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Faculty of Graduate Studies

Kinetic Determination of Bromate in Bread

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Dedication

To my dear father and my beloved mother, who supported me and raised me to be I am today.

To my brothers and sisters who encouraged me all the time.

To my husband, I dedicate this work.

Acknowledgment

First of all, I would like to express my gratitude to ALLAH s.w.t. for the strength and good health to complete my study.

Many thanks and gratitude to my supervisors; prof. Bassem Shraydeh and Dr. Ahmed Abu-Obaid, for imparting their knowledge, expertise, technical guidance and full support during thesis work.

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إقرار

أنا الموقعة أدناه مقدمة الرسالة التي تحمل العنوان:

Kinetic determination of bromate in bread

أقر بأن ما اشتملت عليه هذه الرسالة إنما هي نتاج جهدي الخاص، باستثناء ما تمت الإشارة إليه حيثما ورد، وأن هذه الرسالة ككل، أو أي جزء منها لم يقدم لنيل أية درجة أو لقب علمي أو بحثي لدى أية مؤسسة تعليمية أو بحثية أخرى.

Declaration

The work provided in this thesis, unless otherwise referenced, is the researcher's own work, and has not been submitted elsewhere for any other degree or qualification.

Student Name: **اسم الطالبة:** شذا محمد صلاح أبو حسن

Signature: **التوقيع:**

Date: **التاريخ:** ٧/٦/٢٠١٥

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List of Abbreviations

Symbol	Abbreviation
ppm	Part per million
nm	Nanometer
ICP/ MS	Inductively coupled plasma mass spectrometry
UV	Ultraviolet

Kinetic Determination of Bromate in Bread

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Abstract

Bromate is used in bread making as agent of maturation, however it is classified as a potential carcinogen. A rapid, simple, precise and accurate testing method was developed to determine the level of bromate in bread; this method is based on bromate reaction with iodide ion in acidic medium to produce iodine (I_2). The absorbance of iodine (I_2) was measured at 352 nm. Bromate reacted with iodide during the first 3 minutes after initiation of the reaction. In the first calibration curve, the curve was linear within the range 1-10 ppm of bromate in distilled water. In the second calibration curve, the curve was linear within the range 50-300 ppm of bromate in dough. In the third calibration curve, the curve was linear within the range 50-300 ppm of bromate in bread. The proposed method has been successfully applied to the determination of bromate in commercial bread. In this study we found that the use of two grams (2 g) of potassium bromate per bag flour (60 kg) is safe.

Bromate usually degrades at about 400 ˚C alone but in bread the bromate degrades at 150-200 ˚C due to the presence of other metals which serve as catalysts.

Chapter One

Introduction

1.1 Bread:

Bread has a historical importance in many Western and Eastern societies and a staple food in many countries of the world [1, 2]. In many cultures, and in the West and Middle East during the 1950s, the importance of bread was not only one of nutrition but also of used as a currency instead of money to buy the necessities of life and was the main contributor for families in regards to living and economy [3]. Bread is one of the oldest man-made foods. With the evolution of human bread itself evolved, and with different human needs, bread types would differ according to human needs, such as health conditions and taste preferences.

Methods of preparing bread vary from one society to another in many parts of the worlds. People make bread by simple methods themselves, but in the bakeries bread is produced by machines. The bakeries consume 7-8 bags of flour per day. Many types of bread exist in current markets (300-500 types such as brown bread, toast, etc.). The certain type of bread differs according to many combinations of types of flour and other ingredients, and also according to different traditional recipes and modes of preparation of bread. As a result, there is a wide variety of types, shapes, sizes, and textures of breads in various regions. In the West bank, bread is the one of the most commonly consumed foods in homes and restaurants. Predominant consumption occurs among the poor and youth, in which the individual consumes an average of 9 small loaves of bread daily.

Bread is made from a type of flour (wheat, corn or barley) and some of the basic ingredients include table salt, water, yeast, others flavors, and at least one flour improver [4, 5].

1.2 Flour Improver:

Flour improvers are food additives combined with flour to improve baking functionally. Both dough processing characteristics and loaf volumes are improved with flour improvers [6].

There is a wide range of these conditioners used in factory baking, which fall into four main categories: bleaching agents, oxidizing and reducing agents, enzymes, and emulsifiers. These agents are often sold as mixtures in a soy flour base, as only small amounts are required.

Oxidizing agents are added to flour to help with gluten development. Oxidizing agents affect sulfur-containing amino acids, helping to form disulfide bridges between the gluten molecules. The addition of these agents to flour will create stronger dough [7].

Common oxidizing agents include:

1- Azodicarbonamide.
2- Carbamide.
3- Potassium Bromate (this agent gives bromated flour its name).
4- Phosphates.
5- Potassium iodate.

Reducing agents help to weaken the flour by breaking the protein network. This will help with various aspects of handling strong dough. The benefits

of adding these agents are: reduced mixing time, reduced dough elasticity, reduced proofing time, and improved machinability [7].

Common reducing agents include:

1-Fumaric acid 2-Sodium bisulfate 3-Ascorbic acid

1.3 Bromated Flour:

Bromated flour is flour which has been enriched with potassium bromate and used in bread production. Adding potassium bromate makes the bread stronger and more elastic, and also promotes big rises of bread [8]. The resulting bread tends to be strong and springy, and especially well-suited to commercial production [9].

The mechanism of bromate activity in dough is complex and not well understood. Sullivan et al suggested the mechanism involves the protein fraction of flour [10]. Jorgensen and Balls thought the oxidant inhibits the action of the proteolytic enzymes [11, 12]. Baker et al concluded that a material in gluten that is salt soluble, reacts with oxidants [13]. Although many hypotheses have been proposed, the theory chemists agree upon is that $KBrO_3$ oxidizes thiol groups to disulfide linkages, thus strengthening the protein network. This increases dough expansion capacity and improves the bread's appearance [14, 15].

The presence of bromate in bread may lead to renal failure, anemia, respiratory depression, and cancer in humans, meaning that it may be harmful when consumed [16- 20]. In theory, the substance is supposed to "bake out" of bread dough as it cooks, but if a residue remains behind in the bread, it could be harmful to the consumer in the long term. A careful balance

is required of manufacturers, since they must add enough of the substance to bromated flour to make it perform as expected.

The Food and Drug Administration (FDA) the use of potassium bromate up to a maximum level of 50 mg/kg of flour mass in bread. However, Japan permits the inclusion of only up to 10 mg/kg of flour [21]. In California, a warning label is required when bromated flour is used. Currently in California, it is recognized that it is inappropriate to use potassium bromate in any product or production method, which can be formulated with residues below the level of 20 ppb (i.e. 0.020 mg/kg) in the finished product [22]. The Food and Agriculture Organization (FAO) / World Health Organization (WHO) joint committee's initial recommendation of acceptable level of 0–60 mg $KBrO_3$/kg flour was withdrawn because of long term toxicity and carcinogenicity studies (in vitro and in vivo), which had revealed the development of renal cell tumors in hamsters [23].

1.4 Existing Methods of Bromate Analysis:

Reduction of bromate to bromide occurs because of the reductive properties of bread dough. Reduction begins when ingredients are first mixed and continues during baking. When the reduction process is finished, the amount of bromate residue in the final product is very low. This is the principal reason for which the conventional analytical techniques for bromate determination are only applied in flour and dough [24]. Most analytical methods for the determination of bromate in bread are time consuming, such as: the colorimetric method [25], spectrofluometric method [26], ion

chromatography [27], capillary electrophoresis [28] and oxidation of dyes [4].

Quantitative determination of the levels of potassium bromate in bread samples was done using the spectrophotometric method, which is based on the redox reaction between bromate and promethazine hydrochloride in an acidic medium. The absorbance of the product was read at 515nm. The qualitative test was performed directly on a portion of each bread sample using 2 ml of 0.01M promethazine and 0.6ml of 12 M hydrochloric acid. The change in color of each bread sample to pink indicated the presence of potassium bromate [15].

Methods of analysis for detecting bromate in bread require sophisticated instruments such as ion chromatography with inductively coupled plasma mass spectrometry (IC/ ICP-MS). A method of analysis using IC/ICP-MS was developed and Bromate was extracted from bread using water [29].

A second method of analysis and determining the levels bromate in bread, by wavelength dispersive x-ray fluorescence (WDXRF), was also applied. This method was applied to Bromate determination as an indication of the pre-baking of bread. The calibration of Br in bread obtained showed a low detection limit and a high sensitivity level [6].

A third method of analysis was Flow Injection Analysis (FIA). It was developed for the determination of bromate, based on its reaction with 3,5-dibromo-2-pyridylazo-5-diethylaminophenol (3,5-dibromo-PADAP) and thiocyanate in a strongly acidic medium. This produced an unstable violet product with a maximum absorption at 602 nm. The calibration curve was

linear in the range of $2.00 \times 10^{-6} \pm 2.10 \times 10^{-5}$ mol/l and the detection limit was 8.00×10^{-7} mol/l. The sampling frequency was 90 h^{-1}. The method has been successfully applied to the determination of bromate in commercial bread additives and flours [30].

