Table 6 Odds ratios (ORs) of chemical sensitive population (CSP) cases by Hojo criteria compared to controls categorized by genotype

Variable[a]		Control	Case 1 (Low chemical sensitivity)				Case 2 (Middle chemical sensitivity)				Case 3 (High chemical sensitivity)			
		n=208	n=67				n=38				n=11			
			OR (95% CI) Crude	P[c]	OR[b] (95% CI) Adjusted	P[c]	OR (95% CI) Crude	P[c]	OR[b] (95% CI) Adjusted	P[c]	OR (95% CI) Crude	P[c]	OR[b] (95% CI) Adjusted	P[c]
NAT2														
Genotype	Slow + Inter vs. Rapid	1[c]	0.66 (0.38–1.14)	0.14	0.68 (0.38–1.19)	0.17	0.69 (0.34–1.37)	0.29	0.68 (0.34–1.36)	0.27	0.44 (0.12–1.54)	0.20	0.48 (0.13–1.70)	0.25
	Slow vs. Inter vs. Rapid	1[c]	0.70 (0.43-1.12)	0.13	0.70 (0.43-1.14)	0.15	0.68 (0.37-1.24)	0.21	0.66 (0.36-1.22)	0.18	0.43 (0.13-1.38)	0.16	0.47 (0.14-1.51)	0.20
GSTM1														
Genotype	homozygous-null vs. non-null	1[c]	1.13 (0.65–1.96)	0.67	1.09 (0.62–1.93)	0.76	1.55 (0.77–3.09)	0.22	1.51 (0.75–3.04)	0.25	2.43 (0.69–8.57)	0.17	2.34 (0.66–8.30)	0.19
GSTT1														
Genotype	homozygous-null vs. non-null	1[c]	0.79 (0.45–1.37)	0.40	0.75 (0.42–1.32)	0.32	0.68 (0.34–1.36)	0.27	0.66 (0.33–1.33)	0.24	0.81 (0.24–2.75)	0.81	0.84 (0.25–2.87)	0.78
GSTP1														
Genotype	A/G + G/G vs. A/A	1[c]	1.13 (0.61–2.09)	0.70	1.06 (0.57–1.98)	0.86	1.86 (0.91–3.82)	0.09	1.76 (0.85–3.64)	0.13	2.38 (0.70–8.10)	0.17	2.48 (0.72–8.55)	0.15
	G/G vs. A/G vs. A/A	1[c]	1.08 (0.62-1.89)	0.78	1.02 (0.58-1.81)	0.93	1.88 (1.04-3.41)	**0.04**	1.79 (0.98-3.28)	0.06	1.85 (0.64-5.31)	0.26	1.95 (0.66-5.75)	0.23
CYP2E1														
Genotype	C1/C2 + C2/C2 vs. C1/C1	1[c]	0.92 (0.53–1.61)	0.78	0.96 (0.54–1.71)	0.90	0.52 (0.25–1.11)	0.09	0.50 (0.24–1.08)	0.08	0.48 (0.12–1.87)	0.29	0.52 (0.13–2.06)	0.35
	C2/C2 vs. C1/C2 vs. C1/C1	1[c]	0.89 (0.58-1.38)	0.61	0.94 (0.59-1.47)	0.78	0.67 (0.37-1.20)	0.18	0.65 (0.35-1.18)	0.16	0.47 (0.14-1.57)	0.22	0.50 (0.15-1.67)	0.26
ALDH2														
Genotype	*1/*2 + *2/*2 vs. *1/*1	1[c]	1.08 (0.62–1.89)	0.78	1.02 (0.55–1.89)	0.95	0.78 (0.38–1.62)	0.51	0.61 (0.27–1.36)	0.22	0.34 (0.07–1.59)	0.17	0.26 (0.05–1.40)	0.12
	*2/*2 vs. *1/*2 vs. *1/*1	1[c]	1.00 (0.61-1.65)	1.00	0.95 (0.54-1.67)	0.85	0.97 (0.53-1.77)	0.91	0.78 (0.40-1.53)	0.47	0.34 (0.08-1.53)	0.16	0.27 (0.05-1.35)	0.11
SOD2														
Genotype	Val/Ala + Ala/Ala vs. Val/Val	1[c]	0.94 (0.49–1.81)	0.84	0.89 (0.46–1.75)	0.74	1.16 (0.53–2.55)	0.71	1.09 (0.49–2.42)	0.84	3.90 (1.14–13.31)	**0.03**	4.30 (1.23–15.03)	**0.02**
	Ala/Ala vs. Val/Ala vs. Val/Val	1[c]	1.11 (0.63-1.96)	0.72	1.03 (0.58-1.85)	0.91	1.22 (0.60-2.50)	0.59	1.11 (0.54-2.30)	0.78	3.67 (1.32-10.20)	**0.01**	4.53 (1.52-13.51)	**0.01**

a. OR, odds ratio; CI, confidence interval

b. Odds Ratios were adjusted by age, smoking, and drinking

c. p value <0.05 is considered statistically significant

化学物質への過敏反応に関する質問票

・これらの質問票は、化学物質で過敏反応を示す方々の環境要因を調査、整理する目的でおこなわれるものです。

・この質問票の結果は、化学物質に苦しむ患者さんの診断・治療に役立つのみでなく、国際比較にも使われ、治療法の進歩に役立ちます。ぜひ、空欄を残すことなく、お答えください。

・なお、各個人の秘密は厳守されます.

よろしくお願いいたします。

調査票

職　業

1	2	3	4	5	6	7	8
農林漁業（家族従業を含む）	商工・サービス業（家族従業を含む）	事務職	労務職	自由業 管理職	無職の 主　婦	学生	その他 無　職

性　別

1	2
男	女

年　齢

□□ 歳

氏　名

Q1. ここ１年間についてお聞きします。あなたは、(A)~(J)にあげたものに反応して、頭痛、胃の不調、呼吸が苦しくなる、体がふらふらする、ものが考えられなくなるなどの症状を感じたことがありますか。それぞれについて、その程度を 0~10 の数字でお答えください。

全く何ともない　―　中程度の症状　―　動けなくなったり寝込むほどの症状

回答例: 0

（A）車の排気ガス ---------- □□

（B）タバコの煙 ---------- □□

（C）殺虫剤・除草剤 ---------- □□

（D）ガソリン ---------- □□

（E）ペンキ・シンナー ---------- □□

（F）洗剤類（消毒剤・漂白剤・風呂用洗剤・床用洗剤）---- □□

（G）香水、芳香剤、清涼剤 ---------- □□

（H）コールタール、アスファルト ---------- □□

（I）マニキュア・マニキュアの除光液･整髪剤･オーデコロン ---------- □□

（J）新しいじゅうたんや新しいカーテンなどの新しい家具、または新車とその内装など---- □□

Q２．あなたは、この１年くらいの間に、次にあげた（A）～（J）のようなことを経験されたことがありますか。それぞれについて、その程度を０～10の数字でお答えください。

全く何ともない　　中程度の症状　　動けなくなったり寝込むほどの症状

回答例: 0

(A)水道のカルキ臭などで体調が悪くなる(シャワー、お風呂、お湯の使用時など) --------

(B)特定の食品を食べると体調が悪くなる ----------

(C)ある食品が異常なほど食べたくなったり、または食べてしまったりする。
あるいはその食品がないと体調不良になる ----------

(D)食後、一定時間体調が悪い ----------

(E)コーヒー、紅茶、日本茶、コーラ、チョコレートなどを食べると体調が悪くなる ---

(F)コーヒー、紅茶、日本茶、コーラ、チョコレートなどを食べないと体調が悪くなる ----

(G)ハンバーガー、カップラーメンなどを食べると体調が悪くなる ----------

(H)ハンバーガー、カップラーメンなどを食べないと体調が悪くなる ----------

(Ｉ)少量のビールやワインのような少量のアルコールでも体調が悪くなる ----------

(Ｊ)皮膚に触れる布製品、金属の装飾品、化粧品などで体調が悪くなる ----------

(K)医薬品、インプラント（人口品の体への埋め込み)、入れ歯、避妊器具などで
体調が悪くなる ----------

(L)樹木、草、花粉、家の塵（ちり)、カビ、動物のあか、虫ささされ、特定の食物などで
ぜん息、鼻炎、じんましん、湿しんのようなアレルギー反応が起きる ----------

