App. Envi. Res. 39(3) (2017): 77-90

Applied Environmental Research

Journal homepage : http://www.tci-thaijo.org/index.php/aer

Investigation of Formaldehyde in Gross Anatomy Laboratory: Area-Based and Exposure Levels, Ventilation, Health Risk and Clinical Symptoms

Kritsana Homwutthiwong[1], Maneerat Ongwandee[2,*]

[1] Faculty of Medicine, Mahasarakham University, Mahasarakham, Thailand
[2] Environmental Engineering Program, Faculty of Engineering, Mahasarakham University, Mahasarakham, Thailand
[*] Corresponding author: Email: maneerat.o@msu.ac.th

Article History
Submitted: 1 August 2017/ Accepted: 26 September 2017/ Published online: 29 November 2017

Abstract

Formaldehyde is commonly used in embalming solution in medical fields to preserve tissues. Formaldehyde vapor released from cadavers during dissection practice can adversely affect students and instructors in gross anatomy laboratories. Therefore, this study investigated formaldehyde concentrations in a gross anatomy laboratory of Mahasarakham University, Thailand. Area-based sampling was conducted for three scenarios: (1) a laboratory cleaning period, (2) three periods of teaching classes and (3) a non-teaching class period. Personal samples were also collected for five consecutive working days from two anatomy lab instructors and a non-lab instructor. Measurements were conducted during May to October 2016 using cartridges filled with dinitrophenylhydrazine coated silica gel. Samples were desorbed and analyzed by high performance liquid chromatography. Results showed that the average area-based concentrations during the dissection classes ranged from 9.3 to 17.6 ppb, while the non-teaching class concentration was 5.5 ppb. The concentration increased to 3.1×10^{3} ppb during the laboratory cleaning. The five-day average exposure concentrations were 7.6 ppb and 4.7 ppb for the two lab instructors and 1.1 ppb for the non-lab instructor. The lifetime cancer risk estimates were 5.8×10^{-6}, 2.9×10^{-6} and 8.1×10^{-7}, respectively. The three most reported clinical symptoms by the gross anatomy students were associated with formaldehyde concentrations above 16 ppb at the significance level of 0.05. These included unpleasant odor, general fatigue or fatigue after awakening and dizziness with response rates of 57.5 %, 38.5 % and 33.1 %, respectively. Improvement of the ventilation and source control measures are essential for reducing formaldehyde emissions in the gross anatomy laboratory.

Keywords: Indoor air quality; Formaldehyde-related symptoms; Dissection; Ventilation

Introduction

Formaldehyde is a colorless gas with an irritating pungent odor. It is extremely soluble in water and referred to as formalin when present in solution form. Formaldehyde is a major pollutant in both indoor and outdoor areas especially in workplaces. In a previous study a typical office building had an average formaldehyde concentration of 0.03 ppm, while the outdoor concentration was three times lower [1]. This indicates that in-office formaldehyde emanates mainly from indoor sources such as building materials, furnishings and consumer products. Formaldehyde is used extensively in the wood and plastic industries and is commonly found in medical schools and hospitals where it is used in preserving cadavers for forensic medicine and embalming for teaching [2-5]. Although formalin is widely used in many fields due to its low price and high quality reaction, its toxicity is usually ignored [6-7]. Exposure to low concentrations of formaldehyde can cause irritation to eyes, nose, upper respiratory airway and nerve toxicity [8-9], while exposure to high concentrations results in irritation to the lower respiratory airway, decreasing pulmonary function, and can cause ocular melanoma and leukemia [10-11]. The International Agency for Research on Cancer [12] has categorized formaldehyde in Group 1, as a carcinogenic to humans since sufficient evidence indicates a link to naso-pharyngeal cancer in humans involved in high or prolonged exposure.

Normally, cadavers used in gross anatomy laboratories are embalmed with formaldehyde fixative. Formaldehyde contents vary among the gross anatomy laboratories, typically ranging from 5 to 7 % [13-16]. The fixative is usually infused via the femoral arteries or internal carotid arteries. Thus, the use of concentrated formaldehyde fixative partially contributes to formaldehyde emissions from cadavers during dissection, which in turn inevitably affect students and instructors in the gross anatomy laboratories. Formaldehyde levels in gross anatomy laboratories were reported to be 0.11-1.38 ppm in Japan [2-3, 13], 0.12-9.16 ppm in Korea [17], 0.4-0.5 ppm in Singapore [6], 0.52-1.48 ppm in USA [18] and 0.18-1.69 ppm in Brazil [4]. All concentrations found exceeded the recommended levels by all international guideline limits even though ventilation systems were provided. Beside the influential factors of formaldehyde content in embalming solution and dissection room ventilation, formaldehyde levels depend on other factors such as a number of cadavers in a laboratory, stage of dissection process, room temperature and posture of participants [5,14,19-20].

Despite the number of medical schools and gross dissection units in Thailand, few studies on the investigation of indoor air quality in these facilities have been conducted. Recently, there have been studies on measuring formaldehyde in gross anatomy rooms and at the breathing zone of students and instructors at two medical schools [15, 21-22]. The concentrations were found in the 0.2-1.2 ppm range. However, the relationships of formaldehyde levels and clinical symptoms of working students and staff are still lacking.

Thus, this research aimed to measure formaldehyde concentrations and ventilation rates in a gross anatomy laboratory room at Mahasarakham University, Thailand during different laboratory activities. Furthermore, formaldehyde exposure levels at the breathing zone of instructors were monitored in order to assess their lifetime cancer risks. The prevalence of clinical symptoms was also investigated to determine the association with formaldehyde levels.