1.5 Novelty of This Work:

In this work, we report a very simple and sensitive method for direct determination of bromate in bread and flour, which has been based on bromate's oxidation of iodide to produce an iodine (I_2) product.

1.5.1 Kinetic Method:

In general, kinetic methods enhance the sensitivity of detection to more than a thousand fold [31-33].

Kinetic methods of analysis, qualitative and quantitative chemical analysis, are based on the relationship between the reaction rate and the concentration of the reactants. The kinetics and mechanism of the reaction between potassium bromate and potassium iodide in an acidic medium have been studied extensively, and by many workers [34]. The conclusion drawn from these studies regarding the overall reaction is represented by the following equation:

$$BrO_3^- + 6I^- + 6H^+ \longrightarrow Br^- + 3I_2 + 3H_2O \qquad (1)$$

Takes place with through the following steps:

$$BrO_3^- + I^- + 2H^+ \xrightarrow{k} HBrO_2 + HIO \qquad (2)$$

$$HBrO_2 + I^- \longrightarrow BrO^- + HIO \qquad (3)$$

$$BrO^- + 2H^+ + 2I^- \longrightarrow Br^- + H_2O + I_2 \quad (4)$$

$$HIO + H^+ + I^- \longrightarrow I_2 + H_2O \quad (5)$$

Of which eq. (2) is the slow rate determining step. Thus the overall reaction rate can be represented as follows:

$$\text{Rate} = \frac{-d\,[BrO3-]}{dt} = k\,[BrO_3^-]\,[I^-]\,[H^+]^2 \quad (6)$$

Showing fourth order kinetics.

By using excess of $[I^-]$ and $[H^+]$, the rate of reaction reduce to pseudo first order kinetics and thus eq. (6) become

$$\text{Rate} = k'\,[BrO_3^-] \quad (7)$$

We now have a first order process in bromate. The original iodide and hydrogen ion concentrations are incorporated into the new constant k'.

1.5.2 Kinetic Methods of Analysis:

In this work, the calibration curves were established by using two kinetic methods of analysis:

1) Fixed time method:

   A mode of measurement in a kinetic method of analysis, in which the change of a parameter related to the concentration of a reactant or product, is measured over a predetermined time interval [35, 36].

2) Slope method (tangent method):

   A mode of measurement in a kinetic method of analysis, in which the slope of the response curve at a selected point is measured and related to the concentration of the reactant [35, 36].

1.6 Objective:

The aim of this work is to develop a sensitive kinetic method for the determination of bromate in bread, produced in the Jerusalem area and in different cities in Palestine.

1.7 Methodology:

In this work, the optimum conditions for increasing the production of I_2 were studied. The effect of pH was highlighted and the choice of the exact pH which gave the maximum rate was investigated. The effect of KI concentration on the rate was also studied. Finally, treatment of metal ion catalysis, enhancing the reaction rate, was correlated.

The tangent method for determining the rate or fixed time method will be used, and the choice will be for the method that gives maximum sensitivity. After studying the effect of temperature, the calibration graph was established to analyze $KBrO_3$ in bread. Samples from the Jerusalem area and major cities in the West Bank were analyzed.

1.8 Hypothesis:

It is believed that in the Jerusalem area, bakers are still using $KBrO_3$ in bread and cakes. This unmonitored use of $KBrO_3$ will be reflected in the health of the people consuming the bread in which its used, thereby increasing the rates of kidney failure and cancer. Although $KBrO_3$ use was banned in the West Bank cities, we extended our work to other major cities in the West Bank to scrutinize the concentrations of $KBrO_3$ in various bakeries.

Chapter Tow

Materials and Methods

2.1 Materials:

Flour was bought from markets in Jenin and used for preparing dough and bread. Bread was bought from bakeries from various cities in the West Bank.

2.2 Chemicals:

Hydrochloric acid HCl was purchased from (SDFCL Fine-chem limited). Glycine was purchased from (Riedel). Potassium iodide was purchased from (Frutarom). Potassium bromate was purchased from (Frutarom) used in bread making. Distilled water.

2.3 Apparatus:

A Shimadzu UV-1601 spectrophotometer with 1-cm quartz cell was used for iodine absorbance measurements.

A pH meter (Jenway 3510) was used to measure the pH of HCl.

A hot plate (J lab tech) was used for stirring and heating the solutions.

A Carbolite furnace was used for baking the dough.

Inert gas atmosphere (N_2) was used to expel oxygen (O_2).

Volumetric flasks (10 and 100 ml) were used to prepare reagents.

2.4 Preparation of Required Solutions:

2.4.1 Solutions of Potassium Bromate:

1- Potassium bromate stock solution (1000 ppm) was prepared by dissolving 0.100 g potassium bromate in distilled water then diluting to 100 ml.

2- A solution of 100 ppm was prepared using stock solution (1000 ppm) by dissolving 10.0 ml in distilled water then diluting to 100 ml.
3- A solution of 10 ppm was prepared using 100 ppm bromate solution by dissolving 10.0 ml in distilled water then diluting to 100 ml.
4- A solution of 20 ppm was prepared using 100 ppm bromate solution by dissolving 20.0 ml in distilled water then diluting to 100 ml.
5- A solution of 5 ppm was prepared using 10 ppm bromate solution in a 10.0 ml volumetric flask to study the optimum condition of this concentration.
6- Standard solutions of bromate were prepared ranging from 1 ppm to 10 ppm, by dissolving an appropriate volume of 10 ppm and 20 ppm bromate solutions in 10 ml volumetric flasks to establish the calibration curve.
7- A Potassium bromate solution of 100 ppm was prepared by dissolving 0.010 g of potassium bromate (after placing the bromate in the furnace for 15 minutes at different temperatures) in distilled water then diluting to 100 ml.

2.4.2 Other Solutions:

1- A Glycine buffer (0.1 M) solution was prepared by dissolving 0.7507 g of glycine in distilled water, then diluting to 100 ml.
2- A Hydrochloric acid HCl (2 M) solution was prepared using 32% HCl (10.7 M) by dissolving 18.7 ml of HCl in distilled water, then diluting to 100 ml.

2.4.3 Solutions to Determine the Optimum Conditions:

2.4.3.1 Solutions of HCl:

Hydrochloric acid HCl solutions and 0.1 M glycine buffer solutions of pH values: 1, 1.5, 2, 3 and 4 were prepared from glycine and hydrochloric acid solutions to study the effect of pH on the reaction rate.

2.4.3.2 Solutions of potassium iodide:

Studying the effect of KI, various concentrations of the halide salts were used ranging from 0.025 M to 0.6 M. Each concentration was prepared by weighing the proper amount and diluting to 100 ml.

2.5 Removal of Oxygen from Potassium Iodide:

Besides bubbling nitrogen (N_2) gas throughout the solution, oxygen was eliminated from the reaction vessel by heating the solution and then cooling to room temperature.

2.6 Absorption Experiments:

The product concentration (I_2) was monitored using a UV-Visible spectrophotometry, using the wavelength of maximum absorption of (I_2) at 352 nm. To measure the absorption of iodine (I_2), the experiment was carried out in a 10.0 ml volumetric flask and the following order of events was conducted: solution of potassium bromate was transferred into a flask, then 1.0 ml of buffer solution was added. The solution was diluted to 9.0 ml with distilled water and 1.0 ml of a potassium iodide solution was added. A stopwatch was started just after the addition of the iodide solution. A portion of the solution was transferred into a 1-cm quartz cell to measure the

absorbance at 352 nm during the first few minutes after the initiation of the reaction.

2.6.1 Effect of pH on Reaction:

The suitable pH for the reaction was studied as a function of pH. Three runs were used for each sample. In the first sample, 5.0 ml of 10 ppm bromate was transferred to a 10 ml volumetric flask. Then, 1.0 ml of a pH 1 buffer solution was added. The solution was diluted to 9.0 ml with distilled water and 1.0 ml of 0.1 M of iodide solution was added. Other samples were prepared as the first one, except that the 1.0 ml of a pH 1 buffer solution was used instead of 1ml of pH (1.5, 2, 3 and 4) buffer solutions respectively. Then, we measured the absorbance of iodine I_2 at each pH during the first 4 minutes.

2.6.2 Effect of Potassium Iodide on Reaction:

The concentration of potassium iodide was varied to study its effect on iodine production. Different concentrations were prepared of the iodide solution. Three runs were used for each sample. The order of solutions was performed by using the suitable pH and changing the concentration of potassium iodide (0.025, 0.05, 0.1, 0.2, 0.3, 0.4, 0.5 and 0.6 M) for 5 ml of 10 ppm. Then, the absorbance of iodine I_2 was measured for each concentration of potassium iodide during the first 4 minutes.