Q３．あなたは、この１年くらいの間に、次にあげた（A）～（J）のような症状を経験されたことがありますか。それぞれについて、その程度を０～10の数字でお答えください。

全く何ともない　　中程度の症状　　動けなくなったり寝込むほどの症状

回答例: 0

（A）　筋肉、関節の痛み、けいれん、こわばり、力が抜ける ----------

（B）　眼の刺激、やける感じ、しみる感じ。息切れ、咳のような気管や呼吸症状。たん、鼻汁がのどの奥の方に流れる感じ。風邪にかかりやすい ----------

（C）　どうき、脈のみだれ、胸の不安感などの心臓や胸の症状 ----------

（D）　腹痛、胃けいれん、膨満感、吐き気、下痢、便秘のような消化器症状 ----------

（E）　集中力、記憶力、決断力の低下、無気力などを含む思考力の低下 ----------

（F）　緊張しすぎる、上がりやすい、刺激されやすい、うつ、泣きたくなったり激情的になったりする。以前興味があったものに興味が持てないなどの気分の変調 --------

（G）　めまい、立ちくらみなど平衡感覚の不調、手足の動きがぎこちない、手足のしびれ、手足のチクチク感、目のピントが合わない ----------

（H）　頭痛、頭の圧迫感、一杯にに詰まった感じなどの頭部症状 ----------

（I）　発疹、じんま疹、アトピー、皮膚の乾燥感 ----------

（J）　トイレが近い、排尿困難、尿失禁、外陰部のかゆみまたは痛みなどの泌尿器・生殖器症状（女性の場合：生理時の不快感、苦痛などの症状） ----------

---

Q４．あなたは医療機関で次のような診断をされたことがありますか。あてはまるものすべてに丸を付けてください

1　化学物質過敏症
2　シックハウス症候群
3　気管・呼吸器、皮膚、目、鼻、のど等のアレルギー性疾患
4　どれもない

Q５．お宅では、最近10年以内に、次のようなことがありましたか。あてはまるものすべてに丸を付けてください。

1　家の新築、またはリフォーム（外壁工事、ペンキの塗り替えなども含む）
2　新しい家具、カーペット、カーテンなどの購入
3　新車の購入
4　引越経験（１～２回）
5　引越経験（３～４回）
6　引越経験（５回以上）
7　どれもない

Q６．あなたの生活習慣についておたずねします。次の(A)~(J)の質問を読んで、「はい」または「いいえ」のどちらかに丸を付けてお答えください

(A)週に１回以上タバコを吸ったりしますか　はい　・　いいえ

(B)アルコール飲料（ビール、焼酎、日本酒など）を週１回以上飲みますか　はい　・　いいえ

(C)カフェインの入った飲み物（コーヒー、紅茶など）を週１回以上飲みますか　はい　・　いいえ

(D)香水、ヘアスプレー、香料入りの化粧品を週１回以上使用しますか　はい　・　いいえ

(E)過去数年内に殺虫剤、防カビ剤処理を家や職場で使用しましたか　はい　・　いいえ

(F)最近の仕事や趣味の中で、化学物質や煙、刺激臭にさらされることが週１回以上ありましたか　はい　・　いいえ

(G)あなたの周りに、タバコを吸う家族や同居人はいますか　はい　・　いいえ

(H)ガスストーブや石油ストーブを家庭で使用していますか　はい　・　いいえ

(I)洗濯するとき、繊維を柔らかくする製品（柔軟剤）をよく使いますか　はい　・　いいえ

(J)ステロイド剤、鎮痛剤、抗うつ剤、精神安定剤、睡眠剤などをよく使いますか　はい　・　いいえ

Q７．職場で、最近５年以内に次のようなことがありましたか。あてはまるものすべてに丸を付けてください。

1　事務所（職場）の移転またはリフォーム（外装工事、ペンキ塗り替えなども含む）
2　新しいロッカー、家具、カーペット、カーテンなどの設置、装着
3　新たな薬品の使用
4　新しいトラック、社用車などの使用
5　どれもない

質問はこれで終わりです
ご協力ありがとうございました

III. 研究成果の刊行に関する一覧表

III. 研究成果の刊行に関する一覧表

書籍

著者氏名	論文タイトル名	書籍全体の編集者名	書籍名	出版社名	出版地	出版年	ページ
Kunugita N, Arashidani K, Katoh T.	Investigation of air pollution in large public buildings in Japan and of employees' personal exposure levels.	Sabah A. Abdul-Wahab Al-Sulaiman	Sick Building Syndrome, in Public Buildings and Workplaces	Springer-Verlag	Berlin	2011	269-287

雑誌

発表者氏名	論文タイトル名	発表誌名	巻号	ページ	出版年
Cui X, Lu X, Hiura M, Miyazaki W, Oda M, Katoh T.	Prevalence and interannual changes in multiple chemical sensitivity in Japanese workers.	Environ Health Prev Med.		DOI 10.1007/s12199-014-0378-	2014
Win-Shwe TT, Fujimaki H, Arashidani K, Kunugita N.	Indoor volatile organic compounds and chemical sensitivity reactions.	Clinical and Developmental Immunology	2013	e623812	2013
Uchiyama, S., Tomizawa, T., Inaba, Y., Kunugita, N.	Simultaneous determination of volatile organic compounds and carbonyls in mainstream cigarette smoke using a sorbent cartridge followed by two-step elution.	Journal of Chromatography A	314	31-37	2013
Cui X, Lu X, Hiura M, Miyazaki W, Oda M, Katoh T.	Evaluation of genetic polymorphisms in patients with multiple chemical sensitivity.	Plos One	8	e73708	2013
Uchiyama, S.; Ohta, K.; Inaba, Y.; Kunugita.	Determination of carbonyl compounds generated from the E-cigarette using coupled silica cartridges impregnated with hydroquinone and 2,4-dinitrophenylhydrazine followed by high performance liquid chromatography.	Analytical Sciences	29	1219-1222	2013
Matsumoto, M. Inaba, Y. Yamaguchi, I. Endo, O. Hammond, D. Uchiyama, S. Suzuki, G.	Smoking topography and biomarkers of exposure among Japanese smokers: associations with cigarette emissions obtained using machine smoking protocols.	Environ Health Prev Med.	18	95-103	2013
山田智美; 内山茂久; 稲葉洋平; 瀬戸博; 欅田尚樹	空気中化学物質測定用拡散サンプラーの安定性評価および実試料測定への応用	分析化学	62(7)	603-609	2013

Azuma, K., Uchiyama, I., Okumura, J.	Assessing the risk of Legionnaires' disease: the inhalation exposure model and the estimated risk in residential bathrooms.	Regulatory Toxicology and Pharmacology.	65(1)	1-6	2013
稲葉洋平, 内山茂久	喫煙と室内環境	空衛	66(3)	56-63	2012
Uchiyama, S.; Sakamoto, H.; Ohno, A.; Inaba, Y.; Nakagome, H.; Kunugita, N.	Reductive amination of glutaraldehyde 2,4-dinitrophenylhydrazone using 2-picoline borane and high-performance liquid chromatographic analysis.	Analyst	137	4247-4279	2012
Uchiyama, S.; Inaba, Y.; Kunugita, N.	Ozone removal in the collection of carbonyl compounds in air.	Journal of Chromatography A	1229	293-297	2012
Yamada, T.; Uchiyama, S.; Inaba, Y; Kunugita, N.; Nakagome, N.; Seto, H.	A diffusive sampling device for measurement of ammonia in air.	Atmospheric Environment	54	629-633	2012
Fujimori S, Hiura M, Cui XY, Lu X, Katoh T.	Factors in genetic susceptibility in a chemical sensitive population using QEESI.	Environ Health Prev Med.	17	357-363	2012
Uchiyama, S.; Inaba, Y.; Kunugita, N	A diffusive sampling device for simultaneous determination of ozone and carbonyls.	Analytica Chimica Act	691	119-124	2011
Uchiyama, S., Inaba, Y., Kunugita, N.	Derivatization of carbonyl compounds with 2,4-dinitrophenylhydrazine and their subsequent determination by high-performance liquid chromatography.	Journal of Chromatography B	879	1282-1289	2011
太田和司; 内山茂久; 稲葉洋平; 中込秀樹; 欅田尚樹	ハイドロキノンと2,4-ジニトロフェニルヒドラジンを含浸させた二連シリカカートリッジを用いる電子タバコから発生するカルボニル化合物の分析	分析化学	60(10)	791-797	2011
大澤元毅	健康建築の構図	空衛	65(6)	34-38	2011

