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Assessment of possible variation levels in percentage recovery of Polychlorinated Biphenyls (PCB) using an Automated SPE Extractor

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Abstract
An assessment of percentage recovery using automated Solid Phase extractor (SPE) and traditional vacuum manifold SPE techniques for preliminary polychlorinated biphenyls extraction or clean-up process. The data presented in this paper shows a general interpretation, which reflects the slight variation in the stability of the percentage recovery for different PCBs samples after SPE clean up. From the result, it stated that traditional SPE for the PCB analytical clean-up methods is favourable when compared with the automated SPE methods. The difference in recovery process is clear except in the water sample (9002) and some soil samples (9003 and 9004) where the PCBs result were the same and below the limit of detection (< 0.02 µg/l and < 0.2 mg/l) for the water and soil samples respectively. The result shows that efficiency of both traditional PCB analytical clean-up methods and the automated SPE methods are in ratio of 4:3 respectively. Thus, resulting to a percentage efficiency of 57% and 43% for both traditional PCB analytical clean-up methods and the automated SPE methods respectively. Therefore, the traditional PCB analytical clean-up methods are more efficient when compared with the automated SPE methods, as the difference in recovery process is clear, with higher value across various PCB component. Additionally, the PCBs value for the traditional method, which tends to be higher in most of the samples can be attributed to natural chromatographic flow of the solvents through the SPE matrix without any use of external force or pressure.

Key words: PCBs, Clean up, Sample extracts, Percentage recovery

1. Introduction
Polychlorinated biphenyls (PCBs) are a category of chemicals that were manufactured in the United States between about 1930 and 1977 (Hopf et al., 2009; Obaid and Ruiz, 2016).Polychlorinated biphenyls (PCBs) are a class of aromatic chemical compounds in which some or all hydrogen atoms attached to the biphenyl ring are substituted by chlorine atoms (*m* + *n* = 1–10). The general chemical formula is C12 H (10-m-n Cl(m+n), where (m + n) is the number of chlorineatoms on the two rings. Depending on the positionand number of the chlorine atoms, there aretheoretically 209 individual PCB compounds(congeners). The carbon positions are numbered 1 to 6 on one ring, and 1′ to 6′ on the other. While positions 2,2′,6, and 6′ are called "*ortho*," positions 3,3′,5 and 5′ are named "*meta*" and position 4 and 4′ are called "*para*."
Because of their general chemical inertness and heat stability, PCBs were predominantly used as coolants and lubricants in electrical equipment such as

capacitors and transformers. Because of their non-flammability, chemical stability, high boiling point, and electrical insulating properties, PCBs were used in hundreds of industrial and commercial applications including electrical, heat transfer, and hydraulic equipment; as plasticizers in paints, plastics, and rubber products; in pigments, dyes, and carbonless copy paper; and many other industrial applications (U.S. EPA, 2012).

Depending on the context of the study or investigation, specific congeners may be monitored.For instance, the Stockholm Convention on Persistent Organic Pollutants (POPS)recommends measurement of six indicator PCBs (PCB-28, PCB-52, PCB-101, PCB-138, PCB-153, and PCB-180) to characterize contamination by PCBs. These congeners were chosen for this study, because they are found at higher concentrations in the environment, in food, or in human fluids/tissues.

The physical properties of PCBs are important in understanding their analytical, physiological, and environmental properties. However, the interactions of the various physical properties can be extremely complex (Erickson, 2001). Chemical and physical properties such as solubility, vapour pressure, and Henry's law constant reported for individual congeners (Shiu and Mackay, 1986; Murphy *et al.*, 1987; Sabljić andGusten, 1989; Dunnivant *et al.*, 1992; Falconer and Bidleman, 1994)

PCBs are freely soluble in nonpolar organic solvents and biological lipids (US EPA, 1980), and the shift from water to lipid solubility is in an increasing *K*ow with increased chlorination. PCBs, which were widely used for industrial purposes until 1973 because of their stable physicochemical properties and excellent electric characteristics of high insulation and polarity. However, PCBs have long-term toxicity, neurological and endocrine disruption, and persistency bioaccumulation nature for human health and environment including those characteristics demonstrated in the Yusho poisoning. (Robertson et.al. 2018)

Although our understanding of the extent and controls on PCB fate and transport is still advancing, it is well known that PCBs are dispersed worldwide. Emissions of PCBs are becoming more prominent in the environment of transition and developing countries, with e-wastes representing a relevant contemporary source factor (Robertson et.al. 2018). Remediation of major sources is inefficient in part because of the limitation in analytical methods for identifying sources and environmental transformation. Advances in measurement methods, including enantioselective analysis of chiral PCBs is accelerating the opportunities for reducing emissions and exposure risks. It is no longer appropriate to measure PCBs using calibration sample extract based on historical commercial mixtures. The use of Aroclors, Kanechlors, or other now banned mixtures in analysis results in serious analytical errors and misguided conclusions (Erickson 2017; Robertson et.al. 2018).

