Health Risk Assessment for Bromate in Bottled Drinking Water and Natural Mineral Water

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ABSTRACT Bromate is mutagenic and possibly carcinogenic to humans. It may be found in drinking water as a by-product of water disinfection with ozone. The maximum limit for bromate in drinking water by WHO is 10 µg/L. Despite its toxicity, no regulatory limit has been established in Thailand. This study was conducted to encourage the establishment of regulatory measure to protect consumers from bromate. In this study, an ion chromatographic method for determination of bromate in drinking water was developed and validated, concentrations of bromate in bottled drinking water and natural mineral water were determined, and finally, the health risk from bromate in these waters was estimated. Results of the validation study showed that the performance characteristics of the method were fit for the intended use with the LOD and the LOQ of 1 and 3 µg/L, respectively. By using this method, 100 samples of bottled drinking water and 54 samples of local and imported natural mineral water were analyzed. The results showed that 20% (20 samples) of bottled drinking water and 39% (21 samples) of natural mineral water were ozonated and contained bromate in the range of <3-178 µg/L and <3-133 µg/L respectively, with the median of 14.9 µg/L and 20.5 µg/L respectively. The health risk assessment showed that the bromate levels in 65% of ozonated bottled water and 72% of ozonated natural mineral water were associated with the excess cancer risks higher than the WHO acceptable risk of 5×10^{-5}. It is recommended that the national authority establish regulatory limit for bromate to protect the consumers.

Key words: bromate, disinfection by-product, ion chromatographic method, drinking water, health risk

Introduction

Bottled drinking water sales have increased dramatically in Thailand. People prefer bottled water to tap water for many reasons, among which is the thought that bottled water is safer to health than tap water which is not always true. Bottled water may be produced from various types of water such as surface water, ground water and tap water. The treatment may include physical and chemical processes such as

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filtration followed by disinfection with chlorine, ultraviolet radiation or ozone.

Natural mineral water is normally rich in some kinds of mineral thought to be beneficial to the health. As water sits in contact with minerals, over time, it can absorb some of the mineral content. Natural mineral water is normally bottled at source. Several local and imported brands available in markets indicate the increasing acceptance by consumers in Thailand.

Disinfection of drinking water is essential to destroy microbial pathogens and to maintain microbiological safety. Disinfection involves the use of reactive chemical agents such as chlorine and ozone. For bottled water, ozone is highly effective and preferably used because it does not remain in water or change its taste. The use of disinfectants usually results in the formation of chemical by-products, some of which are potentially hazardous and cause adverse health effects after prolonged period of exposure. For ozonation, one of the disinfection by-products is harmful bromate that is not normally found in water(1). In water containing naturally occurring bromide, ozone oxidizes bromide ion (Br^-) to hypobromous acid (HOBr) and further oxidizes the hypobromite ion (BrO-) to bromate (BrO_3^-)(2). The rate of bromate formation depends on ozone dose, bromide concentration, temperature, contact time, pH and other factors such as the concentration of ammonia, bicarbonate/carbonate and some natural organic matters. Once formed, bromate is difficult to remove. By appropriately controlling disinfection conditions, it is possible to reduce the bromate concentration in water(3).

The concentration of bromate in water can be determined by various techniques. The choice of a method depends mainly on the limit of detection (LOD) and the limit of quantitation (LOQ) required to meet the regulation limits. These include ion chromatography (IC) with conductivity detection(4), IC with UV-Vis detection(5), ICP-MS(6). The LOD may range from <0.1 μg/L to 5 μg/L. In this study a method using IC system with suppressor and conductivity detector available in our laboratory was developed and validated to meet the maximum permissible level (MPL) of 10 μg/L.

For most people, exposure to bromate is unlikely to be significant. If ozone is used to disinfect drinking-water, intake of bromate is not unexpected. Many researches were carried out to study the health effect of bromate by using experiment animals such as rats and mice(7, 8, 9). The studies include, for example, acute toxicity, genetic toxicity, developmental and reproductive toxicity and carcinogenicity. Rats were found to be more sensitive to the effect of bromate than mice. In summary, bromate was found mutagenic both in vitro and in vivo(10).