IV. 研究成果の刊行物・別刷

Environ Health Prev Med
DOI 10.1007/s12199-014-0378-6

REGULAR ARTICLE

Prevalence and interannual changes in multiple chemical sensitivity in Japanese workers

Xiaoyi Cui · Xi Lu · Mizue Hiura · Masako Oda · Aya Hisada · Wataru Miyazaki · Hisamitsu Omori · Takahiko Katoh

Received: 26 December 2013 / Accepted: 14 January 2014

Abstract
Objective We aimed to evaluate the prevalence rates and interannual fluctuations in multiple chemical sensitivity (MCS) in Japanese workers.
Methods We assessed MCS using the Quick Environmental Exposure and Sensitivity Inventory, employing both Miller and Japanese criteria. Workers of two manufacturing companies located in Kyushu, Japan, were assessed, with company A surveyed in 2003, 2006 and 2011, and company B in 2003 and 2011.
Results In company A, the Miller criteria-based MCS prevalence rate was higher in 2011 than in 2003, and according to the Japanese criteria, it was higher in 2011 than 2006. In company B, the Miller criteria-based MCS prevalence rate was lower in 2011 than in 2003.
Conclusion The results indicated that MCS exists among industrial workers in Japan. We found no statistically significant interannual changes in MCS rates.

Keywords Multiple chemical sensitivity · Environmental exposure · QEESI · Sick house syndrome · Allergy · Japanese workers

X. Cui (✉) · X. Lu · M. Hiura · M. Oda · A. Hisada · W. Miyazaki · H. Omori · T. Katoh
Department of Public Health, Faculty of Life Sciences, Kumamoto University, 1-1-1 Honjou, Chuo-ku, Kumamoto 860-8556, Japan
e-mail: bjcxy860508@hotmail.com

H. Omori
Department of Biomedical Laboratory Sciences, Faculty of Life Sciences, Kumamoto University, Kumamoto, Japan

Introduction

Multiple chemical sensitivity (MCS) is an acquired chronic disorder in which exposure to low levels of chemicals causes mild to wholly disabling symptoms [1]. Symptoms are usually vague and nonspecific, involving more than one organ system. In general, the reported symptoms are attributed to previous chemical exposure, and recur on subsequent exposure to chemicals at doses below those known to cause harmful effects in the general population [2]. The etiology of MCS, however, remains unclear. It is difficult to estimate its prevalence because it is derived from self-reports, which differ from case rates diagnosed by medical staff—occupational physicians in particular [1]. MCS patients' clinical characteristics are usually evaluated using questionnaires such as the Environmental Exposure and Sensitivity Inventory (EESI), or clinical interviews that rely on the individual's retrospective self-reports [3]. Miller and Prihoda [4] developed a globally standardized self-administered questionnaire, the Quick Environmental Exposure Sensitivity Inventory (QEESI), designed to assist researchers and clinicians in screening, studying, and evaluating patients with MCS.

People with environmental sensitivities may be susceptible to diverse environmental factors. Some of the more common agents containing chemical compounds that trigger reactions in such people include pesticides and volatile organic compounds (VOCs), such as solvents, perfumes, formaldehyde, and other petrochemicals. These agents may be contained in workplace building structures, furnishings, and cleaning products, among other sources [5]. A definition of work-related MCS was introduced by Cullen [6]. Adverse MCS health effects were observed in workers in subsequent research [7]; the negative effects of chemical hazards are a longstanding part of occupational

Published online: 30 January 2014

health in the industrialized world [8]. While such issues initially tended to arise in industrial workers, similar problems have emerged in residents and workers in tight buildings in air- and water-polluted communities, and in persons exposed to various chemicals [9]. We selected workers employed in two large companies as participants, because this group allowed for easier follow-ups than community residents. The response rate may be higher from the workers who answered the questionnaires as instructed by company leaders. In addition to our aim of evaluating MCS prevalence rates and their interannual change, we assessed the rates of allergies and Sick house syndrome (SHS) in Japanese workers.

Materials and methods

Study characteristics

The present study was carried out at a paper pulp producing company (company A) in 2003, 2006, and 2011, and at an automotive company (company B) in 2003 and 2011, both in Kyushu, Japan. Subjects were asked to complete QEESI questionnaires, which also cover the respondents' diagnostic history of allergies and SHS. After excluding invalid questionnaires, we obtained 832 valid responses in 2003, 729 in 2006, and 144 in 2011 at company A, and 333 responses in 2003 and 426 in 2011 at company B.

Survey instruments

We used the Japanese version of the QEESI questionnaire prepared by Ishikawa and Miyata to assess MCS [10]. Each criterion subscale of the QEESI contains 10 questions rated on a scale from 0 to 10; the total possible score for each subscale, therefore, ranges from 0 to 100.

We used the Miller criteria to define MCS in workers according to the scores yielded by three cut-off subscales: $\geq$40 for chemical sensitivity, $\geq$25 for other chemicals, and $\geq$40 for symptom severity classified as MCS [4]. Hojo et al. [11] designed a study to establish the cut-off value for Japanese criteria using the QEESI as an MCS screening method. We also employed their Japanese criteria of $\geq$40 for chemical sensitivity, $\geq$20 for symptom severity, and $\geq$10 for life impact classified as MCS.

Data analysis

We collected and used anonymous information for data analysis. The distribution differences were examined using Chi square test. The average differences were examined using *t* tests. Statistical analyses were carried out using SPSS version 18 for Windows (SPSS, Japan).

Ethical statement

The ethics review boards of Miyazaki University (no. 82; April 9, 2003) and Kumamoto University (no. 168; May 11, 2011) approved this study, following their ethical guidelines for human research. All participants provided written informed consent to participate, and the complete protection of their personal data was agreed upon in writing.

Results

At the 2003 baseline, SHS diagnostic history rates for companies A and B were 0.1 and 0.3 %, respectively, and the allergy diagnostic history rates for companies A and B were 23.1 and 24.0 %, respectively (Tables 1, 2). The Miller-criteria-based MCS prevalence rate was 1.1 % in company A and 2.4 % in company B in 2003 (Figs. 1, 2).

In company A, SHS diagnostic history rate rose in 2011 (2.1 %) compared to 2003 and 2006 (0.1 %). The allergy diagnostic history rate decreased in 2006 (20.7 %) but rose in 2011 (39.6 %) (Table 1). In company B, the SHS and allergy diagnostic history rates (0.5 and 29.3 %, respectively) increased in 2011 (Table 2). In company A, the Miller-criteria-based MCS prevalence rate rose in 2011 (1.4 %) in comparison to 2003 and 2006 (1.1 %) (Fig. 1). However, in company B, Miller-criteria-based MCS prevalence rates dropped in 2011 (1.6 %) from 2003 (2.4 %) (Fig. 2).

As the life impact subscale was not employed in 2003, company A's QEESI-derived Japanese criteria-based MCS prevalence rate could only be determined between 2006 and 2011 (Fig. 3). The Japanese criteria-based MCS

Table 1 Diagnostic history and characteristics of respondents from company A

	2003	2006	2011
Diagnostic history, % (*n*)			
Sick house syndrome (SHS)	0.1 (1)	0.1 (1)	2.1 (3)
Allergy	23.1 (192)	20.7 (151)	39.6 (57)
None	68.5 (570)	79.2 (577)	58.3 (84)
No answer	8.3 (69)	0 (0)	0 (0)
Total	100 (832)	100 (729)	100 (144)
Sex, % (*n*)			
Female	11.7 (97)	10.0 (73)	16.7 (24)
Male	88.3 (735)	90.0 (656)	83.3 (120)
Total	100 (832)	100 (729)	100 (144)
Average age (years ± SD[a])	42.8 ± 10.34	44.9 ± 10.61	41.5 ± 11.04

[a] Standard deviation

prevalence rate increased in 2011 (4.2 %) from 2006 (3.3 %); these prevalence rates were higher than those derived from the Miller criteria in 2011 (1.4 %) and 2006 (1.1 %) (Fig. 1). Nevertheless, these differences were not statistically significant.