Materials and methods

1) Cadaver embalming

All cadavers used in anatomy laboratory teaching at Mahasarakham University were supported by Khon Kean University, Thailand. Cadavers were perfused via the femoral arteries

with a 48-L preservation fixative solution using a pressure-injection device. The fixative contains 5 % formaldehyde, 23.8 % ethanol, 0.25 % phenol, 3.13 % potassium nitrate and 5 % glycerol in water on a volume basis. Consequently, the cadavers had been preserved in an embalming solution mixture of phenol, glycerol and water in a ratio of 1:3:15 for approximately a year. After the preservative process, the cadavers were encased in plastic bags and delivered to the Faculty of Medicine, Mahasarakham University where they were kept in the gross anatomy laboratory room while not in use.

2) Area-based air sampling in the gross anatomy laboratory

The gross anatomy laboratory of Mahasarakham University is a non-air conditioned room with a total area of 380 m^2. A layout of the room is shown in Figure 1. All windows and doors were kept open and four large standing fans were turned on to assist ventilation during the teaching classes. When the laboratory was not in use, the windows and doors were closed. Area-based air samples were collected at three sampling points for five campaigns as shown in Figure 1. Sampling Point#1 was in the middle of the laboratory since most of the teaching activities were occurred at this area. Sampling Point#2 was in the corner near the washbasins and 20-L plastic buckets with lids containing human organs in 10 % formalin solution in water. This sampling location was also selected due to the potential emission sources of formaldehyde. Meanwhile, Sampling Point#3 was outside the laboratory in the building corridor. The out-lab location was selected for comparative study with in-lab formaldehyde concentrations. Air sampling was conducted from May to October 2016. The cadavers were placed on the dissection tables and they were kept in sealed plastic bags when not in use. The barrels containing formalin were also located in this laboratory. Each barrel contained 20 L of 40 % formaldehyde solution in water stabilized with methanol.

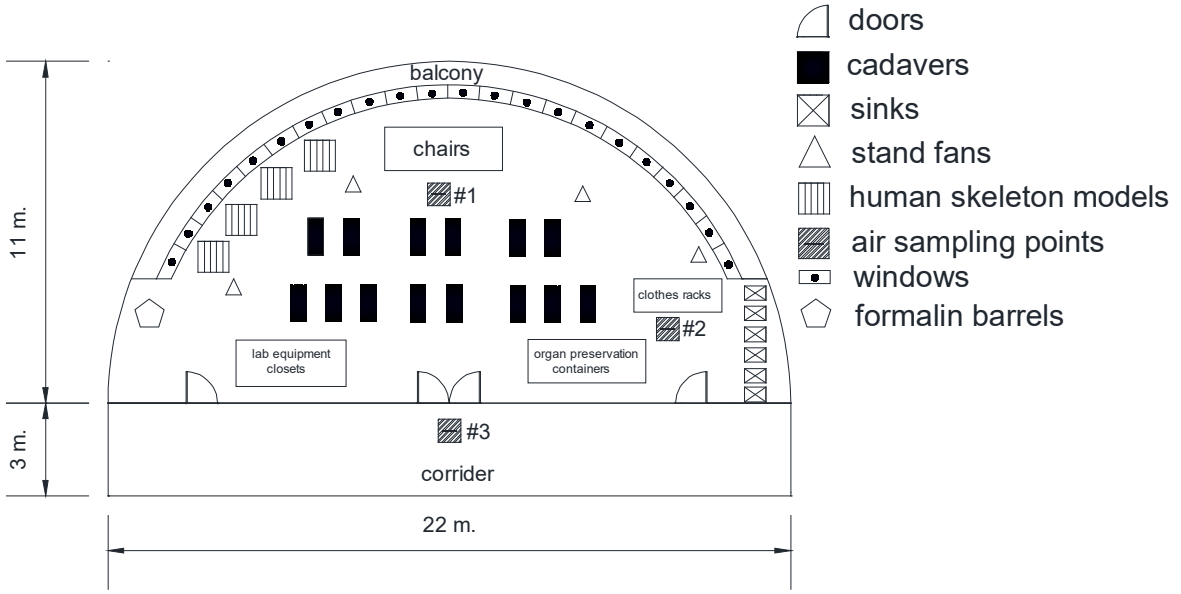

Figure 1 Layout of the gross anatomy laboratory.

The sampling campaigns are summarized in Table 1. The gross anatomy provided medical students with 11 new non-dissected cadavers while nursing students received 4 old previously dissected cadavers which had been kept for study by last year's class. Sampling Campaign A was done during the laboratory room cleaning process, which regularly occurs at the end of each semester. The floor cleaning was done using a commercial cleaning agent with main components of alkyl polyglycoside, polyoxyethylene alkyl ether and sodium laureth sulphate. At that time, specimens and tissue samples were being transferred into new containers with fresh 10 % formalin preservative solution for long-term storage. Therefore, concentrations in Campaign A were expected to be significantly higher than those in the other Campaigns. Sampling campaigns B, C and D were chosen for comparison of formaldehyde concentrations among the different class activities offered for two groups of students, while concentrations measured in Campaign E were considered as backgrounds.