2.6.3 Optimization the Time of Reaction:

The suitable time to complete the reaction was studied. Three runs for each sample were conducted. The absorbance of iodine I_2 was measured for 5 ml

of 10 ppm bromate solution at a suitable pH and suitable iodide concentration for 15 min, to determine the suitable time at 25˚C.

2.7 Calibration Curve:

UV-V spectrophotometry was a fast, simple, and low cost convenient technique. It was used to study the absorbance of iodine (I_2) by changing the concentrations of potassium bromate. The absorbance of iodine (I_2) was measured at 325 nm.

The calibration curve was constructed by measuring the absorbance of iodine (I_2) by changing the concentration of potassium bromate. Three calibration graphs were established for bromate, the first curve is for bromate in distilled water, the second one is for bromate in dough and the third one for bromate in bread.

2.7.1 Calibration Curve of Bromate in Distilled Water:

Standard solutions of bromate were prepared ranging from 1 ppm to 10 ppm (solutions from 1 ppm to 7 ppm using 10 ppm concentration of bromate, solutions from 8 ppm to 10 ppm using 20 ppm).

Three runs for each sample were conducted. In the first sample (1 ppm), 1ml of 10 ppm bromate was transferred to a 10 ml volumetric flask. Then, 1.0 ml of a pH 1 buffer solution was added. The solution was diluted to 9.0 ml with distilled water and 1.0 ml of 0.1 M of iodide solution was added. The absorbance was measured at 352 nm during the first 3 min after mixing (measuring every 30 sec). The other samples (2 ppm -7 ppm) were prepared at first one, except the volume of bromate solution used.

Samples (8, 9 and 10 ppm) were prepared by using a 20 ppm bromate solution. In the sample (8 ppm), 4 ml of 20 ppm bromate was transferred to a 10 ml volumetric flask. Then, 1.0 ml of a pH 1 buffer solution was added. The solution was diluted to 9.0 ml with distilled water and 1.0 ml of 0.1 M of iodide solution was added. The absorbance was measured at 352 nm during the first 3 min after mixing (measuring every 30 sec).

2.7.2 Calibration Curve of Bromate in Dough:

2.7.2.1 Preparation of Dough with Known Amount of Potassium Bromate:

Dough was prepared using the following recipe: flour (100 g), yeast (1g) and water (80 g). Seven dough samples were prepared with different amounts (0, 50, 100, 150, 200, 250, 300 ppm) of potassium bromate. Ingredients were mixed in a beaker using a glass rod for 2 min. Then, the dough was fermented for 20 minutes**.**

2.7.2.2 Sample Pretreatment:

A sample of 10 g of dough was cut and triturated into 100 ml of distilled water with a magnetic stirrer and then filtered. A measured volume of the filtrate solution (5 ml) was transferred into a 10 ml flask.

2.7.2.3 Absorbance Measurements:

In the flask, 1.0 ml of a pH 1 buffer solution was added to the filtrate. Then, the solution was diluted to 9.0 ml with distilled water and 1.0 ml of 0.1M of an iodide solution was added. The absorbance was measured after mixing for each sample (3 runs for each sample). The unknown concentration of

bromate in dough was calculated from the linear regression curve obtained from the standard solutions of bromate.

2.7.3 Calibration Curve of Bromate in Bread:

2.7.3.1 Preparation of Bread with Known Amount of Potassium Bromate:

Bread was prepared using the following recipe: flour (100 g), yeast (1g) and water (80 g). Seven bread samples were prepared with different amounts (0, 50, 100, 150, 200, 250, 300 ppm) of potassium bromate. Ingredients were mixed in the beaker by using a glass rod for 2 min. Then, the dough was fermented for 20 min. Finally, the dough was baked at 270˚C for 15 min.

2.7.3.2 Sample Pretreatment:

A sample of 10 g of bread was cut and triturated into 100 ml of distilled water with a magnetic stirrer and then filtered. A measured volume of the filtrate solution (5 ml) was transferred into a 10 ml flask.

2.7.3.3 Absorbance Measurements:

In the flask, 1.0 ml of a pH 1 buffer solution was added to the filtrate. Then, the solution was diluted to 9.0 ml with distilled water and 1.0 ml of 0.1M of the iodide solution was added. The absorbance was measured after mixing for each sample (3 runs for each sample). The unknown concentration of bromate in bread was calculated from the linear regression curve obtained from the standard solutions of bromate.

2.7.4 Collecting Samples of Bread and Dough from Bakeries:

Bread samples were bought from different bakeries in Jerusalem and from cities in the West Bank. In collaboration with Ministry of National Economy/ Department of Consumer Protection, sample of dough and bread from different bakeries in cities of West Bank were collected.

Then, the absorbance of I_2 for each sample of dough and bread was measured.

2.8 Effect of Temperature on Potassium Bromate in Distilled Water:

The effect of temperature on potassium bromate was investigated from 25 ˚C to the temperature in which all potassium bromate decomposed. 0.5g of potassium bromate was placed in a crucible, then we transferred the crucible to the furnace at the desired temperature for 15 min. After 15 min, 100 ppm of potassium bromate were prepared for each sample by dissolving 0.01g of bromate in 100 ml distilled water, then the absorbance of iodine (I_2) was measured.

2.8.1 Absorption Experiment:

Three runs were used for each sample. The absorbance of iodine (I_2) was measured for 5 ml of 100 ppm bromate solution, transferred into a 10 ml flask, then 1 ml of a suitable buffer solution was added to the flask. The solution was diluted to 9.0 ml with distilled water and 1.0 ml of a suitable concentration of iodide solution was added. Then, the absorbance was measured during the first few suitable minutes.

2.9 Effect of Temperature on Potassium Bromate in Flour:

The effect of temperature on potassium bromate was investigated from 25 ˚C to the temperature in which all potassium bromate decomposed. 0.5g of potassium bromate was placed in a crucible, then we transferred the crucible to the furnace at the desired temperature for 15 min. After 15 min, 100 ppm of potassium bromate were prepared for each sample by mixing 0.01g of bromate in 100 g flour, then the absorbance of iodine (I_2) was measured.

2.10 Analysis Flour Sample by ICP-MS:

Five grams of flour were digested with nitric acid at room temperature and analyzed by inductive coupled plasma-mass spectrometry (ICP-MS) for detecting the metals present (table 3.10).

Chapter Three

Results and discussion

3.1 Determination of Optimum Conditions:

To establish the optimal conditions for the bromate-iodide reaction, series of experiments were carried out. All parameters in which the effect on the production of iodine was studied by altering each variable in turn, while keeping other variables constant.

Bromate reacts with iodide in acidic media to produce iodine according to this equation:

$$BrO_3^-{}_{(aq)} + 6I^-_{(aq)} + 6H^+_{(aq)} \longrightarrow 3I_{2(aq)} + 3H_2O_{(l)} + Br^-_{(aq)} \qquad \text{eq. 3.1}$$

The reaction could be monitored spectrophotometrically by measuring the absorbance of the solution at 352 nm, which is proportional to I_2 concentration. From the equation, iodine production depends on the pH and reagent concentrations (potassium iodide). So it was determined that the suitable optimal condition for producing large amounts of iodine and giving high absorbance.

3.1.1 Effect of pH:

The effect of the pH on the reaction of bromate was studied. Three runs were used for each sample. All conditions were the same for the three runs, except the pH, which was different for each sample. The results are shown in (table 3.1and figure 3.1).

Table3.1: Effect of pH in the range of 4.0 - 1.0 on the reaction of 5 ppm potassium bromate.

pH	Absorbance
4	0.0013
3	0.0027
2	0.0057
1.5	0.0745
1	0.1152

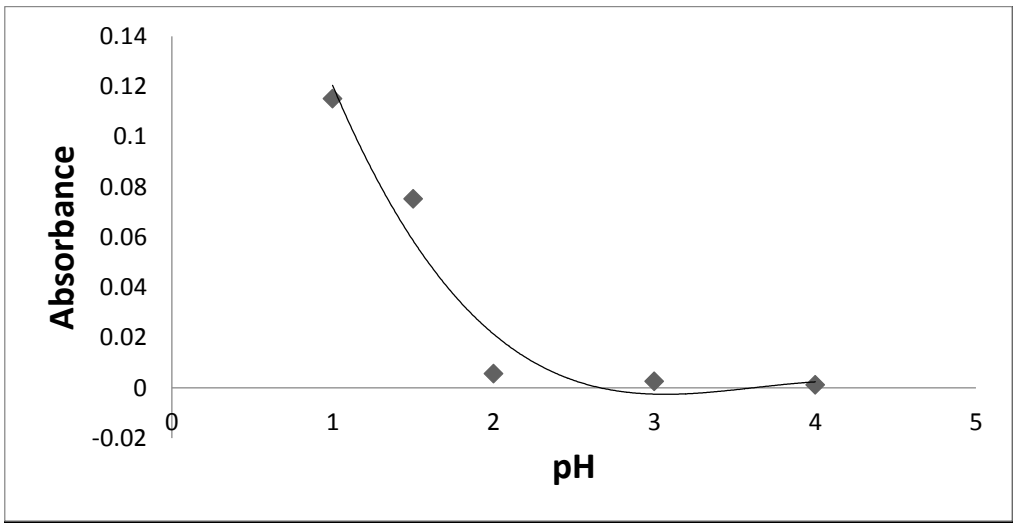

Figure3.1: Effect of pH in the range of 4.0 – 1.0 on the reaction.