In addition, the mean age of company A employees rose in 2006 (44.9 ± 10.61 years old) and decreased in 2011 (41.5 ± 11.04 years old) compared to baseline (42.8 ± 10.34 years old). The proportion of women in company A in 2011 (16.7 %) grew from 2003 (11.7 %) to 2006 (10.0 %) (Table 1). The mean age of employees in company B increased in 2011 (44.8 ± 9.78 years old) when compared to 2003s baseline (40.5 ± 8.65 years old); the proportion of females also grew from 2003 (0 %) to 2011 (3.1 %) (Table 2).

Table 2 Diagnostic history and characteristics of respondents from company B

	2003	2011
Diagnostic history, % (*n*)		
Sick house syndrome (SHS)	0.3 (1)	0.5 (2)
Allergy	24.0 (80)	29.3 (125)
None	67.9 (226)	66.9 (285)
No answer	7.8 (26)	3.3 (14)
Total	100 (333)	100 (426)
Sex, % (*n*)		
Female	0 (0)	3.1 (13)
Male	100 (333)	96.9 (413)
Total	100 (333)	100 (426)
Average age (years ± SD[a])	40.5 ± 8.65	44.8 ± 9.78

[a] Standard deviation

Discussion

This study investigated both Miller and Japanese criteria-based MCS prevalence rates across several years in Japanese general industrial workers.

Miller and Prihoda American study indicated that the MCS rate was 7.1 % in 1999 [4]. A larger-scale investigation by Kreutzer et al. [12] employed a telephone survey, and found MCS rates of 6.3 %. In line with the present findings, studies by Uchiyama in 2000 and by Hojo in 2002 showed Japan's MCS rate to be lower than that of the USA [13, 14]. In both companies, allergy diagnostic history rates rose in 2011 compared to 2003. Our findings were consistent with the increasing trend of allergy epidemic in Japan [15].

In the work environment, Watanabe et al. [16] identified several at-risk categories of chemicals, especially volatile compounds such as organic solvents. Many compounds used daily in manufacturing processes contained such chemicals, and exposed not only the workers who produced them, but also those in areas such as construction, automotive work, textiles, cleaning, and so on [1]. The impact of environmental sensitivities on workers may range from mild to severe, even making work impossible in some cases [5]. However, as this study's subjects were workers from

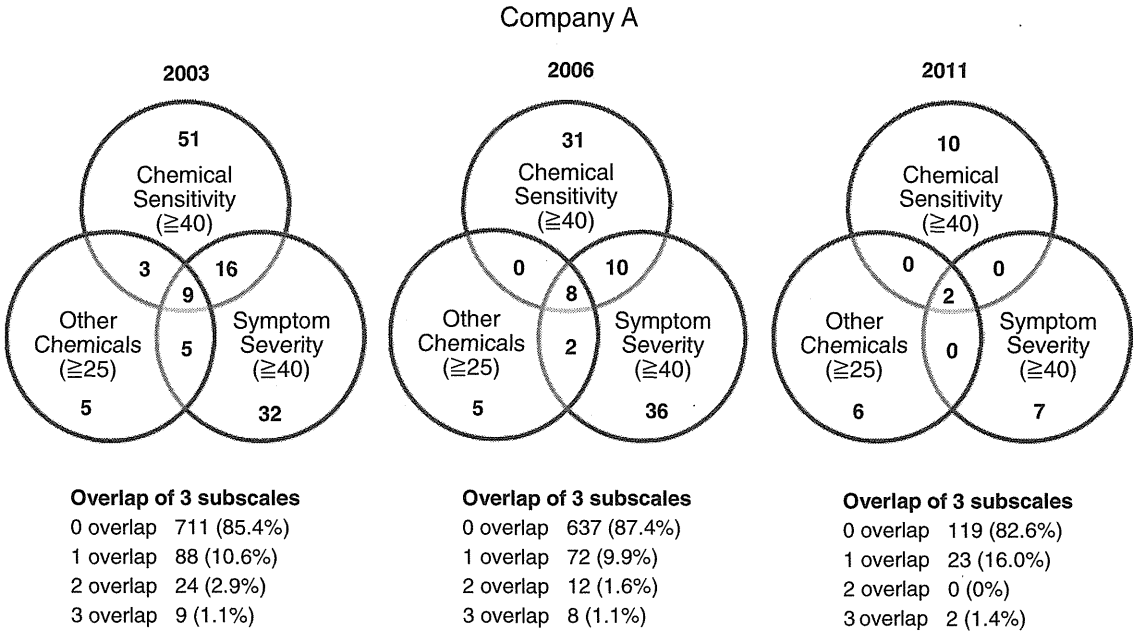
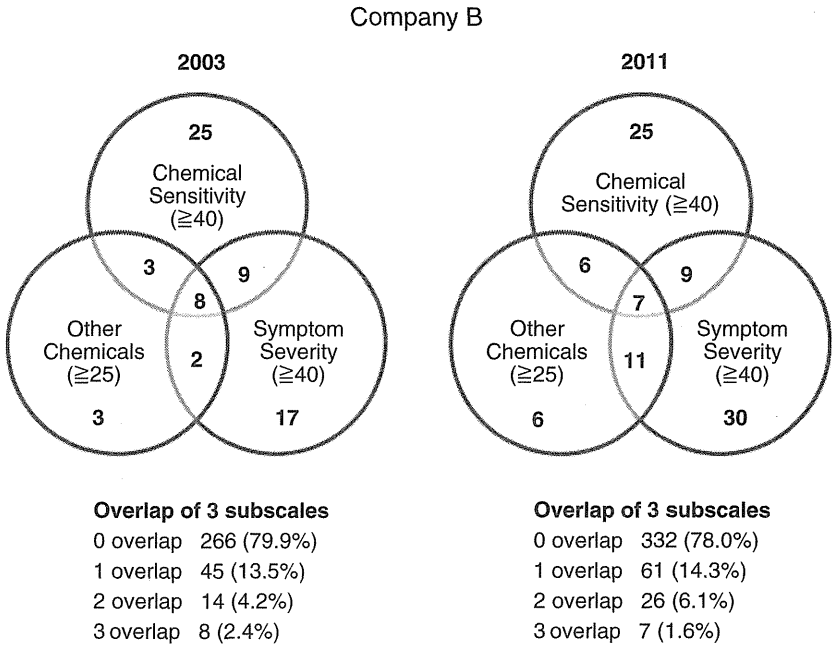


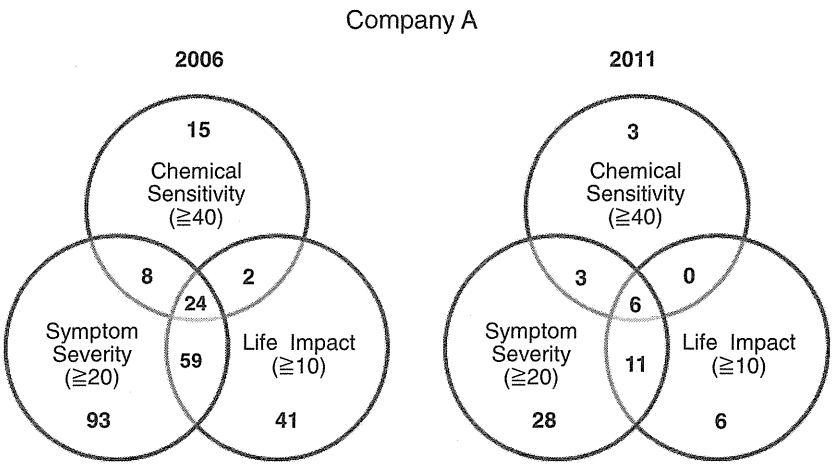
Fig. 1 Interannual changes in company A employee MCS rates, diagnosed through the QEESI using the Miller criteria (3 overlap was considered multiple chemical sensitivity)



Overlap of 3 subscales

	2003	2011
0 overlap	266 (79.9%)	332 (78.0%)
1 overlap	45 (13.5%)	61 (14.3%)
2 overlap	14 (4.2%)	26 (6.1%)
3 overlap	8 (2.4%)	7 (1.6%)