3) Formaldehyde sampling and analysis

Cartridges filled with 60/80 mesh dinitrophenylhydrazine (DNPH) coated silica gel (SKC Inc., USA) were used to collect gaseous formaldehyde at a height of ~0.6 m above the floor in order to avoid hindering the class activities. The air in the laboratory can be considered as a well-mixed core because of the use of four large stand fans. A personal sampling pump (SKC®) was used to draw air at a flow rate of ~0.2-0.9 L min^{-1} for the study period of three hours, except that the air samples of Campaign A were collected for an hour. The sampling flow rates of 0.2-0.9 L min^{-1} provided the sufficient amounts of formaldehyde collected on adsorbents for being quantified by a detector, while they were not too high to cause any breakthrough. Formaldehyde adsorbed in the cartridges were desorbed with 2 mL of acetonitrile (Merck & Co., USA) and a 40- mL aliquot sample was then analysed by high performance liquid chromatography (Shimadzu Co., Japan) using a UV detector operating at 360 nm [23]. A mobile phase of 60 % acetonitrile per 40 % of water was pumped at a flow rate of 0.8 mL min^{-1} for derivative separation in an Inertsil ODS-3 column (46 × 150 mm^2). The method detection limit (MDL) followed the US EPA guideline procedure [24]. The MDL of measured formaldehyde was 0.13 ppb for a sample volume of 162 L. A concentration of formaldehyde in the laboratory air can be calculated as shown in Eq. 1.

$$C_{formaldehyde} = \frac{Mass}{Q \times T} \qquad \text{(Eq.1)}$$

where $C_{formaldehyde}$ = formaldehyde concentration in the laboratory air (μg m^{-3}), *Mass* = amount of formaldehyde adsorbed in the cartridge (μg), Q = sampling flow rate (m^3 min^{-1}) and T = total sampling time (min).

The mass concentration unit (μg m^{-3}) can be converted to a part per billion (ppb) unit at the ambient temperature of 25˚C and pressure of 1 atm by Eq. 2.

$$C_{ppb} = \frac{C_{\mu g/m^3} \times 24.45}{MW} \qquad \text{(Eq. 2)}$$

where C_{ppb} = formaldehyde concentration in ppb, $C_{\mu g/m^3}$ = formaldehyde concentration in μg m^{-3} and *MW* = molecular weight of formaldehyde (=30 g mol^{-1}).

Table 1 Description of the sampling campaigns

Sampling campaign	Sampling date/ class semester	Laboratory conditions	Involving people
A	10/5/2016 Semester 1/2016	Laboratory cleaning at the end of previous semester	Laboratory instructors
B	25/8/2016 Semester 2/2016	Muscular system and nervous system of the dissected cadavers	Nursing students and laboratory instructors
C	30/8/2016 Semester 2/2016	Dissection of superficial back and posterior shoulder region of the new cadavers	Medical students and laboratory instructors
D	13/9/2016 Semester 2/2016	Dissection of forearm and wrist of the new cadavers	Medical students and laboratory instructors
E	30/9/2016 Semester 2/2016	No laboratory session offered	None

4) Ventilation measurement

The room air exchange rates were measured using a tracer gas decay method. Carbon dioxide (CO_2) generated from dry ice was used as a tracer gas. The air exchange measurements were done when no people were in the laboratory to avoid occupant-generated CO_2. We measured the air exchange rates of the laboratory for two conditions. One is the simulated condition similar to when the laboratory session was offered (Campaigns A, B, C and D), while the other was similar to when no laboratory session was offered (Campaign E). CO_2 concentrations were monitored with a Testo® 350 M/XL sampler with an ambient CO_2 infrared detection probe. The time-dependant CO_2 concentrations were then fitted to a mass balance model as given in Eq. 3.

$$C_t = C_0 e^{-\lambda t} + C_{out}\left[1 - e^{-\lambda t}\right] \quad \text{(Eq. 3)}$$

where Ct = in-lab concentration at time t (mg m^{-3}), C_0 = in-lab concentration at t=0, C_{out} = outdoor concentration, λ = air exchange rate (h^{-1}) and t = time (h).

A nonlinear regression method was used to determine the air exchange rate.

5) Health risk assessment

Two instructors teaching the gross anatomy (Instructors#1 and #2) and an instructor not involved with the gross anatomy laboratory (Instructor#3) were selected as study subjects. Personal air samples were collected for eight working hours at the subject's breathing zone using the same area-based equipment. The measurements were done for five working days consecutively from 19/9/2016 to 23/9/2016. Note that the personal air sampling of Instructor#2 was additionally conducted on 10/5/2016 at which time the laboratory cleaning occurred (Campaign A). The lifetime cancer risks for the instructors exposed to formaldehyde were estimated according to US Environmental Protection Agency guidelines as given in Eq. 4 [25].

$$R = CDI \times IUR \quad \text{(Eq. 4)}$$

where R = lifetime cancer risk (unitless), IUR = Inhalation Unit Risk (1.3×10^{-5} m^3 μg^{-1} for formaldehyde) [26] and CDI = chronic daily intake averaged over 70 years (μg m^{-3}). It was calculated as given in Eq. 5.

$$CDI = \frac{C \times ET \times EF \times ED \times CF}{AT} \quad \text{(Eq. 5)}$$

where C = five-day average concentration of formaldehyde obtained from the personal air sampling (μg m^{-3}), ET = exposure time (h d^{-1}), EF = exposure frequency (d a^{-1}), ED = exposure duration (years), CF = con-version factor (year per

8760 hours) and AT = averaging time (70 years for lifetime cancer risk).

