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## Determination of Boric Acid Levels in Food Samples Using the UV-Vis Spectrophotometry Method

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**Abstract.** Food is a basic need for every human being to grow and sustain life. In food, there is what is called BTP or Food Additives. According to the Regulation of the Minister of Health of the Republic of Indonesia No. 033 of 2012 [1], Regarding food additives, boric acid or borax is prohibited from being used in food products. However, many people still use boric acid as BTP in producing food. This study aimed to identify and determine boric acid levels in food samples. The research method used was qualitative analysis with the addition of liquid curcumin and quantitative analysis using UV-Vis spectrophotometry at a maximum wavelength of 540 nm. The results of identification and determination of boric acid levels showed that sample A had a color change from purplish-red to orange, indicating that the food sample was positive for boric acid. The average level of boric acid in sample A equals 986.947 ppm. The %RSD value obtained is 1.583%, indicating that this test has good precision.

**Keywords :** Boric acid, Borax, UV-Vis Spectrophotometry

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## Introduction

Food is a basic need for humans. Every human being needs food to grow and sustain life. In addition, food also serves as a source of energy for humans to move. According to Drug Control Agency Regulations and Food No. 28 of 2019 [2], regarding processing aids food, food is everything that comes from biological sources of agriculture, plantation, forestry, fishery, animal husbandry, aquatic and water products, both processed and unprocessed, which are intended as food or drink for human consumption including food additives, raw materials food, and other materials used in the process of preparing, processing, or making food or drink. Food safety is a problem that must get special attention, especially in developing countries like Indonesia. One of the causes of the lack of food safety in Indonesia is the common knowledge, skills, and responsibility of food producers regarding the quality and safety of food, especially in small industries or home industries. The tendency to misuse some hazardous chemicals in food is the main trigger. One of the dangerous chemicals that is often misused in food is borax or in the form of boric acid water.

Boric acid is a chemical compound in the form of a white powder that can dissolve in water and is present as a colorless crystalline solid. Boric acid is widely used in the glass industry, waterproof wood coatings, cement, porcelain pelican, cleaners, preservatives, and ant exterminators [3]. Boric acid has a molar mass of 61.832 gram/mol and a density of 1.435 g/cm<sup>3</sup>. Boric acid dissolves in water with a solubility of 5.7 grams per 100 mL of water at 250°C. The crystalline phase of boric acid consists of layers of B(OH)<sub>3</sub> molecules held together by hydrogen bonds [4]. Boric acid has anti-septic or antibacterial properties that can inhibit the growth of decomposing microorganisms so that food stays fresh and lasts longer. In addition, the addition of boric acid can control the gelatinization of starch, causing increased elasticity and giving a savory taste to starch foods [5].

Since 1982 the government has banned the use of boric acid and reinforced by Permentkes No. 33 of 2012 [1] regarding Food Additives, compounds that are prohibited from be-

ing added to food include boric acid. However, even though it is prohibited, some people are still stubborn to add borax on the grounds of seeking profit. The adverse effects can last a long time, even if it is used in small amounts. The main danger posed by boric acid if exposed continuously is that it can irritate the respiratory tract if inhaled, cause skin blisters if in contact with skin, nausea, vomiting, diarrhea, headaches, and hypotension. Further symptoms are characterized by weakness in the body, kidney damage, even shock and death if ingested, with a fatal dose of 15-20 grams for adults and 3-6 grams for children [6].

Due to the significant impact of boric acid on public health, the government has taken several actions against the misuse of this food additive (BTP). One manifestation of the government's particular action and attention to this phenomenon of boric acid abuse is conducting surveillance activities on the circulation of products containing boric acid to consumers by conducting tests on food products circulating in the community. Testing the borax content in food was conducted qualitatively and quantitatively using UV-Vis Spectrophotometer instrumentation.

