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The Use of Dithizone for Lead Analysis in Blush

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***Abstract*—Objectives: In this research, lead analysis was carried out on the blush cosmetic preparations using the UV-Vis Spectrophotometry method and dithizone complex. The purposes of this study are to determine lead levels in cosmetics blush preparations and to determine the safety of the blush preparations that are circulating concerning the provisions of BPOM. The results obtained by the regression equation y = 7.895x + 0.0701 with a correlation coefficient of 0.9984; the detection limit was 0.00596 ppm and the limit of quantitation was 0.0198 ppm. Accuracy tests result the percent recovery of 80, 100, and 120% concentrations respectively 94.08; 97.86; and 98.77%. The results of consecutive precision tests are 0.9297; 0.5098; 0.3351; 0.1565; and 0.1842%. The blush sample preparation process is carried out with the dry destruction method, which goes through the drying and graying stages. The destroyed ash was added with 6 M hydrochloric acid, then drying again. The drying product was dissolved with 0.1 M nitric acid. Dry destruction digestion sample solution was used for the analysis of lead content in the blush sample. The test result showed that the lead content in the sample was 0.0599 ppm. Based on the BPOM's Head Regulation No.17 of 2014 on lead contamination in cosmetics must not exceed 20 ppm, so the blush cosmetic is still below the threshold set by BPOM and still safe to use.**

Keywords: dithizone, limit of detection, limit of quantification, lead, blush on availability

I. INTRODUCTION

Lead is toxic to humans, which can originate from the habit of consuming food, drinks, or through inhalation of air, lead contaminated dust (Pb), contact through the skin, contact with the eyes, and parenteral. In the human body lead can inhibit the activity of enzymes involved in the formation of hemoglobin (Hb) so that it can cause anemia, lead can also cause brain damage, aminosiduria, cause increased vascular permeability, disrupt the reproductive and endocrine systems, and are carcinogenic in high doses [7].

Every woman would want to always look beautiful. This desire can be realized by using various kinds of cosmetics such as powder, lipstick, eye shadow, blush, etc. But many women do not realize that among these beauty products may contain hazardous materials such as heavy metals. Eye shadow preparations from the Kiaracondong Market, that in one of the eye shadow preparations that was not registered contained lead of 127,356 ppm [2], which exceeded the requirements set by the BPOM's Head Regulation No 17 of 2014. .

Regulation of the Head of the Drug and Food Supervisory Agency of the Republic of Indonesia Number 17 of 2014 concerning Amendments to the Regulation of the Head of the Drug and Food Supervisory Agency Number HK.03.1.23.07.11.6662 of 2011 concerning the Requirements of Microbial and Heavy Metal Contamination in Cosmetics, states that the limit of lead contamination in cosmetics is no more than 20 mg / kg or 20 mg / L (20 ppm). Metal analysis is usually carried out by the Atomic Absorption Spectrophotometry (AAS) method used for the determination of metal elements contained in solutions with very small concentrations. Although this method has been validated, the availability of the instrument is still limited, so it can use another method, the UV-Vis Spectrophotometry method as an alternative. Measurement of lead content using the UV-Vis Spectrophotometry method can be done using a dithizone complexing so that it can produce Pb-ditizonate complex compounds that can absorb radiation at UV-Vis wavelengths [6]. However, to use the UV-Vis Spectrophotometry method must be validated first, so that the method must be validated with parameters of linearity, accuracy, precision, limit of detection, and limit of quantitation.

Based on this background, a study was carried out on the analysis of lead in blush preparations using the UV-Vis spectrophotometric method using the dithizone complex. Thus we can find out how and how much lead content is contained in blush on.

II. MATERIAL AND METHOD

A. Preparation of $Pb(NO_3)_2$ Solution

Make a $Pb(NO_3)_2$ solution with a concentration of 1000 ppm. Pipette as much as 10 mL of $Pb(NO_3)_2$ solution of 1000 ppm, put into a 100 mL volumetric flask. Then add with aqua d.m until the boundary mark. The concentration of $Pb(NO_3)_2$ 100 ppm was obtained.