6) Assessment of clinical symptoms with a questionnaire

The questionnaire survey was conducted while the area-based concentrations were measured. The number of questionnaire respondents was 120, of whom 33 were men and 87 were women. The respondents were chosen as random sampling. The inclusion criteria were first year nursing students and second year medical students while exclusion criteria were smokers and asthmatic persons. The questionnaire was modified slightly from the questionnaire used in the study of Saowakon et al. [15]. It included three parts, i.e., respondent background information protective equipment use and formaldehyde-related clinical symptoms. The symptom response scale had five-point rating scales: 0 - not recognizable, 1 - a few recognizable, 2 - moderate irritating, 3 - high irritating and 4 - intolerable. According to the five-point rating scales, we calculated the response percentage of each reported symptom as given in Eq. 6.

The prevalence of symptoms was then analysed in relation to the measured formaldehyde concentrations. The National Institute for Occupational Safety and Health, USA recommends a time-weighted average concentration of formaldehyde of 16 ppb for prevention of work-related injury and illness [27]. Thus, we used Chi-square test to analyse the relationship between the prevalence of symptoms reported at different response scales and the formaldehyde levels to which the students were exposed during the laboratory class. The exposure concentrations were classified in two levels, i.e., < 16 ppb and > 16 ppb.

$$response\quad percentage = \left[\frac{(n_0 \times 0) + (n_1 \times 1) + (n_2 \times 2) + (n_3 \times 3) + (n_4 \times 4)}{4}\right] \times 100 \qquad \text{(Eq. 6)}$$

where $n_{0, 1, 2, 3, 4}$ = proportions of the numbers of respondents reporting a rating scale, i.e., 0, 1, 2, 3 and 4, respectively.

Results and discussion

1) Formaldehyde levels

Table 2 summarizes formaldehyde concentrations measured inside and outside the laboratory at three sampling points as depicted in Figure 1. Ratios of the inside to outside concentrations (In/Out ratio) are also presented.

The average concentrations in the laboratory ranged from 9.25 to 17.59 ppb during the class sessions (Campaigns B, C and D), while the average background concentration was 5.50 ppb (Campaign E - no class). The practical dissections appear to be the main sources of formaldehyde emitted into the laboratory air. However, the formaldehyde levels in this study were relatively lower than those measured in the previous studies [3, 5, 15, 21-22]. It is worth noting that the concentrations measured in the previous semester but at the different dissections, i.e., perineum and pelvis, were 46 ppb for inside the lab and 19 ppb for outside the lab. Thus, this may be due to different regions of the dissected cadavers, the number of cadavers and the time spent on dissection.

Comparing different dissection activities indicates that Campaigns C and D had in-lab concentrations almost two times higher than Campaign B. The higher concentrations were found in the lab sessions offered for the medical students, which are mainly attributed to the dissections of all fresh cadavers similar to the study of Sugata et al. [16], which reported a significant increase in formaldehyde levels in the laboratory after skin incision. In contrast, the

nursing students did not have a practical dissection during the Campaign B period. The instructors provided the nursing students with old dissected cadavers for the muscular and nervous systems lab, while the new cadavers were kept in sealed plastic bags. Thus, the main emission of formaldehyde from the cadavers was expected to be lower in the nursing student class rather than in the medical student class. It should be noted that the concentration at Sampling Point#2 was two times higher than that at Point#1. On that sampling day, most of the teaching activities were occurred near Point#2 where the students studied the preserved human organs e.g. brains, brainstems, spinal cords, meninges and cerebellums.

The lowest concentrations were detected when no class was offered since all cadavers were kept in the sealed plastic bags. The cadavers appear to be the major sources of released formaldehyde into the room air due to the use of embalming fluids containing concentrated formaldehyde. Thus, embalming methods are one of the key mitigation measures for formaldehyde in gross anatomy laboratories. Several chemical combinations have been used to reduce the percentage of formaldehyde or even to replace formaldehyde in embalming solution [28-31]. For example, Thiel and ethanol-glycerin techniques have been adopted for specimen fixation [28-29]. A saturated salt solution (SSS) consisted of sodium chloride has also been employed to embalm cadavers for surgical skills training [30]. Furthermore, Kawamata and Kodera [31] reported that a perfusion of ammonium carbonate solution into formaldehyde-fixed cadavers was able to reduce formaldehyde emitted into the air since the reaction of ammonium carbonate with formaldehyde results in harmless hexamethylenetetramine.

During the end-semester laboratory cleaning campaign (Campaign A) the formaldehyde concentrations increased significantly by 200 times above those measured in regular classes. The major sources of emitted formaldehyde were due to the transferring procedure of specimens and tissue samples into the new fresh 10 % formalin preservative solution. These activities were done under the same ventilation condition as class ventilation. Therefore, one of the measures for lowering formaldehyde vapor is to perform the transferring procedure of anatomic specimens in a fume hood.

In this study, the average formaldehyde concentrations measured during the regular classes were found to be 10-1000 times lower than those measured in other anatomy laboratories of the Thai medical schools [15, 20-21]. This may be because our studied laboratory had five times smaller numbers of the cadavers than the other schools. Moreover, our sampling campaigns did not involve morphological studies of formalin-preserved organs, which are expected to cause high emissions of formaldehyde. These morphological studies for the medical students are usually taken place in another classroom.