Rahma and Hidjrawan (2021) [7] have researched the analysis of boric acid using the natural ingredient turmeric in food samples. The results showed that 10 test samples were positive for boric acid which was indicated by the change in the color of the test samples to brownish red. The sample that contains the most boric acid is salted fish. In this study, the method used was only a qualitative method where the value of the boric acid content obtained was not known with certainty. On this basis, it is necessary to analyze using a quantitative method so that the positive sample is known how much boric acid is. Based on Munandar's research (2022) [8], quantitative analysis using a UV-Vis spectrophotometer is more selective. It has high accuracy with a relative error of 1-3%, so even minimal quantities of substances can be determined quickly and precisely. This study was used to observe the use of boric acid in food.

## Method

### Research Design

This test method refers to MA PPOMN years 2000 No. 07/MM/00.

### **Time and Place of Research**

This research was conducted from June to August 2022 at the Nutrition and Food Laboratory of the Center for Drug and Food Control in Padang, West Sumatra.

### **Tools and materials**

#### **Tool**

The equipment used in this study were a porcelain crucible, crucible lid, AL204 Analytical Balance, Memmert WNB14RING water bath, C-MAG HS 7 IKA hotplate magnetic stirrer, furnace (Muffle Furnace Ceramic Fiber), IWAKI beaker glass, measuring cup, dropper pipette, funnel, filter paper, volumetric flask, stir bar, volume pipette, PYREX Erlenmeyer, quartz cuvette, and AMV11 UV-Vis spectrophotometer.

#### **Material**

The materials used in this study were sample A, sample B, borax, 1% sodium carbonate ( $\text{NaCO}_3$ ), hydrochloric acid (HCl), curcumin, glacial acetic acid, standard solution of boric acid, aqua dest, sulfuric acid ( $\text{H}_2\text{SO}_4$ ), ammonium acetate.

### **Research Stages**

The stages of the research carried out were identification of borax, preparation of standard solutions, preparation of 0.125% curcumin solution, preparation of concentrated sulfuric acid-acetic acid solution (1:1), preparation of ammonium acetate solution and determination of boric acid.

### **Research Implementation**

#### **Identification of Boric Acid**

Crush 1 gram of sample and put it in a porcelain crucible, add 4 mL of 1%  $\text{Na}_2\text{CO}_3$ , and stir until homogeneous. Evaporate the mixture in a water bath until dry, charcoal with a hot plate ( $\pm 500^\circ\text{C}$ ) until no smoking, and heat in a  $500^\circ\text{C}$  furnace until complete ashing. Add 2 mL of hydrochloric acid (1:4) to the ashes, then heat over a water bath. Rinse with hot water and filter, then put into a 50 mL volumetric flask. After it cools down, add water up to the mark.

#### **Standard Solution**

Weigh 25 mg of the standard boric acid solution, put it into a 25 mL volumetric flask, and dissolve it with distilled water up to the mark (1000 mL solution  $\mu\text{g}/\text{mL}$ ). Pipette 2.5 mL into a 25

mL volumetric flask and dissolved with distilled water up to the mark (100 mL solution  $\mu\text{g}/\text{mL}$ ). Dilute it into a standard borax solution 1, 2, 4, 6, 16 g/mL.

#### **Curcumin Solution 0.125%**

Weigh 0.0625 grams of the curcumin powder into a 50 mL volumetric flask. Add glacial acetic acid up to the mark. Stir using a magnetic stirrer until the solution dissolves completely.

#### **Concentrated Sulfuric Acid-Acetic Acid Solution (1:1)**

Measure 50 mL of concentrated acetic acid solution in a beaker. Add 50 mL of concentrated sulfuric acid little by little and stir gently until homogeneous.

#### **Ammonium Acetate Solution**

Weigh 25 grams of ammonium acetate, then dissolve it with 30 mL of glacial acetic acid. Add aqua dest up to the mark. Stir using a magnetic stirrer until the solution dissolves completely.