Th is paper is therefore, aimedat assessing the possible variationin percentage recovery of Polychlorinated Biphenyls (PCB) using an automated SPE extractor and a traditional vacuum manifold SPE technique for preliminary polychlorinated biphenyls extraction or clean-up process

2. Methodology

2.1 Sample preparation

Water samples extraction

1 litre water sample is measured in an Erlenmeyer flask, to it is added 10 ml cyclohexane and then 100µl surrogate (PCB-209) and 1 ml of ISTD. This is extracted using liquid /liquid extraction technique with the aid of a magnetic stirrer and the inserted magnetic rod for a minimum period of one hour. The solvent phase (extract) is then separated from the water phase with the aid of a separating

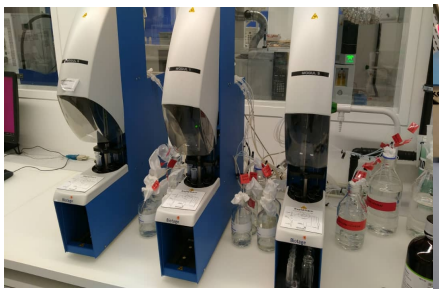
funnel. Extract is then concentrated under nitrogen gas to 1ml and is ready for clean-up.

Solid sample extraction

20 grams of soil or solid sample is weighed in a clean extraction bottle. To this is added approx. 10g sodium sulphate as a drying agent. 40 ml of extracting solvent solvent (Aceton/ Cyclohexane 1:1) mixture is the then added and with the aid of a shaker at 230rpm the combination is extracted for two hours. After two hours, the extract is allowed to settle before an aliquot is taken out. To this aliquot is 100µl surrogate (PCB-209) and 1 ml of ISTD. Extract is then concentrated under nitrogen gas to 1ml and is ready for clean-up.

For the automated SPE extractor and the Traditional Vacuum Manifold SPE Techniques,SPE cartridges were prepared and condition with approx.3 – 5 ml of cyclohexane. 1 mlof the PCB samplesextract is introduced into the cartridge and eluted with approximately 9 ml of cyclohexane effluent in a 10 mlvails and was finally concentrated under a nitrogen gas stream from 10 ml to 1 ml ready for analysis.

Please see below Pictures for the different SPE techniques. (Figure 1. Figure 2 and Figure 3)



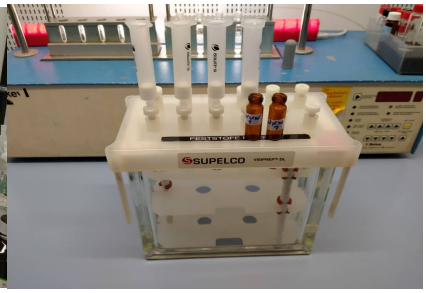


Figure1: An automated SPE ExtractorFigure2: A traditional manifold SPE Techniques

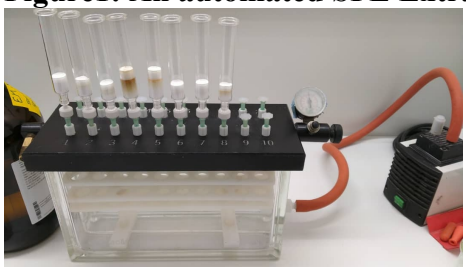


Figure 3: A traditional SPE with a vacuum pump

2.2 Analytical Methods

Gas Chromatography was coupled with an electron capture detector (GC/ECD) which have been previously used to determine PCBs by Burse et al., 1989; ATSDR, 2000, but confirmation by mass spectrometry (MS) was also used for multiple individual congener measurements are required as recommended by Koopman-Esseboom et al., 1994; ATSDR, 2000).

