Interlaboratory Results for Blood Lead Proficiency Testing Program in Thailand

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Abstract

Between December 1994 to 1995. Faculty of Medical Technology, Mahidol University have a thirthteen laboratory to paticipate in Blood Lead Proficiency Testing Program (BLPT). Every-month the Toxicology Unit in Faculty of Medical Technology Mahidol University which served for Reference Laboratory Center for BLPT send the three lots of whole blood sample which made from bovine blood to the participants by EMS. After evaluation in term of accuracy : using 75 per cent correctly when compared with the target value. There are nine laboratory which have successful in blood lead below 20 μg/dL and three laboratory which passing the analysed when the concentration of blood lead at 21-50 μg/dL and 51-80 μg/dL.

In term of precision evaluation the planning process will show that laboratory using Graphite Furnace Atomic Absorption Spectrophotometer (GFAAS) reported reliability than using Flame Atomic Absorption Spectrophotometer (FAAS). From joining with this program the participants will get three advantages, First the target value from the reference laboratory in Thailand and Singapore are similar with the Center for Disease Control (CDC) from the United State of America which known world wide for Reference Laboratory. In the second participants will develope the analysis and solve the problem by himself, The third advantage : will showing the superior, methodology and instruments which garantee for quality assurance and guideline to standardized blood lead analysis in the International level.

Key word : Blood Lead, Interlaboratory, Proficiency Testing, Reference Laboratory Center, Graphite Furnace Atomic Absorption Spectrophotometer (GFAAS), Flame Atomic Absorption Spectrophotometer (FAAS)

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Lead is an environmental pollutant that impairs health especially the central nervous system, reproductive organs, immune system and kidney. Lead affects both men and women including infertility, hypertension and premature delivery. Long term lead exposure at level 70 µg/dl may generate irreversible functional and morphological renal change. A low level of lead may induce colic due to its effect on the gastrointestinal tract. Inhibition of heme synthesis and shortened life span of the erythrocytes may cause anemia as a chronic system of lead exposure(1).

Blood lead analysis may be used for screening health programmes and making diagnostic evaluation. The programme should assess industrial workers who have been exposed to lead and its compounds and should study the effects of environmental pollution of lead in the population.

Proficiency testing is a requirement as a routine part of hospital and clinical laboratories to perform tests for medicare patients. So screening or investigation by blood lead determination are needed for evaluation of the current analytical method. The toxicology laboratory units, Department of Clinical Chemistry, Faculty of Medical Technology, Mahidol University have provided a Blood Lead Proficiency Testing Programme : (BLPT) for the participants, designed to monitor and assist laboratory performances for lead analysis in Thailand.

MATERIAL AND METHOD

1. Whole blood lead samples (Specimen Preparation)

Fresh blood was collected from a bovines and preserved with dipotassium EDTA as anticoagulant (final concentration 1.5 g/l). This process is prefered to heparin to ensure that the clotting does not occur during haemolysis(2). The homogenous EDTA blood was designed for appropiate lead concentration and collected in 5.0 ml. of venoject tube from Terumo(R). Those specimens were kept in a deep freeze (-30°C) before shipping (approximately 24-72 hours). The home made, three difference concentration samples (three lots) were transported to the participants every month in boxes and sent by EMS (Emergency Mail Services) without temperature control. In general, samples reached the destination within 24 hours in Bangkok and the outskirts but for provincial laboratories it took 2 days.Thirty samples were sent out, including seven duplicates for precision study. Between the programme, one lot of specimens was not evaluated because two laboratories complained about the characteristics of unsuitable specimens, due to some fibrin in the specimens and it took about 2 weeks for transportation.

2. Criteria for analysis

The criteria used for estimation of accuracy and precision of each lot (determination) followed that of the Centre for Disease Control (CDC) (3), United States of America which accepted ± 4 µg/dL from target value for a lead level that was lower than 40 µg/dL and ± 10 per cent from target value for a lead level that was higher than 40 µg/dL.