Bromate produces tumors at multiple sites in male rats with kidney as the major target (adenomas and carcinomas). There is inadequate evidence of carcinogenicity of bromate in humans. In 1995, the Joint FAO/WHO Expert Committee on Food Additives (JECFA) concluded that the use of potassium bromate in food processing was not appropriate(11). The International Agency of Research on Cancer (IARC) has classified potassium bromate as a possibly carcinogenic to human (Group 2B)(12). The U.S. Environmental Protection Agency (EPA) has classified bromate as a probable human carcinogen (Group B2) by the oral route of exposure(13). The conclusion that bromate has potential to be a human carcinogen is based on the adequate evidence of carcinogenicity in experimental animals, especially rats. The International Program on Chemical Safety (IPCS) developed the tolerable daily intake (TDI) based on the non-linear approach for carcinogenicity of bromate. A TDI of 1 μg/kg bw-day was calculated based on a non-effect level of 1.3 mg/kg bw-day in a study by Kurokawa et. al.(14) and the use of the uncertainty factor (UF) of 1000(15).

The World Health Organization (WHO) has derived the guideline value of 10 μg/L for bromate in drinking water based on the study by DeAngelo(16). An upper-bound estimate of cancer potency for bromate is 0.19 per mg/kg body weight per day, based on low-dose linear extrapolation. This potency estimate corresponds to a drinking water unit risk of 5×10^{-6} per μg/L, assuming a daily consumption of 2 L/day for a 70 kg adult. The concentrations of bromate in drinking water associated with upper-bound excess lifetime cancer risks of 10^{-4}, 10^{-5} and 10^{-6} are 20, 2 and 0.2 μg/L respectively. The U.S. EPA and Health Canada have recommended the value of 10 μg/L as a MPL for bromate in drinking water(17, 18). For bottled water, the U.S. FDA has set the maximum residual disinfectant level for bromate at 10 ug/L(19) This value is set instead of the health-protection value due to the limitations in available analytical and treatment methods. According to WHO, this value is associated with excess lifetime cancer risks of 5×10^{-5}(20). Since 2003, the European Union (EU) has reduced the maximum limit for bromate in natural mineral waters and spring waters treated with ozone to 3 μg/L(21).

In Thailand there is no regulatory limit for bromate in bottled drinking water and natural mineral water. This study aimed at enhancing the consumer protection as well as raising public awareness of the health risk from bromate. A method for the determination of bromate was developed and validated, concentrations of bromate in bottled drinking water and natural mineral water were determined and health risk due to the ingestion of bromate in these waters was estimated. It is expected that the results of this study will encourage the national authority to establish the regulatory limits for bromate to protect consumers.

Materials and method

Instruments

Ion chromatography (IC) system (Dionex ICS-3000) with suppressor (ASRS 300) and conductivity detector (112 mA), autosampler with injection volume of 500 µL, IonPac AS19 Analytical column (4 × 250 mm) with guard column AG 19 (4 × 50 mm), ultrasonic bath, plastic tubes, and membrane filter (0.45 µm).

Standard solution:

Bromate standard solution 1000 µg/mL ± 10 µg/mL (95% confidence interval) (Environmental Express) for preparation of calibration curve and bromate standard solution 1000 µg/mL ± 1% (k = 2) (AccuIon Reference Standard) for preparation of spiked samples.

The working standard solutions were prepared by diluting the stock solution of bromate with deionized water (18.2 megohm-cm).

Mobile phase

10 mM KOH gradually increasing to 45 mM in 30 minutes.

Samples

One hundred samples of bottled drinking water and 54 samples of natural mineral water (19 local and 35 imported products) were collected from super markets and food shops. They were of different commercial brands or same brands but from different production locations. The production locations were in 21 provinces, mainly, in the central part of Thailand. These samples included products with and without declaration about ozonation in the labelling. For imported natural mineral water, they were from 14 countries such as Indonesia, Malaysia, Taiwan, Korea, Japan, Australia, Canada, France, Italy, United Kingdom, Germany, Iceland, Slovenia and Norway.

Procedure

The standard solutions of 2 - 20 µg/L were injected to the IC system for preparation of a calibration curve. The samples were injected to the IC system without any preparation step except for filtration with membrane filter. Sample might be diluted if the bromate concentration was beyond the upper limit of the calibration curve.

Method validation

The validation study of this method was carried out according to Eurachem Guide[22] and A Practical Guide for Single Laboratory Method Validation of Chemical Methods[23].

Calibration curve

A calibration curve was established from the injection of standard solutions of 2, 5, 8, 10, 12, 15 and 20 μg/L bromate (3 replicates for each level) to the IC. The Pearson correlation coefficient r, slope and intercept were calculated and the residuals were plotted.

The acceptance criteria: $r > 0.995$ and the residuals should distribute randomly around the zero line.

Linearity of the method

The same data as for the recovery and repeatability study were used to evaluate the matrix effect and linearity of the method. By plotting the average concentration values on the y- axis and the theoretical concentration on the x axis, the slope and intercept were calculated.