Fig. 2 Interannual changes in company B employee MCS rates, diagnosed through the QEESI using the Miller criteria (3 overlap was considered multiple chemical sensitivity)



Overlap of 3 subscales

	2006	2011
0 overlap	487 (66.8%)	87 (60.4%)
1 overlap	149 (20.4%)	37 (25.7%)
2 overlap	69 (9.5%)	14 (9.7%)
3 overlap	24 (3.3%)	6 (4.2%)

Fig. 3 Interannual changes in company A employee MCS rates, diagnosed through the QEESI using the Japanese criteria (3 overlap was considered multiple chemical sensitivity)

two large companies, it is important to note the selection bias known as the healthy worker effect (HWE), the process wherein unhealthy individuals are excluded from the workplace, as healthy workers are more likely to continue to work than those who are sick [17]. Some studies have suggested that incomplete follow-ups with workers who leave employment and migrate away from their workplace could be a source of the HWE [18, 19]. Terr comments that some MCS patients change their jobs because of their symptoms [20], Lax and Henneberger later reported a similar conclusion [21]. A 2-year follow-up of 50 subjects with MCS showed that most were unchanged or worse by their final assessment [22]. Follow-up studies show that people with MCS frequently suffer symptoms for many years, but may show gradual improvement over time [16]. As the present study anonymized participants, it was impossible to confirm how many workers were continuously checked across several years. We also have found that it was impossible to follow-up with subjects who ceased working due to MCS. However, many patients were

overlap in different years, which may indicate that they did not leave the workplace and were not recovered from MCS symptoms. The HWE may influence the MCS prevalence rate found in this study; however, this bias may not have had a significant impact on the results.

In occupational settings, exposures are often chronic. This suggests that controlling chemical exposure in the early phases of MCS may prevent more serious developments. Even for healthy people, numerous aggravating exposures may be below legal limits but not within adequate safety margins to prevent symptoms [23]. Improving environmental quality in the workplace can promote worker's health and productivity. Such workplace accommodation may include behavior changes, including the use of the least toxic cleaning products and pest control practices, and avoidance of scented products [5]. Furthermore, comparing estimated onset factors between male and female patients revealed that workplace chemical exposure was markedly higher in males [24].

Acknowledgments This work was supported in part by Grants-in-Aid for Research on Health Security Control (H24-Kenki-jpan) from the Ministry of Health, Labor and Welfare of Japan (http://www.mhlw.go.jp/). We express our sincere appreciation to the staff members of the Department of Public Health, Faculty of Life Sciences, Kumamoto University.

Conflict of interest No conflict of interest.

References

1. Martini A, Iavicoli S, Corso L. Multiple chemical sensitivity and the workplace: current position and need for an occupational health surveillance protocol. Oxid Med Cell Longev. 2013; 351457.
2. Graveling RA, Pilkington A, George JP, Butler MP, Tannahill SN. A review of multiple chemical sensitivity. Occup Environ Med. 1999;56:73–85.
3. Saito M, Kumano H, Yoshiuchi K, Kokubo N, Ohashi K, Yamamoto Y, et al. Symptom profile of multiple chemical sensitivity in actual life. Psychosom Med. 2005;67:318–25.
4. Miller CS, Prihoda TJ. The Environmental Exposure and Sensitivity Inventory (EESI): a standardized approach for measuring chemical intolerances for research and clinical applications. Toxicol Ind Health. 1999;15:370–85.
5. Sears ME. The medical perspective on environmental sensitivities. Can Hum Rights Commission. 2007;16–60.
6. Cullen MR. The worker with multiple chemical sensitivities: an overview. Occup Med. 1987;2:655–61.
7. Gibson PR. MCS: a survival guide. New Harbinger: Oakland; 2000.
8. Moen BE. Chemical sensitivity and the work place environment: research needs. Psychoneuroendocrinology. 2005;30:1039–42.
9. Ashford NA, Miller CS. Chemical exposures: low levels and high stakes. New York: Van Nostrand Reinhold; 1997.
10. Ishikawa S, Miyata M. Multiple chemical sensitivity-criteria and test methods for diagnosis. Allergol Immunol. 1999;6:990–8.
11. Hojo S, Sakabe K, Ishikawa S, Miyata M, Kumano H. Evaluation of subjective symptoms of Japanese patients with multiple chemical sensitivity using QEESI(c). Environ Health Prev Med. 2009;14:267–75.
12. Kreutzer R, Neutra RR, Lashuay N. Prevalence of people reporting sensitivities to chemicals in a population-based survey. Am J Epidemiol. 1999;150:1–12.
13. Uchiyama I, Murayama R. Multiple chemical sensitivity as seen from the public health. In: Heisei 11 Welfare grant-in-aid for scientific research report. 2000. pp. 1–5 (in Japanese).
14. Hojyo S. Epidemiological study using QEESI in Japan. In: Heisei 13 Welfare grant-in-aid for scientific research report. 2002. pp. 134–152 (in Japanese).
15. Akasawa A. The development factors and medical system evaluation based on epidemiological study about national age prevalence of allergic diseases and treatment guidelines spread effect. In: Heisei 22 Labor and welfare grant-in-aid for scientific research report. 2011. pp. 245–250 (in Japanese).
16. Watanabe M, Tonori H, Aizawa Y. Multiple chemical sensitivity and idiopathic environmental intolerance (part two). Environ Health Prev Med. 2003;7:273–82.
17. Li CY, Sung FC. A review of the healthy worker effect in occupational epidemiology. Occup Med (Lond). 1999;49:225–9.
18. Monson RR. Observations on the healthy worker effect. J Occup Med. 1986;28:425–33.
19. Bell CM, Coleman DA. Models of the healthy worker effect in industrial cohorts. Stat Med. 1987;6:901–9.
20. Terr AI. Clinical ecology in the workplace. J Occup Med. 1989;31:257–61.
21. Lax MB, Henneberger PK. Patients with multiple chemical sensitivities in an occupational health clinic: presentation and follow-up. Arch Environ Health. 1995;50:425–31.
22. Terr AI. Environmental illness: a clinical review of 50 cases. Arch Intern Med. 1986;146:145–9.
23. Ziem G, McTamney J. Profile of patients with chemical injury and sensitivity. Environ Health Perspect. 1997;105:417–36.
24. Hojo S, Ishikawa S, Kumano H, Miyata M, Sakabe K. Clinical characteristics of physician-diagnosed patients with multiple chemical sensitivity in Japan. Int J Hyg Environ Health. 2008;211:682–9.

Hindawi Publishing Corporation
Clinical and Developmental Immunology
Volume 2013, Article ID 623812, 8 pages
http://dx.doi.org/10.1155/2013/623812

Review Article

Indoor Volatile Organic Compounds and Chemical Sensitivity Reactions

Tin-Tin Win-Shwe,[1] Hidekazu Fujimaki,[2] Keiichi Arashidani,[3] and Naoki Kunugita[4]

[1] *Center for Environmental Health Sciences, National Institute for Environmental Studies, Tsukuba, Ibaraki 305-8506, Japan*
[2] *Center for Environmental Risk Research, National Institute for Environmental Studies, Tsukuba, Ibaraki 305-8506, Japan*
[3] *University of Occupational and Environmental Health, 1-1 Iseigaoka, Yahatanishi-ku, Kitakyushu, Fukuoka 807-8555, Japan*
[4] *Department of Environmental Health, National Institute of Public Health, 2-3-6 Minami, Wako City, Saitama 351-0197, Japan*

Correspondence should be addressed to Naoki Kunugita; kunugita@niph.go.jp

Received 28 April 2013; Accepted 17 September 2013

Academic Editor: Carlos Barcia

Studies of unexplained symptoms observed in chemically sensitive subjects have increased the awareness of the relationship between neurological and immunological diseases due to exposure to volatile organic compounds (VOCs). However, there is no direct evidence that links exposure to low doses of VOCs and neurological and immunological dysfunction. We review animal model data to clarify the role of VOCs in neuroimmune interactions and discuss our recent studies that show a relationship between chronic exposure of C3H mice to low levels of formaldehyde and the induction of neural and immune dysfunction. We also consider the possible mechanisms by which VOC exposure can induce the symptoms presenting in patients with a multiple chemical sensitivity.