B. Determination of Maximum Wavelength

From a 100 ppm $Pb(NO_3)_2$ solution pipetted as much as 5 mL, put into a 50 mL volumetric flask, add aqua demineralization up to the mark (obtained $Pb(NO_3)_2$ solution with a concentration of 10 ppm) in a $Pb(NO_3)_2$ 10 ppm solution NH_4OH 1 N was added to pH 10 and 5 mL of 10% KCN. Extraction was carried out in a separating funnel with 0.001% dithizone solution in chloroform until it was perfect (until it was not red). Then the maximum wavelength of the Pb-dithizonate complex was measured with a UV-Vis spectrophotometer in the wavelength range of 400-800 nm.

C. Determination of Complex Stability

Standard solution $Pb(NO_3)_2$ with a concentration of 10 ppm, which has been extracted with 0.001% dithizone solution in chloroform at pH 10, then absorbance is measured in an interval of 5 minutes until there is a change in absorbance.

D. Sample Preparation

Weigh 5 grams of sample, then spiked with a standard solution of $Pb(NO_3)_2$ concentration of 0.02 ; 0.04 ; 0.06 ; 0.08 and 0.1 ppm. The spiked sample is transferred into a porcelain crucible for destruction. Then the spiked sample is dried on a hot plate while adding concentrated HNO_3, then the temperature is raised to 80-100 ^oC to dry. Then the spiked sample is blushed in the furnace, the furnace temperature is set to 250 ^oC, then slowly the temperature is raised to 350 ^oC with each increase of 50 ^oC. Then the temperature is raised to 500 ^oC spiked sample is allowed to ash. The furnace is turned off, and let it cool, the porcelain crushing is removed from the furnace and allowed to cool and store in a desiccator. The resulting ash was dissolved with 5 mL 6 M HCl, then dried on a hot plate. The drying product was dissolved with 0.1 M HNO_3, then filtered with Whatmann paper into a 10 mL volumetric flask, and set it to the limit mark with 0.1 M of HNO_3.

Solution in a pipette, add NH4OH 1 N to pH 10 and 5 mL of 10% KCN then extracted with 0.001% dithizone solution in a separating funnel then shake strongly, allowed the layer to separate and form a red color on the chloroform layer. Extraction is done until the chloroform layer is no longer red. The extraction product is collected in a 10 mL volumetric flask and adjusted to the limit mark with chloroform.

E. Determination of Calibration Curves

Spiked sample extraction results from standard solution $Pb(NO_3)_2$ 0.02; 0.04; 0.06; 0.08; and 0.1 ppm which has been extracted with 0.001% dithizone in chloroform, measured absorbance at a wavelength of 516 nm.

F. Determination of Linearity, Limit of Detection, and Limits of Quantification

Linearity determination is done from spiked concentration of standard solution $Pb(NO_3)_2$ 0.02; 0.04; 0.06; 0.08; and 0.1 ppm was extracted in a separating funnel with a dithizone solution in chloroform. Measure the absorbance at a wavelength of 516 nm, then a calibration curve is made between concentration (x) and absorbance (y), so that the regression equation is obtained $y = bx + a$, the value of the correlation coefficient (r) and the coefficient of variation of function (VX0). Next, calculate the limit of detection (LoD) and the limit of quantification (LoQ) of the linear equation obtained

G. Precision Determination

The precision was determined from the absorbance of the sample spiked standard solution $Pb(NO_3)_2$ 0.02; 0.04; 0.06; 0.08; and 0.1 ppm. From the absorbance results the standard deviation (SD) was calculated, then calculate the coefficient of variation value.

H. Determination of Accuracy

Accuracy is determined from the concentration of spiked $Pb(NO_3)_2$ 0.04 ppm as a concentration of 80%, 0.05 ppm as a concentration of 100% and 0.06 ppm as a concentration of 120%. Measured as percent recovery (% recovery) of analytes added to the measurement

I. Measurement of Lead Content in Samples

The extracted sample solution was analyzed using UV-Vis spectrophotometry at a wavelength of 516 nm, then a calibration curve was made between concentration (x) and absorbance (y). To get the lead content in the sample, the absorbance value is substituted into the equation $y = bx + a$

III. RESULTS

Based on UV-Vis spectrophotometric measurements, the Pb-ditizonate complex of standard solution $Pb(NO_3)_2$ forms a red complex that has a maximum wavelength of 516 nm (Figure 1).