As the figure shows, investigated anion (bromate) did not react with iodide in the range of pH from 2.0 to 4.0 during the first 4 min after mixing, even when the concentration of bromate was 100-fold in excess over 5 ppm. Also as the obvious figure shows, bromate reacted with iodide in the range of pH from 1.0 – 1.5 during the first 4 min after mixing, but the reaction rate increased at pH values lower than 1.5. Therefore, a pH of 1.0 was selected as the optimum pH for the reaction and the absorbance change during the first 4 min after mixing was proportional to the bromate concentration.

3.1.2 Effect of Potassium Iodide Concentration:

The effect of the iodide concentration on the rate of the reactions was investigated at a concentration of 5 ppm of bromate and by varying the concentration of iodide in the range of 0.025 - 0.6 M. The results are shown in (table 3.2 and figure 3.2) at pH 1 (optimal pH).

Table3.2: Effect of iodide concentration at pH 1.

Iodide concentration /M	Absorbance
0.025	0.0634
0.05	0.0852
0.1	0.1152
0.2	0.1373
0.3	0.1524
0.4	0.1536
0.5	0.1556
0.6	0.1582

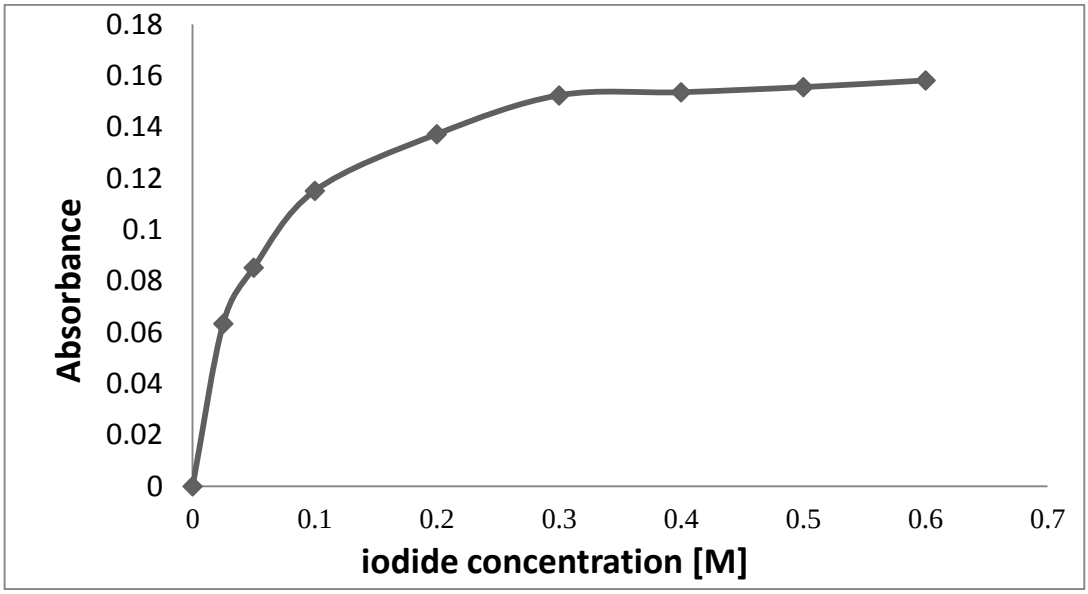

Figure3.2: Effect of iodide on the reaction at pH 1.

As figure 3.2 shows, at pH 1, the absorbance during the first 4 min after initiation of a reaction for bromate increased (linear) upon increasing the iodide concentration up to 0.3 M, and remained nearly constant at higher

concentrations (zero order and constant rate). Therefore, the maximum absorbance during the first 4 min was obtained with iodide concentration at 0.1 M to suppress air oxidation.

3.1.3 Optimization the Time of Reaction:

The suitable time to complete the reaction was studied for 5 ppm of bromate. The absorbance of iodine I_2 was measured at pH 1 and 0.1 M iodide concentration for 15 min. The results are shown in figure 3.3.

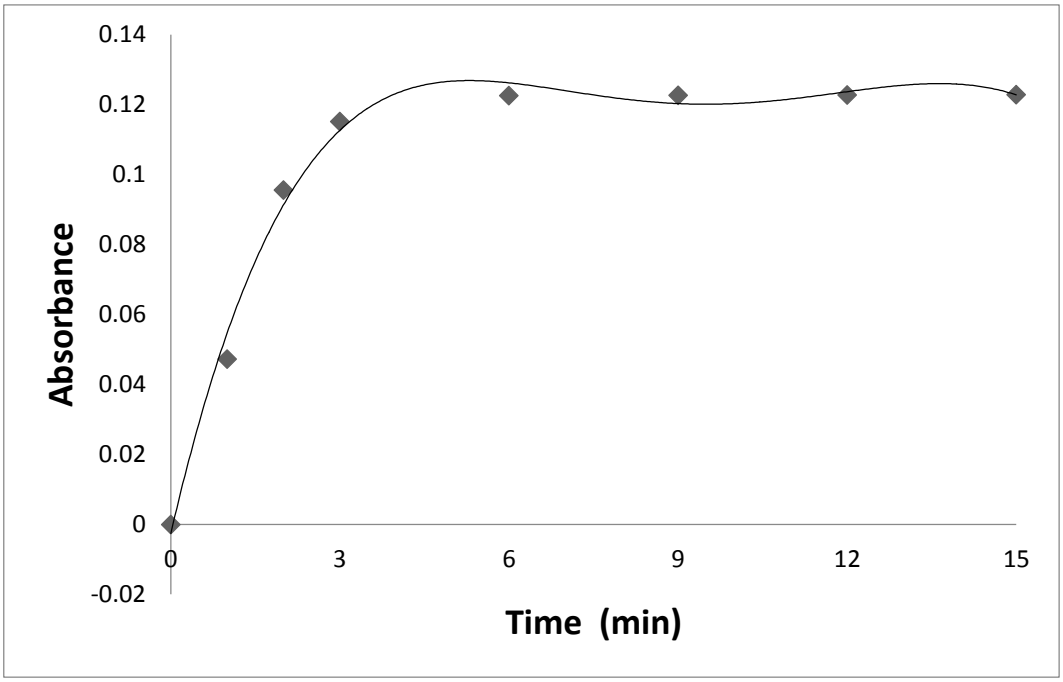

Figure3.3: Optimization the time of reaction at pH 1 and 0.1M KI.

As figure 3.3 shows, the complete reaction required 3 min. Therefore, the absorbance of iodine (I_2) was measured during the first 3 min and the optimum conditions were applied in further experiments.

3.2 Calibration Curve:

Calibration graphs for the determination of bromate in distilled water, dough and bread were obtained under the optimum conditions.

3.2.1 Calibration Graph of Bromate in Distilled Water:

The tangent method for determining the rate or fixed time method was used to choose the method that gave maximum sensitivity.

Tangent method (slope method):

In this method, the curves were established between the measuring absorbance versus time for each concentration of bromate. Then, the slope for each graph was measured at a selected point and related to concentration.

Table3.3: Absorbance of I_2 for 1 ppm of bromate during 3 min at pH 1 and 0.1 M KI.