Table 2 In- and out-laboratory concentrations (ppb) and In/Out ratio

Sampling campaign	In-lab concentration		Out-lab concentration	In/Out ratio
	Sampling point#1	Sampling point#2	Sampling point#3	
A	3023	3216	664	4.70±0.21
B	6.49	12.01	7.37	1.26±0.53
C	17.44	17.74	8.76	2.01±0.02
D	15.02	16.36	7.53	2.08±0.13
E	4.44	6.56	3.83	1.44±0.39

The in-lab to out-lab (In/Out) ratios of formaldehyde concentrations are also shown in Table 2. Generally, an In/Out ratio of contaminant concentrations indicates where the contaminant is mainly from outside the room or it is originally from a source(s) inside the room. In this study, the ratios of all sampling intervals are greater than 1, indicating that the important sources of formaldehyde were indeed inside the laboratory. Comparing the out-lab concentrations between Campaign E (background) and the other Campaigns (class activities) may imply that the in-lab formaldehyde was brought to the building corridor due to the room ventilation.

During the lab sessions, the room ventilation, driven by electric standing fans along with natural ventilation, was found to be 0.2 air change per hour (0.7 m^3 h^{-1} m^{-2}), while the ventilation was as low as 0.05 air change per hour (0.17 m^3 h^{-1} m^{-2}) when the laboratory was not in use. The Engineering Institute of Thailand [32] has released a draft of a new standard for air-conditioning and ventilation systems. An air conditioned laboratory should have a ventilation rate of 2 m^3/h-m^2, while the Department of Disease Control of Thailand [33] suggests a fresh air intake rate for a laboratory or autopsy room of at least 2 air changes per hour using an air-conditioning system. In other words, an autopsy room requires at least 2 times the air within the room to be replaced by new fresh air within an hour. In the present study, the gross anatomy lab is non-air conditioned, thus comparing the measured air exchange rates to the standard may not be appropriate. However, the air exchange rate during class time indicates obviously insufficient ventilation. The current ventilation method by natural ventilation can be regarded as an ineffective and unsuitable method for exhausting contaminated air out of the dissection area and for supplying fresh air into the laboratory. Thus, improvement of the ventilation system is essential. The natural ventilation should be replaced with a mechanical ventilation system that is designed to move out and bring in a certain amount of air at a specific speed. To be specific to this type of the workplace, a local exhaust system equipped at the dissection table should be used to control air contaminants by trapping them at or near the source (cadaver). A local exhaust system is mainly composed of a hood or opening that captures the contaminant, ducts, air cleaning device and exhaust stack. Additionally, the improvement of the ventilation system will combine with the other purpose of ventilation that is to maintain temperature and humidity at comfortable levels. During the measurements, the temperature inside the laboratory varied in relation to the outdoor ambient temperature due to the natural ventilation. The laboratory temperature ranged from 30 to 34 ˚C, which were relatively higher than the comfort temperature of 26 ˚C [34]. Controlling the room temperature not only maintains thermal comfort for the occupants, but also reduces emissions of formaldehyde from the in-lab sources such as cadavers and containers filled with formalin solution [35].

2) Life time cancer risk among instructors

Table 3 summarizes daily formaldehyde levels at the breathing zone of three instructors and their occupational activities over five sampling days. Note that the personal air sampling of Instructor#2 was additionally conducted on 10/5/2016 at which time the laboratory cleaning occurred (Campaign A). Among the five studied days, the Instructors#1 and #2 experienced the highest concentrations of 16.82 and 10.27 ppb, respectively, on 23/9/2016. The instructors taught the morphology of brain structure using formalin-preserved organs in the classroom. Comparing between the laboratory preparation days (19/9 and 21/9) and the laboratory teaching days (20/9 and 22/9) indicated that Instructor#1 experienced the higher

concentrations when preparing the laboratory materials. The laboratory preparation usually requires more closely work tasks with the cadavers and preserved organ than those on the teaching days. It is worth noting that Instructor#2 had the lower concentrations than Instructor#1 since she served as a lab assistant whom was not required to dissect cadavers. Meanwhile, the non-lab Instructor#3 experienced the lowest concentrations of < 2 ppb for all five days. The five-day average concentrations were 7.63, 4.73, and 1.06 ppb for Instructors#1, #2 and #3, respectively. The average concentrations did not exceed the guideline value for occupational exposure of 16 ppb [27]. However, the two instructors involved in teaching gross anatomy were exposed to formaldehyde at levels 4-7 times greater than the instructor who was not involved. Ohmichi et al. [3] also indicated a serious occurrence for people who work in the dissection room. Long-term exposures have been associated with cancers of the lung and nasal passageways. The anatomy laboratory instructors are prone to formaldehyde causing illnesses via activities other than teaching such as laboratory preparing and laboratory cleaning. The concentration measured during the laboratory cleaning event was found to be 174 ppb, which was 40 times greater than regular work exposure levels. Exposure to a high concentration of formaldehyde even in short time may cause severe mucous membrane irritation, burning and lacrimation. It also causes significant inflammation of the lower respiratory tract, resulting in swelling of the throat, inflammation of the trachea and bronchi, narrowing of the bronchi, inflammation of the lungs, and accumulation of fluid in the lungs. Pulmonary injury may continue to worsen for 12 hours or more after exposure [36].

