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Relationship between Exposure to Formaldehyde and Immunoglobulin E (IgE) Production during the Gross Anatomy Laboratory

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In the gross anatomy laboratory, which is one of the compulsory subjects in most medical and dental schools, participants cannot avoid exposure to formaldehyde (FA), which is emitted from cadavers during dissection. FA has been recognized as a harmful chemical and we have previously reported that symptoms felt by participants in a gross anatomy laboratory are similar to those of allergic diseases. Although immunoglobulin E (IgE)-mediated sensitization to FA is a matter of controversy, it is possible that IgE production is evoked during a gross anatomy laboratory and is responsible for the reported symptoms. In order to test this hypothesis, we examined the relationships between the personal FA exposure levels and plasma IgE levels in a gross anatomy laboratory. In the laboratory, the personal FA exposure levels ranged from 0.33 to 1.47 ppm. Total blood IgE levels did not increase significantly and specific IgE to FA was negative during the laboratory sessions. Thus, from this study, we cannot support the hypothesis that the exposure to FA triggers an IgE-mediated reaction in this study. In conclusion, exposure to FA does not induce IgE production during gross anatomy laboratories at our school.

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INTRODUCTION

Recently, people have been concerned about the effect of formaldehyde (FA) on the human body. The International Agency for Research on Cancer (IARC) has reported that exposure to FA "would probably cause cancer in human beings."[1] FA is also classified by the Japan Society for Occupational Health as "a substance that is considered to cause cancer in human beings and has evidence of causing cancer."[2] Also, FA is a potent contact sensitizer and can elicit contact dermatitis and respiratory symptoms, probably by irritant mechanisms.[3] Therefore, FA has been recognized as a harmful chemical.

Gross anatomy laboratory is one of the compulsory subjects in most medical and dental schools for the study of the normal structure of the human body. Cadavers for the gross anatomy laboratory are generally injected with embalming fluid which contains FA as a principal component. Since it requires long periods in the gross anatomy laboratory to learn human body structures in detail, it is necessary to fix and preserve the cadavers. Antisepsis using FA is used for this purpose because no better method exists at present. The gross anatomy laboratory is so important that medical and dental students cannot proceed to the next step without this practice, and consequently they cannot avoid exposure to the FA, which is emitted by cadavers. To reduce exposure to FA, the Japanese Ministry of Education, Culture, Sports, Science and Technology has released "The improvement plan for the dissection course in medical and dental schools"[4] requiring a reduction in FA concentrations in gross anatomy laboratories.

In the gross anatomy laboratory at Chiba University in 2003, average indoor FA concentrations during the 4th, 10th and 18th laboratory sessions were 0.45, 0.38 and 0.68 ppm, respectively, ranging from 0.23 to 1.03 ppm.[5] FA concentration levels in the laboratory always exceeded the guidelines for indoor FA concentration of both WHO (0.08 ppm for the general indoor environment)[6] and the Ministry of Health, Labour and Welfare (0.25 ppm for specific workplaces).[7] Some levels even exceeded the guidelines of the Japan Society for Occupational Health, 0.5 ppm.[2] Further, our previous report[5] showed that, if a person is close to the cadavers during the gross anatomy course, his/her personal ex-

posure levels are 2 to 3-fold higher than the mean indoor FA concentration. These results suggest that medical students are exposed to higher concentrations of FA than the guideline values given above. Under these circumstances, we have previously reported physical symptoms such as eye, throat and/or nasal irritation induced by exposure to FA.[8] These symptoms described by participants in the gross anatomy laboratory were similar to those of allergic diseases.