3. Reference laboratories for target value

This process was carried out from December 1994 to December 1995. Two reference laboratories performed lead analysis to confirm the target ranges by using the Zeeman Graphite Furnace Atomic Absorption Spectrophotometer : Toxicology Units, The Department of Clinical Chemistry, Faculty of Medical Technology, Mahidol University, Bangkok, Thailand and the Department of Science Service, Institute of Science and Forensic Medicine, Singapore.

4. The participants

There were 13 participating laboratories for this programme which used methodologies to analyse samples of blood lead. However, only 4 categories of methods were used: Anodic Stripping Voltametry (ASV) and Atomic Absorption Spectrometry (AAS) in which either extraction with MIBK (EAAS), or digestion and dryashing (DAAS), and Graphite Furnace (GFAAS).

5. Reporting system

Five pages of report results were returned to the participants every month, three pages for the analysis and status of general performance; the fourth page for the accuracy and precision including the target value and target range of all the participants. The last page demonstrated distribution of all laboratory results. The acceptable range is shown in Fig. 1.

RESULTS

This programme used code lab no. 1-14 instead of the name of the laboratory. After three

months, laboratory code no. 7 did not participate in the programme. So there were only 13 laboratories for the programme evaluation. The distribution of laboratories location, method and status of the participants in blood lead Proficiency Testing : eight laboratories were in the centre of Bangkok, two laboratories in the outskirts of Bangkok and three laboratories from other provinces.

