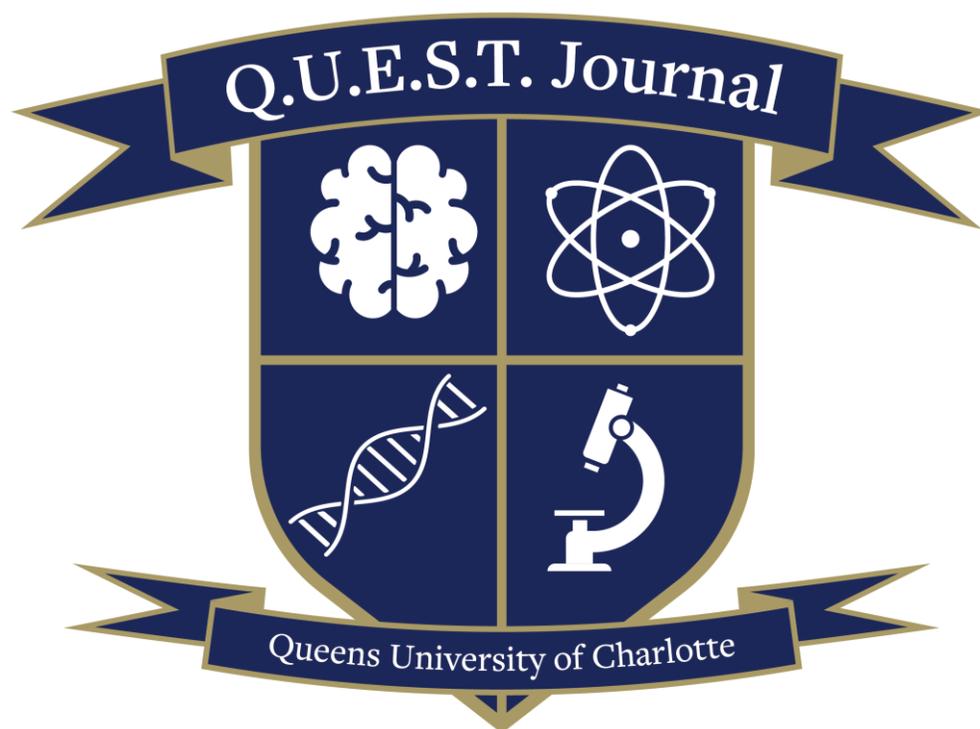


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Editor in Chief notes

As the Editor – in – Chief of the QUEST Journal for my second and final year, it is an amazing feeling to know that support for this journal has not only continued but expanded. While support has grown mostly at our home institution, Queens University of Charlotte, seeds of support are beginning to take root at other colleges and universities. These seeds were planted from the hard work of my staff, whom embody the culture we strive to sustain. Our culture is of the Red Queen variety as we have made improvements in different areas of publication. While our journal may not yet have the weight of more established journals, we are still making waves in the fabric of undergraduate research. To the current and future undergraduate students, I challenge you to be proactive in your research endeavors. Go to your professors and tell them you want to do research. Forge your trail to success till it is as defined as connection between neurons.

As always, a big thank you to our journal advisor in helping us throughout the year.

May your QUEST bring you success,

Marcus Ramos-Pearson

Editor – in – Chief of the QUEST Journal, 2018

Determination of the Presence of Benzene in Incense, Mosquito Coils, and Candles

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ABSTRACT: The popularity of scented candles is well known as candle sales surpass \$2 billion annually. Recent studies have reported that the combustion of candle wax, specifically those made from paraffin, may emit harmful chemicals such as benzene and toluene. Other potentially harmful components such as acrolein are used as solidifying agents, plasticizers, fragrances, solvents, and dyes. This study investigated the presence of benzene in candle wax as well as the vapor from candles, incense, and mosquito coils. Samples were tested in a glass chamber using the OSHA ORG-12 method and gas chromatography mass spectroscopy. Benzene was not found in the candle wax of paraffin and non-paraffin candles. Vapor collected from the combustion of candles made with beeswax, soy wax, and paraffin wax did not contain concerning levels of benzene. Vapor samples collected from incense and mosquito coils contained benzene with concentrations ranging from 0.1 ppm to 0.5 ppm within the 50-minute collection period. While these concentrations are below the recommended exposure limits, benzene is a suspected human carcinogen and exposure should be reduced to the lowest feasible amount.

