The degreening of “Selayar” orange using ethephon: The color peel changes and ethephon residue

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Abstract. The degreening technology using etilen to improve the color of “Selayar” orange peel is impracticable to apply because this exists the gases form, so it can be substituted with working use Ethephon. Ethephon is more practical to use because this is a liquid. However, its use requirement prioritizes the principle of caution because it leaves residual. This study aimed to analyze the colour changes of the orange peel and Ethephon residues in degreening applications. This study used a completely randomized design with three concentration treatments, namely 0 ppm, 500 ppm, and 1000 ppm, with four repetitions. This research showed that the Ethephon in concentration 500 ppm was not significantly different from the 1000 ppm etaphon treatment on fruit peels colour based on L *, a *, b *, colour index (CCI), and hue (h). Both concentrations exist to develop the orange peel colour, while the control fruits (0 ppm) are solely green on the 7th day of observation and greenish-yellow on the 14th day. Ethephon residue on orange fruit peels for 1000 ppm treatment approximately 0.30 ppm while at the group of 500 ppm ended 0.03 ppm. Degreening with etaphon concentrations of 500 ppm and 1000 ppm has the identical ability to improve the peel colour of “Selayar” orange. The ethaphon residue in both concentrations (500 and 1000 ppm) is more moderate than the maximum admitted by the Indonesian government and Codex, namely 0.05 ppm.

1. Introduction

Increasing public awareness about health has led to an increase in demand for fresh orange fruits in Indonesia. Orange fruits are known to contain vitamin C (ascorbic acid) and phytonutrients. It such as lycopene, potassium, bioflavonoids and folic acid as antioxidants, anti-cancer [1–7], antimutagenic and positively associated with bone, heart, and immune system [7,8]. Consumption of orange fruit also contributes to lowering the incidence of death from cardio-cerebrovascular disease [5,6].

Consumers prefer orange-coloured oranges [9] which are generally imported oranges. Meanwhile, the peel of orange fruit produced by farmers in Indonesia tends to be yellowish-green even though it is ripe [2]. This is because it is grown in the lowlands so that the fruit is not exposed to cold temperatures [10]. One of the technologies that can be used to change the colour of orange peels is degreening. Orange degreening using ethylene (C2H4) is commonly used to improve the skin colour of fresh oranges [10–13]. However, ethylene is a gas that is colourless [14]. Its use requires special knowledge and equipment, the price is high, so it is difficult for orange farmers to apply in Indonesia.

Ethephon (2-chloroethyl phosphonic acid) can be used as an ingredient that substitutes ethylene in degreening and has been widely used by fresh orange fruit producers in several countries. Ethephon is very easy to use; oranges are only dipped in ethephon solution with a specific concentration and duration.
of time. Ethephon is the fruit tissue that will be hydrolyzed to produce ethylene, chlorine ions, and phosphate. Ethephon is a compound that spontaneously releases ethylene upon contact with water [15].

Several previous degreening studies have used Ethephon at different concentrations. Shamouti, Washington Navel Oranges, Eureka and Taiwan Lemon use Ethephon at 500–12,000 ppm concentrations. In India, Mosambi, Hamlin and Valensia oranges use Ethephon at 1000–3000 ppm [10]. Common mandarin orange degreening uses concentrations of 1000 ppm [9,16–18]. However, currently, no studies look at lower concentrations and the amount of Ethephon residue that can contaminate the fruit’s skin, even though this aspect is essential in food safety.

Ethephon can affect the inhibition of cholinesterases in blood plasma and red blood cells for a long time in dogs and rats. Therefore, the presence of Ethephon can accumulate and is very dangerous for human health [19] The maximum tolerable amount of Ethephon residue is 0.5 ppm.

The application of degreening technology still has to prioritize the principle of precaution. Therefore, it is necessary to research the use of Ethephon mandarin oranges at concentrations lower than 1000 ppm and measuring the amount of Ethephon residue on the fruit’s skin. This research aimed to study low concentration degreening of Ethephon against changes in orange peel colour and to determine the residual levels of Ethephon in degreened orange peels.