1. Introduction

Indoor pollutants sometimes induce health problems. The increase in neurological symptoms and immunological abnormalities in some sensitive populations that live in buildings with relatively high concentrations of volatile organic compounds (VOCs) has been recognized as the sick building syndrome (SBS) or multiple chemical sensitivity (MCS). Recent prevalent research suggests that MCS has a physiological and not a psychological etiology [1]. The features of chemical sensitivity overlap with those of addiction, allergy, and toxicity [2]. Because the induction of MCS might be related to VOCs in houses and offices, there may be a relationship between immunological and neurological abnormalities and inhalation of VOCs. Plausible mechanisms of action to explain MCS and its susceptible subpopulations are lacking. However, there is rapidly accumulating evidence that environmental agents can modify health by disrupting the homeostatic mechanisms that regulate the nervous, endocrine, and immune systems [3].

Neurogenic inflammation can be induced by air pollutants [4], and exposure to either an allergen or chemical irritant leads to a sensory nerve impulse. Chemical irritants directly trigger peripheral nerve receptors. Once the impulse reaches the central nervous system (CNS), it is redirected to another peripheral location leading to the release of neuropeptides that produce inflammation at the second site [5]. Neurogenic switching explains how antigen, stress, chemical exposure, or damage at one body site might lead to diverse symptoms at multiple distant sites. Sensory neuron sensitivity to irritants and its interactions with airway tissues and other peripheral tissues may be related to neurogenic disorders. Toxic components and related compounds in air pollutants can be transported from the nasal mucosa to the olfactory bulb [6]. Therefore, we speculate that VOCs as indoor pollutants can stimulate the sensory nerve terminal and olfactory system release of neuropeptides and cytokines that modulate neuroimmune interactions. Neurotoxic reactions to air pollutants may secondarily result from the distraction caused by sensory stimulation [7]. Common chemical sense irritancy refers to sensations that arise from stimulation of the trigeminal nerve endings in the skin and mucosal surfaces of the head [8]. Primary irritant gases, such as formaldehyde (FA), may evoke pungent sensations at subtoxic exposures.

Subsequently, local nerves activate local reflexes, such as the neurogenic reflex, whereby impulses transmitting toward the CNS produce activity in the periphery along unstimulated branches. This leads to the release of possible mediators of irritation, such as substance P, or to impulses traveling to more remote sites that induce reflexes such as momentary dyspnea or cough [7].

MCS is an amplification of the nonspecific immune response to low-level irritants in the upper airways [9]. Incidence of immunological symptoms such as asthma or wheezing, susceptible to infections, increased in MCS patients. It is suspected that exposure to low levels of indoor air pollutants modulates allergic and neurogenic inflammation [10]. In high-responder rats, the cholinergic-response rate increases upon exposure to organophosphate; however, there are no appropriate animal models that describe the mechanisms of MCS. FA is a toxic indoor air pollutant derived from furniture and construction materials that is found at relatively high concentrations in indoor environments [11, 12]. In this review, we focus on the effects of low-level FA inhalation on neuronal and immunological parameters and discuss the relationship between MCS and FA inhalation. Although chronic exposure to FA is not known to damage neurological functions, this may be due to a lack of study.

2. Effect of Long-Term Low-Level FA Exposure on the Mouse Olfactory System

MCS is a disease of unknown etiology. It has been hypothesized that MCS occurs from repeated exposure to low levels of chemicals, particularly VOCs; therefore, the olfactory system probably plays an important role in the initial expression of MCS symptoms [13, 14]. Olfactory stimulation probably represents the most likely route of exposure [15], and it is reasonable to assume that a significant activation of the olfactory epithelium (OE) cells can provide sufficient input into the CNS limbic circuits to induce sensitization. There may be a mechanism by which repeated olfactory stimulation and other sensitization processes induced by some other physiological process are exacerbated by prior, repeated olfactory stimulation. The olfactory system consists of the OE, the main olfactory bulb (MOB), and higher brain centers such as the piriform cortex and the amygdala. FA is aversive to mice at concentrations that approximate sensory irritation in humans [16].

In our previous study, morphological analysis of the mouse olfactory system was conducted after a long-term exposure to low-level FA to examine the association between olfactory function and the induction of MCS [17]. In that study, the mice were divided into four groups and exposed to 0, 80, 400, or 2000 ppb of FA vapor. After a long-term exposure (3 months), the mice were anesthetized and perfused with fixative solutions. The OE and the brains were removed and processed for morphological analysis. When the ultrastructure of the OE surface was examined in mice exposed to 2000 ppb of FA vapor, there were a slight degeneration and injury of microvilli of the supporting cell in several mice; however, most of the mice had no characteristic OE degeneration. Histological and immunocytochemical analyses of the OE indicated that there was no difference in the thickness and the expression of the olfactory marker protein (a marker of mature olfactory neurons) between the mice exposed to FA and control mice. Our results indicated that the long-term exposure to low-level FA induced no severe effect on the OE and that the OE was still functionally capable of detecting and transmitting olfactory information. We also found that there was no significant difference of olfactory bulb size between the control group and FA exposed groups [17].

Tyrosine hydroxylase (TH), which initiates dopamine synthesis, is abundant in periglomerular (PG) cells in the MOB, and its expression is a useful marker for monitoring olfactory function [18, 19]. Immunocytochemical analysis of TH-positive PG cells showed that long-term exposure to low-level FA induces an increase in the number of TH-expressing PG cells [17]. These results suggest that MOB activity is modulated by long-term exposure to low levels of FA.

The axons of olfactory neurons form synaptic contacts with the dendrites of secondary neurons (mitral and PG cells) in the olfactory bulb glomerulus, and it is well known that a change in the size of a synapse is activity dependent. Results of an electron microscopic analysis revealed a decrease in the size of a synapse in mice subjected to a long-term exposure of low levels of FA. Further studies are necessary to clarify the biological significance of the synaptic changes in the present study.

In the central olfactory system, MOB neurons mainly project to the piriform cortex and amygdala. In these areas, gamma-aminobutyric acidergic (GABAergic) inhibitory neurons are colocalized with the calcium-binding proteins, calbindin and parvalbumin [20]. Our preliminary results indicated that a long-term exposure to low levels of FA induced an increase in the number of parvalbumin and calbindin expressing cells in the amygdale (unpublished results). These results suggest that long-term exposure to a low level of FA modulates the GABA inhibitory system in the central olfactory areas. The functions of the amygdala are variable but include the emotional, motivational, and homeostatic functions of the brain [21]. The piriform cortex is a higher center for processing of odor sensory information, especially odor memory [22]. It may be that these brain functions are altered by repetitive, low doses of FA. By the repetition of low-dose chemical stimulation via olfactory bulb, abnormal response in signal pathway of central nervous system induced the persistence of activation.

The results of this morphological study indicate that the olfactory system is influenced by a long-term exposure to low levels of FA. Physiological and behavioral analyses are necessary to clarify the relationship between MCS and the olfactory system.

3. Estimation of the Effect of FA and Toluene Inhalation on the HPA Axis

The hypothalamo-pituitary-adrenal gland (HPA) axis responds to stress by initiating a cascade of endocrine events

including the hypothalamic secretion of corticotrophin releasing hormone (CRH), the release of adrenocorticotropic hormone (ACTH) from the anterior pituitary, and the secretion of corticosteroids from the adrenal gland [23–26]. Activation of the HPA axis by stress is dependent on the characteristics of the stressor. We examined the effects of FA inhalation on the HPA axis in female mice. Our study showed that long-term exposure to low levels of FA and the allergic condition induced by ovalbumin (OVA) sensitization may act as an HPA axis stressor [27]. Briefly, we discovered a dose-dependent upregulation (0, 80, 400, and 2000 ppb) of the number of CRH-immunoreactive (ir) neurons and ACTH-ir cells and ACTH mRNA expression in nonallergenic mice exposed to FA. Furthermore, the allergic reaction and exposure to FA acted in a synergic manner on the hypothalamus and pituitary gland. These values were all significantly higher ($P < 0.05$) in control (unexposed) allergenic mice than in control nonallergenic mice. These values were significantly higher in the 80 ppb allergenic group mice compared with the 80 ppb nonallergenic mice but significantly less in the 2000 ppb allergenic mice than in the nonallergenic mice, suggesting that the 2000 ppb allergenic group mice suffered from impaired HPA axis function. The appearance of SBS in humans may result from a depressed HPA axis that is unable to react to the secondary stress (headache, mental fatigue, nausea, etc.) induced by FA [28]. Therefore, there may be a synergistic effect between low-level FA-induced and antigen-stimulated lesions of the hypothalamus and pituitary gland.