In allergic individuals, immunoglobulin E (IgE) antibodies trigger allergic responses through allergen-mediated cross-linking on effector cells followed by mediator release.[9] Evidence of specific allergic sensitization to FA has been reported. Wilhelmsson and Holmstrom found that long-term inhalational exposure to FA may sensitize and trigger classical IgE-mediated allergy in atopics.[10] Wantke *et al*.[11] suggested that gaseous FA, besides its irritant action, leads to IgE-mediated sensitization among children. However, Kramps *et al*.[12] stated that exposure to FA rarely evokes the production of specific IgE. Dykewicz *et al*.[13] suggested that clinical IgE-mediated allergy to gaseous FA does not exist, or if it does, is extremely rare. Also, a study by Liden *et al*.[14] did not support the hypothesis that specific IgE antibodies are active in the pathogenesis of contact sensitivity to FA either in atopic or nonatopic patients. Thus IgE-mediated sensitization to FA is controversial, as mentioned above. Furthermore, little is known about whether IgE-mediated sensitization relates to the level of FA exposure such as that found in the gross anatomy laboratory.

Since participants in the gross anatomy laboratory are exposed to higher concentrations of FA than the general population,[5] and the physical symptoms are similar to allergic symptoms induced by allergens,[8] it is possible that IgE production is evoked during the gross anatomy laboratory. In order to test this hypothesis, we examined the relationships between the personal FA exposure levels and plasma IgE levels in a gross anatomy laboratory course.

MATERIALS AND METHODS

Site of this Study —— This study was conducted in a dissection room at Chiba University. The room dimensions were $15 \times 25 \times 3$ m and thus the room volume was 1125 m^3. During the study there were 48 cadavers under dissection, each of which was on a separate table. The dissecting tables were spread evenly throughout the dissection room.

Twelve supply diffusers, 4 air conditioners and 8 air return grills provided general ventilation to the laboratory. The diffusers were arranged in three rows on the ceiling of the room. The air conditioners were in the corners and the air returns were at floor level. This ventilation system supplied a total airflow of 60 m^3/min. There was no local ventilation. The room temperature was set at 24°C by the air conditioners.

The gross anatomy laboratory was evaluated in the 1th and 10th sessions of 20 laboratory sessions in total over a period of 10 weeks.

FA Sources —— In the gross anatomy laboratory, FA is emitted from cadavers that have been prepared with a FA solution. The solution is a mixture containing 31.1% 'masked-form 2A' (Japan Tanner Corporation, Osaka, Japan), 38.8% ethanol and 13.8% glycerin in water on a volume basis. The masked-form 2A contains 7.4% FA, and thus the final concentration of FA is 2.3%. When not in use, each cadaver was wrapped in cloth and enclosed in a vinyl bag equipped with a fastener.

Air Sampling for FA Exposure Levels —— 8 subjects who participated in the gross anatomy laboratory course from April 21 to June 16 2003 were investigated. Air sampling and analysis were carried out according to the methods of Uchiyama and Hasegawa,[15] the U.S. Environmental Protection Agency (EPA; TO-11A),[16] and the National Institute for Occupational Safety and Health (NIOSH; NIOSH 2016),[17] with minor modifications. Air samples were collected at the 10th session using a diffusive sampling device containing a 2,4-dinitrophenylhydrazine cartridge (DSD-DNPH; Sigma-Aldrich, MO, U.S.A.). This sampling device was pinned on each person's lapel during the laboratory session, which was 1.1 to 6 hr in duration, with an average of 3 hr. Within three days of collection, DNPH derivatives were extracted by acetonitrile from the cartridges. The DNPH derivatives were then measured by high performance liquid chromatography (HPLC).