The acceptance criteria: The slope should be 1 and the intercept should be zero.

Recovery and repeatability

The spiked samples were prepared by adding bromate standard solution to give 3, 5, 11, and 17 μg/L into deionized water and mixed well. The bromate concentrations in the spiked samples were measured against the standard curve. For each level, 10 replicates were measured. The average value and standard deviation and %CV for each level were calculated. The acceptance criteria for recovery (%R) and repeatability (%CV) are 60-115 and 22, respectively.

Limit of detection (LOD)

The LOD was calculated from the intercept of the matrix calibration curve. The LOD is the concentration of bromate that gives the response of y-intercept plus 3 times standard deviation of the y-intercept.

The acceptance criteria: LOD should be < 2 μg/L (1/5 of the WHO guideline value for bromate of 10 μg/L)

The acceptance criteria for recovery, precision and LOD are based on the criteria adopted by Codex for food analysis[24].

Limit of quantitation (LOQ)

The LOQ was first estimated from the intercept of the matrix calibration curve. The estimated LOQ is the concentration of bromate that gives the response of y-intercept plus 10 times standard deviation of the y-intercept.

The concentration of bromate at the LOQ level was confirmed by measuring the spiked sample prepared at the concentration close to the estimated LOQ.

Measurement uncertainty

The measurement uncertainty of a result obtained by using this method was estimated according to the Eurachem Guide[25].

Determination of bromate in water samples

Each of the samples was injected to the IC system. The peak area was converted to the concentration by using the calibration curve.

Health risk assessment

In this study, the individual lifetime excess cancer risk (ECR) was estimated. The excess cancer risk is the probability that an exposed individual will develop cancer because of that exposure by age 70. Cancer risks increase with increasing bromate levels. The excess lifetime cancer risk can be estimated from the cancer potency factor (CPF) (risk per μg/L drinking water) as follows:

$$ECR = CPF \times C$$

CPF for bromate in drinking water is 0.5×10^{-5} per μg/L

C = concentration of drinking water or natural mineral water (μg/L)

Results

Method validation

The validation data showing the performance characteristics of the developed method are summarized. (Table 1) This method is fit for the intended use for the determination of bromate in drinking water.

Table 1 The validation data showing the performance characteristics of the method

Characteristics	Value	Criteria
Linearity range (3.0 - 20 μg/L)	r = 0.9975 Residuals distribute randomly around the zero line	r ≥ 0.995 Residuals distribute randomly around the zero line
Accuracy % (at 3.0, 5.0, 11.0 μg/L)	99 - 112%	60 - 115 %
Precision (RSD: repeatability)	2.4 - 8.4%	Predicted %RSDr = 22
LOD μg/L	1.0	≤ 2.0
LOQ μg/L	3.0	-
Measurement uncertainty (95% Confidence level)	13.5%	44%

The chromatograms of a bromate standard (10 μg/L), sample blank and sample blank spiked with 15 μg/L bromate, and water sample containing bromate at 10 μg/L are shown. (Figure 1a, 1b and 1c, respectively).

