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Detection of ethyl chloride abuse in urine sample

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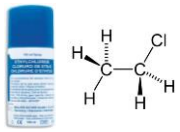
GRAPHICAL ABSTRACT



(a) Ethyl chloride spray and (b) structure of ethyl chloride

ABSTRACT

Glue sniffing become popular with young adolescents and may have slipped down the list of worried for parents because it is a bigger killer for teenagers than hard drugs. Now, ethyl chloride spray was use as a sniffing alternative since it has the same characteristic with solvents and also categorized as a volatile organic compounds. Ethyl chloride normally used to prevent pain caused by minor surgical procedures and in sports used for temporary relief of minor injuries. Their high lipophilicity provides easy access to the central nervous system, where they produce global neuronal dysfunction through diverse and poorly understood means. This research was done to detect ethyl chloride presence in urine due to the wide abuse of this compound among teenager ultimately leads to addiction. In this study, the detection of ethyl chloride was analyzed using UV – Visible Spectroscopy. Thymol blue is used as an indicator to estimate the amount of ethyl chloride present. The result demonstrated that ethyl chloride can be detected in both distilled water and urine sample at 212nm with 0.9592 and 0.9966 linear regression respectively. Standard deviation for both distilled water and urine sample were 7.0090×10^{-4} and 8.8690×10^{-5} respectively. Limit of Detection for distilled water sample was 2.1027×10^{-3} g and for urine sample was 2.6607×10^{-4} g respectively. Finally the percentage of error for distilled water and urine sample were 0.19 % and 9.88 % respectively.

Keywords: Glue sniffing, ethyl chloride, volatile organic compounds, UV – Visible Spectroscopy, thymol blue, urine sample.

1. INTRODUCTION

Huffing or inhaling glues, gases, aerosols and other substances that categorized as volatile organic compounds is not a new phenomenon. These substances will make someone to get high, extra energetic and super strong. Besides, will make loss of muscular movement, coordination, slurring of speech, hallucinations and can cause sudden sniffing death even it is first time inhaling these volatile solvent [1].

Nowadays, a substantial number of children and adolescents world-wide abuse volatile substances which is ethyl chloride, which comes in the form of a muscle spray. The stout spray cans are inexpensive, legal, was readily available, easily concealed and may be bought at pharmacies. The main use of ethyl chloride spray is as a spray to reduce pain for athletes [2]. Ethyl chloride is a vapocoolant local anesthetic that evaporizes rapidly and when applied as fine spray it wills produces freezing of superficial tissues and resulting in insensitivity of peripheral nerve endings and local anesthesia.

Human exposure to volatile substances can occur through different routes, including ingestion, inhalation and dermal contact. The toxicological impacts of these compounds have been carefully assessed, and the results demonstrate the potential associations of cancer incidence with exposure to volatile substances [3].

This research was done to detect ethyl chloride presence in urine sample as urine is noninvasive nature and availability in large volumes, urine as a matrix for volatile organic compounds analysis also has an advantage over other biofluids in that analytes are concentrated by the kidney before being excreted from the body [4].

Thymol blue indicator was used to detect ethyl chloride in urine sample as it compound has a longer chain of conjugated double bonds that able to absorb light at a longer wavelength by using UV – Visible Spectroscopy.

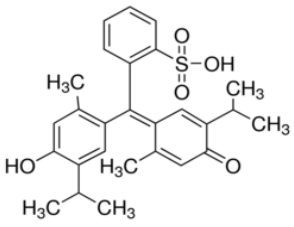


Figure 1: Chemical Structure of Thymol Blue [5]

2. EXPERIMENTAL

Standard solution was prepared by adding 20mL of methanol to 400mL distilled water in 500mL beaker. The solution is stir by using glass rod until homogeneous. Urine sample solution was prepared by adding urine in standard solution with ratio 1:20 of urine and distilled water. In this method, the calorimetric can be divided into two categories in which, category one, calorimetric of different mass of ethyl chloride with urine sample and thymol blue indicator. Category two was colorimetric of different mass of ethyl chloride with distilled water and thymol blue indicator. Detection of ethyl chloride in distilled water was used as a reference to compare the result with the detection of ethyl chloride in urine sample. By these categories, the results that are get more specific and accurate.

Ethyl chloride spray (88g/100mL) is difficult to be measure by volume, so, ethyl chloride was measured based on puff as references with other medication spray. One puff is equal to 0.2 drop. So, one drop equal to 0.05mL and one puff equivalent to 0.01mL. Therefore, one puff will equal to 0.0088g. The mass of ethyl chloride used for both categories were 0.0264g, 0.0528g, 0.0792g, 0.1056g, 0.1320g and 0.1584g. The sample was run using UV – Visible Spectroscopy in range of 190nm to 350nm.