3. Results and Discussion

Table 1: Tabular representation of the PCBs values across ten samples extract

	PCB - 28	PCB - 52	PCB – 101	PCB - 118	PCB - 138	PCB – 153	PCB – 180
9001 Normal (µg/l)	<0.002	0.002	0.007	0.003	0.026	0.023	0.014
9001 SPE-Extractor (µg/l)	<0.002	<0.002	0.0057	0.0025	0.019	0.017	0.011

9002 Normal (µg/l)	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002
9002 SPE-Extractor (µg/l)	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002
9003 Normal	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2
9003 SPE-Extractor (mg/l)	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2
9004 Normal (mg/l)	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2
9004 SPE-Extractor (mg/l)	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2
9005 Normal (mg/l)	<0.2	<0.2	0.5	0.8	1.3	1.1	0.5
9005 SPE-Extractor	<0.2	<0.2	0.43	0.55	0.78	0.64	0.29
9006 Normal (mg/l)	0.53	0.93	0.72	0.5	1.37	1.04	0.52
9006 SPE-Extractor (mg/l)	0.54	1.13	0.81	0.6	1.77	1.33	0.66
9007 Normal (mg/l)	<10	<10	63.1	20.1	151	131	99.1
9007 SPE-Extractor (mg/l)	<2	8.64	50.8	16.8	135	102	74
9008 Normal (mg/l)	0.84	2.02	4.49	5.37	6.12	4.04	1.74
9008 SPE-Extractor (mg/l)	0.73	2.29	4.73	5.68	7.24	4.72	2.01
9009 Normal (mg/l)	0.64	0.58	0.28	0.25	0.21	<0.2	<0.2
9009 SPE-Extractor (mg/l)	0.71	0.71	0.33	0.3	0.29	<0.2	<0.2
9010 Normal (mg/l)	<0.2	0.40	2.50	1.00	5.1	1.3	2.3
9010 SPE-Extractor (mg/l)	<0.2	0.39	1.21	0.98	4.67	3.94	1.95

Note:
Normal = Sample results: Extracted using traditional SPE vacuum technique.
SPE Extractor =Sample results: Extracted using automated SPE technique.

The table above which deduce the various normal and SPE-extractor(PCB sample extract after SPE clean)of the recommendedsix indicator by Stockholm Convention on Persistent Organic Pollutants (POPs) to characterize contamination by PCBs. These measurementindicators of PCBs are (PCB-28, PCB-52, PCB-101, PCB-118, PCB-138, PCB-153, and PCB-180). From the table, when the variousnormal SPE were compared with their automatedSPE-extraction the following were observed: as 9001 and 9002 sample extracts are water samples and the others (9003, 9004, 9005, 9006, 9007, 9008, 9009, and 9010) are soil samples which were measured in µg/l and mg/l respectively. In sample extract 9001,

PCB-28 was observed to have an extraction value of <0.002 which corresponded with the 9001 Normal as they were undetected. Although other PCBs were observed to have contrary values, with a few slight difference. For instance, PCB-52 extraction value < 0.002 which signifies below the detection limit. Thus, less than the 9001 Normal of 0.002, PCB-101 with an extraction value of 0.0057 was 0.0013 less than the normal, andPCB-118 with an extraction value of 0.0025 is lower when compared to the 0.003 for 9001 normal.PCB-138 was observed to follow the same trend with an extraction value of 0.019 which is less than the 9001 Normal of 0.026. Likewise PCB-153 and PCB-180, with PCB-153 having extraction value of 0.017 which is 0.006 less than the 9001 sample extracts value, and PCB-180 with an extraction value of 0.011 which is equally less than 9001 Normal of 0.014. From the result, it could be said that the percentage margin ratio when the extraction value of the various PCBs are compared to the 9001 Normal are as follows:0% (PCB-28), cannot be determined (PCB-52), 18.57% (PCB-101), 16.6% (PCB-118), 26.92% (PCB-138), 26.09% (PCB-153), 21.43% (PCB-180).The highest percentage margin ratio for 9001 was observed at PCB-138 with a value of 26.92%.