Time (s)	Abs 1	Abs 2	Abs 3	Average
0	0	0	0	0
30	0.0062	0.0074	0.0071	0.0069
60	0.0093	0.0101	0.0106	0.01
90	0.0125	0.0134	0.0148	0.013567
120	0.0161	0.0171	0.0178	0.017
150	0.0201	0.0206	0.0212	0.020633
180	0.0227	0.0242	0.0255	0.024133
210	0.0230	0.0245	0.0255	0.024343
240	0.0233	0.0247	0.0256	0.02453
270	0.0234	0.0247	0.0256	0.02456
300	0.0248	0.0256	0.0234	0.02458
330	0.0247	0.0255	0.0237	0.02465
360	0.0247	0.0256	0.0237	0.02468
390	0.0248	0.0257	0.0237	0.02473
420	0.0248	0.0257	0.0237	0.02473

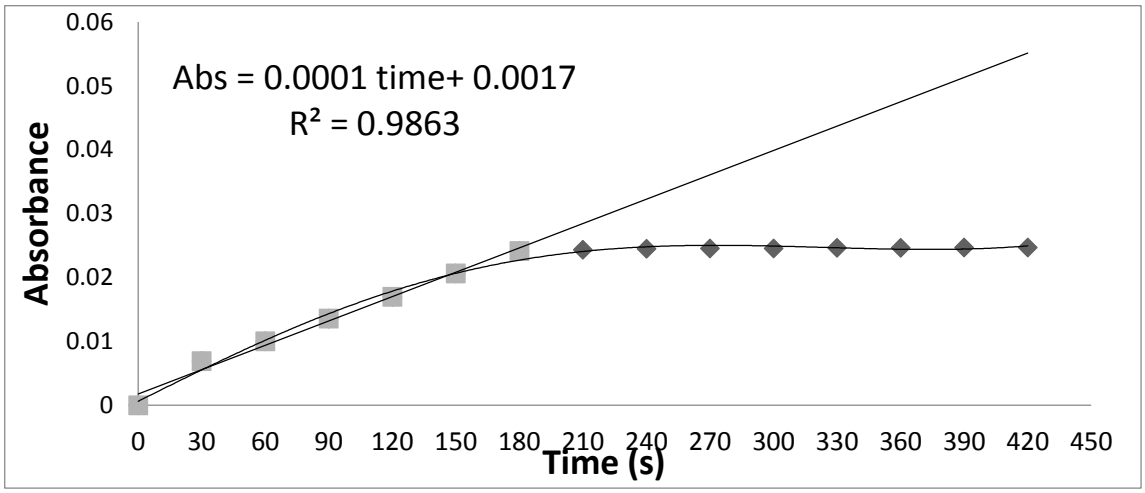

Figure3.4: Absorbance of I_2 for 1 ppm of bromate during 3 min**.**

2 ppm of potassium bromate:

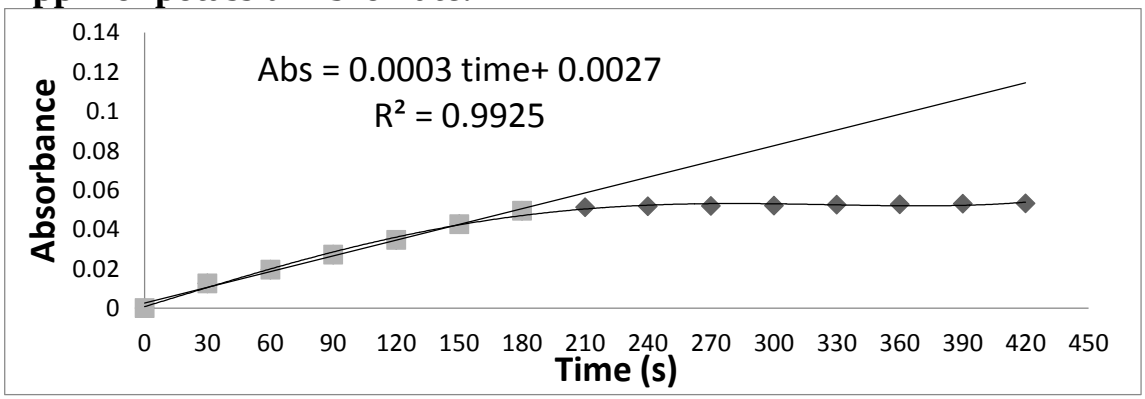

Figure3.5: Absorbance of I_2 for 2 ppm of bromate during 3 min.

3 ppm of potassium bromate:

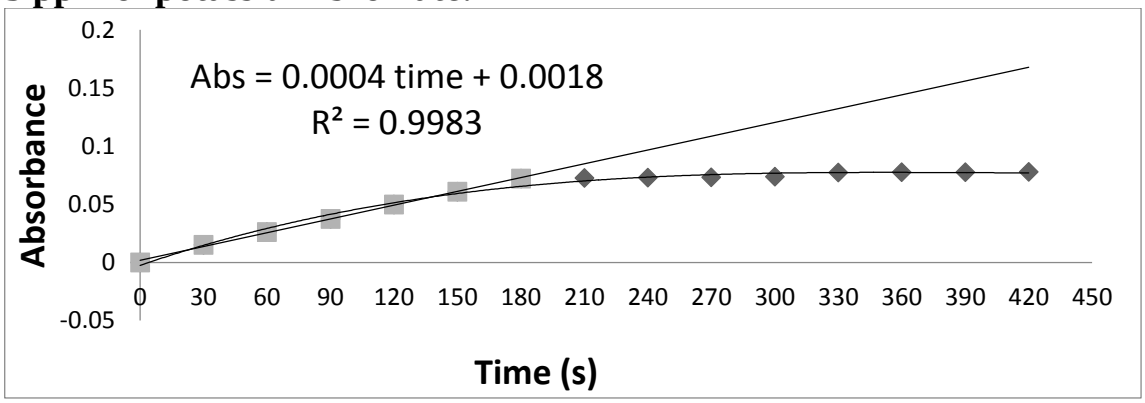

Figure3.6: Absorbance of I_2 for 3 ppm of bromate during 3 min.

4 ppm of potassium bromate:

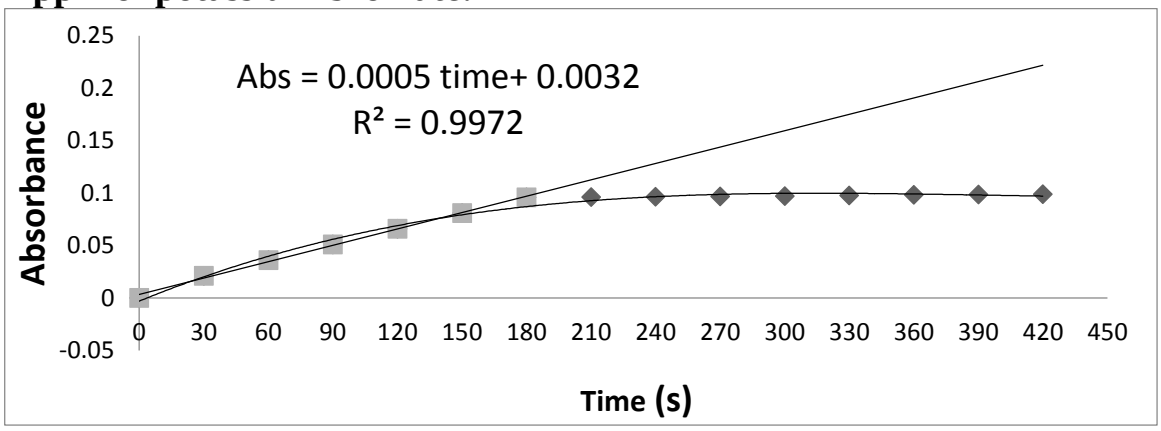

Figure3.7: Absorbance of I_2 for 4 ppm of bromate during 3 min.

5 ppm of potassium bromate:

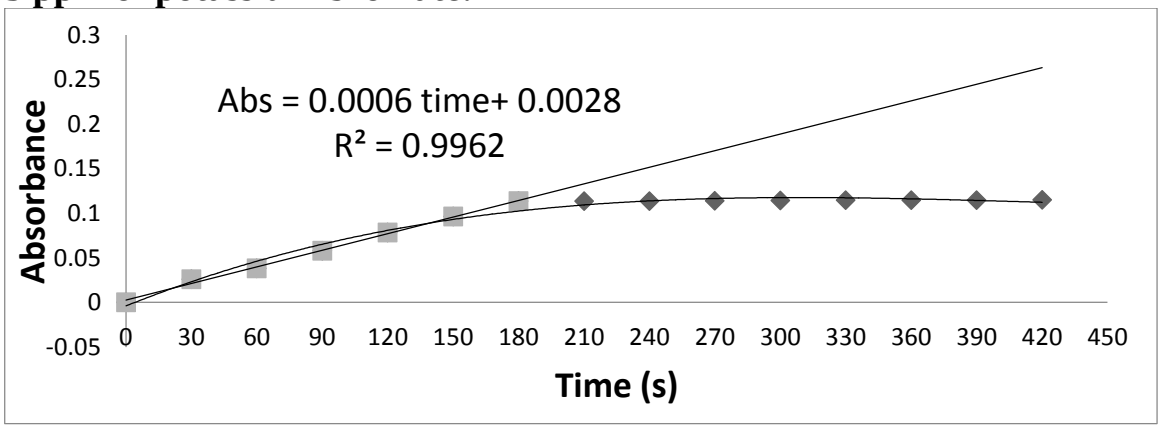

Figure3.8: Absorbance of I_2 for 5 ppm of bromate during 3 min.