Table 3 Daily exposure concentrations of formaldehyde and occupational activities for each studied instructor

Instructor	Concentration (ppb) and occupational activities						
	19/9/2016	**20/9/2016**	**21/9/2016**	**22/9/2016**	**23/9/2016**	**10/5/2016***	**Average**
#1	8.70 Lab preparation for cadaver dissection	2.82 Cadaver lab teaching	6.98 Lung dissection and respiratory lab preparation	2.81 Respiratory lab teaching	16.82 Brain and spinal cord dissection and lab teaching	nd	7.63± 5.75
#2	2.37 Lab preparation	2.77 Lab preparation	5.53 Working with preserved organs	2.71 Lab preparation	10.27 Assisting in lab teaching	174 Lab cleaning and preparing for preservative solution	4.73± 3.35**
#3	1.81 Class teaching	0.21 Office working	1.95 Class teaching	0.69 Office working	0.63 Office working	nd	1.06± 0.77

Note: * Date 10/5/2016 was Sampling Campaign A – Laboratory cleaning at the end of previous semester, ** Excluding date 10/5/2016 and "nd" denotes not determined.

Table 4 presents the CDI and lifetime cancer risk of the three instructors based on the average formaldehyde concentrations measured over five working days. Note that the exposure concentration measured on the laboratory cleaning day was excluded. All instructors had the lifetime cancer risk below the suggested range of 1×10^{-6} to 1×10^{-4} by USEPA [37]. The risk assessment indicated that the two instructors who were responsible for the gross anatomy course were more severely harmed than the non-lab instructor. However, the risks for the lab instructors may be underestimated since the chronic daily intakes were based on the activities that occurred only in the five-day measurement period. During the academic year, the lab instructors provide different laboratory topics which require different dissection conditions for cadavers and organs such as brains, spinal, heart and kidneys. These organs have been preserved in a 10 % formalin solution. Therefore, the laboratory preparation and teaching with these specimens can lead to significantly high exposure to strong formaldehyde vapor.

3) Formaldehyde-related clinical symptoms

The response percentage for all 17 clinical symptoms is shown in Table 5. The Chi-square test was used to analyse the association between the prevalence of the symptoms reported at different response scales and the exposure level of formaldehyde at a significance level of 0.05. The *p*-value of the hypothesis test is presented in Table 5.

Table 4 CDI and lifetime cancer risk of the instructors exposed to formaldehyde

Instructor	CDI (μg m^{-3})	Risk
#1	4.5×10^{-1}	5.8×10^{-6}
#2	2.2×10^{-1}	2.9×10^{-6}
#3	6.2×10^{-2}	8.1×10^{-7}

Table 5 Prevalence of formaldehyde-related clinical symptoms

Clinical symptom	Response percentage	*p*-value
Unpleasant odor	57.50	<0.001*
General fatigue	38.54	0.001*
Dizziness	33.12	0.012*
Burning nose	24.79	0.315
Unusual thirst	23.97	0.154
Headache	22.50	0.154
Burning eyes	22.09	0.154
Eye irritation	21.24	0.315
Skin irritation	13.55	0.079
Wheezing	13.54	0.315
Tearing	13.33	0.315
Vomiting	10.00	-
Itchy nose	9.79	-
Blurred vision	8.96	0.315
Chest pain	7.50	-
GI irritation	6.46	0.315
Faint	2.92	-

Note: * *p*-value less than 0.05 and "-" denotes that the symptom was so rarely observed, that most levels had '0' for the expected value and Chi-square values could not be reliably computed.

The Chi-square test shows that the percentages of students in the laboratory with formaldehyde concentrations greater than 16 ppb reporting the following symptoms that were statistically different among the five response scales (0-4). These symptoms included unpleasant odor (57.50 %), general fatigue (38.54 %) and dizziness (33.12 %), corresponding to previous studies on the clinical symptoms related to formaldehyde exposure in the gross anatomy laboratory [5, 15, 21]. The most reported symptom was unpleasant odor since the major uptake of gaseous formaldehyde is inhalation which causes respiratory tract irritation, obstruction of airways and impaired lung function [38]. Prolonged exposure has caused bronchial asthma and DNA damage in lymphocytes and buccal cells [39-40] Furthermore, formaldehyde exposure during dissection might lead to Immunoglobulin E (IgE)-mediated sensitization and specific IgE antibodies against formaldehyde but not Immunoglobulin G (IgG) against formaldehyde-albumin [41]. Additionally, exposure to formaldehyde vapor or liquid formalin may sensitize skin reaction even below 1 ppm and it causes major allergic symptoms and intensified chemical sensitivities [20].

The other two most reported symptoms were general fatigue and dizziness, which might be related to inhalation of formaldehyde after dissection. Saowakon et al. [15] indicated an increase in means of FVC and FEV1 in pulmonary function test among the anatomy-lab students and instructors. Although the associations between the prevalence of eye and nose irritations and unusual thirst and levels of formaldehyde were not statistically significant at a level of 0.05, their response rates were still greater than 20 %. Eye irritation is a common complaint that has been reported at formaldehyde concentrations of 0.5 to 1 ppm in healthy volunteers [42]. Besides the eyes symptoms, burning nose and unusual thirst were found in anatomy-lab students [5, 15]. These indicate upper respiratory tract irritation.

Additionally, we conducted the questionnaire survey among the two lab-instructors. The four symptoms that were reported with 100 % response included unpleasant odor, general fatigue, burning nose and burning eyes, while skin irritation was reported with 87.5 %. Apart from the similar students' response symptoms for unpleasant odor and general fatigue, the lab-instructors also complained about burning nose and eyes and skin irritation. These responses could be due to the prolonged working period with the cadavers and formalin-preserved organs.