From the table, it could be deduced that there is 0% percentage recovery margin ratio in that of 9002 which is a water sample extract, this percentage recovery margin cut across all the PCBs determined in this study as the PCBs components of this sample extract was below the detection limit. As this was similar to results observed in the soil sample extract ofsample extract 9003 and 9004 whose margin ratio is equally 0%. The values which cut across the different PCBs are as follow < 0.002(9002), < 0.2 (9003) and < 0.2 (9004). In 9005, PCB-28 and PCB-52 were observed to have similar Normal and SPE-extraction value of < 0.2, thus have a percentage margin error of 0, which was quite different from the other PCBs. For instance, the extraction value0.43 for PCB-101 was observed to be less than the 9005 sample extract of 0.5, thus having a percentage margin of 14% which is less than the 18.57% of PCB-101 in sample extract 9001.For that of PCB-118, the 9001 Normal was observed to be 0.25 more than the extraction value of 0.55, thus having a percentage margin of 31.25%. When compared with PCB-118 of 9001, it could be said to be approximately 50% more. As the margins were 16.6% and 31.25% for 9001 and 9005 respectively. While for that of PCB-138, the extraction value 0.78 which is less than Normal value of 1.3 for 9005. The margin between both values was spotted to be 0.52, thus having a percentage margin of 40%, which is much higher than the 26.92% margin observed at sample extractof 9001. The PCB-153 extraction value was observed to be 0.64, which is 0.46 less than the 9005 Normal value of 1.1. In other words, the percentage margin is 41.81%, when the margin was compared to that of 9001 sample extract was found to be higher. For PCB-180, the 9005 extraction value was observed to be 0.29, which is 0.21 less than the 9005 Normal value of 0.5. The percentage margin is 42% can be said to be similar to that of PCB-153 which is 41.81%, but when the percentage margin was compared to that of 9001 sample extract was found to be higher with a 100% difference, as the latter has a margin of 21.43%.

From the table above, sample extract 9006 was generally lower than the extraction value across all the seven PCBs in this study. For PCB-28, the extraction value was slightly higher than the sample extract value with 0.01, the latter was 0.53 and the former was 0.54. The percentage recovery factor for PCB-28 is 1.89%. The same trend was observed with PCB-52, as the extraction value was 0.20 more than the 9006 value of 0.93, thus having a total

value of 1.13 and a percentage recovery margin is slated at 21.5%. This is the first indication of percentage margin for PCB-52, as the previous value was0% due to the occurrence of similar value between the traditional and automated SPE extraction values as 9001, 9002, 9003, 9004 and 9005 were below detection limit. As indicated in the table above, the 9006 for PCB-101 was 0.09 lesser than the extraction value of 0.81. thus having a percentage recovery margin of 12.5%, when compared to the previous percentage recovery margin, it could be said to be one of the least value as others such as 9001 and 9005 had more values of 18.57% and 14% respectively. The information for that of PCB-118 for 9006shows that the normal value is 0.1 less than the extraction value of 0.6 and a percentage recovery margin of 20%, which is greater than that of 9001 and less than 9005 as 16.6% and 31.25% were calculated for both 9001 and 9005 respectively. And for that of PCB-138, the extraction value was 0.40 more than the normal value of 1.37, of which the value was indicated at 1.77. The percentage recovery margin was 29.2%,which is greater than the percentage margin of 9001 and less than that of 9005, as both recorded percentage recovery margin of 26.92% and 40% respectively. For PCB-153, the 9006 extraction value was found to be 0.29 greater than the 9006 Normal value of 1.04, therefore having a total value of 1.33Thus, the percentage ratio margin for PCB-153 is 22.11%. Although a similar approach was noticed with that of PCB-180, which has an extraction value with 0.14 greater than the normal value of 0.5, thus, an extraction value of 0.66 and a percentage recovery margin of 26.92%. The percentage recovery margin observed was less than 9005 sample extract but more than that of 9001 sample extract.