Below were sample preparation, procedure of category 1, and procedure of category 2.

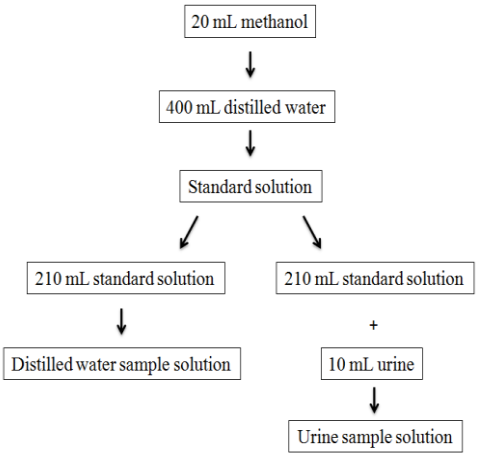


Figure 2: Sample preparation

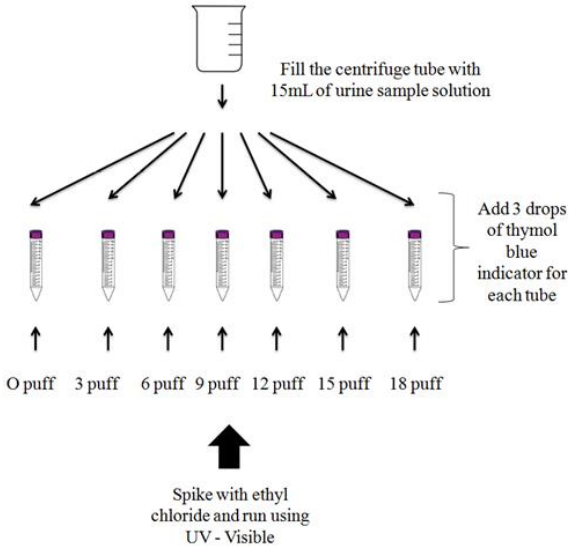


Figure 3: Procedure of category 1

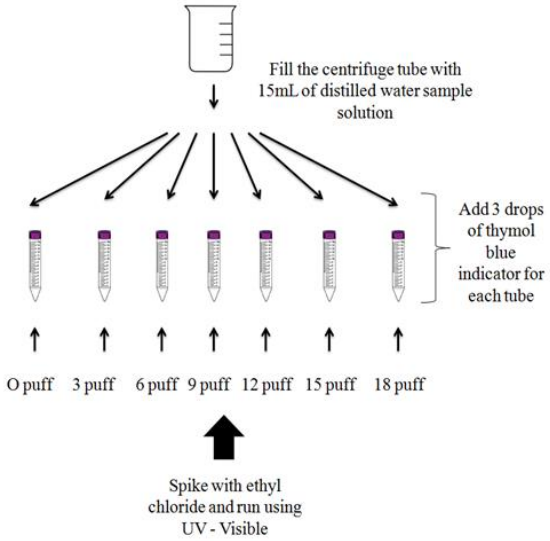


Figure 4: Procedure of category 1

3. RESULTS AND DISCUSSION

3.1. Quantitative analysis of ethyl chloride

Ethyl chloride is one of volatile organic compound. Usually volatile organic compound are excreted by urine as hydrocarbon. Hydrocarbon does not dissolve in water because they are non-polar compounds and water is a polar solvent. The use of general solubility rule "like dissolves like" which is polar solvents dissolve polar solutes and non-polar solvents dissolve non-polar solutes. Thus, ethyl chloride does not form stable solution in water and urine. A methanol solution was added to trap the ethyl chloride in the urine and distilled water. So, in this experiment, a mixture of distilled water and methanol been used instead of pure distilled water [2].

3.2. Calibration curve of ethyl chloride in distilled water using UV - Vis

The distilled water sample was prepared using standard solution and it's were spike with different mass of ethyl chloride. The graph demonstrated method of correlation value of ethyl chloride mass and absorbance from the UV spectrum method at wavelength 212nm.