#### **Determination of Borax**

Pipette 0.5 mL of each solution 1, 2 and blank solution into a different 50 mL Erlenmeyer, add 3 mL of 0.125% curcumin solution in glacial acetic acid and mix until homogeneous. Add 3 mL of acetic acid mixture:  $\text{H}_2\text{SO}_4$  (1:1) mix until homogeneous, and leave for several hours. Add 15 mL of ammonium acetate solution in glacial acetic acid (see reagent preparation). Measure the orange-red solution in a 1 cm quartz cuvette at a 500-600 nm wavelength range. Perform a blank solution using water in the same way as the test solution. Create a standard curve between absorption and boric acid content. Calculate the boric acid content in the sample using the boric acid content formula.

$$\text{Boric acid content (ppm)} = \frac{(\text{absorbance-a})}{b} \times \frac{\text{dilution factor}}{(\text{final weight-initial weight})} \dots (1)$$

### **Results and Discussion**

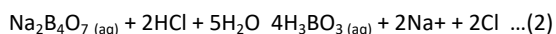
#### **Qualitative Analysis of Boric Acid**

This qualitative analysis aims to identify the presence of boric acid in food samples. This method can be done by testing using liquid curcumin. The preparation of the boric acid solution was reacted with curcumin because the boric acid solution is colorless and does not have a chromophore group [8]. Boric acid will be bound by curcu-

min to form a boron cyanon curcumin complex.

The prepared sample was added with 1% sodium carbonate in this qualitative analysis test. The function of adding sodium carbonate is to bind or form borax salts which do not evaporate during the ashing process. In this process, sodium is released, and the tetraborate ion where the tetraborate ion will react with water to form acids orthoborate. The sample containing the orthoborate is charred on a hotplate at room temperature 500°C until the sample turns into charcoal to obtain black charcoal containing metaboric acid. This coagulation process aims to remove some of the organic compounds in the sample, the rest of which will be removed during the ashing process [3].

The furnace functions as a tool used to turn charcoal into ashes. The ashing of the sample aims to remove organic compounds so that what remains in the ashing process are metals and salts that do not evaporate at high-temperature conditions. The ash that had cooled was then identified by adding 2 ml of HCl (1:4). The purpose of adding HCl is to dissolve the borax salt in the remaining sample of the ashes, as well as to provide an acidic atmosphere in the sample solution to make it easier to identify it.

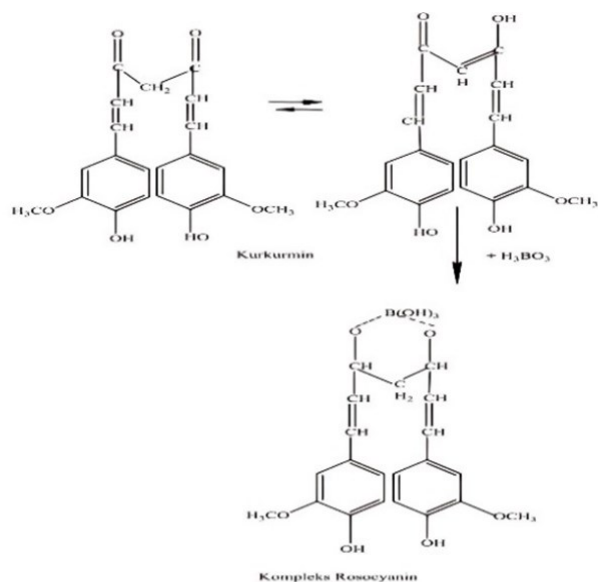


Based on previous research, the concentration of curcumin used is 0.125% based on the range of 0.100% - 0.150% curcumin can dissolve entirely in acetic acid without filtering [9]. The curcumin reagent was chosen because this rea-