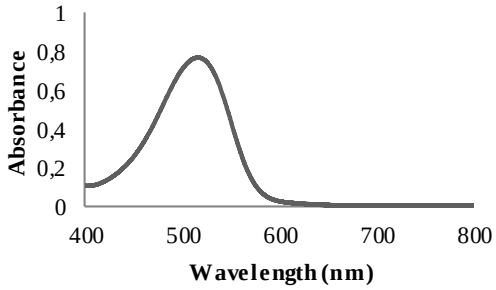

Figure 1. UV-Vis Spectrum of Pb-ditizonate complex

Absorbance was stable at 30 minutes for 20 minutes (Figure 2). Measurement of the stability of the Pb-ditizonate complex is done every 5 minutes for 1 hour.

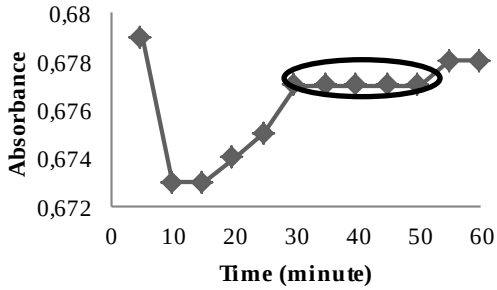

Figure 2. Pb-ditizonate complex stability curve to time

Calibration curves are made by means of absorption measurements with a concentration range of 0.02; 0.04; 0.06; 0.08; and 0.1 ppm. Based on the measurement results from the spiked uptake calibration curve obtained a regression equation $y = 7.895x + 0.0701$ with a correlation coefficient (r) of 0.9984 (Figure 3).

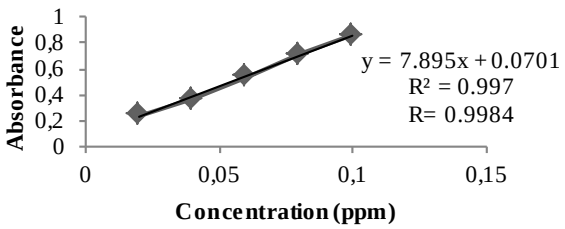

Figure 3. Pb-ditizonate Complex Calibration Curve

IV. DISCUSSION

Determination of maximum wavelength is the basis for qualitative and quantitative analysis of the UV-Vis Spectrophotometry method. The maximum wavelength is the wavelength that can provide maximum absorbance at the time of measurement [4]. The Pb-ditizonate complex is formed because the dithizone compound has the -NH group as a donor of electron pairs, which can bind to the Pb metal. This complex has chromophore and auxochrome groups. The maximum wavelength of this complex is 516 nm.

The measured complex stability shows the relationship between absorbance and time. Determination of complex stability aims to get the measurement time when the reaction has been running optimally which is marked by a stable absorbance.. Samples before destruction were spiked with a standard solution of $Pb(NO_3)_2$. This spiked method is used to increase the concentration of the analyte in order to increase the reading of the instrument, so as to reduce errors [5]. Separation of analytes from organic compounds in blush samples was carried out by dry destruction.

This destruction process is carried out by means of two stages, namely the process of charcoal and graying. In the charcoal process, the sample is added to the concentrated HNO_3 solution. The use of concentrated HNO_3 is because concentrated HNO_3 is a strong oxidizing agent that can decompose organic matrices in a sample, so the reaction of lead breakdown of organic compounds in the sample takes place quickly [1].

Then, the sample is heated on a hot plate to vaporize the concentrated HNO_3 added. After drying, the next stage is the graying process. The graying process is carried out with a furnace with an initial temperature of 250 ^oC which is then gradually raised to a temperature of 500 ^oC until the sample becomes ash. Conditioning temperature of 500 ^oC is done to prevent the evaporation of analytes, because lead can evaporate at temperatures of 550-600 ^oC and can react with oxygen in the air to form Pb oxide [2]. Samples that have turned to ash are added to a solution of hydrochloric acid with a concentration of 6 M which serves as a catalyst to accelerate the reaction of lead breakdown in organic compounds present in the sample.