Introduction

Benzene

Benzene is a colorless, volatile liquid that is made from petroleum and used in the manufacturing of many commercial goods (Toxicological, 2007). Every day the general population is exposed to benzene through inhaling air, the primary route of exposure (Toxicological, 2007). Benzene can enter the body through the lungs, gastrointestinal tract, and skin (Toxicological, 2007). Low concentrations of benzene have been found to cause health problems (Rinsky, 1989). The Ames test has suggested that benzene is a mutagenic compound (Orecchio, 2011). Benzene has been connected to respiratory irritation (Ho et al., 2005) and one report indicated a correlation between benzene and leukemia (Rinsky, 1989). Organizations differ on their exposure recommendations for benzene. The Occupational Safety and Health Administration's (OSHA) permissible exposure limit (PEL) is 1 ppm per eight-hour period, the National Institute for Occupa-

tional Safety and Health's (NIOSH) recommended exposure limit (REL) is 0.1 ppm per eight-hour period, and the American Conference of Government Industrial Hygienist's (ACGIH) threshold limit value (TLV) is 10 ppm per eight-hour period (OSHA, 1988). Short-term exposure limits are recommended by OSHA as 5 ppm within a 15-minute time frame and by NIOSH as 1 ppm within a 15-minute time frame (OSHA, 1988). NIOSH and ACGIH both recognize carcinogenic properties of benzene (OSHA, 1988) and the International Agency for Research on Cancer (IARC) considers benzene a class 1 human carcinogen (IARC et al., 1987). This study examined the vapors of three common household items that potentially contained benzene - candles, incense, and mosquito coils.

Candles

The National Candle Association (NCA) defines wax as a primarily hydrocarbon, water resistant, smooth, low toxic compound that is solid at room temperature and liquid at higher

temperatures (NCA et al., 2013). This study examined beeswax, soy wax, and paraffin wax candles. The concept of candles releasing harmful chemicals in the vapor comes from the way that the wax is manufactured. Waxes contain many organic volatile molecules, which makes it easy for these chemicals to be released during combustion. Beeswax is secreted by bees and is obtained by melting honeycombs (NCA et al., 2013). Soy wax is manufactured by hydrogenating soybean oil into a solid that contains esters, acids, alcohols, and hydrocarbons (NCA et al., 2013). The main candle wax used in the United States is paraffin wax (NCA et al., 2013). Paraffin wax is a synthetic wax composed of saturated, straight-chained hydrocarbons and made by chilling petroleum (NCA et al., 2013). Both paraffin wax and benzene are derived from petroleum resulting in the concern that benzene may be present in paraffin wax. Studies have shown conflicting results for the presence of benzene in candles. One study suggested that burning candles increased the concentration of polycyclic aromatic hydrocarbons (Orecchio, 2011). However, another study tested the worst-case scenario by burning 30 candles for three hours and determined that the emissions presented no health concern (Lau et al., 1997). It has been suggested that most of the emission compounds were from unburned candle wax (Fine et al., 1999). This experiment tested wax as well as candle vapor to determine the presence of benzene. If benzene was present in candle wax, this would support the presence of benzene in candle vapor because vapor is composed of mostly unburned wax. If wax samples did not contain benzene but benzene was present in candle vapors, it would suggest that benzene was generated from incomplete combustion of larger compounds with aromatic rings. Therefore, determining the presence of benzene in wax suggested how benzene was produced during combustion.