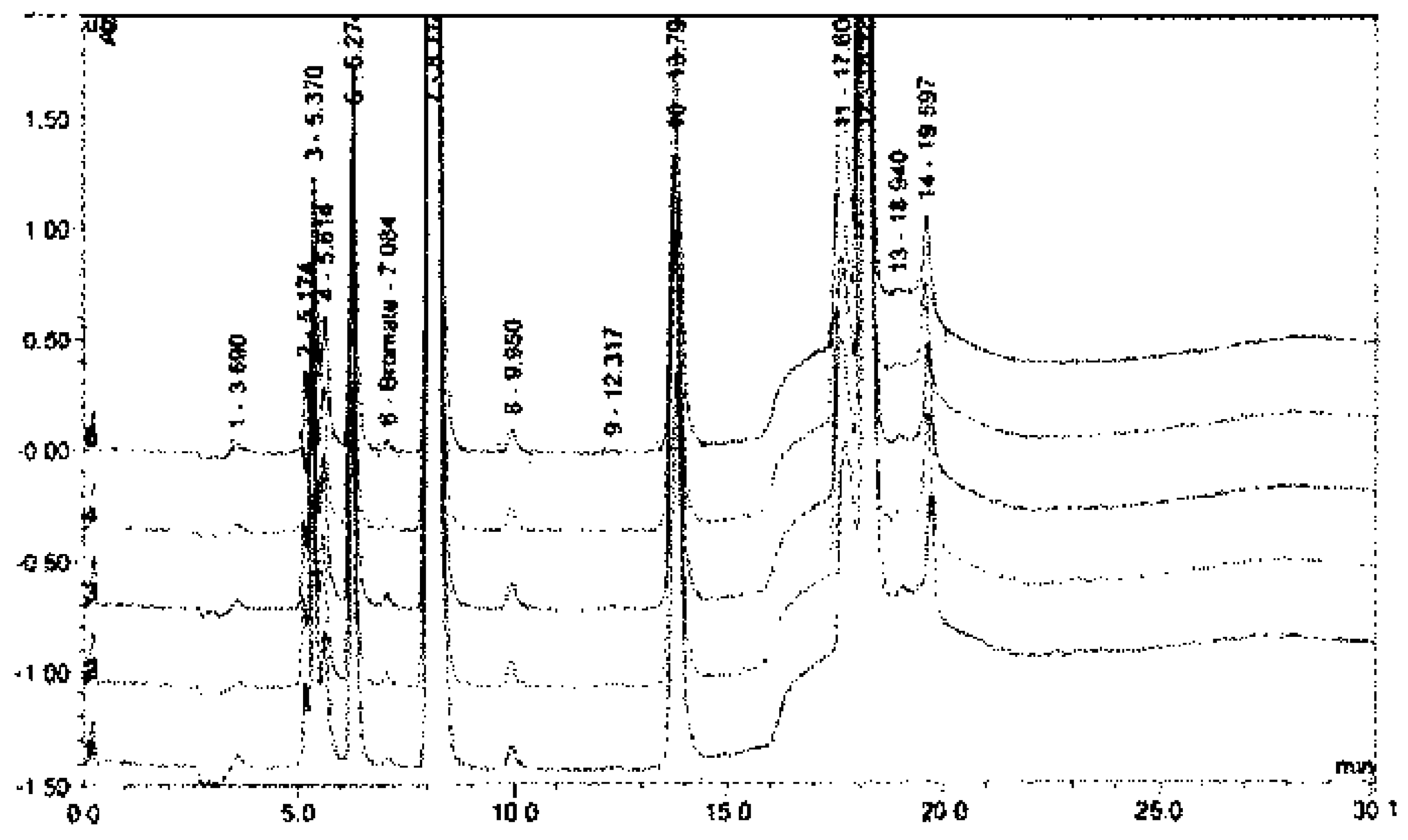

Figure 1 a

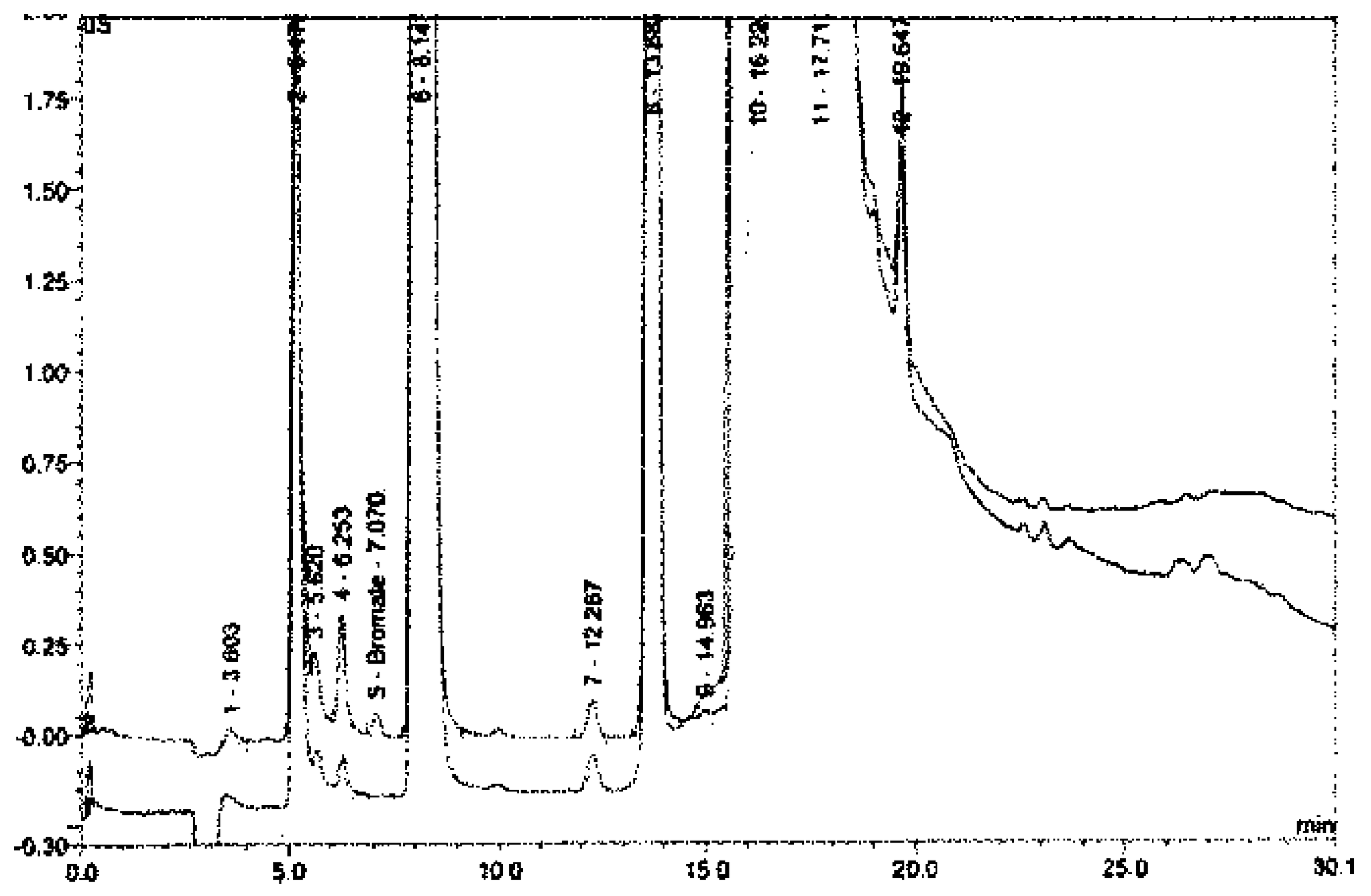

Figure 1 b

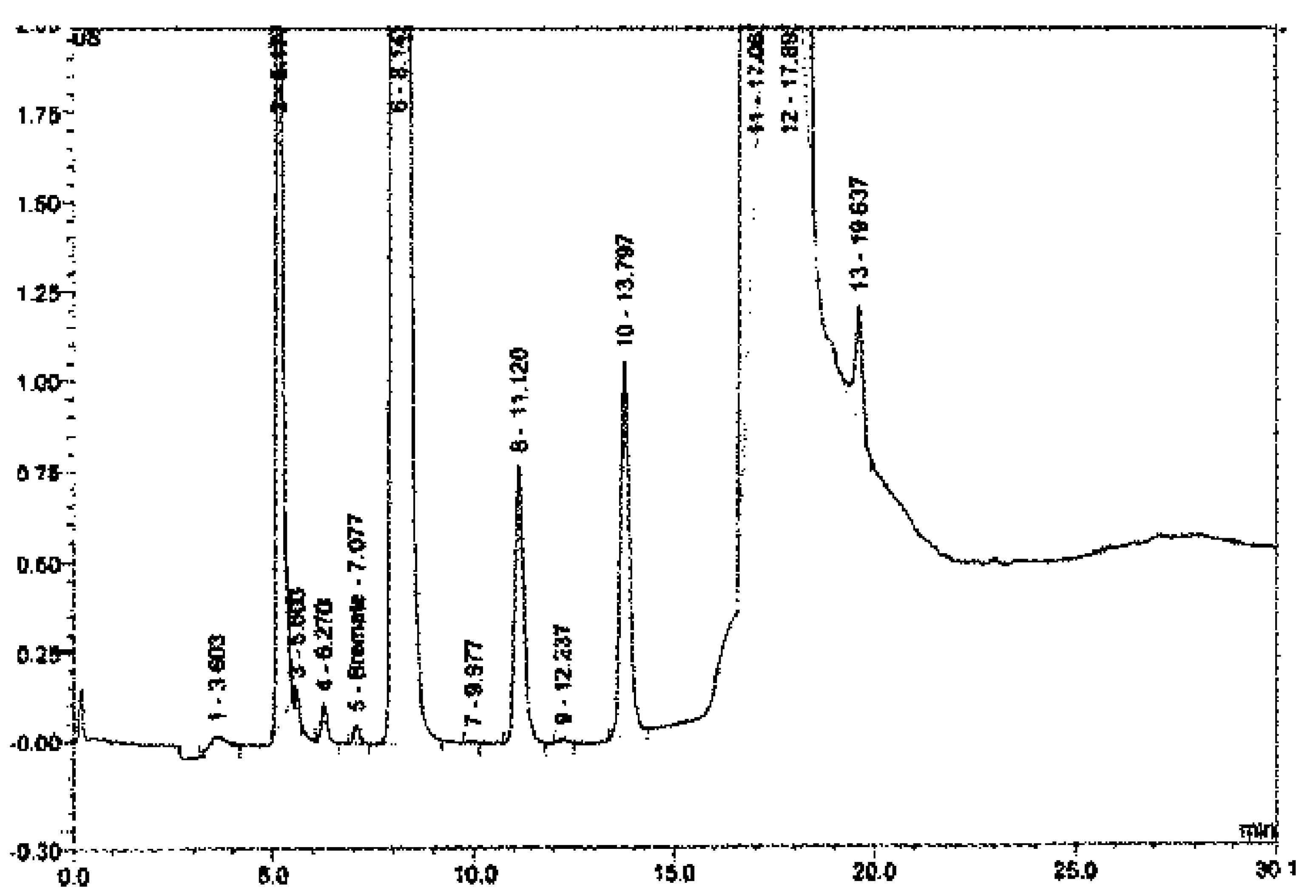

Figure 1 c

Figure 1 Chromatograms (conductivity vs retention time) of (1a) standard solution of bromate: 10 μg/L (5 injections), (1b) sample blank and sample blank spiked with 15 μg/L bromate and (1c) ozonated water sample containing bromate at 10 μg/L.

Bromate concentrations in bottled drinking water and natural mineral water

From the total of 100 samples of bottled drinking water analyzed, the analytical results showed that 20 samples (20%) contained bromate in the range of <3 - 178 μg/L with the median of 14.9 μg/L. More details are given. (Table 2) For natural mineral water, the analytical results showed that 21 from 54 samples (39%) contained bromate in the range of <3 - 133 μg/L with the median of 20.5. These were 15 local products and 6 imported products. The details are shown. (Table 3)