Conclusions

Dissection of cadavers is considered an important activity where formaldehyde emitted in the anatomy lab due to the use of embalming fluids containing concentrated formaldehyde. Thus, the lab instructors and students are prone to formaldehyde-causing symptoms. Proper personal protective equipment should be worn during work periods in the laboratory. However, the personal protection can only reduce the exposure level to a certain degree. Embalming techniques are one of the most critical issues for controlling formaldehyde emitted from the cadavers in the lab. Therefore, formaldehyde-free or low-formaldehyde embalming techniques should be considered as an alternative method for preserving anatomical specimens in order to reduce formaldehyde emissions. Another source control is to keep the cadavers and preserved organs in a separated storage room when not in use. Improvement of the laboratory facilities is also essential for controlling good indoor air quality in a gross anatomy laboratory. Use of an effective room ventilation system or dissection tables equipped with local exhaust ventilation can remove contaminated air from the area.

Acknowledgements

This study was financially supported by the Faculty of Engineering, Mahasarakham University. The author thanks Mr. Kan Nameepol, Miss Atchara Panthapol and Miss Suwanlee Hongsiri for assistance in air sampling and sample analyses. The authors also appreciate the cooperation of the students for questionnaire survey.

References

[1] Ongwandee, M., Moonrinta, R., Panyametheekul, S., Tangbanluekal, C., Morrison, G. Investigation of volatile organic compounds in office buildings in: Concentrations, sources, and occupant symptoms. Building and Environment 2011, 46, 1512-22.

[2] Tanaka, K., Nishiyama, K., Yaginuma, H., Sakaki, H., Maeda, T., Kaneko, S. …, Tanaka, M. Formaldehyde exposure levels and exposure control measures during an anatomy dissecting course. Acta Anatomica Nipponica 2003, 78, 43-51. (English abstract).

[3] Ohmichi, K., Komiyama, M., Matsuno, Y., Takanashi, Y., Miyamoto, H., Kadota, T. …, Mori, C. Formaldehyde exposure in a gross anatomy laboratory. Environmental Science and Pollution Research 2006, 13, 120-124.

[4] Ochs, S. M., Grotz, L. O., Factorine L. S., Rodrigues. M. R., Pereira, N. A. D. Occupational exposure to formaldehyde in an institute of morphology in Brazil: A comparison of area and personal sampling. Environmental Science and Pollution Research 2011, 19, 2813-2819.

[5] Azari, M. R., Asadi, P., Jafari, M. J., Soori, H., Hosseini, V. Occupational exposure of a medical school staff to formaldehyde in Tehran. Tanaffos 2012, 11, 36-41.

[6] Chia, S. E., Ong, C. N., Foo, S. C., Lee, H. P. Medical students' exposure to formaldehyde in a gross anatomy dissection laboratory. Journal of American College Health 1992, 41, 115-119.

[7] Brenner, E. Human body preservation – Old and new techniques. Journal of Anatomy 2014, 224, 316-344.

[8] Pala, M., Ugolini, D., Ceppi, M., Rizzo, F., Maiorana, L., Bolognesi, C. …, Vecchio, D. Occupational exposure to formaldehyde and biological monitoring of research institute workers. Cancer Detection and Prevention 2008, 32, 121-126.

[9] Songur, A., Ozen, O. A., Sarsilmaz, M. The toxic effects of formaldehyde on the nervous system. Reviews of Environmental Contamination and Toxicology 2010, 203, 105-118.

[10] Dufresne, A., Infante-Rivard, C., Malo, J. L., Gautrin, D. Exposure to formaldehyde among animal health students. American Industrial Hygiene Association Journal 2002, 63, 647-650.

[11] Noisel, N., Bouchard, M., Carrier, G. Evaluation of the health impact of lowering the formaldehyde occupational exposure limit for Quebec workers. Regulatory Toxicology and Pharmacology 2007, 48, 118-127.

[12] IARC. Formaldehyde. In: A review of human carcinogens. Part F: Chemical agents and related occupations. International Agency for Research on Cancer, France. 2009, 401-436.

[13] Kurose, T., Kodera, H., Aoyama, H., Kawamata, S. Formaldehyde concentration in the air and in cadavers at the gross anatomy laboratory in Hiroshima University. Hiroshima Journal of Medical Sciences 2004, 53, 33-37.

[14] Vohra, M. S. Personal formaldehyde exposure level in the gross anatomy dissecting room at College of Medicine King Saud University Riyadh. International Journal of Occupational Medicine

and Environmental Health 2011, 24, 108-113.

[15] Saowakon, N., Ngernsoungnern, P., Watcharavitoon, P., Ngernsoungnern, A., Kosanlavit, R. Formaldehyde exposure in gross anatomy laboratory of Suranaree University of Technology: A comparison of area and personal sampling. Environmental Science and Pollution Research 2015, 22, 19002-19012.

[16] Sugata, Y., Miyaso, H., Odaka, Y., Komiyama, M., Sakamoto, N., Mori, C., Matsuno, Y. Levels of formaldehyde vapor released from embalmed cadavers in each dissection stage. Environmental Science and Pollution Research 2016, 23, 16176-16182.

[17] Kim, H., Kim, Y. D., Cho, S. H. Formaldehyde exposure levels and serum antibodies to formaldehyde-human serum albumin of Korean medical students. Archives of Environmental Health: An International Journal 1999, 54, 115-118.

[18] Keil, C. B., Akbar-Khanzadeh, F., Konecny, K. A. Characterizing formaldehyde emission rates in a gross anatomy laboratory. Applied Occupational and Environmental Hygiene 2001, 16, 967-972.