6 ppm of potassium bromate:

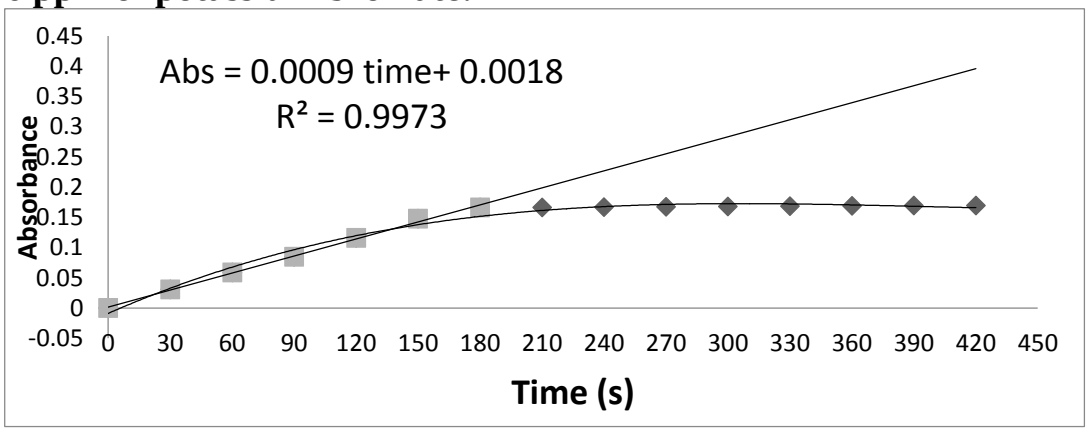

Figure3.9: Absorbance of I_2 for 6 ppm of bromate during 3 min.

7 ppm of potassium bromate:

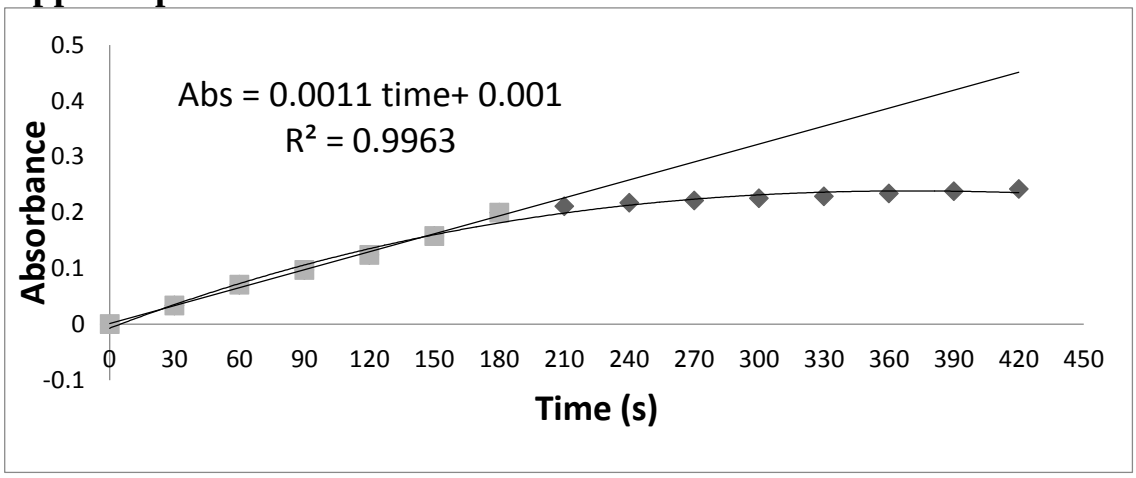

Figure3.10: Absorbance of I_2 for 7 ppm of bromate during 3 min.

8 ppm of potassium bromate:

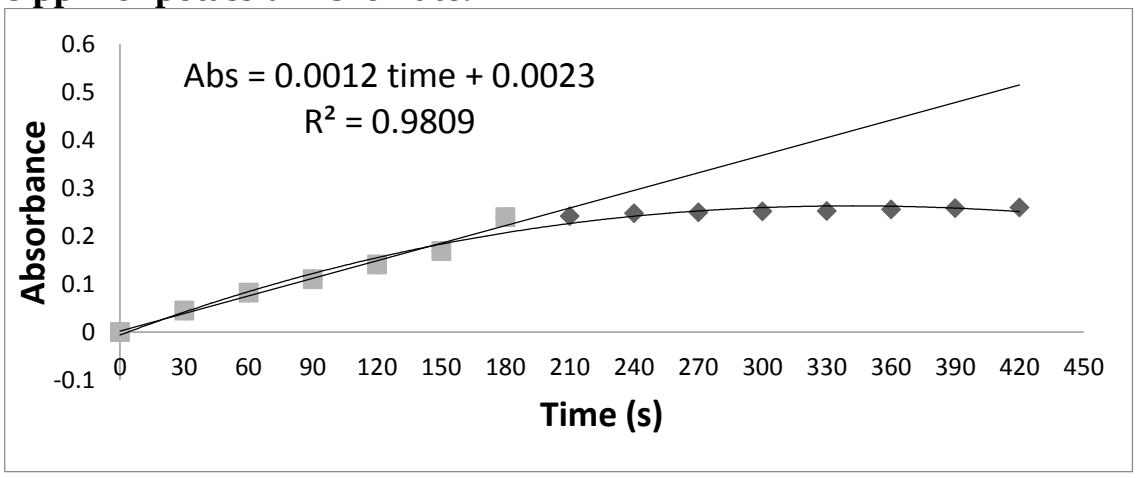

Figure3.11: Absorbance of I_2 for 8 ppm of bromate during 3 min.

9 ppm of potassium bromate:

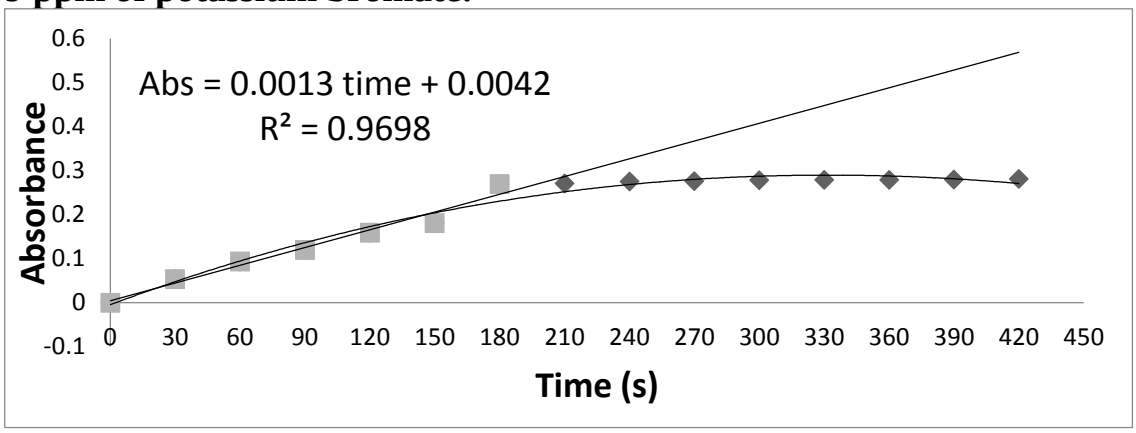

Figure3.12: Absorbance of I_2 for 9 ppm of bromate during 3 min.

10 ppm of potassium bromate:

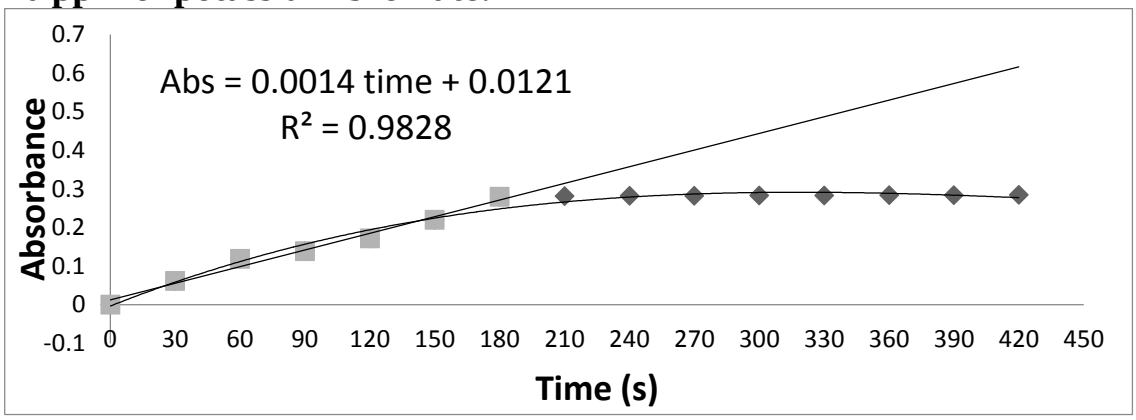

Figure3.13: Absorbance of I_2 for 10ppm of bromate during 3 min.

Calibration curve by tangent method:

Table3.4: Calibration curve of $KBrO_3$ in water by tangent method.