Blood Sampling —— Blood sampling and analysis were conducted in accordance with the recommendations outlined in the Declaration of Helsinki and performed in accordance with bioethical guidelines established at Chiba University. Blood sampling was performed to determine whether the presence of IgE was associated with exposure to FA. Ten ml of blood was collected from each subject at the following time; 90 min before the first laboratory session (April 21), 90 min before and after starting

Table 1. Characteristics of Subjects, Total Blood IgE Levels and Formaldehyde Exposure Levels

Subject number	Sex	Age	Total blood IgE (IU/ml)				Formaldehyde exposure level (May 14) (ppm)
			90 min before the 1st session (April 21)	90 min before starting of the 10th session (May 14)	90 min after starting of the 10th session (May 14)	22–23days after the last session (July 9,10)	
1	Male	43	73	59	59	53	0.52
2	Male	44	19	24	23	22	0.43
3	Female	39	286	259	257	252	1.47
4	Male	38	296	273	262	297	0.68
5	Male	37	5620	5840	5650	5810	0.33
6	Female	28	< 19	< 19	< 19	< 19	0.98
7	Male	25	195	183	178	200	1.17
8[a)]	Male	28	57	54	51	48	0.16
9	Male	46	Not measured	Not measured	Not measured	< 19	Not measured
10	Female	unknown	Not measured	Not measured	Not measured	39	Not measured
11	Female	29	Not measured	Not measured	Not measured	235	Not measured

a) This subject was in the dissection room but did not participate in the laboratory at the 10th session.

of 10th session (May 14), and 22 or 23 days after the last session (July 9 or 10). Age and sex of the subjects are shown in Table 1. In addition to the 8 subjects, we collected blood samples from 3 subjects who had never attended laboratory sessions as controls at the last sampling (22 or 23 days after the last session). Total IgE in the collected samples was measured by latex agglutination nephelometric immunoassay at the Division of Laboratory Medicine, Chiba University Hospital.

Further, we selected 6 samples (subjects 1–6) and measured specific IgE to FA by fluorometric enzyme immunocapture assay (FEIA) at SRL Inc. (Tokyo, Japan). These samples were selected because the four staff members (subjects 1, 2, 4 and 5) had been exposed to FA for a longer time than other subjects for occupational reasons and the other two (subjects 3 and 6) had complained of sensitivities to chemicals before sessions.

RESULTS

FA Exposure Levels

At the 10th session (May 14), minimum and maximum FA exposure levels among subjects who participated were 0.33 and 1.47 ppm, respectively (Table 1, Fig. 1). Thus, FA exposure levels of all subjects who participated in the session exceeded not only the guideline limit of 0.25 ppm as an average for specific workplaces in Japan,[7)] but also the ACGIH ceiling limit of 0.3 ppm.[18)] The exposure level of subject 8, who was in the dissection room but did not participate in the laboratory, was lower than the others.

Blood IgE Levels

Total blood IgE levels of the 8 subjects did not increase significantly during the laboratory course (Table 1, Fig. 1). Also, we could not find a clear relationship between FA exposure levels and total blood IgE levels (Table 1, Fig. 1). In the 6 samples collected from subjects1, 2, 3, 4, 5, and 6, specific IgE to FA was negative (under 0.34 UA/ml).

The total IgE levels of control subjects 9, 10, and 11 were under 19, 39, and 235 IU/ml, respectively (Table 1).

DISCUSSION

FA exposure levels of all subjects that participated in the laboratory sessions exceeded not only the guideline of 0.25 ppm for specific workplaces in Japan,[7)] but also the ACGIH ceiling limit of 0.3 ppm.[18)] For people with hypersensitive respiratory systems, the WHO is considering a value of 0.008 ppm for a 30-min exposure period.[19)] This should be considered in the risk assessment of FA in the gross anatomy laboratory.

In a previous study,[8)] it was found that some students who did not have symptoms before the labo-