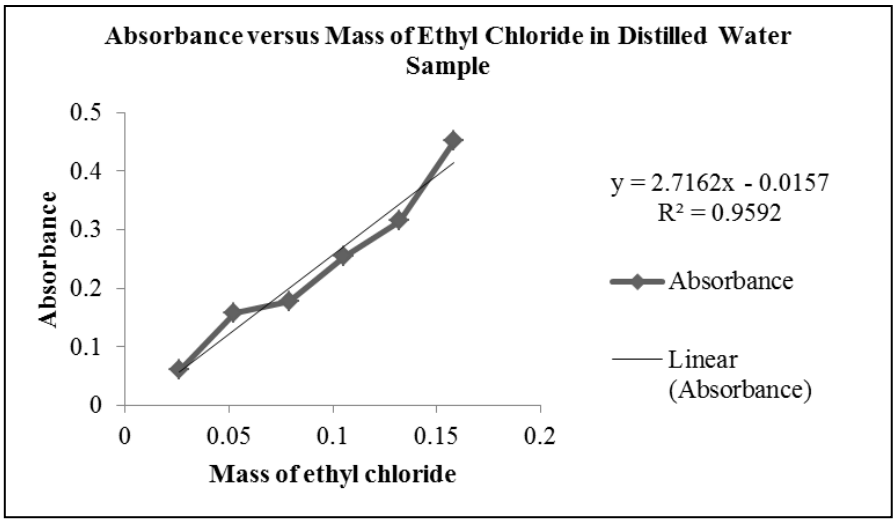


Figure 5: Calibration curve of ethyl chloride in distilled water sample solution

The regression equation of the calibration curve for ethyl chloride in distilled water was y=2.7162x – 0.0157 with correlation of 0.9592. Calibration curve for ethyl chloride was linear up to 0.1584g. The results of ethyl chloride in this experiment show the percentage of error in experiments are in small value. For precision, the experiment was repeated ten times to get more accurate result.

Standard deviation, Limit of Detection and the percentage of error for detection of ethyl chloride in distilled water were 7.0090×10^{-4} g, 2.1027×10^{-3} g and 0.19 % respectively.

3.3. Calibration curve of ethyl chloride in urine using UV - Vis

The UV – Vis spectra of the mixtures were recorded at 190nm to 350nm using Shimadzu UV – 1800. The calibration curve of ethyl chloride in urine sample was shown in figure 6.

Determination of ethyl chloride was prepared using urine standard solution which spike with different mass. The regression equation of the calibration curve for ethyl chloride in urine was y = 1.7019x + 0.0476 with correlation of 0.9966. Calibration curve was linear up to 0.1584g for ethyl chloride. The result of ethyl chloride in urine sample show the percentage of error in expected value and for precision, the experiment was repeated ten times to produce more accurate result.

Standard deviation, Limit of Detection and the percentage of error for detection of ethyl chloride in urine were 8.8690×10^{-5} g, 2.6607×10^{-4} g and 9.88 % respectively.

Table 1: Analysis of ethyl chloride in distilled water sample

Experiments Number	Absorbance (y)	Mass of Ethyl Chloride (x) (gram)	$\lvert x - x\ bar \rvert$	$(x - x\ bar)^2$
1	0.0625	0.0288	3.7×10^{-4}	1.369×10^{-7}
2	0.0625	0.0288	3.7×10^{-4}	1.369×10^{-7}
3	0.0604	0.0280	4.3×10^{-4}	1.849×10^{-7}
4	0.0612	0.0283	1.3×10^{-4}	1.69×10^{-8}
5	0.0612	0.0283	1.3×10^{-4}	1.69×10^{-8}
6	0.0646	0.0296	1.17×10^{-3}	1.369×10^{-6}
7	0.0592	0.0276	8.3×10^{-4}	6.89×10^{-7}
8	0.0625	0.0288	3.7×10^{-4}	1.369×10^{-7}
9	0.0582	0.0272	1.23×10^{-3}	1.513×10^{-6}
10	0.0628	0.0289	4.7×10^{-4}	2.209×10^{-7}
		x bar = 0.02843	$\Sigma = 5.5\times10^{-3}$	$\Sigma = 4.4213\times10^{-6}$