Incense and Mosquito Coils

Incense vapor has been found to contain volatile organic compounds such as benzene, toluene, and xylene (Lin et al., 2008). Incense sticks are composed of a bamboo stick with wood and QUEST

herbal powder, adhesive powder, and fragrance material (Lin et al., 2008). A mosquito coil is an incense that traditionally contains pyrethrum powder paste and botanical extracts (Liu et al., 2003). Incense are lit for a short period of time to get a smoldering burn that produces smoke. Incense emissions have been linked to respiratory system dysfunction and elevated levels of Immunoglobulin E, which may cause an allergic reaction (Lin et al., 2008). Adverse health effects such as coughing and irritation of the nose and throat were found in a study performed on temple workers who were continuously exposed to high concentrations of incense smoke (Ho et al., 2005). One study found that emissions from incense contained benzene (Eggert et al., 2004). Benzene, toluene, and methylene chloride were found in the emissions from mosquito coils in high concentrations (Lee et al., 2006). It is known that many herbal plants contain aromatic rings so it was suggested that incomplete combustion of these compounds could be responsible for benzene production in incense and mosquito coils. This is in agreement with a study that stated that incense emissions are due to incomplete combustion (Eggert et al., 2004).

Summary

This study determined whether benzene was present in candle wax and vapors from candles, incense, and mosquito coils. The purpose of this experiment was to identify common household items that may release benzene and contribute to unhealthy living conditions. Based off of previous research, it was hypothesized that mosquito coils and incense vapors would contain benzene and that concentrations would exceed the recommended exposure limits. It was hypothesized that benzene concentration would exceed the limits suggested for the paraffin wax candles, and the benzene concentration would remain within a safe limit for the beeswax and soy wax candles. This hypothesis was based on the manufacturing procedures for each type of wax because paraffin wax is derived from petroleum whereas beeswax and soy wax are not.

Materials and Methods

Occupational Safety and Health Administration (OSHA) is an organization that is responsible for promoting safe working conditions. OSHA recommends two standard methods for sampling benzene concentration in the workplace. The OSHA Method ORG-12 used charcoal sorbent tubes and an air pump to collect vapors (Benzene et al., 1979). The OSHA Method ORG-1005 used passive samplers, which eliminated the air pump and was easier to use in the workplace (Benzene et al., 2002). This study used the OSHA Method ORG -12 to analyze vapors because the air pump guaranteed the vapors were being collected and the glass sorbent tubes were better suited for collection with an open flame (Benzene et al., 1979). The OSHA ORG – 12 method used a gas chromatography flame ionization detector, but a Thermo gas chromatography mass spectrometer (GC-MS) was used in this experiment (Benzene et al., 1979). Mass spectrometry has similar performance outcomes compared to flame ionization detection. Calibrated standards were used in order to ensure that the mass spectrometer results would be comparable to the flame ionization detection results.

A calibration curve, using reagent grade benzene, was made using a GC-MS. The calibration curve used dichloromethane instead of carbon disulfide, the desorbing agent, in order to reduce cost. All calculations were based off of the OSHA ORG -12 Method given standard stating that 1 ppm benzene in air is equivalent to 36.5 nanoliters of benzene per milliliter of solvent (Benzene et al., 1979). Standards were made in concentrations ranging from 0.05 ppm to 100 ppm.

This experiment used a glass chamber to collect vapor in order to simulate burning in a small-unvented area and to protect samples from disturbances while burning. Samples were burned in a 0.0437 cubic meter glass chamber that was lifted 2.7 cm off of the lab bench to allow for an air inlet (Figure 1). The chamber was 51.5 centimeters wide by 26.5 centimeters deep with a height of 32.0 centimeters. Benzene is less

dense than air so benzene was collected at the top of the chamber where there was no ventilation. Vapor samples were collected on SKC Inc. coconut charcoal sorbent tubes using a LaMotte air sampling pump (Benzene et al., 1979). The pump was positioned outside of the chamber and was connected to a sorbent tube that was held 23.2 centimeters from the bottom of the chamber and positioned directly above the flame. Ten-liter samples were collected at a flow rate of 0.2 liters per minute (L/min) (Benzene et al., 1979).



