. Brodowska postulated that many dyes present in plants are glycosides [23]. The lower TFC value can possibly promoted by the degradation of flavonoid due to the glycosides breaking, however this breaking compound did not produce the complex compound that should be occure on the natural dyes. The TAC seems to affect color appearance more than the TFC as evidenced by the study of response surface methodology. Significant relationship between nutritional value (including anthocyanin content) provides the color and maturity indices of mango fruits [24].

***Fitting models***

Since the value of TAC and TFC treated with $Al_2(SO_4)_3$ as fixative agent were found as the middle value between the value of TAC and TFC treated with $CaCO_3$ and $FeSO_4$, therefore, the $Al_2(SO_4)_3$ application was not used for the RSM study. Moreover, the $Al_2(SO_4)_3$ treated cotton provided pale shades on the dyed cotton, which provided low quality of color appearance for commercial purposes.

Tabel 3 shows the result of the second order response surface model fitting in the form of Analysis of Variance (ANOVA) of dependence variables. The ANOVA of the regression model of TAC and TFC demonstrates that the Fisher's F-test of TAC (111.91) provided higher value than the Fisher's F-test of TFC (78.81). The significance of each coefficient to the dependent variables was determined by p-values ($p < 0.05$). Its indicated that concentration of rambutan peel ($X_1$) have significance to the TAC ($Y_1$), while type of fixative ($R_0$) significantly affect the TFC ($Y_2$). These results were also strengthened by the value of standardized effects.

**Table 3:** Analysis of variance of process variables as linear, quadratic and interactive terms on response variables treated by applying $FeSO_4$ and $CaCO_3$

| Source of variation | Sum of squares | Degree of freedom | Mean squares | F-value | p-value probe > F | Standardized effects |
|---|---|---|---|---|---|---|
| **Total Anthocyanin Content (TAC)** | | | | | | |
| $R_o$ (blocks) | 13.3008 | 1 | 13.3008 | 2.7050 | 0.1986 | 1.6447 |
| $X_1$ | 469.3611 | 1 | 469.3611 | 95.4546 | *0.0023 | 9.7701 |
| $X_2$ | 38.3152 | 1 | 38.3152 | 7.7922 | 0.0683 | 2.7915 |
| $X_1X_2$ | 14.3682 | 1 | 14.3682 | 2.9221 | 0.1859 | 1.7094 |
| $X_1^2$ | 10.3451 | 1 | 10.3451 | 2.1039 | 0.2428 | 1.4505 |
| $X_2^2$ | 4.5978 | 1 | 4.5978 | 0.9351 | 0.4049 | 0.9670 |
| ε | 14.7513 | 3 | 4.9171 | | | |
| Total sum of squares | 574.7279 | 9 | | | | |
| R-squared | 0.9743 | | | | | |
| Adj R-squared | 0.9230 | | | | | |
| **Total Flavonoid Contenct (TFC)** | | | | | | |
| $R_o$ (blocks) | 1245.1180 | 1 | 1245.1180 | 54.5965 | *0.0051 | 7.3389 |
| $X_1$ | 0.0880 | 1 | 0.0880 | 0.0038 | 0.9545 | 0.0619 |
| $X_2$ | 0.5470 | 1 | 0.5470 | 0.0240 | 0.8868 | 0.1549 |
| $X_1X_2$ | 27.6010 | 1 | 27.6010 | 1.2102 | 0.3516 | 1.1001 |
| $X_1^2$ | 1.7750 | 1 | 1.7750 | 0.0778 | 0.7984 | 0.2790 |
| $X_2^2$ | 522.2540 | 1 | 522.2540 | 22.9000 | 0.0174 | 4.7854 |
| ε | 68.4170 | 3 | 22.8060 | | | |
| Total sum of squares | 1577.6750 | 9 | | | | |
| R-squared | 0.9566 | | | | | |
| Adj R-squared | 0.8699 | | | | | |

*$p < 0.05$

The value of standardized effects were presented as an absolute value, and the highest value indicated the highest influenced parameters on the experimental design [25]. The concentration of rambutan peel gave the highest influenced parameters for TAC, while the quadratic value of fixative content performed the lowest influenced parameters. For TFC, the fixative type performed the highest influenced parameter, while the concentration of rambutan peel plays the lowest influenced parameters.