Concentration	Slope
1	0.0001
2	0.0003
3	0.0004
4	0.0005
5	0.0006
6	0.0009
7	0.0011
8	0.0012
9	0.0013
10	0.0014

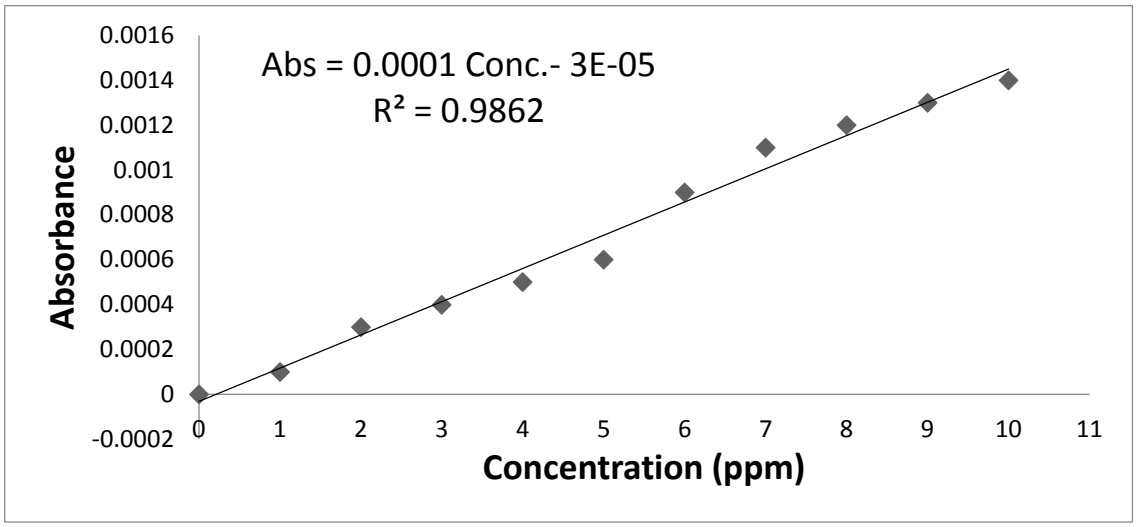

Figure3.14: Calibration curve of bromate in distilled water by tangent method.

Calibration curve by fixed time method:

In this method, the curve was established between the measuring absorbance over a predetermined time interval and related to concentration.

Table3.5: Calibration curve of $KBrO_3$ in water by fixed time method.

Concentration	Absorbance at 3 min
1	0.024133
2	0.049533
3	0.072133
4	0.0958
5	0.113533
6	0.1659
7	0.1995667
8	0.239433
9	0.2696
10	0.279667

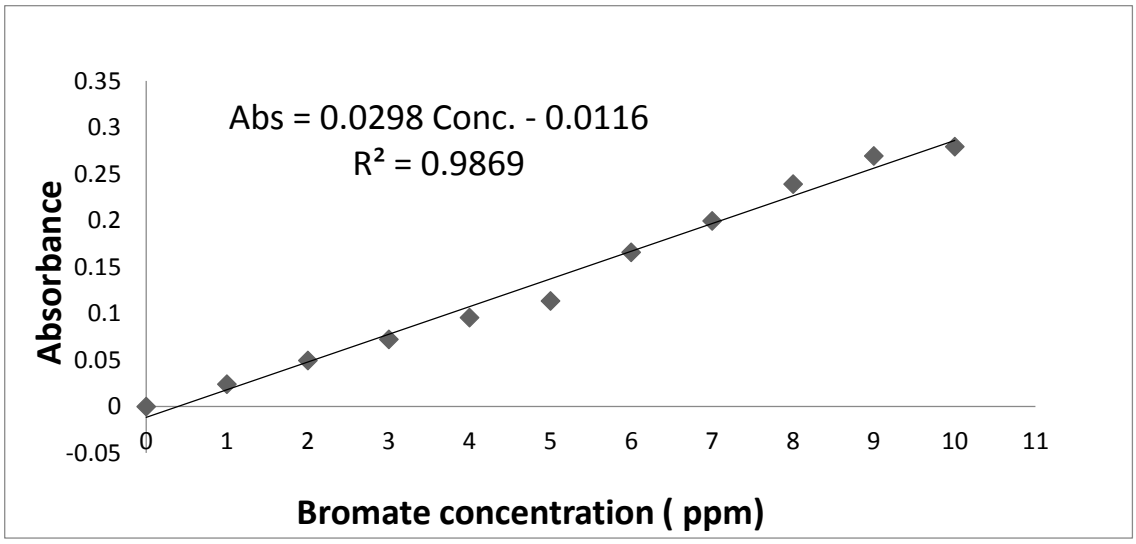

Figure3.15: Calibration curve of $KBrO_3$ in water by fixed time method.

As figure 3.14 and 3.15 show, the standard calibration curves was linear over the concentration range of 1-10 ppm and from this curves the fixed time method gave more reproducible results.

3.2.2 Calibration Graph of Bromate in Dough:

Table3.6: Calibration curve of $KBrO_3$ in dough by fixed time method.

Concentration of BrO_3^- added (ppm)	Absorbance
0	0.003
50	0.1438
100	0.2878
150	0.4318
200	0.5756
250	0.7196
300	0.8634

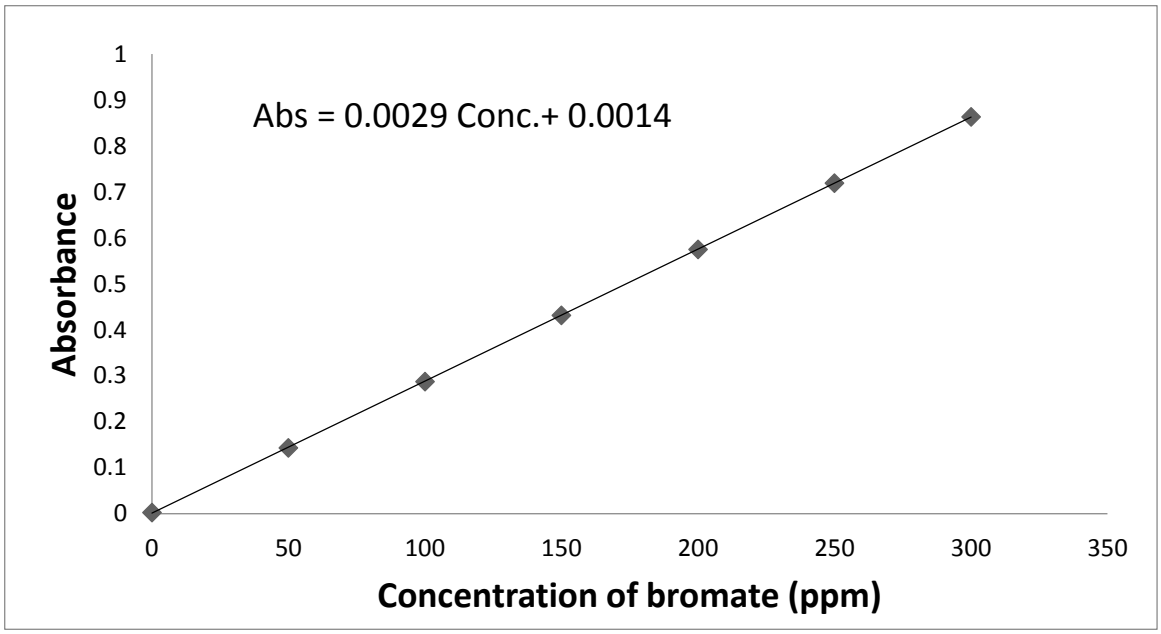

Figure3.16: Calibration curve of bromate in dough by fixed time method.

3.2.3 Calibration Graph of Bromate in Bread:

Table3.7: Calibration curve of $KBrO_3$ in bread by fixed time method.

Concentration of bromate added (ppm)	Absorbance
0	0.001
50	0.013
100	0.0332
150	0.0597
200	0.0847
250	0.0999
300	0.1321

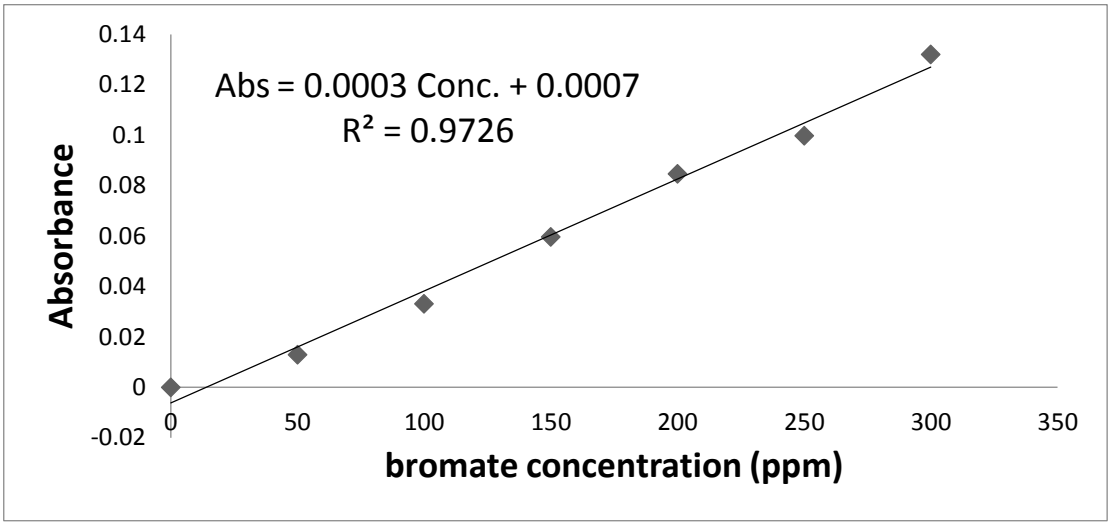

Figure3.17: Calibration curve of bromate in bread fixed time method.