Figure 1 - Burning chamber set up with the sorbent tube positioned directly above the flame and the pump positioned outside of the chamber

Five candles, one mosquito coil, and two types of incense were used in this study (Table 1). Candle 1 was a white paraffin wax that had sticky consistency similar to glue. Candle 2 was a pink paraffin wax that was very dry and flakey. Candle 3 was a white soy wax with a sticky consistency. Candle 4 was dark yellow beeswax with a dry consistency. Candle 5 was a white paraffin wax with mosquito repellent, and it had a sticky consistency. Two scents of incense were used in this study (Table 1). Incense 1 was a dragon blood scent and had a red color. Incense 2 was a

Table 1 - Candles, incense, and mosquito coil utilized in this study. Concentrations of benzene produced during a 50 minute burn at a rate of 0.2 L/min.

Samples	Brand	[Benzene] (ppm)	Benzene (mg)
Candle 1	Glade clean linen candle	BD*	BD
Candle 2	Yankee pink sands candle	ND**	ND
Candle 3	Meyers Day lavender soy candle	ND	ND
Candle 4	Bluecorn Naturals Beeswax candle	ND	ND
Candle 5	Cutter Citro Guard candle	BD	BD
Incense 1	Flora Classique INC. – Dragon Blood	0.529	5.29
Incense 2	Flora Classique INC. – Spiritual	0.438	4.38
Mosquito Coil	Pic mosquito repelling coil	0.132	1.32

* BD = below reporting limit (0.050 mg/L), **ND = none detected

spiritual scent and had a green color. The mosquito coil had a green color.

The ends of the tube were broken prior to sampling and the tube was connected to the air pump hose with the wool closest to the open end. The sample was lit outside of the glass chamber and then placed inside. All candle samples were burned in their circular glass containers. Incense were burned in canoe incense burners and two incense were burned at a time. The mosquito coil was burned in a Terra Cotta burner without the cover. The pump was started once the sorbent tube and lit sample were placed in the chamber. After the 50-minute collection period, the pump was shut off and the lit sample was removed and extinguished outside of the chamber. The absorbent tube was removed and covered with caps to prevent contamination.

Samples were analyzed within 24 hours of being collected (Benzene et al., 1979). The absorbing tubes were broken above the wool and the charcoal was transferred into test tubes. Only the main absorbing section was used and the break through charcoal was discarded. The samples were desorbed for 30 minutes in 1.5 milliliters of Sigma- Aldrich low benzene carbon disulfide with intermittent shaking. Samples were analyzed on a Thermo Finnigan Trace GC and ITQ-QUEST

700 MS with a TR-5MS 30 meter column and an inner diameter of 0.25 millimeters. The samples were analyzed using selective ion monitoring (SIM) between 1.50 and 3.00 minutes and a 2 microliter injection with a 10:1 split ratio. The target ion was 78 and the qualifying ion was 52 (Jacq et al., 2008). Benzene had a retention time of 1.87 minutes and was confirmed in the electron ionization mass spectra

at m/z 78.92. A sorbent tube without air drawn through it was exposed with the same breaking and sealing method mentioned above and was used as a control for the study. The chromatograph was used to determine the presence of benzene in the sample. All samples were stored in the refrigerator with parafilm to prevent evaporation, as carbon disulfide is very volatile.

The extraction efficiency and recovery percentage was tested by placing the absorbing charcoal into a test tube with 5.5 microliters of benzene. The benzene was allowed to absorb onto the charcoal for 30 minutes and was desorbed with 1.5 milliliters of carbon disulfide. The sample was analyzed using GC-MS.

Candle wax was analyzed for the presence of benzene. Hydrocarbons were used to dissolve nonpolar wax. Wax samples ranging from 0.049 and 0.166 grams were taken from each candle and were dissolved in 3 milliliters of hexane. Dissolved wax samples were analyzed using GC-MS. Many of the wax samples did not dissolve completely. Samples were allowed to settle and the liquid was transferred into new vials until the sample was clear enough to be analyzed.