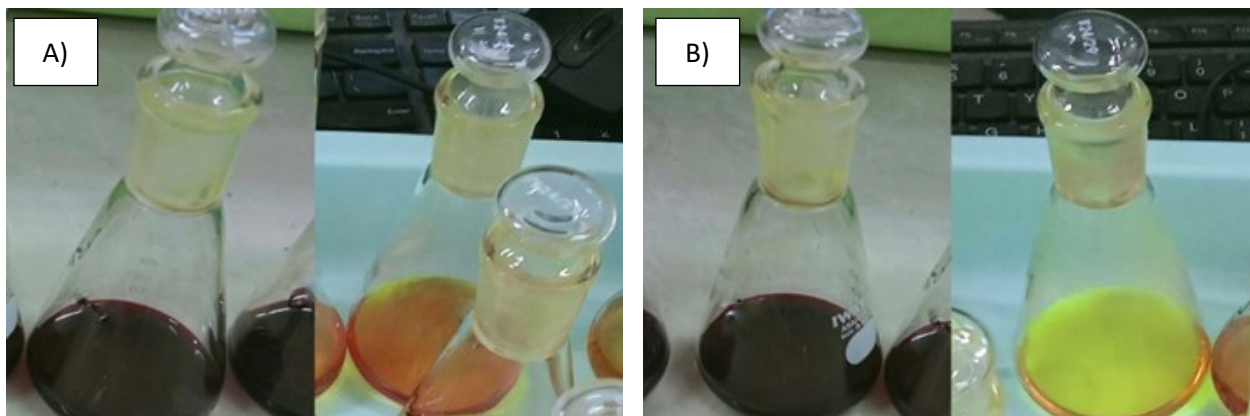
gent is very sensitive in determining the amount of boron, even in small amounts, by forming a colored complex orange [10]. Adding this 0.125% curcumin solution binds boric acid and forms a rosa color complex, often called the Boron Cyano Curcumin compound. After cooling the solution, 3 mL of sulfuric acid and acetic acid (1:1) were added while stirring until there was no yellow color. The purpose of adding strong acid is to provide an acidic environment to the sample so that it can help dissolve boric salts into boric acid, and the boric acid is bound by curcumin to form a complex of orange-colored rosa. This color complex is used to measure borax levels using a UV-Vis spectrophotometer.

Furthermore, the addition of ammonium acetate to the solution has cooled. Boron Cyanon complex compounds, when reacted with ammonia will form the anion. Making ammonium acetate solution must always be made fresh. This is caused by the use of alcohol as a solvent with volatile properties that will affect the solution's concentration. The stability of the color complex can only be maintained for 2 hours after the color complex is formed in an acidic condition. This color reaction is specific for borax and boric acid. If the sample contains positive borax, it will produce an orange color.

**In Figure 1.** Curcumin has two tautomeric forms, namely ketones, and enols. If the food sample is positive for boric acid, the ketone and hydroxyl groups will interact with boric acid to produce rosocyanin compounds. The formation of an orange color indicated it in the sample.



**Figure 1.** Reaction of Boric Acid with Curcumin



**Figure 2.** Color Change of Samples A and B

Based on the results obtained, it can be seen in sample A that the color changed from purplish-red to orange after adding ammonium acetate which indicates that sample A is positive for the boric acid test. Meanwhile, sample B changes color from purplish red to deep yellow, indicating no boric acid in the sample. From the results obtained, it can be seen that the darker the color (orange to brown) produced, the higher the boric acid content in the sample.

### Quantitative Analysis of Boric Acid

The boric acid analysis method can be done in several ways, namely qualitatively and quantitatively. One method of quantitative analysis is by using the UV-Vis spectrophotometer method. This method has high sensitivity and specificity and can be used to determine highly concentrated samples small [11]. Quantitative analysis in this test aims to determine the level of boric acid in the

**Table 1.** Determination of Boric Acid Levels in Samples A and B

Substance Name	Weight		Dilution Factor (mL)
	Container + Sample	Container + Remnants	
Comparison Standard Acidium Baricium	41,880 mg	16.8670 mg	0.5mL/50mL 1mL/50mL 2mL/50mL 3mL/50mL 8mL/50mL
Blank			50mL
Test Substance			
(A-1)	52.4966 gr	51.2449 gr	50mL
(A-2)	52.7310 gr	51.7102 gr	50mL
(B-1)	51.0720 gr	50.0651 gr	50mL
(B-2)	50.7047 gr	49.6940 gr	50mL