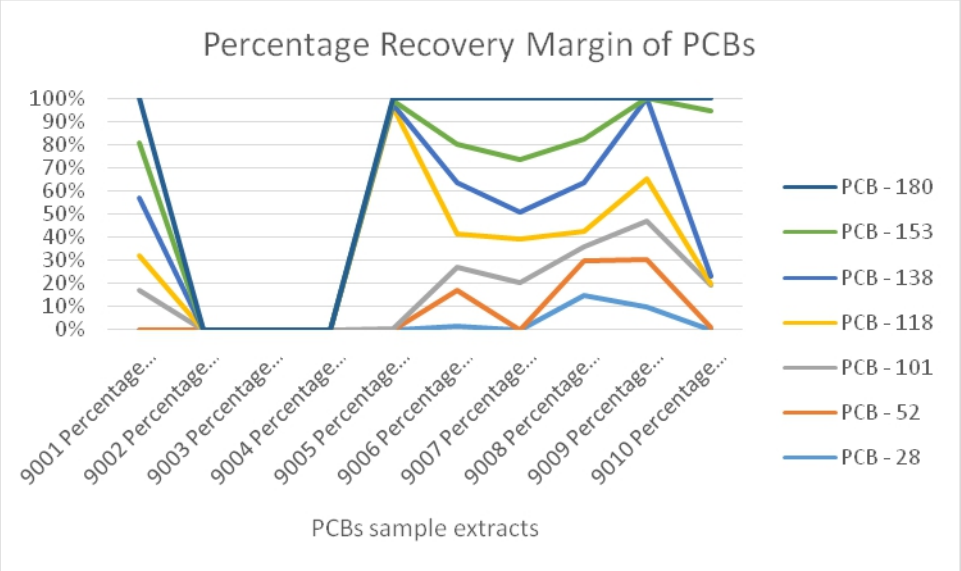


Figure 4: A line presentation of the PCBs percentage recovery margin across ten (10) Sample extracts

The percentage recovery margin for 9007 sample extract could only be determine for five out of the seven PCBs analysed for. On a general note, the normal value for 9007 was observed to be greater than the extraction value across the seven. For instance, PCB-28 have an extraction value of < 2 and a normal value of <10, thus the percentage recovery margin cannot be determined and this is similar to PCB-52

whose percentage recovery margin could not be determined as the extraction value is 8.64 while the normal value is <10. PCB-101, which have a 19.49% recovery margin can be attributed to the 12.3 margin between the normal value and the extraction value, with the normal value of 63.1 which is 12.3 greater than the extraction value of 50.8. And for that of PCB-118, where the extraction value of 16.8 is 3.8 less than the normal value of 20.6, thus have a percentage margin of 18.45% which is one of the least percentage margin observed for 9007 sample extract. PCB-138 was observed to have a percentage recovery margin of 10.6% and this can be attributed to the 16.0 margin between the normal value of 151 and the extraction value of 135. As earlier stated, the normal value had greater impact in the percentage value recorded, as well as the margin recorded for PCB-153,which have an extraction value of 102 that is less than the normal value of 131 with 29.0. Thus establishing a percentage value of 22.14%, which when compared with other PCBs of 9007 sample extract could be said to be the second highest after PCB-180 which have a percentage recovery margin of 25.33%, and a normal value of 99.1 which is 25.1 higher than the extraction value of 74.0.

For 9008, the extractive values for the PCBs were higher than the 9008 normal value except for that of PCB-28 whose 9008 Normal value is 0.11 higher than that of extractive value of 0.73. Thus having a total of 0.84,on the other hand, PCB-52 recorded an extraction value that is 0.27 greater than the normal value of 2.29 and this resulted to a percentage recovery margin of 13.37%. The result observed for PCB-101 was similar to that of PCB-52, as the margin between the extraction value and the normal value is 0.24, with a normal value of 4.49 and extraction value of 4.73, thus having a percentage ratio margin of 5.34%. It could also be deduced that PCB-118 have a percentage margin ratio of 5.77%, as the extraction value can be said to be 0.31 greater than the normal value of 5.37, thus the extraction value is 5.68. In addition, it could said that the percentage margin of 9008 sample extract is one of the least after that of 9010 recorded 2% in PCB-118.For PCB-138, the extraction value was observed to be 1.12 higher than the normal value of 6.12. Thus having a percentage recovery margin of 18.3%, which is quite high when compared to the value observed in sample extract 9010 (9.22%), 9007 (10.6%), 9003 (0%), 9004 (0%) and 9002 (0%), but it was quite low when compared with other sample extract such as 9009 (38.09%), 9006 (29.2%) and 9001 (26.92%). The PCB-153 for 9008 sample extract shows that the extraction value is 4.72 which is 0.68 greater than the normal value of 4.04 and a percentage recovery margin of 16.83%, which is quite low when compared to the other sample extract. A similar approach was observed with that of PCB-180, as the normal value is 0.27less than the extraction value of 2.01, thus led to a percentage recovery margin of 15.52%. For 9009 sample extract, the extraction value was higher than the normal value across the PCBs analysed except for PCb-180, of which the normal value was slightly higher than the extraction value. PCB-28 was observed to have a percentage margin of 10.93%, as there was a 0.07 margin between the extraction value (0.71) and the normal value of 0.64, and for PCB-52, the percentage recovery margin is 22.41%, as the extraction value of 0.71 is 0.13 greater than the normal value of 0.58. PCB-101 which margin of 0.05 was slightly less than that of PCB-28 had an extraction value of 0.33 as against the normal value of 0.28, thus resulted to a percentage recovery margin of 17.86%, which is the third highest value recorded for PCB-101. Likewise PCB-118 have a percentage recovery margin of 20%, which is as a result of the 0.05 margin between the normal value of 0.25 and the extraction