Applying multiple regression analysis on the experimental data, the following second order polynomial equations were found to represent the TAC ($Y_1$) treated by FeSO4, which provided the optimum value, on Equation (5), as follows:

$$\mathrm{TAC} = -58.8355 + 5.8817\,X_1 - 0.0910\,X_1^2 + 0.3159\,X_2 + 0.0038\,X_2^2 - 0.0190\,X_1X_2 + 2.7292 \quad (5)$$

and the TFC ($Y_2$) treated by CaCO3, which provided the minimum value, on Equation (6), as follows:

$$\mathrm{TFC} = 84.0069 - 2.1881\,X_1 + 0.0377\,X_1^2 - 2.5302\,X_2 + 0.0404\,X_2^2 + 0.0263\,X_1X_2 + 13.2029 \quad (6)$$

The accuracy of those models were performed by the coefficient of determination (R-square/$R^2$). The closer the value $R^2$ to 1, indicates the better of the predicted model to the experimental observed. In this study, the value of the coefficient of determination showed 0.9743 (97.43%) for TAC and 0.9566 (95.66%) for TFC (Figure 2). These result indicated that the TAC observed values provided a better accuracy of model than the TFC observed values.

Figure 3 represent the isoresponse contour and surface plots for the optimization of TAC and TFC by using $FeSO_4$ and $CaCO_3$ as fixative agent, respectively. The effects of the concentration of rambutan peel and fixative on the TAC by applying FeSO4 showed in Figure 3a. The increasing of fixative content did not effect the TAC content, while the increasing of concentration of rambutan peel up to 30 % increased the TAC. Further increasing of rambutan peel concentration, conversely, decreased the TAC. The optimum value for maximum TAC passed from the design of experimental parameter applied for rambutan peel concentration. It indicated that more mature shades possibly obtained by applied 30 % of rambutan peel concentration regardless to the fixative content by using $FeSO_4$ as fixative agent. The contrary effect on the response was observed for TFC (Figure 3b). The minimum value of TFC was observed on the rambutan peel concentration at 20 % and fixative content at 25 % by applying $CaCO_3$ as the fixative agent.