Health risk from bromate in ozonated water

The excess cancer risks due to the ingestion of bromate in ozonated bottled drinking water varied in a range of 1.5×10^{-5} – 9.0×10^{-4} with the median of 7.5×10^{-5}. The range for ozonated natural mineral water was 1.5×10^{-5}–7.0×10^{-4} and the median was 1.0×10^{-4}. The percentage of samples found to be associated with the risks higher than WHO accepted risk of 5×10^{-5} was 65 % (13 from 20 samples) for bottled water and 72% (15 from 21 samples) for natural mineral water. In the worst cases at 95th percentile, the risks were 7.0×10^{-4} and 6.0×10^{-4} for drinking water and natural mineral water respectively. The details of the excess cancer risk as well as descriptive statistics are shown. (Table 2 and 3 respectively).

Table 2 Bromate content in ozonated bottled drinking water, excess cancer risk due to ingestion of bromate in these waters and basic statistics.

Source/Location	pH	BrO_3(µg/L)	Excess cancer risk (1×10^{-5})
Pathum Thani (D1)	8.1	28.5	14.2
Pathum Thani (D2)	8.6	9.1	4.5
Pathum Thani (D3)	6.8	6.0	3.0
Pathum Thani (D4)	8.2	138	69
Pathum Thani (D5)	7.8	9.5	4.8
Pathum Thani (D6)	7.7	9.3	4.7
Pathum Thani (D7)	7.3	3.2	1.6
Bangkok (D8)	7.2	<3	1.5
Bangkok (D9)	7.8	60.2	30.1
Bangkok (D10)	8.0	15.0	7.5
Bangkok (D11)	8.1	16.3	8.2
Ayutthaya (D12)	7.4	68.4	34.2
Ayutthaya (D13)	7.5	29.5	14.8
Ayutthaya (D14)	8.0	14.9	7.4
Sara Buri (D15)	8.3	9.2	4.6
Samut Prakan (D16)	6.9	18.0	9.0
Sing Buri (D17)	8.3	178	89
Prachin Buri (D18)	7.8	13.6	6.8
Prachin Buri (D19)	7.8	12.8	6.4
Rayong (D20)	7.5	85.6	42.8
Min-Max		<3 - 178	1.5 - 89
Mean		36.4 ± 47.8	18.2
Median		14.9	7.5
5th percentile		3.2	1.6
95th percentile		140	70.0

Table 3 Bromate content in natural mineral water, excess cancer risk due to ingestion of bromate in these waters and basic statistics.