[19] Shiraishi, N. Levels of formaldehyde, phenol and ethanol in dissection room air and measures for reduction. Japanese Journal of Occupational Medicine and Traumatology 2006, 54, 1-9.

[20] Takahashi, S., Tsuji, K., Fujii, K., Okazaki, F., Takigawa, T., Ohtsuka, A., Iwatsuki, K. Prospective study of clinical symptoms and skin test reactions in medical students exposed to formaldehyde gas. The Journal of Dermatology 2007, 34, 283-289.

[21] Lakchayapakorn, K., Watchalayarn, P. Formaldehyde exposure of medical students and instructors and clinical symptoms during gross anatomy laboratory in Thammasat University. Journal of the Medical Association of Thailand 2010, 93, S92-S98.

[22] Watchlayann, P., Tapyai, A., Lakchayapakorn, K. Area and personal exposure levels to formaldehyde and its variation among undergraduate students during gross anatomy laboratory practice. EnvironmentAsia 2014, 7, 54-59.

[23] US EPA. Compendium method TO-17 determination of volatile organic compounds in ambient air using active sampling onto sorbent tubes. US Environmental Protection Agency, USA. 1999, EPA/625/R-96/010b.

[24] US EPA. Revised assessment of detection and quantitation approaches. US. Environmental Protection Agency, Engineering and Analysis Division Office of Science and Technology Office of Water (4303T), Washington DC, USA. 2004, EPA-821-B-04-005.

[25] US EPA. Risk assessment guidance for superfund volume I human health evaluation manual (Part A). US Environmental Protection Agency, USA. 1989, EPA/540/1-89/002.

[26] US EPA. Integrated Risk Information System (IRIS). US Environmental Protection Agency, USA. 2017. [Online] Available from: https://www.epa.gov/iris.

[27] NIOSH. Pocket guide to chemical hazards: Formaldehyde. National Institute for Occupational Safety and Health, USA. 2016. [Online] Available from: https://www.cdc.gov/niosh/npg/npgd0293.html

[28] Hammer, N., Löffler, S., Feja, C., Sandrock, M., Schmidt, W., Bechmann, I., Steinke, H. Ethanolglycerin fixation with thymol conservation: A potential alternative to formaldehyde and phenol embalming. Anatomical Sciences Education 2012, 5, 225-233.

[29] Hammer, N., Löffler, S., Bechmann, I., Steinke, H., Hädrich, C., Feja, C. Comparison of modified Thiel embalming and ethanol glycerin fixation in an anatomy environment: Potentials and limitations of two complementary techniques. Anatomical Sciences Education 2015, 8, 74-85.

[30] Hayashi, S., Homma, H., Naito, M., Oda, J., Nishiyama, T., Kawamoto, A. …, Itoh, M. Saturated salt solution method: a useful cadaver embalming for surgical skills training. Medicine (Baltimore) 2014, 93, 1-10.

[31] Kawamata, S., Kodera, H. Reduction of formaldehyde concentrations in the air and cadaveric tissues by ammonium carbonate. Anatomical Science Interna-tional 2014, 79, 152-157.

[32] EIT. Draft of standard for air-conditioning and ventilation systems. The Engineering Institute of Thailand under H.M. The King's Patronage, Bangkok, Thailand. 2016. [Online] Available from http://eit.or.th/DownloadDocument.

[33] Department of Disease Control. Manual for indoor air quality improvement for hospitals. Bamrasnaradura Infectious Diseases Institute, Bangkok, Thailand. 2007.

[34] Yamtraipat, N., Khedari, J., Hirunlabh, J. Thermal comfort standards for air conditioned buildings in hot and humid Thailand considering additional factors of acclimatization and education level. Sol Energy 2005, 78, 504-517.

[35] Godish, T. Indoor air pollution control. Lewis, Michigan, USA. 1989.

[36] US Department of Health and Human Services. ASTDR – Toxicological profile for formaldehyde. Public Health Service Agency for Toxic Substances and Disease Registry. 1999.

[37] US EPA. National oil and hazardous substances pollution contingency plan – final rule. Fed. Reg. 55 CFR Part 300-66. US Environmental Protection Agency, USA. 1990.

[38] Khaliq, F., Tripathi, P. Acute effects of formalin on pulmonary functions in gross anatomy laboratory. Indian Journal of Physiology and Pharmacology 2009, 53, 93-96.

[39] Souza, A. D., Devi, R. Cytokinesis blocked micronucleus assay of peripheral lymphocytes revealing the genotoxic effect of formaldehyde exposure. Clinical Anatomy 2014, 27, 308-312.

[40] Lorenzoni, D.C., Pinheiro, L.P., Nascimento, H.S., Menegardo, C.S., Silva, R.G., Bautz, W.G. …, da Gama-de-Souza, L.N. Could formaldehyde induce mutagenic and cytotoxic effects in buccal epithelial cells during anatomy classes? Medicina Oral Patologia Oral y Cirugia Bucal 2017, 22, e58-e63.

[41] Wantke, F., Focke, M., Hemmer, W., Tschabitscher, M., Gann, M., Tappler, P. …, Jarisch, R. Formaldehyde and phenol exposure during an anatomy dissection course: A possible source of IgE mediated sensitization? Allergy 1996, 51, 837-841.

[42] Lang, I., Bruckner, T., Triebig, G. Formaldehyde and chemosensory irritation in humans: A controlled human exposure study. Regulatory Toxicology and Pharmacology 2008, 50, 23-36.