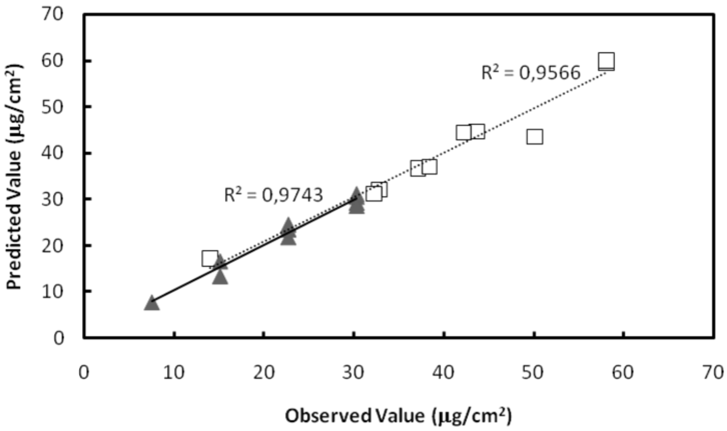


**Figure 2:** Observed vs predicted values of TAC, ▲, and TFC, □

*Response Surface Fitting on Observed Parameters*

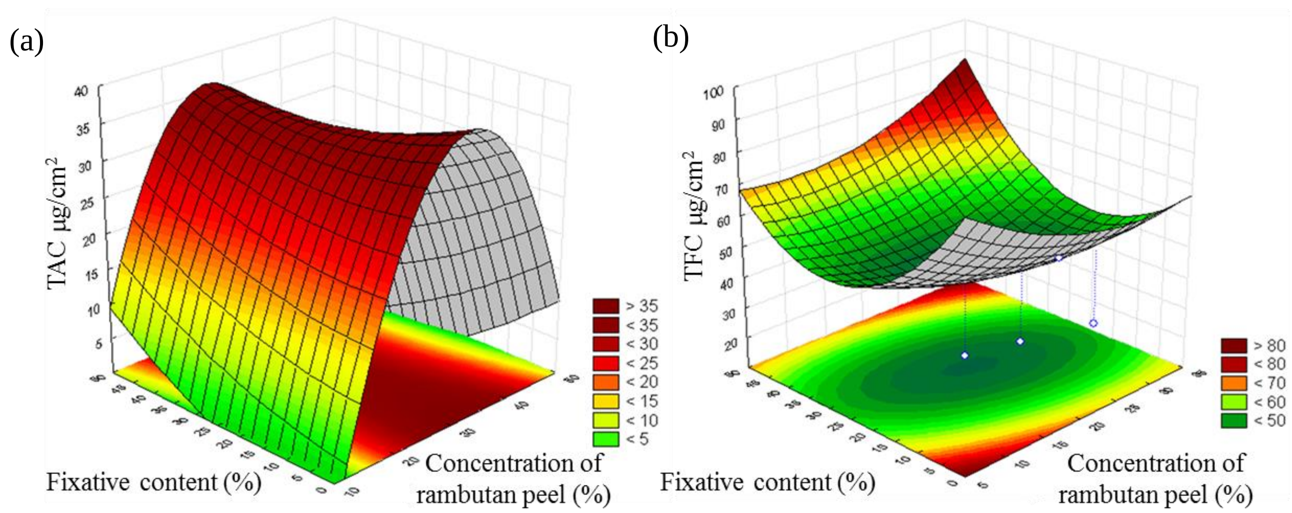


**Figure 3:** 3D-estimated response surface by plotting TAC (a) and TFC (b) versus: fixative content (%) and concentration of rambutan peel (%).

***Optimum dyeing parameters***

The predicted values of optimum value of TAC and TFC on its critical parameter values are shown on Table 4. The critical value for optimum TAC (30.26 μg/cm$^2$) was obtained at 29.09 % of rambutan peel concentration and 31.06 % of fixative content. The predicted critical value of minimum TFC (17.24 μg/cm$^2$) was found at 20.43 % of rambutan peel concentration and 24.67 % of fixative content. These results were strengthened the efficacy of TAC on the color appearance of cotton fabric application more than the TFC. Yin *et al.* optimized purple sweet potato powder for silk fabric dyeing and found 191.09 mg.L$^{-1}$ of natural colorant anthocyanin under 52.48 $^0$C of ultrasound temperature and 48.19 min of ultrasound time [26].

**Table 4:** Predicted values of optimum TAC and TFC on critical values of concentration of rambutan peel and fixative content

| **Factor** | **Critical values** | |
|---|---|---|
| | **TAC** | **TFC** |
| Concentration of rambutan peel (%) | 29.0909 | 20.4349 |
| Fixative content (%) | 31.0606 | 24.6718 |
| Active compound value (μg/cm$^2$) | 30.2569 | 17.2350 |

## CONCLUSION

The concentration of rambutan peel gave the highest influenced parameters for TAC, while the fixative type performed the highest influenced parameter for TFC. The TAC provided a better accuracy of model than the TFC. The predicted optimum values of TAC obtained by using fixative of $FeSO_4$ were found at 29.09 % of rambutan peel's concentration and 31.06 % of fixative content with the coefficient of determination ($R^2$) obtained was 0.9743 (97.43 %).

## ACKNOWLEDGMENT

This work was supported by the Diponegoro University Research Grant [grant numbers 33/UN7.5.13/SK/2017, 2017].

## REFERENCES

[1] Tsatsaroni, E., Liakopoulou-Kyriakides, M., and Eleftheriadis, I., 1998, "Comparative study of dyeing properties of two yellow natural pigments—effect of enzymes and proteins," Dyes Pigm., 37(4), pp. 307-315.

[2] Win, Z. M., and Swe, M. M., 2008, "Purification of the natural dyestuff extracted from Mango bark for the application on protein fibers," World Acad. Sci. Eng. Technol., 46, pp. 536-540.

[3] Saravanan, P., and Chadramohan, G., 2011, "Dyeing of silk with ecofriendly natural dye obtained from Barks of Ficus religiosa L," Univers. J. Environ. Res. Technol., 1(3), pp. 268-273.

[4] Uddin, M. G., 2014, "Effects of Different Mordants on Silk Fabric Dyed with Onion Outer Skin Extracts," Journal of Textiles, 2014, pp. 1-8.

[5] Rather, L. J., Shabbir, M., Bukhari, M. N., Shahid, M., Khan, M. A., Mohammad, F., 2016, "Ecological dyeing of Woolen yarn with Adhatoda vasica natural dye in the presence of biomordants as an alternative copartner to metal mordants" J. Environ. Chem. Eng., 4(3), pp. 3041-3049.

[6] Kumar, B., Smita, K., Cumbal, L., and Angulo, Y., 2015, "Fabrication of silver nanoplates using Nephelium lappaceum (Rambutan) peel: A sustainable approach," J. Mol. Liq., 211, pp. 476-480.