value of 0.30, the percentage margin was similar to that of 9006 which was also 20%. And for PCB-138, the extraction value which is 0.29 is 0.08 greater than the normal value of 0.29, thus resulted to a percentage recovery of 38.09% which is the largest recovery margin for the sample extract and the second largest margin of PCB-138across all the ten sample extract. On the contrary, PCB-153 AND PCB-180 were below detection limit. Thus, there was no percentage recovery margin for both PCBs.

From table 1, it could also be said that for 9010, the three of the PCBs exhibited a greater value in their normal as against their extraction value, and two of the PCBs have greater extraction value as against their normal value. While PCB-28 have similar extraction and normal values. For instance, PCB-28 have a percentage recovery margin of 0% which can be attributed to the zero margin between the normal value and the extraction value of <0.2. PCB-52, which have a percentage recovery margin of 2.5%, which was a result of 0.01 difference between the normal value of 0.4 and the extraction value of 0.39. As this was one of the least determined percentage margin recovery.On the other hand, PCB-101 has a percentage recovery of 51.6%, which was basically as a result of the 1.29 difference between the extraction value of 1.21 and the normal value of 2.50. For PCB-118, the percentage recovery margin is 2%, which is as result of the difference between the extraction values of 0.98 as against the normal value of 1.00. And for that of PCB-138 the extraction value of 4.67 was 0.47 less than the normal value of 5.1, thus resulting in a percentage recovery margin of 9.22% which happens to be the least percentage recovery margin across the ten sample extract for PCB-138. It was observed that PCB-153 has a massive percentage recovery margin of 203.78%, which was depended on the difference between the external value of 3.94 and the normal value of 1.3. In addition, the margin between both values were the highest recorded across the various PCBs and sample extract in this study. On the other hand, PCB-180 recorded a percentage recovery of 15.21%, which is clearly a result of the 0.35 margin between the external value of 1.95 and the normal value of 2.3

A similar observation was also reported by Lahmanov and Varakina (2019) on a short review of sample preparation method for the pesticides residue analysis in fatty acid, where the comparison between the traditional and other automated methods were clearly highlighted. Bjorklund et. al. 2002 also observed 60% recoveries from milk samples having compared several extraction process. Although the recoveries observed from this research were between 2% to slightly above 200% for the pre-treatment and clean-up process of polychlorinated biphenyls (PCBs) in soil and water samples. There was also no significant differences in recoveries as observed for the three different samples (9002, 9003, 9004) as they were below detection limit while some other PCB components across the ten samples and seven indicators had recovery margins and the average recoveries for the investigated polychlorinated biphenyls (PCBs) also varies across the seven indicators and ten samples.

Conclusion

From the general interpretation of the data, it reflects the instability of the percentage recovery between the PCB sample extract using thetraditional and the automated SPEclean up techniques. The results obtained varies across the different PCB compound and sample extracts. The results showed that the efficiency of both traditional PCB analytical clean-up methods and the automated SPE methods are in ratio 4:3 respectively. Thus resulting to a percentage efficiency of 57% and 43%of traditional PCB analytical clean-up

methods and the automated SPE methodsrespectively. Although the margin in efficiency is relatively equivalent, with traditional PCB analytical method been more efficient in 4 out of the 7 sample extract above the detection limit as against the automated SPE method which is 3.Thus, it can be concluded that traditional PCB analytical clean-up methods is more efficiently reliable when compared with the automated SPE methods, as the difference in recovery process is clear, with higher value across various PCB component in this study. Additionally, it should be noted that the traditional method values which tends to be higher in most samples,frequently requires the use of more organic solvent during sample preparation and extraction process when compared to the automated SPE methods. While the automated SPE technique is precise, saves time and solvent. This is why it is a preferred technique for the analytical industry as the traditional method can be laborious and time consuming with a higher tendency of human errors.This study recommends appropriate methodology in the analysis of PCBs using ISO certified calibration standards.

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