Results

The calibration curve was accepted when R^2 was 0.99 or greater (Figure 2). Since 0.05 ppm of benzene was the lowest standard it was defined as the reporting limit for the study.

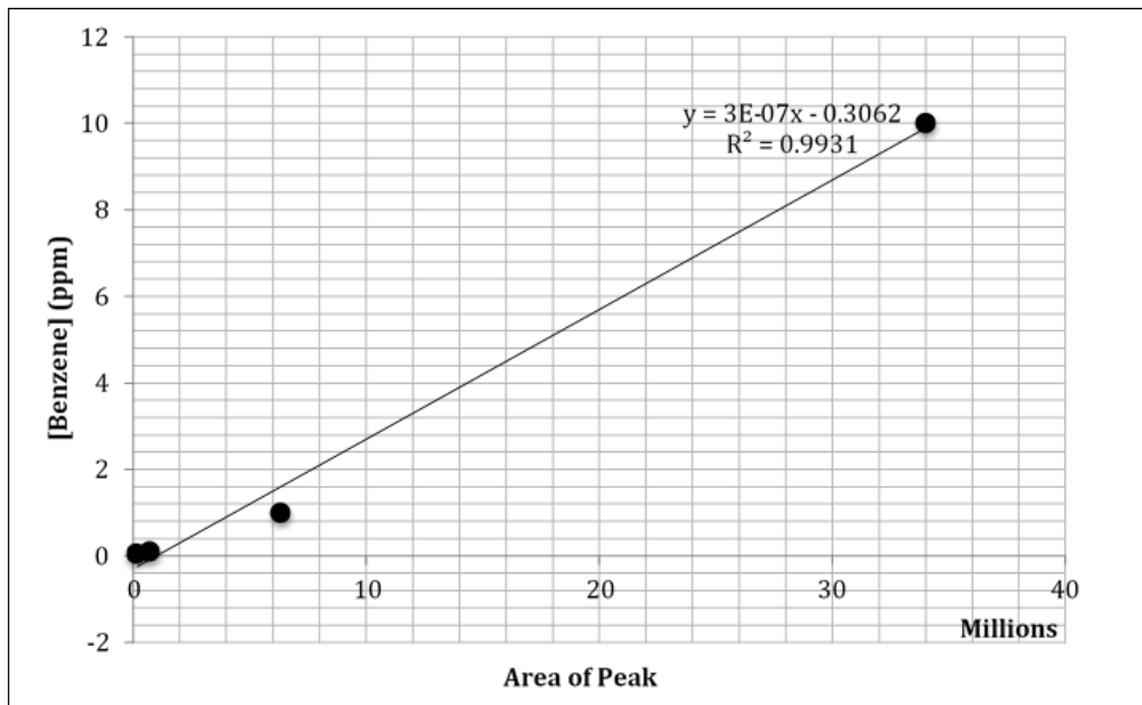


Figure 2 - Standard curve with concentrations of benzene ranging from 0.05 to 10 ppm. Solutions were made with reagent grade benzene and dichloromethane.

The spiked sample theoretically had a concentration of 100 ppm benzene. This sample was compared to the 100 ppm standard to determine that 90.9% extraction efficiency was achieved using the OSHA ORG – 12 method. For quantifiable samples, the extraction efficiency was accounted for in the calculations in order to state the full concentration of benzene emitted.

Wax from each of the five candles did not contain a detectable amount of benzene. There was no detectable benzene present in the vapor samples from candles 2, 3, and 4. Vapor samples from candles 1 and 5 contained benzene concentrations that were below the reporting limit of 0.050 mg/L (Figures 3, 4). The incense vapor contained 0.438 ppm benzene for the spiritual scent and 0.529 ppm benzene for the dragon

blood scent. The mosquito coil vapor contained 0.132 ppm benzene.