The factors that influence the increase in selectivity and stability of dithizone compounds in forming the Pb-ditizonate complex are pH, the presence of other metals and the formation time of the complex. In this study, the optimum pH at 10. The presence of other metals in the sample at the time of measurement needs to be considered, because other metals that can be treated with dithizone, especially metals which have properties and characteristics that are almost the same as the Pb to be analyzed, with the addition of potassium cyanide (KCN) as a masking agent that can suppress the influence of other metals, where cyanide ions in the pH range 9-12 can mask metal ions other than Pb such as nickel (Ni), zinc (Zn), copper (Cu), and cadmium (Cd) which may be in the sample forming a stable and strong complex.

The spiked method was chosen because the analyte content in the sample was very small, so as to increase the sensitivity in the measurement with the addition of a standard solution concentration. Based on the above data (figure 3) it can be concluded that the method meets the acceptable linearity requirements because the correlation coefficient (r) value is greater than 0.995. Thus, the method produces good linearity.

After obtaining a calibration curve that meets the analysis requirements, then the data obtained from the concentration of each analyte that provides different absorbance is processed to obtain the detection limit and the limit of quantification. The limit of detection is the lowest analyte concentration in a sample that can still be detected [3].

Based on the calculation results of the limit of detection and the limit of quantification using the standard solution spiked method, the detection limit value is 0.00596 ppm and the quantitation limit is 0.0198 ppm. The limit of quantitation is defined as the lowest analyte concentration in the sample. The limit of quantitation was carried out to determine the level of the smallest analyte in the sample.

Precision is usually expressed by the coefficient of variation. Precision can be seen from the repetition of standard lead levels added to the sample by the spiked process, which will later be shown by the coefficient of variation values where the conditions for a good variation coefficient value of more than 2% [3].

The coefficient of variation in the individual spiked lead ion method were 0.9297%, 0.5098%, 0.3351%, 0.1565%, and 0.1842%, respectively. Based on these results it can be seen that the spiked method meets the permitted limits. Careful criteria are given if the method provides a relative standard deviation value or coefficient of variation of 2% or less, indicating that the precision parameter provides a well-accepted repeatability [3].

Accuracy is a measure that shows the degree of closeness of the analysis results with the actual level of the analyte. Accuracy is expressed as a percent recovery (% recovery) of analytes added 80%, 100%, and 120%. The concentration used is 0.04 ppm as a concentration of 80%, 0.05 ppm as a concentration of 100%, and 0.06 ppm as a concentration of 120%. The results of the recovery test (% recovery) with the spiked concentrations were 94.08%, 97.86% and 98.77%, respectively. This shows that the data obtained can meet the recovery criteria, namely 80-110%.

The validated method was applied to blush on samples that could potentially contain lead, which was carried out by spiked a series of concentrations of standard $Pb(NO_3)_2$ solution. Measurement of lead content from spiked samples obtained a regression equation y = 4.26x + 0.408 with a value of r = 0.9975. Lead content in the sample is calculated by entering the absorbance value of the sample in the linear regression equation y = 4.26x + 0.408.

Based on the determination of lead levels in blush on preparations with a UV-Vis spectrophotometer calculated when y = 0 so that the levels of lead obtained in the sample of 0.0599 ppm. Judging from the Head of BPOM Regulation No. 17 of 2014, regarding the limit of lead heavy metal contamination in cosmetics must not be more than 20 ppm, so that the cosmetic blush is still below the lead contamination threshold, so that the cosmetic blush is still safe for use by the public.

V. CONCLUSION

Validation of the analytical method that has been done on the analysis of lead content by the UV-Vis Spectrophotometry method obtained linear regression equation y = 0.7895x + 0.0701 with a correlation coefficient (r) 0.9984. The limit of detection value is 0.00596 ppm and the limit of quantification is 0.0198 ppm. Accuracy test results as percent recovery at 80%, 100%, and 120% respectively 94.08%; 97.86%; and 98.77%. Precision is usually expressed by the coefficient of variation value. The coefficient variations in the method of spiked concentration 0.02-0.1 ppm were 0.9297%; 0.5098%; 0.3351%; 0.1565%; and 0.1842%. The result of the determination of the lead content obtained from the blush on sample is equal to 0.0599 ppm. Based on this result, the lead content in the blush on sample still met the maximum limit requirements for lead heavy metal contamination in cosmetics.

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