2. Research Method

2.1. Research Location
The research was conducted from August to October 2019. The sample from Selayar oranges with the same age and maturity stage. Oranges are harvested from smallholder gardens in Bantaeng Regency, South Sulawesi. The degreening treatment was carried out at the PKP Laboratory of Hasanuddin University, Makassar. Observation of fruit colour used 48 samples of orange fruit, where each treatment used four samples of orange fruit which was repeated four times. Observation of the Ethephon residue was carried out using the mixed method by taking four oranges each for each replication for each treatment so that the whole orange fruit on the Ethephon residue test was 48 oranges.

2.2. Degreening Procedure
The ethephon used is a formulation of Ethrel 480 SL (a product from Bayer CropScience) equivalent to 2-chloroethyl phosphonic acid, 480 ppm diluted according to research treatment (0 ppm, 500 ppm and 1000 ppm) with the following equation:

\[ v_1n_1 = v_2n_2 \].

Orange fruits are immersed in a solution according to treatment (0 ppm, 500 ppm and 1000 ppm) for 60 s, then dry to air at room temperature. After that, it was placed in a cold room at 20 °C [16,17] for three days [2], then it was removed at room temperature for observation.

2.3. Colour change measurement
Changes in the colour quality of orange peels were measured using a Color Reader (KONICA MINOLTA, Japan, CR-400). This tool has a colour notation system (colour system L, a, and b). The L notation states the brightness parameter with a colour range of 0 to 100. The value of L 0 means black, and the more it points to the value 100, the whiter or brighter it means. The a’s notation states the chromatic colour of the red-green mixture with a value of + a (positive), the higher the red colour, and vice versa. The value is 0 to -80 for the green colour. The b * notation represents a blue-yellow mixed chromatic color with a -b (negative) value from 0 to -70 for blue and a + b * (positive) value from 0 to +70 for yellow [20].

The orange colour change movement can be measured by converting a* and b* values into chromatic units of degrees h (hue) and expressed from 0 to 360. The hue value describes the dominant colour in a mixture of several colours. Based on Manera et al. (2012) the hue value used the equation [21]:

\[ \text{Hue (°h)} = \tan^{-1}(b/a) \].
Colour measurement was also observed by measuring the orange colour index (CCI) for fruit skin colour based on [22] Jimenez-Cuesta, et. al. (1981) with the equation:

\[ \text{CCI} = \frac{1000 \times a}{b \times L} \]

2.4. Ethephon Residue Determination
The procedure for determining Ethephon Residue is based on the Indonesian Directorate of Plant Protection (2006), where pure Ethephon in acetone is used as the standard (98%). The orange peel sample was extracted as much as 25 g and then added with 5 ml of acetone and 5 ml of chloromethane. Stir for 30 minutes, then filtered using filter paper. Then evaporated until it reaches ± 50 mL. The evaporation results are then purified, bypassing the sample to a chromatography column filled with phosphoric ± 3 g and anhydrous sodium sulfate and evaporated to a volume of only ± 1 mL. Next, the tube is rinsed with 65% methanol gradually. Finally, the tube is accommodated in the test tube up to a volume of 10 mL. The samples were injected and detected used a photodiode array (PDA) at wavelength UV-VIS 254nm. The determination is made by comparing the retention time on the standard and the sample.

2.5. Data Analysis
Data were analyzed using Excel 2013 and SPSS 16.0 software, and the results were expressed as means. Statistical comparisons were made with a one-way analysis of variance (ANOVA) followed by Duncan’s multiple range test and significant differences at P <0.05.

3. Result and Discussion

3.1. Discoloration
Table 1 shows that the citrus fruits before treatment (at 0 HSP) had compact green skin. Based on colour measurements in the notation L (brightness), a (green-red) and b (blue-yellow), there were no significant differences (P <0.05). This proves that the samples used have a uniform skin colour.

The colour change of tangerine was visible at storage day 7 to day 14 for all treatments based on the notation L (brightness), a * (green-red) and b * (blue-yellow). Based on Chakraborty et al. (2021) [20], the higher the value of a *, the indication that the product is getting redder and vice versa. The value from 0 to minus (-) of a product means that it is green. On the other hand, the b* notation indicates a blue-yellow mixed chromatic colour; if the value of b * is positive (+) means yellow and minus (-) means blue. Table 1 shows that the brightness notation (L *) in the 500-ppm treatment group was not significantly different from the 1000 ppm treatment but substantially different from the control. Likewise, in a* and b * notations in the two treatment groups, 500 ppm and 1000 ppm concentrations showed that they were not significantly different. However, the values of both were significantly higher than those without treatment.