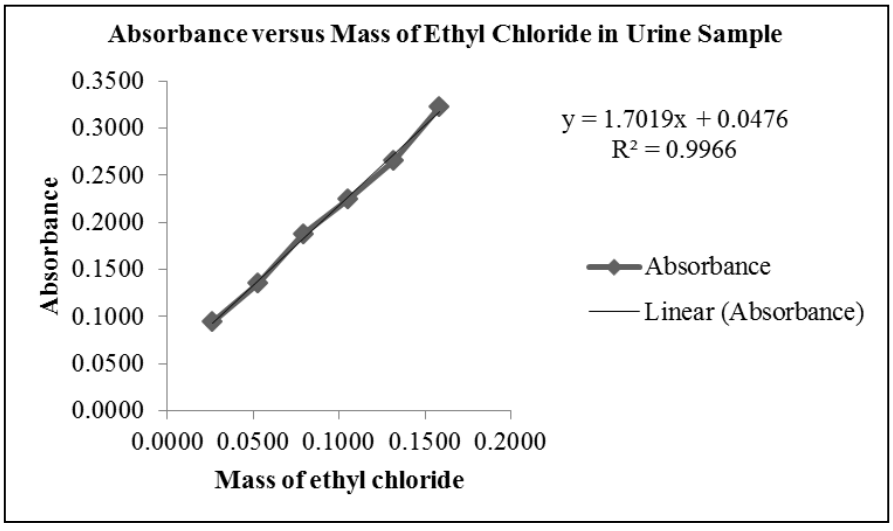


Figure 6: Calibration curve of ethyl chloride in urine sample solution

Table 2: Analysis of ethyl chloride in Urine sample

Experiments Number	Absorbance (y)	Mass of Ethyl Chloride (x) (gram)	$\lvert x - \bar{x} \rvert$	$(x - \bar{x})^2$
1	0.0463	7.639×10^{-4}	4.2018×10^{-3}	1.7655×10^{-5}
2	0.0498	1.293×10^{-3}	3.6727×10^{-3}	1.3489×10^{-5}
3	0.0496	1.175×10^{-3}	3.7907×10^{-3}	1.4369×10^{-5}
4	0.0474	1.175×10^{-4}	4.8482×10^{-3}	2.3505×10^{-5}
5	0.0486	5.876×10^{-4}	4.3781×10^{-3}	1.9168×10^{-5}
6	0.0498	1.293×10^{-3}	3.6727×10^{-3}	1.3489×10^{-5}
7	0.0696	0.01293	7.9643×10^{-3}	6.3430×10^{-5}
8	0.0640	9.636×10^{-3}	4.6703×10^{-3}	2.1812×10^{-5}
9	0.0628	8.931×10^{-3}	3.9653×10^{-3}	1.5724×10^{-5}
10	0.0696	0.01293	7.9643×10^{-3}	6.3430×10^{-5}
		$\bar{x} = 4.9657\times10^{-3}$	$\Sigma = 0.0491$	$\Sigma = 2.66071\times10^{-4}$

3.4. Factor interfere result

The correlation value from the result obtained not very accurate to 1.0000 due to the presence of some error. As assumption the mass parameter for ethyl chloride might be the reason for the error presence. The assumptions have done 1 puff equal to 0.0088g, but there are other factors that will affect result. The factor such as presence of ethyl chloride that does not really show it characteristic after spray has been done.

Loss of the volatile substances by evaporation is one of the errors in quantitative analytical procedures. Therefore analysis of the sample should start as soon as possible. Volatile substances diffuse from the sample into the atmosphere until equilibrium is reached. Every time the container is opened, losses occur due to the replacement of air. Every time fresh air comes inside the container, diffusion of the compounds will create a new equilibrium [6].

3.5. Discussion on sample preparation

Urine sample solution must be dilute 22 times dilution factor because too concentrated. The concentrated urine sample solution cannot be detected by UV spectroscopy as urine contains other metabolite. The peak on UV graph will overlap each other and produce inaccurate results. Thus, only 10 mL of urine is used with 1:20 ratio with distilled water solution.

First sample was spike with 0.0264g of ethyl chloride that equal to 3 puffs and follow by 6, 9, 12, 15, 18 puffs because spiking ethyl chloride with lower mass will produce imprecise value for absorbance. The peak may overlap if the gap value of mass of the ethyl chloride used to spike on each sample is too small.

4. CONCLUSION

In the detection of Ethyl Chloride Abuse in Urine Sample, the entire stated objectives have been accomplished. The distilled water sample solution was prepared and calibrates to perform calibration curve for the absorbance with different mass of ethyl chloride. This study also successfully detects the presence of the different mass ethyl chloride in urine sample solution at 212nm by using UV – Visible spectroscopy. The results show that ethyl chloride can be detected in urine sample by using proposed method in this research. Absorbance of ethyl chloride in both distilled water and urine sample increased as mass of ethyl chloride use increase.

Finally, the error of analysis in both sample were calculated. The lower percentages of error show that the research was done successfully using proposed method although some error need improvements in future study.

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