Source/Location	pH	BrO_3(µg/L)	Excess cancer risk (1×10^{-5})
Pathum Thani (M1)	7.6	70.0	35.0
Pathum Thani (M2)	7.9	108	54.0
Pathum Thani (M3)	7.6	133	66.5
Pathum Thani (M4)	7.7	85.9	43.0
Pathum Thani (M5)	7.7	53.0	26.5
Pathum Thani (M6)	8.0	100	50
Pathum Thani (M7)	8.3	102	51
Tak (M8)	7.5	18.3	9.2
Tak (M9)	7.9	22.4	11.2
Tak (M10)	5.7	21.7	10.8
Chiang Mai (M11)	7.5	12.3	6.2
Sing Buri (M12)	7.6	119	59.5
Kanchanaburi (M13)	7.5	<3	1.5
Kanchanaburi (M14)	8.1	20.5	10.2
Chumphon (M15)	8.1	<3	1.5
Taiwan (M16)	8.0	4.9	2.5
Indonesia (M17)	7.3	14.2	7.1
Malaysia (M18)	7.9	7.1	3.6
Koria (M19)	6.9	<3	1.5
Australia (M20)	6.7	12.0	6.0
Canada (M21)	7.6	<3	1.5
Min-Max		<3 - 133	1.5 - 66.5
Mean		45.7 ± 45.7	21.8
Median		20.5	10.3
5^{th} percentile		<3	1.5
95^{th} percentile		119	59.5

Discussion

The analytical method in this study has the LOD and the LOQ of 1.0 and 3.0 µg/L, respectively. This low LOD was attained by using large injection volume of 500 µL. It is found appropriate to be used for a purpose of determination of bromate in bottled drinking water and natural mineral water to see whether they comply with the WHO limit of 10 µg/L. However, if the expected target maximum limit for bromate in natural mineral water is 3 µg/L as set by the EU, a more sensitive method with lower LOD will be required.

The concentrations of bromate in ozonated bottled drinking water found in this study (<3 - 178 µg/L, median 14.9 µg/L,) are in agreement with those found in a study previously conducted in Thailand in 2008 by Ketkaew *et. al.* (8 - 216 µg/L)[26]. A study in India shows that the bromate concentrations in ozonated bottled drinking water from various regions are in the range of 2 - 30 µg/L[27]. Neither India nor Thailand has set a regulatory limit for bromate but bromate levels in our bottled water are much higher. They are higher than the maximum permissible level for drinking water as recommended by WHO and as regulated by US EPA, Canada and EU of 10 µg/L. High levels of bromate are also found in natural mineral water (range <3 - 133 µg/L, median 20.5 µg/L). Since there is no regulatory limit, ozone is used without proper control. This leads to high level of bromate in water and consequently, high health risk to consumers. It was observed in this study that the pH values, one of factors affecting bromate formation, was 7.5 and above in most samples containing high bromate concentration. By adjusting the pH value, among other factors, the formation of bromate can be controlled.

For natural mineral water, according to the Ministry of Public Health Notification No. 199, the Codex Standard and the EU Directive for natural mineral water, disinfection is not permitted[28, 29, 30]. Use of ozone or ozone-enriched air is permitted only for the purpose of removal of some unstable element such as arsenic, iron and manganese before filtration. For this purpose the dose of ozone is much lower than for disinfection purpose and the level of bromate, consequently, must be very low. This might be a reason why the EU lowered the limit of bromate in natural mineral water and spring water to 3 µg/L in 2003. In this study bromate was not detected in imported natural mineral water from EU countries. This finding strongly indicates that the formation of bromate can be controlled to the required level. High level of bromate found in natural mineral water is, therefore, the result of ozone misuse. Some manufacturers even declare the use of ozone for disinfection in the labelling. Thus the existing regulation should be more effectively enforced to protect the consumers before regulatory limit for bromate is established.

It is also observed that in some provinces, there were more products that did not complied with WHO guideline value than in other provinces. It is because some manufacturers produce more than one commercial brand. Among these are manufacturers in Patum Thani and Tak.

In Thailand potassium bromate is not permitted for manufacturing, importing or sale as food additive(31). Therefore, the primary route of exposure to bromate is drinking water. The estimated excess cancer risks due to ingestion of bromate in bottled drinking water and natural mineral water were very high. According to the U.S. EPA, the excess cancer risk that is below 1×10^{-6} (1 chance in 1,000,000) is so small as to be negligible and risks above 1×10^{-4} is sufficiently large that some sort of remediation is desirable. Excess cancer risk that ranges between 1×10^{-6} and 1×10^{-4} is generally considered to be acceptable(32). In this study, 35% and 50% of ozonated bottled drinking water and natural mineral water, respectively, are associated with excess cancer risk at level that needs remediation. When compare with WHO acceptable risk value of 5×10^{-5}, only 35% of ozonated bottled drinking and 28% of ozonated natural mineral water are considered safe from health risk. In the worst case at 95th percentile, the risk for bottled drinking water is 7.0×10^{-4}. This risk value means that 7 in 10,000 people regularly drinking ozonated bottle drinking water are expected to develop cancer at age 70 which is 14 times higher than the acceptable risk. For natural mineral water, the 95th percentile risk is 6.0×10^{-4} which is 12 times higher than the acceptable risk.