| Storage Time, days | Concentration, ppm | Color measurements parameters |
|-------------------|-------------------|-------------------------------|
| 0 day             |                   |                               |
| 0                 | 0                 | 46.72 a -3.11 a 34.72 a       |
| 500               |                   | 50.86 a -2.18 a 33.55 a       |
| 1000              |                   | 49.98 a -2.77 a 33.10 a       |
| 7th day           | 500               | 47.65 b 0.95 b 36.62 b        |
|                   | 1000              | 58.86 a 18.81 a 58.86 a       |
|                   | 0                 | 60.00 a 17.9 a 54.37 a        |
| 14th day          | 500               | 52.25 b 10.12 b 39.38 b       |
|                   | 1000              | 61.35 a 23.27 a 61.55 a       |
|                   |                   | 58.10 a 22.14 a 56.26 a       |
Fruit skin colour on the colour index (CCI) measurement based on Jimenez-Cueata et al. (1981) showed that there were differences in skin colour between those treated with Ethephon (500 ppm and 1000 ppm) and control. The Ethephon treatment with the concentrations of 500 ppm and 1000 ppm were seen changing together to orange-yellow, while the control was still green on the 7th day of observation. The CCI value at the two concentrations drastically increased together while the control tended to be stable (Figure 1). This is also reinforced based on the measurement of the hue angle (h) (Figure 2). It can be seen that the two concentrations move sharply downward from green to orange-yellow simultaneously, while the decrease control does not show a significant colour movement during the 7th day of storage. At storage on the 14th day, the fruit colour became brighter orange for both Ethephon treatments, while the control colour was starting to change but only formed a pale yellow colour.

This study shows that based on the CCI and hue values, the degreening treatment using Ethephon at a concentration of 500 ppm was not significantly different from the treatment with a concentration of 1000 ppm. Therefore, it can be said that the Ethephon treatment with a concentration of 500 ppm has the same ability as a concentration of 1000 ppm to improve the colour of the orange peel.

This research also shows that naturally, chlorophyll degradation can occur in Selayar tangerines. However, the skin only forms a yellow colour and is not compact because there are still green patches. With degreening treatment using Ethephon at a concentration of 500 ppm and 1000 ppm, the degradation of chlorophyll becomes faster, and the colour formed becomes bright orange. Dегreening causes exposure to carotenoids previously dominated by chlorophyll [9]. Specific carotenoids can be synthesized before degradation of the chlorophyll structure, and also particular carotenoids can be synthesized when the chlorophyll structure is degraded [10]. In this study, oranges with Ethephon treatment can form the orange peel of Selayar to become bright orange, while without Ethephon, it is greenish-yellow. It appears that there is a specific carotenoid pigment (such as red pigment) in citrus,
which is formed after chlorophyll degradation occurs. One of the particular pigments that formed is the red pigment (β-imageurin). This pigment causes the colour of the fruit to turn red [10,23].

3.2. Ethephon residue on orange peels

Observation of Ethephon residue on the skin in degreening the citrus fruit showed a difference in each concentration treatment. This study proves that the higher the Ethephon concentration used, the greater the residual level.

| Ethephon concentration, ppm | Sample Residue, ppm | Standard concentration, ppm |
|-----------------------------|--------------------|-----------------------------|
| 0                           | 234                | 134186                      |
| 500                         | 53043              | 134186                      |
| 1000                        | 48543              | 134186                      |

The degreening treatment of oranges generally uses Ethephon to change the skin colour that consumers prefer, namely orange. However, over a long period, Ethephon can cause inhibition of cholinesterases in the blood and accumulate in the body so that it is harmful to human health, as in tests that have been done on dogs and rats [19]. One effort that can be made is to reduce the concentration used. The Indonesian government and world food security (Codex) have set a threshold for Ethephon residue for Mandarin oranges at 0.5 ppm. This study indicates that degreening at a concentration of 1000 ppm. There is a residue on the skin of 0.310 ppm, while at a concentration of 500 ppm. The residue of Ethephon on the skin is lower than 0.033 ppm. This proves that the two concentrations do not pass the recommended threshold.

4. Conclusions

Degreening of citrus fruits using Ethephon with a concentration of 500 ppm resulted in the same orange peel colour with a concentration of 1000 ppm. Ethephon residue on the orange peel that was degreened at these two concentrations did not exceed the threshold that endangers human health.

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