We used toluene as a chemical stressor to study whether these effects of FA exposure on the HPA axis are specific to FA. Toluene is an organic solvent that is widely used in industrial glues, lacquers, and paint removers and induces MCS in humans [29–31]. The toluene inhalation experiment consisted of two related studies. To mimic the preinhalation trigger produced by high-dose toluene, mice were exposed to 500 ppm toluene for three consecutive days prior to FA inhalation [32]. After the three days, groups of mice were exposed to various doses of (0, 80, 400, and 2000 ppb) FA as before [33]. The results showed that the number of CRH-ir neurons, the proportion of ACTH-ir cells, and ACTH mRNA expression were upregulated according to the inhaled dose of FA. The same results were found for the number of ACTH cells with the exception that the number of ACTH cells in the 2000 ppb subgroup was reduced compared with the 400 ppb subgroup. We previously described that abundant dilatation of the sinusoids was found in the anterior pituitary of the 80, 400, and especially the 2000 ppb subgroups compared with the control group [33]. Although the proportion of ACTH cells increased with the dose of FA inhaled, these results explain the reduction in the number of ACTH cells in the 2000 ppb subgroup.

To compare the effect of FA inhalation with that of toluene, each group was further divided into control and 50 ppm subgroups of 0 and 50 ppm with and without OVA sensitization [33]. ACTH mRNA expression in the 50 ppm toluene subgroup of nonallergenic mice was greater than in the control nonallergenic group, and ACTH mRNA expression in the control allergenic group was greater than in the control nonallergenic group. Furthermore, the ACTH mRNA expression in the 50 ppm toluene subgroup of allergenic mice was greater than in the control allergenic mice. We are currently investigating the number of hypothalamic CRH-ir cells and the proportion and number of pituitary ACTH-ir cells in these mice. It follows that the inhalation of low-level toluene may have the same effect on the HPA axis as FA, with the exception that there was an increase in number and size of the sinusoids in the anterior pituitary of mice that inhaled toluene.

4. Effect of Low-Level FA Inhalation on the Expression of Neurotransmitter mRNAs in the Mouse Brain

Patients suffering from MCS report hypersensitivity to a wide variety of environmental chemicals including VOCs. Because most of the common symptoms are extreme fatigue, headache, gastrointestinal problems, depression, anxiety, irritability, and sensitivity to perfumes, it can be postulated that these symptoms accompany brain dysfunction. Moreover, recent studies indicate that neural plasticity is involved in the initiation and the development of MCS [34]. This kindling causes deterioration of long-term potentiation, which is a mechanism of efficient learning and memory. Glutamate-responsive neurons are common in the limbic system, a site of short-term memory and associated functions, and glutamate levels correlate with kindling [35]. When toxic chemicals are bound to the GABAα receptor, their response to glutamate is disinhibited.

Thus, we studied the effect of low-level FA exposure, which is highly suspected to initiate MCS and its symptoms, on the expression of neural transmission-related mRNAs in the mouse brain (400 ppb, daily for 16 h, 5 days per week for 12 weeks). Semiquantification of mRNAs by RT-PCR revealed that FA exposure caused an increase in the glutamate receptor epsilon 1 subunit mRNA in the neocortex and hippocampus and in the epsilon-1 and epsilon-2 mRNA in the amygdala [36]. Exposure to FA also decreased the epsilon-2 mRNA in the neocortex and hippocampus. In the hypothalamus, FA increased 5-hydroxytriptamine 1A-receptor mRNA and GABA receptor α-1 subunit mRNA. The continuous disruption of GABA and unstable ACTH action through the synergistic effects of indoor chemical pollutants lead to unanticipated threshold changes, time-dependent sensitization [37] and may cause kindling.

5. Immunological Alteration in Mice Exposed to Low Levels of FA

Exposure to FA elicits a variety of allergic signs and symptoms and irritates the upper respiratory tract. There is a positive relationship between worker exposure to FA and asthma or asthmatic bronchitis [38, 39]. Long-term exposure to FA significantly increases the numbers of CD26-IL-2-positive cells, B cells, and autoantibodies in some patients with multiple-organ symptoms that involve the CNS, upper- and lower-respiratory systems, and gastrointestinal tract [40]. An increase in the CD4/CD8 ratio, due to a low percentage

of CD8 positive cells, has also been observed among MCS patients [41]. However, another study found no indication of immunologically mediated respiratory disease in a group of workers exposed to FA, but some appeared to experience respiratory or ocular symptoms caused by an irritant mechanism [42].

Exposure to low levels of FA affects various immune functions in animals. In guinea pigs, an 8 h exposure to 300 ppb FA increased airway reactivity to acetylcholine [43], and, after five consecutive days of exposure to 250 ppb FA, OVA sensitivity was significantly enhanced, but there was no effect of a 130 ppb exposure [44]. Exposure to 1600 ppb FA for 10 days increased OVA-specific IgE production in mice intranasally sensitized with OVA [45]. Therefore, short-term FA exposure may directly enhance sensitization in airways and aggravate allergic inflammatory reactions. However, the long-term effects of low-level FA exposure on allergic inflammation are largely unknown. We tried to identify the presence or absence of hypersensitivity reactions in mice exposed to low doses of FA. However, exposure of mice to 80 or 400 ppb FA alone resulted in no significant changes in proinflammatory cytokine production in bronchoalveolar lavage fluid and in the hippocampus, total antibody production and lymphocyte subpopulations in peripheral blood, or infiltration of inflammatory cells to the lung; however, OVA-immunized mice exposed to 2000 ppb FA had significantly more inflammatory cells in the bronchoalveolar lavage fluid.

Exposure to FA significantly increased hippocampal nerve-growth factor (NGF) mRNA (80 and 400 ppb) and NGF content (400 ppb) in OVA-immunized mice [46]. In contrast, low levels of FA may act in concert with OVA stimulation to suppress NGF production in the plasma and bronchoalveolar lavage fluid [47]. Neurotrophins, including NGF, and brain-derived neurotrophic factors play an important role in the development of airway inflammation and hyperresponsiveness by inducing the production of tachykinins, such as substance P, which are involved in allergic responses [47]. It is likely that exposure to FA stimulates vagus nerve endings to release neuropeptides that can activate immunocompetent cells and modulate allergic inflammation. Immunological inflammation is induced at the local site by invading antigenic substances or chemicals, whereas neurogenic inflammation is induced at sites distant from stimulation. Although FA is a potent contact allergen, it lacks the ability to cause sensitization of the respiratory tract [48]. Our data suggest that exposure to FA disrupts the regulatory mechanisms of neurogenic and immunological inflammation that contribute to host homeostasis. In animal models, neurogenic inflammation may contribute to the inflammatory response to allergens, whereas chronic inflammation, which causes the release of neurotrophins from inflammatory cells, may lead to changes in innervation patterns [49].

6. Modulation of Neuroimmune Response in Mice Exposed to FA

Recently, an investigation of the effect of repeated low-level FA on escape or cocaine-induced behavior in mice showed that 1000 ppb FA inhalation is aversive and produces behavioral changes [16, 50]. FA inhalation induces tachykinin release from sensory nerve endings in rats [51]. Sneezing can be provoked by FA and can be evoked from C-fiber stimulation [52]. We found that sneezing frequency in mice dose dependently increased by FA inhalation [53]. Because we have shown the influence of long-term exposure to low levels of FA in the olfactory neurons and in the Ca-binding proteins in the amygdala and piriform cortex, low-level FA exposure may stimulate olfactory and trigeminal pathways and result in an abnormal response of CNS signaling pathways.