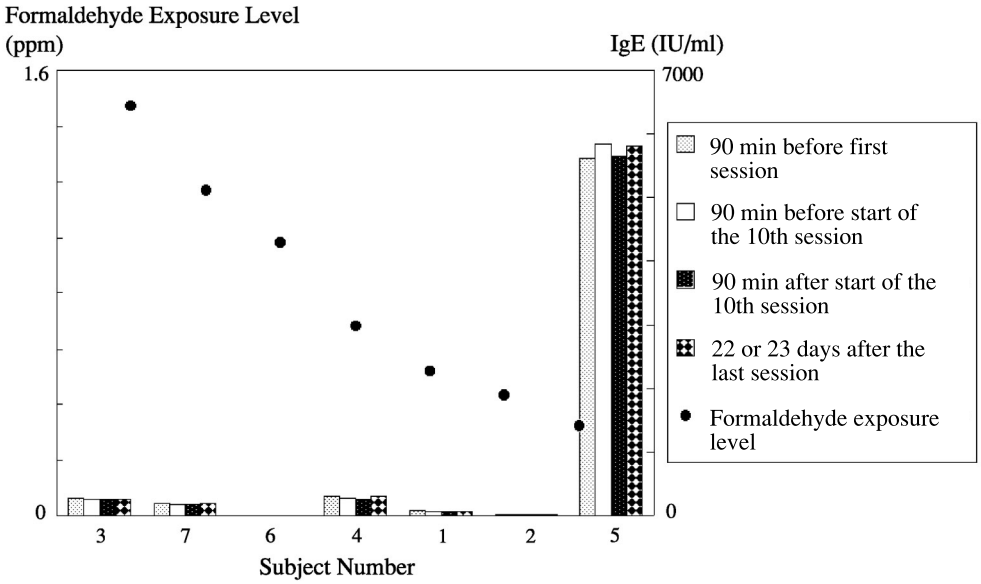

Fig. 1. Relationship between Total Blood IgE and Formaldehyde Exposure Levels

The bar graph shows total blood IgE levels. The solid circles show formaldehyde exposure levels at the 10th laboratory session, the topic of which was dissection of the anterior aspect of the abdomen.

ratory sessions began to experience symptoms afterwards, probably due to FA exposure. Therefore, changes in total blood IgE and specific IgE to FA were expected. Contrary to this assumption, no significant changes in IgE levels were observed during laboratory course. In our laboratory, FA exposure levels ranged from 0.33 to 1.47 ppm among participants (Table 1, Fig. 1). These results show that IgE-mediated allergic reactions due to sensitization to FA did not occur in our gross anatomy laboratory.

From an occupational health point of view, specific IgE to FA has rarely been found in workers[20] and pathologists.[21] Also in the present study, specific IgE to FA was negative (under 0.34 UA/ml) in all the subjects examined. Even comparing the total IgE levels, those of the 8 subjects who participated in the laboratory showed no significant difference from controls who had not attended laboratory sessions. Thus in this study, we can give no support to the hypothesis that exposure to FA triggers an IgE-mediated reaction.

On the other hand, Garrett *et al*. reported that among children suffering from respiratory symptoms, more frequent symptoms occurred in those exposed to higher FA levels.[22] Another report showed that, in children, asthma was related to higher concentrations of FA.[23] Thus, it is possible that an increase in allergic disease among children could be brought about by FA exposure. It was reported that the specific IgE to FA of school children (average age, 8) in a classroom considered to have a high indoor FA concentration (0.075 ppm) decreased significantly when they moved to a classroom with a 0.029 ppm indoor FA concentration.[11] Also, Wantke *et al*. have reported that FA exposure (indoor FA range 0.11–0.33 ppm) during dissections over a 43-day period may induce specific IgE against FA-albumin.[24] The indoor FA level reported by Wantke *et al*.[24] was lower than our laboratory level, which ranged from 0.23 to 1.03 ppm[5] during the course of dissection. However, dissections were conducted over a period of 43 days in the Wantke *et al*. study,[24] which was twice as long as our dissection sessions that covered a period of 20 days. This indicates long-term exposure to FA may induce IgE production.

Although the FA level in our gross anatomy laboratory did not induce IgE production in this study, FA could lead to IgE-mediated sensitization and specific IgE antibodies against FA-albumin have been detected.[25] Our results suggest that IgE production due to FA exposure may be rare in our gross anatomy laboratory and the relationship *in vivo* between FA exposure and the development of physical symptoms remains unclear.

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