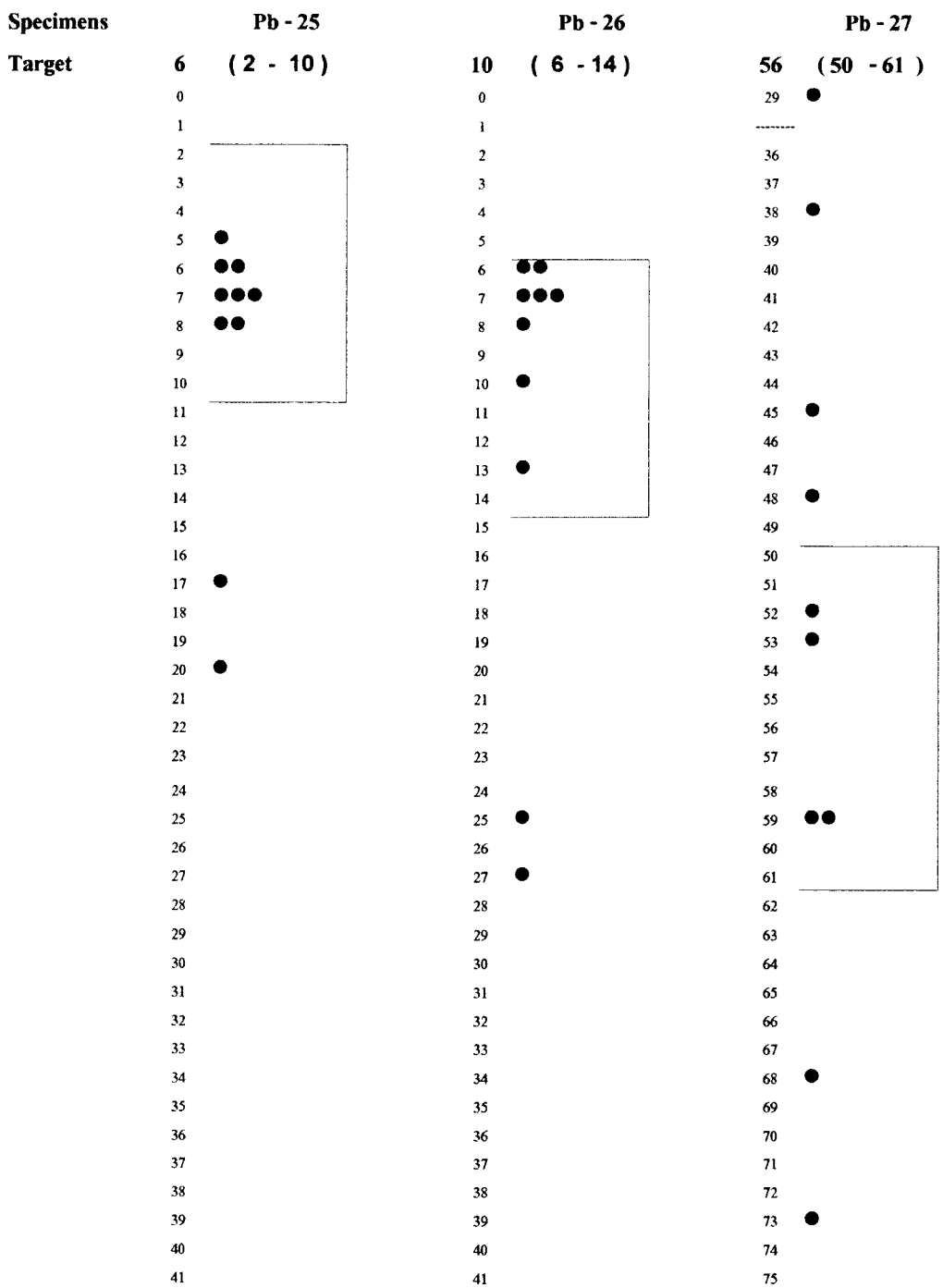

Fig. 1. Reporting system of each month (three lots) shows the distribution of all laboratories results with the acceptable range and target value.

The samples were sent out from December 1994 to December 1995. The three lead levels were classified : less than 20 µg/dL fourteen lots, range 20-50 µg/dL and 50-80 µg/dL four lots and eleven lots respectively. The distribution of these results is shown in Fig. 2.

The accuracy of reports which were received from the participants ranged from zero to