Concerning the roles of substance P, an increase in the number of immunoreactive nerve fibers was found in asthmatics compared to nonasthmatics, but less immunoreactivity was found in lung tissues of the asthmatics patients [54]. Short-term inhalation of irritants exacerbates allergic inflammation due to the release of substance P and related neuropeptides. However, the contribution of persistent inhalation of low-level irritant agents to neuroimmune inflammation remains unknown. FA can activate the sensory irritant receptor by reacting with a thiol group in the receptor and produce a more potent effect than substances physically adsorbed to the receptor [52]. During toluene diisocyanate (TDI) exposure, TDI-induced release of NGF may mediate substance P upregulation in airway-sensory neurons [55]. Not only the production of NGF but also the release of substance P in CNS and immunogenic inflammation were also modulated by low-level FA as well as TDI exposure.

Because ACTH suppresses the immune system [56], chronic stress can modulate the HPA axis and alter the innate and acquired immune response to infection [57]. CRH release from the hypothalamus stimulates the production of neuropeptides that irritate sensory organs [58], and activation of the HPA axis in FA-exposed mice directly and indirectly affects innate and acquired immune responses in the brain.

A proposed central mechanism of MCS induced by organic solvents involves the widespread stimulation of NMDA activity in the limbic system followed by a widespread increase in nitric oxide and peroxynitrite [59]. Our results stating that low levels of FA increase hippocampal NMDA-mRNA expression are in agreement with recent reports showing the association of NMDA stimulation in formalin-induced pain [60, 61]. Treatment with the NMDA receptor-antagonist AP5 produced analgesia in a formalin pain test [62]. Although our experimental design called for a different time course and route of stimulation than the formalin pain test, the phenomenon of NMDA receptor activation is a common response to FA.

It is suggested that the interaction between the neural and immune systems is an underlying cause of MCS. We conducted an experiment to examine the effects of FA exposure in OVA-immunized mice [63]. In that study, the mice (C3H females) were injected intraperitoneally with 10 μg OVA and 2 mg alum before their exposure to FA (0 or 400 ppb). On days 21, 42, 63, and 77 of the exposure period, each mouse was booster challenged with OVA, and the brains were sampled one day following the final FA inhalation. To determine the effects of low-level FA exposure on the expression of neuronal synaptic plasticity related genes in the hippocampus, we

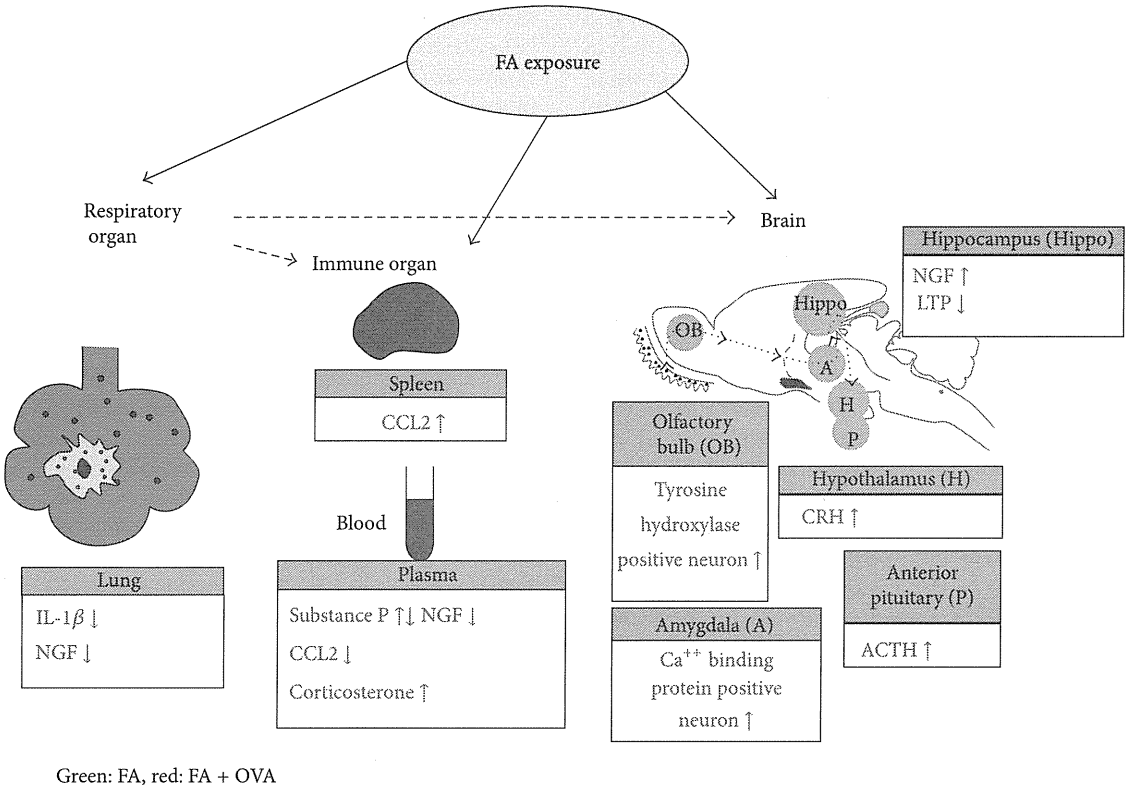


FIGURE 1: Possible target organs and biomarkers affected by formaldehyde exposure. FA exposure with or without OVA affects respiratory, immune, and central nervous systems by modulating the cytokines (IL-1β, CCL2), neuropeptides (NGF, substance P), hormones (CRH, ACTH, and corticosterone), and enzymes (TH) and intracellular calcium-binding protein in the mouse model (green: FA, red: FA + OVA).

examined the mRNA expression of the NMDA receptor-subtypes NR2A and NR2B, dopamine D1 and D2 receptors, cAMP response element-binding (CREB)-1 and -2, and FosB/ΔFosB. The mRNA levels of NR2A, dopamine D1 and D2 receptors, and CREB-1 significantly increased in 400 ppb FA-exposed mice compared to the control mice, but the levels of NR2B, CREB-2, and FosB/ΔFosB were not changed [63]. We also found that treatment with MK-801, a noncompetitive NMDA receptor antagonist, normalized the NR2A, CREB-1, and dopamine D1 and D2 receptor mRNA levels that were induced by FA exposure [63]. These results suggest that stimulation by low-level FA exposure and OVA immunization selectively affects synaptic plasticity-related genes in the hippocampus and that the effects are mediated by glutamatergic neurotransmission through NMDA receptors.

To the best of our knowledge, our serial reports showing that exposure to low levels of FA alone can alter the molecular basis of brain neural transmission and that a combination of OVA immunization and FA exposure induces a significant increase in hippocampal NMDA receptor mRNA levels. It is well known that NMDA receptors are a key molecule for the formation of neuronal memory. Because neuronal memory formation is a major phenomenon in neural sensitization, and neuroimmune interaction is thought to participate in the development of MCS, it follows that the increase in hippocampal NMDA after OVA immunization and FA exposure reflects, at least in part, the hypersensitivity underlying MCS.

We previously described the effects of FA on the expression of the Bcl-2 family, which regulate survival and death of cells, and the NMDA receptors, which are associated with hippocampal function, in mice [64]. In that study, Western blot analyses were performed for Bcl-2, Bax, NMDA receptors types 2A and 2B (NR2A and NR2B) of the hippocampus taken from C3H mice exposed to 0 or 400 ppb of FA with or without OVA immunization [64]. We found that the ratio of Bcl-2/Bax expression in OVA-immunized mice significantly increased with 400-ppb FA exposure, although the differences in the expression level of each protein were not significant between the control and FA-exposed groups. The NR2A and NR2B expression of FA-exposed OVA immunized mice were sustained at levels comparable to the control; however, there was no difference in NR2A, NR2B expression, or Bcl-2/Bax expression ratio in mice that were not OVA immunized and exposed to 400 ppb FA [64]. These results suggest that the changes in the Bcl-2/Bax expression ratio that occur with low-level FA exposure and OVA immunization may follow enhanced NGF production and exert a protective effect against apoptosis. We speculate that our putative defense mechanisms are initially exerted in the hippocampus in response to FA exposure and that adverse FA effects successively arise when the FA exposure reaches an intolerable level. This mechanism does not account for the transition of the influence of FA from "protective" to "adverse"; however, it may be a key for understanding FA sensitivity in individuals