**Table 2.** Absorbance Measurement Results of Standard Boric Acid Solution

No.	Dilution	Content ( $\mu\text{g}/\text{mL}$ )	absorbent
1	0.5mL/50mL	1	0.065
2	1mL/50mL	2	0.099
3	2mL/50mL	4	0.174
4	3mL/50mL	6	0.293
5	8mL/50mL	16	0.696

The quantitative analysis begins with the preparation of a standard or standard solution. Preparation of this standard solution is made to determine the maximum wavelength. In making standard solutions, boric acid solutions are taken in different volumes. The purpose of taking these different volumes is to create a standard concentration with which to compare the results of this test.

Then the sample and standard solution which had changed color when the ammonium acetate was added were put into the quartz cu-

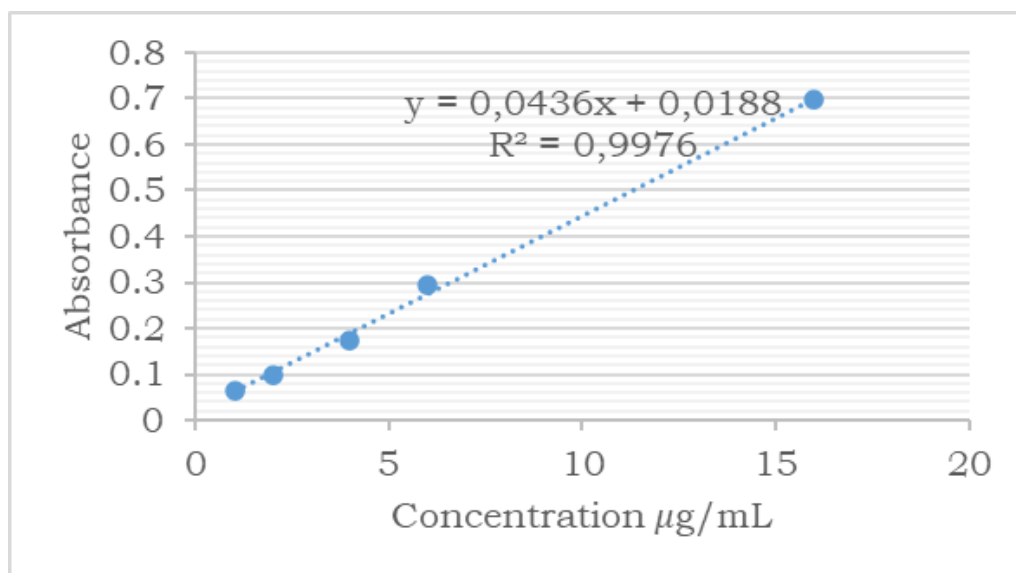
vette alternately and measured using a UV-Vis spectrophotometer at a maximum wavelength of approximately 540 nm. The maximum wavelength is used because the substance provides the highest absorption at that wavelength, and there is the absorption of boric acid at that wavelength. According to previous research, rosacyanin can be observed at a UV-Vis spectrophotometer at a maximum wavelength of 547 nm [12]. The difference in the maximum wavelength depends on the sensitivity of the tool. The spectrum produced by the UV-Vis spectrophotometer is :

**Table 3.** Sample Absorbance Value

Sample Name	Absorbance Value
Sample A-1	1.085
Sample A-2	0.908
B sample	0.086
Raw 8	0.696

Table 3 shows the absorbance values of samples A, B and standards measured with a UV-Vis spectrophotometer at a maximum wavelength of 540 nm. It can be seen in the table that samples A-1 and A-2 have absorbance values that exceed the standard absorbance values and have LOD values above 1.23 ppm which indicates that these samples contain positive borax. Where as sample

B has an absorbance value that is less than the standard absorbance value, and the sample LOD value is below the value of 1.23 ppm, indicating that the sample is adverse to the boric acid test. The limit of detection (LOD) is the smallest amount of analyte in a sample that can be detected which still gives a significant response compared to the blank.