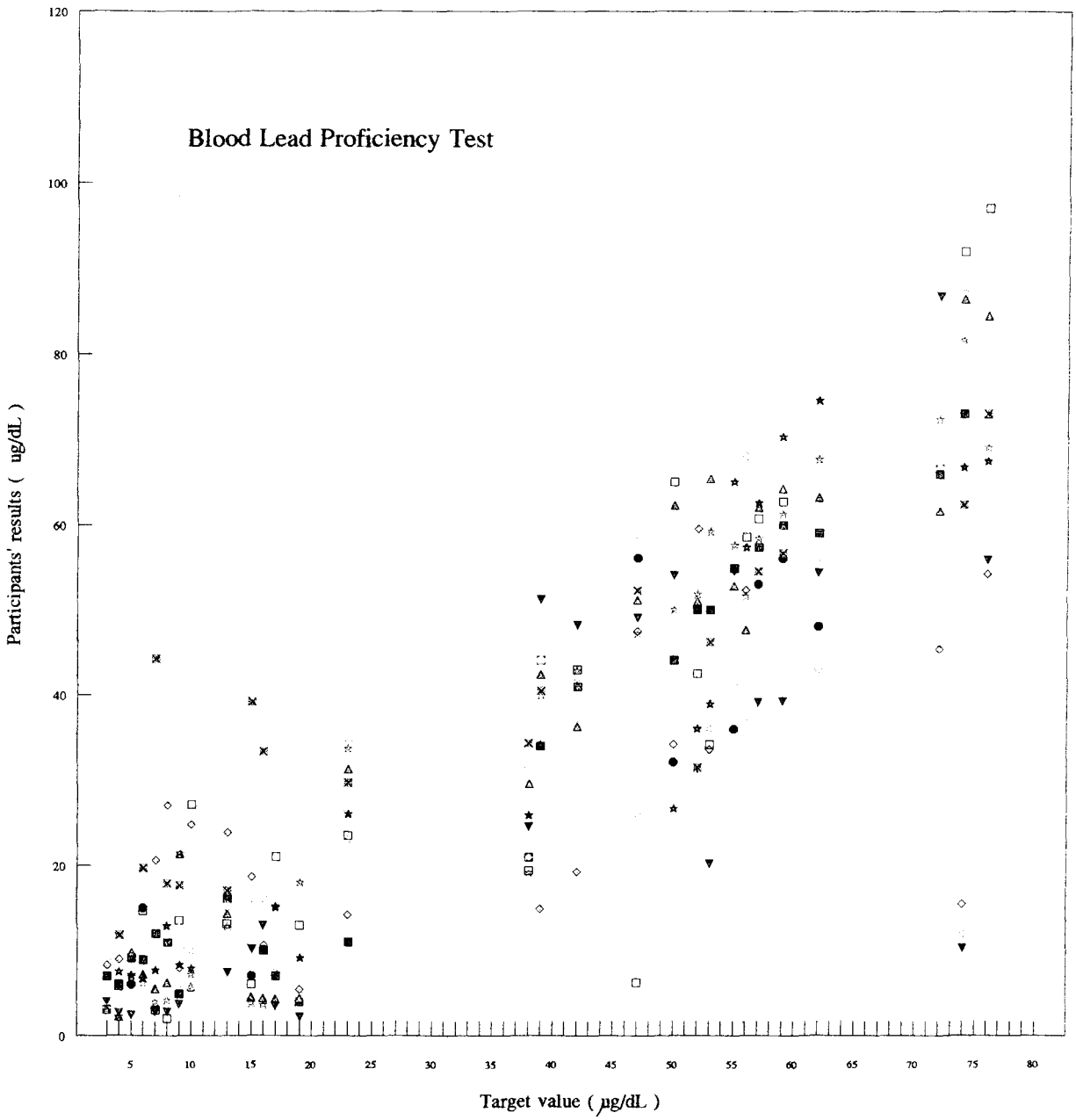

Fig. 2. Distribution of whole blood lead control materials, analysing by 13 participant labs in BLPT program during December 1994 to December 1995.

Table 1. Show the distribution of participant laboratories, report and percentage of acceptable in variable concentration of blood lead (* acceptable score over 75%).

Lab Code Number	Lead level < 20 μg/dL Total 14 Lots		Lead level 21-50 μg/dL Total 4 lots		Lead level 51-80 μg/dL Total 11 lots		Methodology
	No. Report	Accept(%)	No. Report	Accept(%)	No. Report	Accept(%)	
1	10	10 (100)*	3	2 (66.7)	10	9 (90)*	ASV
2	12	4 (33.3)	4	1 (25.0)	7	1 (14.3)	EAAS
3	14	11 (78.6)*	4	2 (50.0)	11	6 (54.5)	GFAAS
4	13	6 (46.2)	4	2 (50.0)	9	3 (33.3)	GFAAS
5	14	13 (92.9)*	4	1 (25.0)	10	0 (0)	EAAS
6	11	11 (100)*	-	-	9	2 (22.2)	GFAAS
8	11	9 (81.8)*	2	0 (0)	11	5 (45.5)	GFAAS
9	9	5 (55.6)	4	2 (50)	9	1 (11.1)	GFAAS
10	10	10 (100)*	4	0 (0)	7	4 (57.1)	GFAAS
11	14	14 (100)*	4	4 (100)*	11	11 (100)*	GFAAS
12	14	14 (100)*	4	4 (100)*	11	11 (100)*	GFAAS
13	11	2 (18.2)	4	3 (75.0)*	10	5 (50)	DAAS
14	4	3 (75.0)*	-	-	3	0 (0)	GFAAS

Note : Lab 7 withdrawn after 2 months

Table 2. Precision performance study for duplicate determination of blood lead in various concentrations from 13 participants (✔ = accept ✖ = Imprecision ND = no result) (acceptable range using the difference value of duplicate sample ±4 μg/dl and ±10% for lead was lower than 40 μg/dl and higher than 40 μg/dl respectively).