3.3 Collecting Samples of Bread and Dough from Bakeries:

Table3.8: Collecting sample of dough and bread.

City	No. of Sample	Type of Sample	Result
Jerusalem	Sample 1	Bread	Bromated Free
	Sample 2	Bread	Bromated Free
	Sample 3	Bread	Bromated Free
	Sample 4	Bread	Bromated Free
	Sample 5	Bread	Bromated Free
	Sample 6	Bread	Bromated Free
	Sample 7	Bread	Bromated Free
Hifa	Sample 1	Bread	Bromated Free
Nablus	Sample 1	Bread	Bromated Free
	Sample 2	Bread	Bromated Free
	Sample 3	Bread	Bromated Free
	Sample 4	Bread	Bromated Free
	Sample 5	Dough	Bromated Free
	Sample 6	Bread	Bromated Free
Jenin	Sample 1	Bread	Bromated Free
	Sample 2	Dough	Bromated Free
	Sample 3	Dough	Bromated Free
	Sample 4	Bread	Bromated Free
	Sample 5	Bread	Bromated Free
	Sample 6	Bread	Bromated Free
Tulkarm	Sample 1	Dough	Bromated Free

The proposed method was used to investigate the level of bromate in bread done by some bakers in different regions in West Bank.

3.4 Effect of Temperature on Potassium Bromate:

The effect of temperature on bromate was studied in the range from 50˚C to the temperature in which all potassium bromate decomposed. The effect of temperature was studied at 50 ppm of bromate solution for each sample. The results are shown in table 3.9 (the absorbance measured at pH 1 and iodide concentration 0.1 M).

Table3.9: Effect of the temperature on $KBrO_3$.

Temperature/ ˚C	**Absorbance of I_2 in D.W.**	**Absorbance of I_2 in flour**
25	1.2431	1.2153
50	1.2114	1.0254
100	1.1091	0.6563
150	0.9874	0.2512
200	0.8641	0.0934
250	0.6534	0.0314
300	0.3932	0.0014
350	0.0123	
400	0.0012	

As table 3.9 shows, the absorbance of iodine (I_2) decreased upon increasing the temperature during the first 3 min after mixing the bromate. The absorbance was in the range from 350-400 ˚C ≈ zero, this meaning that the bromate decomposes and converts to KBr and O_2 from heating as in equation:

$$2\ KBrO_3 \longrightarrow 2\ KBr + 3\ O_2 \qquad \text{eq. 3.2}$$

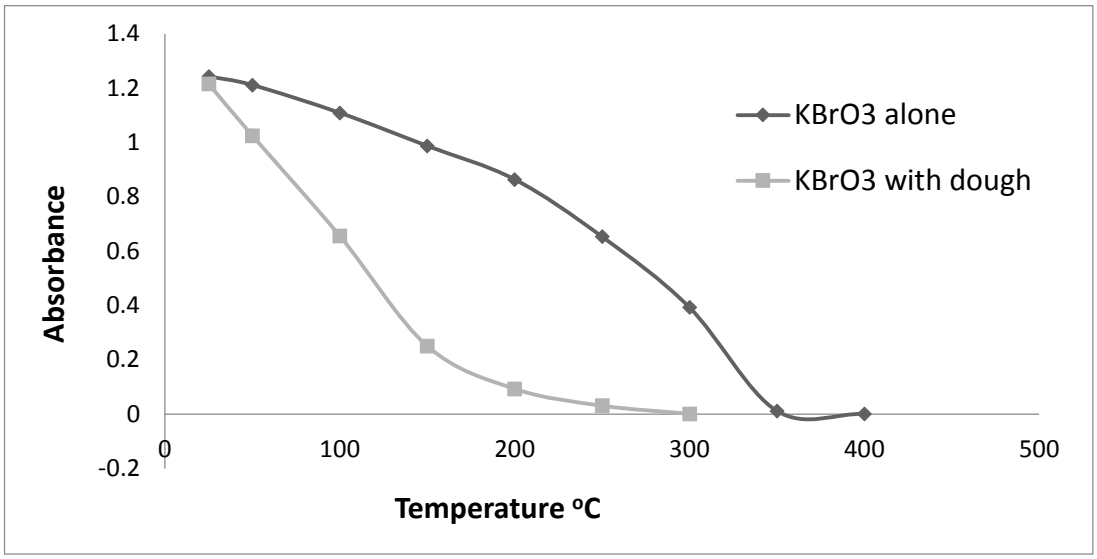

Figure3.18: Effect of the temperature on the potassium bromate.

Bromate decomposes in distilled water at (350-400 ° C) but in bread decomposes at (200-250 ° C). The decomposition of bromate in bread at low temperatures is due to the presence of metal ions in flour, which acts as a catalyst.

Metal ions in flour analyzed by ICP/MS:

Table3.10: ICP/ MS of flour.

Analyte	Mass	Net Intens. Mean	Conc. Mean (ppb)	Conc. (ppm)
Ag	107	40.333	0.057	0.01
Al	27	20300.24	13.253	2.65
Ba^{-1}	138	5402.539	3.539	0.71
Be	9	0	0	0
Bi	209	38	0.029	0.01
Ca	43	4455.736	726.405	145.28
Cd	111	5	0.031	0.01
Co	59	59.334	0.043	0.01
Cr	52	11372.421	7.512	1.5
Cs	133	8.333	0.005	0
Cu	63	5085.444	7.146	1.48
Fe	57	2802.848	74.154	14.83
Ga	69	260.338	0.185	0.04
In	115	12.373	0.007	0
K	39	35388241.17	5915.11	1183.02
Li	7	-5.667	-0.01	0
Mg	24	709924.108	785.069	157.01
Mn	55	42062.332	22.402	4.48
Mo	98	1075.544	1.43	0.29
Na	23	241316.041	106.546	21.31
Ni	60	643.691	2.189	0.44
Pb	208	52.667	0.061	0.01
Rb	85	6892.623	3.271	0.65
Sr	88	12075.594	4.468	0.89
V	51	-38.337	-0.019	0
Zn	66	6107.83	31.445	6.29

Conclusion:

The present work describes a simple, rapid and validated kinetic method for determining bromate. This method is based on bromate's reaction with iodide ions in an acidic medium to produce iodine (I_2). The absorbance of iodine was measured at 352 nm.

Recommendations for Future Work:

The determined method can be recommended for the routine control of bromate in bread bakeries. This method will be an alternative for laboratories, not equipped with expensive materials necessary for the procedure.

Can be recommended, that using bromate in bakeries should be allowed at a certain ratio (2 g per bags 60 Kg).

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كلية الدراسات العليا

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الملخص

مادة البرومات تستخدم في صناعة الخبز كعامل محسن ولكنها تصنف كمادة مسرطنة. قمنا بإيجاد طريقة سريعة و بسيطة و دقيقة لتحديد كمية البرومات في الخبز تعتمد على تفاعل البرومات مع الايوداید في وسط حمضي لإنتاج اليود و قد تم قياس امتصاص اليود على 352 نانومتر. وقد تفاعلت البرومات مع الايوداید خلال اول ثلاث دقائق بعد خلط المتفاعلات. في منحنى المعايرة الاول، كان منحنى خطي من 1الى 10 جزء من المليون من البرومات في الماء. في منحنى المعايرة الثاني، كان منحنى خطي من 50 الى 300 جزء من المليون من البرومات في العجين. في منحنى المعايرة الثالث، كان منحنى خطي من 50 الى 300 جزء من المليون من البرومات في الخبز. وقد تم تطبيق الطريقة المقترحة بنجاح في تحديد كمية البرومات في الخبز التجاري. في هذه الدراسة وجدنا أن استخدام 2 جم من برومات البوتاسيوم في الدقيق كيس (60 كلغ) هو آمن. لأن البرومات يتكسر عند درجة حرارة 400 درجة مئوية لوحدها ولكن في الخبز تتكسر البرومات عند درجة حرارة 200 درجة مئوية و ذلك لوجود العناصر في الطحين التي تعمل كعامل مساعد.