**Figure 6.** Boric Acid Regression Curve

Furthermore, the absorbance measurement data results for sample A (positive sample) were used in the quantitative test using a UV-Vis spectrophotometer. The first stage is to make a standard borax calibration curve to calculate the levels of substances present in samples that positively contain boric acid. Making this regression curve aims to see whether concentration and absorp-

tion have a linear relationship. Determining the standard borax solution calibration curve, the regression equation is  $y = 0.0436x + 0.0188$  with a correlation coefficient ( $r$ ) of 0.9976. Based on the results obtained, it can be seen that the value of the correlation coefficient ( $r$ ) is close to 1. It means that there is a linear relationship between concentration and absorption.

**Table 4.** Analysis Data of Boric Acid Levels

Sample	Sample Weight		Dilution	absorbent	Content (ppm)	Content (ppm)	SD	%RSD
	Container Weight + Sample	Container Weight						
A-1	52,497	51,245	50	1.085	975,903	986,947	15,619	1,583
A-2	52,731	51,710	50	0.908	997,991			

Following what is in table 3, where to get accurate data results, two replications were carried out, and then the average level results were taken. After calculating the boric acid level in sample A, the boric acid level in sample A-1 is 975.903 ppm, and the level in sample A-2 is 997.991 ppm. The average level of the two samples is 986.947 ppm. It can be concluded that sample A contains boric acid which indicates that this sample should not be consumed. Based on the results of the data obtained the Relative Standard Deviation (RSD) value is 1.583%. The magnitude of this RSD indicates the level of accuracy of the analyst, the smaller the % RSD produced, the higher the accuracy of the data results. Because the results of these data obtained RSD2%, the data has good precision. Precision is a measure that indicates the degree of agreement between individual test results, measured by the spread of individual results from the average if the procedure is applied repeatedly to samples taken from a homogeneous mixture. From the data obtained, it means that food sample A is not suitable for consumption because it can have a harmful effect.

Based on the Regulation of the Minister of Health of the Republic of Indonesia No. 033/Menkes/Per/2012 [1] regarding Food Additives, said borax is a dangerous and toxic substance and, therefore, cannot be used as a food additive and cannot be used as a preservative. The detection of boric acid in food sample A indicates that many traders still add boric acid as a food additive to their food products to make them last longer and

chewier. The physical characteristics of foods containing borax are identified by their chewy, drier, bright color, long-lasting, and odorless [13].

The impact of using this boric acid can last a long time even though the levels of boric acid are used in small amounts. If ingested, boric acid can cause adverse effects on the central nervous system, kidneys, and liver. The highest concentration is reached during excretion. The organ that suffered the most damage compared to other organs was the kidney. The fatal dose for using boric acid in adults is 15-20 grams, and for children it is 3-6 grams [14]. Based on RI Minister of Health No. 33 of 2012 [1] concerning Food Additives, the acute hazard caused by consuming boric acid is the body feeling unwell (malaise), nausea, severe pain in the upper abdomen, gastro-enteritis bleeding accompanied by vomiting blood, diarrhea, weakness, drowsiness, fever, and headache. The chronic or long-term danger is loss of appetite, weight loss, mild irritation accompanied by indigestion, skin rash and red-red, dry skin and mucous membranes and cracked lips, red tongue, inflammation of the eye membranes, anemia, damaged kidney, acute circulatory system failure, and even death.

## Conclusion

Based on the research that has been done, it shows that sample A positively contains boric acid which is characterized by a change in color from purplish red to orange with an average boric acid

level of 986.947 ppm. The %RSD value obtained is 1.583%, indicating that this test has good precision. The detected boric acid in sample A indicates that the food sample is unsuitable for consumption because it can harm health.

### Thank-you note

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