Lab Code Number	Blood lead level (μg/dL)							Methodology or Instrument
	5	10	15	40	50	60	80	
1	✔	✔	ND	✔	ND	✔	✔	ASV
2	✔	✖	✖	✖	✖	ND	✖	EAAS
3	✔	✔	✔	✔	✖	✔	✖	GFAAS
4	✔	✔	✔	✔	✔	✔	✔	GFAAS
5	✔	✖	✔	✖	✖	✖	✖	EAAS
6	✔	✔	ND	ND	ND	✖	ND	GFAAS
8	✔	✔	ND	ND	ND	✔	✔	GFAAS
9	✔	✔	ND	✖	✔	✔	✖	GFAAS
10	✔	ND	✔	✔	✔	✔	✖	GFAAS
11	✔	✔	✔	✔	✔	✔	✔	GFAAS
12	✔	✔	✔	✔	✔	✔	✔	GFAAS
13	✔	✖	✖	✖	✔	✔	✖	DAAS
14	ND	ND	ND	ND	ND	ND	ND	GFAAS

Note : Lab 7 withdrawn after 2 months

one hundred per cent as shown in Table 1. There were 5 participants who reported absolutely correct one hundred per cent for blood lead levels less than 20 μg/dL (0-20 μg/dL). However, nine laboratories reported 75 per cent correct at the blood lead less than 20 μg/dL. Only three laboratories reported correctly an acceptable data for blood lead concentration over 20 μg/dL : 21-50 μg/dL and 51-80 μg/dL respectively.

The precision of performance was studied by sending the duplicated samples at different times and after the results were evaluated we found

that most of the laboratories had good precision but lacked accuracy, especially at higher lead levels when using MIBK or Dryashing technique for samples preparation. The data showed higher imprecision and inaccuracy than those by graphite furnace atomic absorption spectrophotometer as shown in Table 2.

DISCUSSION

From the summaries in Table 1, thirteen laboratories participated in this programme. Fourteen lots of low lead level (less than 20 μg/dL) were distributed and nine participants (64.2%) performed successful goals in which the results were correct for the score accepted (being over 75 per cent) but for the remaining 15 lots of high lead level (21-80 μg/dL) most of the participients showed unsuccessful performance except three laboratories achieving an accepted score.

However, the evaluation of the proficiency testing programme of blood lead in Thailand correlated with the programme that belongs to the Blood Lead Proficiency Testing programe (BLPT) which was conducted by the Centre for Disease Control[4].

Table 2 shows the performance of participating laboratories using various methodology and instruments for blood lead determination. The imprecision from blood lead performances is passibly cause by the poor calibration curve. The most likely sources of error that would account for the overestimate of lead at the lower concentrations and underestimation of lead at the higher concentration should be identified. The overestimation may be a problem of lead contamination of glassware, reagents or equipment and inadequate blank correction. The underestimation may be involved in the incomplete analytical recovery in extraction or concentration step and non linearity of calibration curves assumed and extrapolated analytical range.

In terms of precision, most performances were accepted as shown by the results of samples at all levels of blood lead determination. The organizer of BLPT sent three duplicate lots of low lead level (5 μg/dL, 10 μg/dL and 15 μg/dL) two duplicate lots of 40 μg/dL and 50 μg/dL respectively and higher levels of 60 and 80 μg/dL which were evaluated as shown in Table 2. We found that most imprecision was due to the problem of the preparation of samples and operating the Atomic Absorption Spectrophotometer. Preparation of sample fluctuation in the sample line which decreased the sensitivity of the instrument. So cleaning of instruments and check with the control material for 5-10 samples analysed solved the problem of insensitivity and imprecision. This conclusion is consistent which the report from CAP (the College of American Pathologists) proficiency survey which suggested that graphite furnance instruments have excellent sensitivity[5].

In future, laboratories should be concerned with quality assurance in order to improve the quality of the laboratory for both routine survices and research in the area of toxicology. The director of the clinical laboratory of toxicology must consider three factors: selection of instruments in laboratory performance; well trained technologists and in house maintenance of instruments ; and vision of the total quality management for planning and standardizing of laboratories to international accreditation.