Discussion

This study analyzed candles, incense, and a mosquito coil to determine if benzene was emitted during the burning process. The OSHA ORG – 12 method was used to collect vapor samples and the samples were analyzed with GC-MS. All candle vapor samples in this study were below the reporting limit of 0.05 ppm. This project determined the presence of benzene in vapor samples from incense and mosquito coils while they were burned in a small glass chamber.

Candle wax did not contain benzene. This suggests that if benzene was present in vapors it would be due to incomplete combustion. Candles 2, 3, and 4 did not emit a detectable amount of benzene. This supported previous findings that stated that beeswax candle vapors do not contain benzene (Lee et al., 2006). Candles 1 and 5 emitted benzene concentrations below the reporting limit. The reporting limit was 0.05 ppm, which is half of the lowest threshold value of 0.1 ppm set by NIOSH. Although these candles released a detectable amount of benzene, the concentrations were not concerning to human health and were below all threshold standards. It was hypothesized that paraffin candles would emit concentrations of benzene over the 0.1 ppm threshold. However, it was found that one of the paraffin

wax candles (candle 2) released no detectable amount of benzene and the other paraffin wax candles (candle 1 and 5) released negligible amounts of benzene. This was consistent with previous studies that found that candles did not release benzene (Lee et al., 2006).

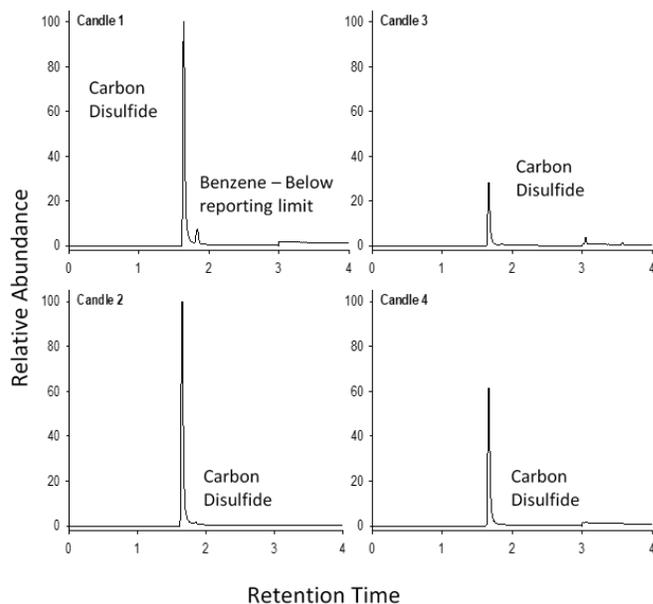


Figure 3 - GC-MS Total Ion Chromatogram (TIC) of fumes collected from 4 candles. Fumes from candle #1 contained benzene below the reporting limit while no benzene was detected from candles #2, #3 and #4. Carbon disulfide was used as the desorbing agent in the sample preparation process.

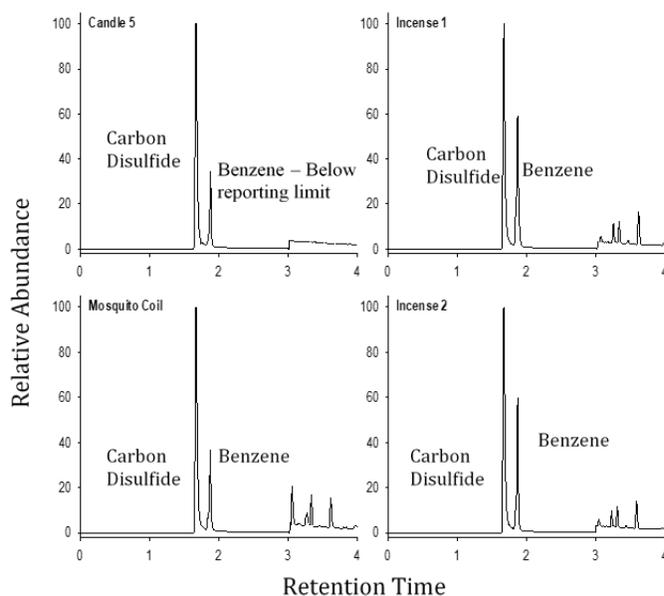


Figure 4 - GC-MS chromatogram Total Ion Chromatogram (TIC) of fumes collected from a mosquito candle, coil, and incense. Fumes from all 4 products contained benzene. Benzene from the mosquito candle was below the reporting limit. Carbon disulfide was used as the desorbing agent in the sample preparation process.