Conclusion

The validated method by using IC with suppressed conductivity detector provides the required sensitivity and detection limit that meet the maximum permissible level of 10 μg/L bromate in drinking water as recommended by WHO. This study showed that ozonated bottled drinking water and natural mineral water contained bromate at the levels varying in a wide range with most levels much higher than 10 μg/L. These levels are associated with the excess cancer risk from less than 1 to 14 times higher than the acceptable risk of WHO. It is obvious that use of ozone without proper regulation has led to high health risk to consumers. Since bottled drinking water and natural mineral water are the main sources of water for consumption in various groups of people in Thailand, and since they are the primary sources of exposure to bromate, establishment of regulatory limits for bromate is strongly recommended. In the meantime, the information about the risk should be communicated to consumers so that they become aware of their health and take care of themselves before regulation can be enforced. For manufacturers, social responsibility should be raised taking into account safety aspect when producing water for drinking purpose.

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การประเมินความเสี่ยงต่อสุขภาพจากโบรเมตในน้ำดื่มบรรจุขวด และน้ำแร่ธรรมชาติ

ทิพวรรณ นิ่งน้อย กัญญา พุกสุ่น และกรรณิกา จิตติยศรา

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บทคัดย่อ โบรเมตเป็นสารก่อกลายพันธุ์และอาจก่อมะเร็งในคน ในน้ำดื่มอาจพบโบรเมตได้โดยเกิดจากกระบวนการฆ่าเชื้อโรคด้วยโอโซน องค์การอนามัยโลกกำหนดให้มีโบรเมตในน้ำดื่มได้ไม่เกิน 10 ไมโครกรัมต่อลิตร แต่ในประเทศไทยยังไม่มีการกำหนดค่ามาตรฐาน งานวิจัยนี้มีวัตถุประสงค์เพื่อกระตุ้นให้มีมาตรการคุ้มครองผู้บริโภคให้ปลอดภัยจากสารโบรเมต โดยได้พัฒนาวิธีวิเคราะห์โบรเมตในน้ำดื่มด้วยเทคนิคไอออนโครมาโตกราฟ สำรวจปริมาณโบรเมตในน้ำดื่มบรรจุขวดและน้ำแร่ธรรมชาติ และประเมินความเสี่ยงจากโบรเมตในน้ำ ผลการทดสอบความถูกต้องของวิธีที่พัฒนาพบว่ามีความเหมาะสมต่อการใช้งานโดยมีขีดจำกัดของการตรวจพบ และการวัดปริมาณที่ 1 ไมโครกรัมต่อลิตรและ 3 ไมโครกรัมต่อลิตรตามลำดับ ผลของการวิเคราะห์น้ำดื่มบรรจุขวด 100 ตัวอย่าง และน้ำแร่ธรรมชาติที่ผลิตในประเทศและนำเข้าจากต่างประเทศ 54 ตัวอย่าง พบว่าร้อยละ 20 (20 ตัวอย่าง) ของน้ำดื่มบรรจุขวดและร้อยละ 39 (21 ตัวอย่าง) ของน้ำแร่ธรรมชาติมีการใช้โอโซน และพบโบรเมตอยู่ในช่วง <3 - 178 ไมโครกรัมต่อลิตร และ <3 - 133 ไมโครกรัมต่อลิตร โดยมีค่ามัธยฐานที่ 14.9 และ 20.5 ไมโครกรัมต่อลิตรตามลำดับ ผลการประเมินความเสี่ยงจากโบรเมตพบว่า ร้อยละ 65 และ ร้อยละ 72 ของน้ำดื่มบรรจุขวดและน้ำแร่ธรรมชาติที่ผ่านโอโซนมีค่าความเสี่ยงต่อการเป็นมะเร็งสูงกว่า 5×10^{-5} ซึ่งเป็นค่าที่องค์การอนามัยโลกยอมรับ ดังนั้นหน่วยงานที่รับผิดชอบจึงควรกำหนดค่ามาตรฐานสำหรับโบรเมตเพื่อคุ้มครองผู้บริโภคให้มีความปลอดภัย