[7] Kumar, B., Smita, K., Angulo, Y., and Cumbal, L., 2016, "Valorization of rambutan peel for the synthesis of silver-doped titanium dioxide ($Ag/TiO_2$) nanoparticles," Green Process Synth., 5, pp. 371-377.

[8] Kulkarni, S. S., Bodake, U. M., and Pathade, G. R., 2011, "Extraction of natural dye from chili (*Capsicum Annum*) for textile coloration," Univers. J. Environ. Res. Technol., 1, pp. 58-63.

[9] Dweck, A. C., 2002, "Natural ingredients for colouring and styling," Int. J. Cosmet. Sci., 24(5), pp. 287-302.

[10] Nam, K. Y., Go, Y. E., Lee, S. Y., and Lee, J. S., 2013, "A Study on Natural Dye Having the Effects on the Atopic Dermatitis – *Juniperus chinensis* Heartwood Extract –," Fibers and Polymers, 14(12), pp. 2045-2053.

[11] Liu, Y., Xin, J. H., and Choi, C. H., 2012, "Cotton Fabrics with Single-Faced Superhydrophobicity," Langmuir, 28(50), pp. 17426-17434.

[12] Cheng, G. W., and Breen, B. J., 1991, "Activity of phenylalanyl ammonialyase (PAL) and concentrations of anthocyanins and phenolics in developping strawberry fruit," J. Am. Soc. Hort. Sci. 116, pp. 865-868.

[13] Giusti, M. M.. and Wrolstad, R. E., 2001, "Anthocyanins characterization and measurement with UV-visible spectroscopy," In: Current protocols in Food Analytical Chemistry Wrolstad R.E. (ed.). J. Wiley, New York, pp. F1.2.1–F1.2.13.

[14] Cao, S., Hu, Z., Zheng, Y., Yang, Z., and Lu, B., 2011, "Effect of BHT on antioxidant enzymes, radical scavenging activity and decay in strawberry fruit," Food Chem., 125, pp. 145-149.

[15] Tonutare, T., Moor, U., and Szajdak, L.., 2014, “Strawberry anthocyanin determination by pH differential spectroscopic method – How to get true results?,” Acta Sci. Pol., Hortorum Cultus, 13(3), pp. 35-47.

[16] John, B., Sulaiman, C. T., George, S., and Reddy, V. R. K., 2014, “Total phenolics and flavonoids in selected medicinal plants from Kerala,” Int. J. Pharm. Pharm. Sci., 6(1), pp. 406-408.

[17] Montgomery, D. C., 1997, “Design and Analysis of Experiments,” 4th edition, John Wiley & Sons, New York.

[18] Box, G. E. P., Hunter, W. G., and Hunter, J. S., 1978, “Statistics for experimenters: an introduction to design, data analysis, and model building,” John Wiley & Sons, New York.

[19] Wang, H., Hussain, M., and Zhou, W., 2016, “A novel method for natural dyeing of cotton fabrics with anthocyanin pigments from Morus rubra fruits,” Textile Research Journal, 87(15), pp. 1896-1911.

[20] Bechtold, T., Mahmud-Ali, A., and Mussak, R, 2007, “Anthocyanin dyes extracted from grape pomace for the purpose of textile dyeing,” J. Sci. Food. Agric., 87(14), pp. 2589-2595.

[21] Bechtold, T., and Mussak, R., 2009, “Handbook of Natural Colorants,” JohnWiley & Sons,West Sussex, UK.

[22] Wang, H., Li, P., and Zhou, W., 2014, “Dyeing of silk with anthocyanins dyes extract from Liriope platyphylla fruits,” Journal of Textiles, 2014, pp. 1-9.

[23] Brodowska, K. M., 2017, “Natural flavonoids: classification, potential role, and application of flavonoid analogues,” Eur. J. Biol. Res., 7(2), pp. 108-123.

[24] Uddin, M. J., Robbani, M., Hasan, M. F., and Rahman, H., 2017, “Development of fruit maturity color chart of Bari Aam-3,” Int. J. Agric. Environ. Res., 3(4), pp. 3544-3560.

[25] Box, G. E. P., and Draper, N. R., 1987, “Empirical model-building and response surfaces,” John Wiley & Sons, New York.

[26] Yin, Y., Jia, J., Wang, T., and Wang, C., 2017, “Optimization of natural anthocyanin efficient extracting from purple sweet potato for silk fabric dyeing,” J. Clean. Prod., 149, pp. 673-679.