The incense released 0.529 ppm and 0.438 ppm of benzene, and the mosquito coil released 0.132 ppm of benzene. This was consistent with the results of previous studies that found levels ranging from 0.004 to 0.117 ppm benzene (Orecchio, 2011). Samples were analyzed within a few hours of being desorbed in order to prevent evaporation. Carbon disulfide is very volatile so it is possible that some of the desorbing agent could have evaporated which would concentrate the benzene in the sample. Additionally, two incense were burned in order to meet the 50-minute collection period and this could have raised the benzene concentrations measured in this study. While these benzene concentrations were above the 0.1 ppm threshold set by NIOSH, it should be noted that the threshold measures continuous exposure for an 8-hour time frame. Moreover, the short-term exposure limit set by NIOSH is 1 ppm per 15-minutes (OSHA, 1988). The concentrations of benzene in this study were well below the 1 ppm short-term exposure limit. Additionally, it should be noted that these samples were collected in a 0.0437 cubic meter chamber with a 50-minute burn time. These conditions do not reflect the typical burning environment. These results were in agreement with the hypothesis since incense and mosquito coil vapors contained benzene; however, the benzene concentration was not above the threshold limits. Incense and mosquito coil samples were not analyzed before burning to determine if benzene was present in the material itself or was a byproduct of combustion. However, it is known that the powders used to make in-

cense and mosquito coils contain aromatic rings. It is suggested that benzene is a combustion by-product considering that incense are composed of primarily dried herbs and wood, and mosquito coils are similar to incense with the addition of pyrethrum powder (Eggert et al., 2004).

Benzene is a suspected human carcinogen so any amount of benzene can potentially cause adverse health effects. Recommended exposure levels vary between 0.1 ppm to 10 ppm over an 8-hour period of time. This study found that all vapors released during the 50-minute collection time were within safe limits as defined by OSHA, NIOSH, and ACGIH. Studies have noted that the concentration of benzene being produced may not be a concerning level, but humans are further exposed to benzene when pumping gas and from cigarette smoke (Plebani et al., 1999). Levels may not be concerning alone, but when looking at all possible exposures over a duration of time, a person may be over the recommended threshold for benzene exposure.

The measurable levels of benzene that were found in incense and mosquito coils in this study suggested that further investigation is needed to determine the extent to which harmful compounds may be emitted during the use of candles, incense, and mosquito coils. This project can be further researched in many ways. Testing incense and mosquito coils for a longer duration of time and under different conditions would be beneficial to simulate real-life burning experiences. Mosquito coils are typically burned outside in a ventilated area so burning conditions could be altered to simulate real-life burning. However, incense are normally burned in small rooms with multiple incense being burned in a short duration of time. This allows many factors to influence the concentration of benzene being emitted from incense. Many individuals burn these materials on a daily basis making this topic relevant and suitable for further research.

Future studies should look into other chemicals that may be released from candles. Other studies have noted that candles may release toluene, xylene, acrolein, and methylene

chloride (Lee et al., 2006). These chemicals are known to be dangerous to humans so candles, incense, and mosquito coils should be investigated to see if their emissions are threatening to human health. Sooting is another problem that is caused by a disturbance to a burning candle. Past studies have reviewed the particle emissions and detected soot on surfaces surrounding burning candles (Fine et al. 1999). Although candles may not release harmful concentrations of benzene, the health effects of inhaling soot should be further studied.

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