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REFERENCES

1. Pidetcha P, Intaramanee S, Suwanthon L. Lead poisoning and quality assurance. Faculty of Medical Technology, Mahidol University 1997: 7-19.
2. Stephen TW, Frank P. The stability of human blood lead in storage. J of Anal Toxic 1985; 9: 85-8.
3. Licensure and Proficiency Testing Division, Bureau of Laboratories, Centre for Disease Control, Public Health Service, U.S Dept. of HEW. Atlanta, GA 30333.
4. Boone J, Hearn T, Lewis S. Comparision of laboratory results for blood lead with results from a definitive method. Clin Chem 1979; 25: 389-93.
5. Porter WH, Moyer TP. Lead In : Burtis CA, Ashwood ER,eds Teitz textbook of Clinical Chemistry 2nd ed, Philadelphia, Pa, WB Saunders : 1994; 1220-2.

ศึกษาผลการตรวจสอบประสิทธิภาพการวิเคราะห์สารตะกั่วในเลือดของห้องปฏิบัติการชันสูตรในประเทศไทย

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ในระหว่างเดือนธันวาคม 2537 ถึงเดือนธันวาคม 2538 หน่วยพิษวิทยา ภาควิชาเคมีคลินิก คณะเทคนิคการแพทย์ มหาวิทยาลัยมหิดล ได้จัดโครงการตรวจสอบประสิทธิภาพ (Proficiency Testing Program) ของการวิเคราะห์สารตะกั่วในเลือด ให้กับห้องปฏิบัติการที่สนใจในประเทศไทย 13 แห่ง โดยการนำส่งเลือดในรูปเลือดครบส่วน (whole blood) ซึ่งเตรียมจากเลือดวัวและใส่หลอดสูญญากาศไปให้ห้องปฏิบัติการชันสูตร เดือนละ 3 หลอด ซึ่งมีความเข้มข้นหลายระดับผลการประเมินพบว่ามีห้องปฏิบัติการที่ทำการวิเคราะห์ได้ค่าที่ถูกต้องว่า (accuracy) โดยถือการประเมินผลความถูกต้องมากกว่า 75% ในระดับความเข้มข้นตะกั่วในเลือดต่ำกว่า 20 μg/dL ทั้งหมด 9 แห่ง (64.2%) และพบว่ามีความถูกต้องทั้งสองระดับ ในระดับความเข้มข้น 21-50 μg/dL และ 51-80 μg/dL มีอยู่ 3 แห่ง (23.1%) ในด้านของความแม่นยำ (precision) ของการวิเคราะห์สารตะกั่วในเลือดพบว่าการใช้ระบบของเครื่องอะตอมมิคแอบซอบชั่นสเปคโตรโฟโตเมตรี โดยหลักของแกรไฟต์เฟอนาสหรือไม่มีเปลวไฟจะให้ผลดีกว่าการใช้อะตอมมิคแอบซอบชั่นสเปคโตรโฟโตเมตรี โดยหลักของการย่อยสาร (extraction) หรือการเผาสารให้เป็นเถ้า (dryashing) แล้วนำมาวัดโดยมีเปลวไฟ อย่างไรก็ตามจากการเข้าร่วมโครงการตรวจสอบประสิทธิภาพของการวิเคราะห์สารตะกั่วในเลือดครั้งนี้ นอกจากการประเมินผล ใช้ระบบของ Centre for Disease Control ของประเทศสหรัฐอเมริกาแล้วยังมีประโยชน์สำหรับห้องปฏิบัติการ 3 ประการคือ เป็นข้อมูลในการเลือกเครื่องมือสำหรับการวิเคราะห์ สองมีการเตรียมบุคลากรเพื่ออบรมเทคนิคการทำงาน และการบำรุงรักษาเครื่องมือได้อย่างมีประสิทธิภาพ และประการสุดท้ายคือ การเตรียมวางแผนงานเพื่อบริหารจัดการให้ห้องปฏิบัติการพิษวิทยามีมาตรฐานในระดับนานาชาติ

คำสำคัญ : สารตะกั่ว, ระหว่างห้องปฏิบัติการ, โพรพีเซียนซี เทสติ้ง, ศูนย์ห้องปฏิบัติการอ้างอิงแกรไฟต์เฟอร์นาสอะตอมมิคแอบซอบชั่นสเปคโตรโฟโตเมตรี, เฟลมอะตอมมิคแอบซอบชั่นสเปคโตรโฟโตเมตรี

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