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Abnormal liver function associated with occupational exposure to dimethylformamide and glutathione *S*-transferase polymorphisms

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Abstract

Dimethylformaide (DMF) is a major solvent predominately used in synthetic leather and resin production. Many human and animal studies have linked the cause of hepatoxicity to DMF. Previously, the authors demonstrated the significant dose–response relationship between abnormal liver function tests and DMF exposure and the interaction with hepatitis B virus (HBV) infection in Taiwanese workers. Because the toxic effect of various chemicals can be modified by metabolic traits, the study also investigated the influence of the glutathione *S*-transferases (GSTM1 and GSTT1) on the toxic effect of DMF. The average DMF exposure concentration was 23.87 ppm (range 5.2–86.6 ppm) in the high-exposure ($\geq$5 ppm) group and 2.41 ppm (range 0.9–4.3 ppm) in the low-exposure ($<$5 ppm) group. There were 13 of 44 (29.6%) abnormal liver function tests (elevations of either glutamate oxaloacetate transaminase (GOT) or glutamate pyruvate transaminase (GPT)) among the high DMF exposure workers, two of 22 (9.1%) abnormal liver function tests among the low DMF exposure workers. Chronic liver disease as determined by ultrasonography was present in seven of 44 (15.9%) high DMF exposure workers, and 0 of 22 (0%) low DMF exposure workers. There were 11 of 34 (32.4%) abnormal liver function tests among the GSTT1 null genotype workers, and four of 32 (12.5%) abnormal liver function tests among the GSTT1-positive genotype workers. Compared with the low DMF exposure workers, the adjusted odds ratio and 95% confidence intervals for abnormal liver function tests was 6.78 (0.94–48.7) for the high DMF exposure workers. Compared with the GSTT1-positive genotype workers, the adjusted odds ratio and 95% confidence intervals for abnormal liver function tests was 4.41 (1.15–16.9) for the GSTT1 null genotype workers. Compared with the low DMF group with GSTT1-positive genotype workers, the odds ratio (adjusted for HBV status) of abnormal liver function test was 12.38, 95% CI = (1.04–146.9) for the high DMF group with GSTT1 null genotype workers. This study indicates that abnormal liver function and chronic liver disease are associated with DMF exposure, and there are more than multiplicative interaction effects on abnormal liver function tests between the DMF exposure and the GSTT1 genotype.

Keywords: *Abnormal liver function, DMF, GSTT1*

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Introduction

N,N-dimethylformamide (DMF) is an excellent colourless liquid solvent used for numerous organic compounds when a solvent low volatility is necessary. Used in the manufacture of synthetic leader and resins, DMF becomes absorbed into the body by inhalation and skin contact (Finkel 1983). Acute or long-term exposure to DMF affects the liver, as the target organ, of both animals (Massmann 1956, Tanaka 1971) and humans (Potter 1973, Redlich et al. 1988, Wang et al. 1991, Cai et al. 1992, Wrbitzky 1999). Abnormal liver function and liver damage with steatosis, spotty necrosis and diffuse regeneration have been associated with occupational exposure to DMF (Potter 1973, Redlich et al. 1988, 1990, Wang et al. 1991, Cai et al. 1992, Wrbitzky 1999).

Recent studies have indicated that metabolic traits might modulate the toxic effects of various chemicals. Genetic polymorphisms have been detected in a variety of enzymes involved in the metabolism of exogenous chemicals and have been associated with varying degrees of environmental disease causation. DMF is metabolized by cytochrome P4502E1 to monomethyl and formamide derivatives such as N-methyl formamide (NMF), N-(hydroxymethyl)-N-methylformamide (HMMF) and N-acetyl-*S*-(N-methylcarbamoyl)cysteine. Methyl isocyanate, a highly toxic reactive metabolic intermediate of DMF, may be conjugated with glutathione to become *S*-(N-methylcarbamoyl) glutathione (SMG) (Mraz et al. 1993). Epoxides have been reported to be substrates for glutathione *S*-transferases (Ketterer 1988), and GSTM1 and GSTT1 polymorphisms have been reported (Nelson et al. 1995). The present authors' previous study demonstrated the significant dose–response relationship between abnormal liver function tests (LFTs) and DMF exposure and the interaction with hepatitis B (HBV) infection in Taiwanese workers (Luo et al. 2001). The present study furthermore investigated the modulatory effects of two glutathione *S*-transferases polymorphisms on DMF-induced liver damage.

Materials and methods

In October 1996, 66 males from three workstations (artificial leather — workstation I; printed circuit (PC) boards — workstation II; and epoxy resin — workstation III) of a resin synthesis factory were recruited for this study. Personal and area air sampling was performed to determine DMF exposure of workers. Details are published by Kuo et al. (2000). Briefly, 21 area sampling points were selected throughout the plant based on the proximity to sources of solvent emissions. Sampling time ranged from 30 to 180 min. Forty-five workers were also selected at random for personal sampling.

Most workers engaged in the procedures of synthetic leather manufacturing and PC board manufacturing were considered to have DMF exposure, whereas epoxy resin manufacturing workers were considered to have no DMF exposure. Study subjects were divided into two groups according to DMF exposure concentrations (Table I). The cut-off point for high and low exposure to DMF was defined arbitrarily as 5 ppm. High DMF exposure was defined according to job descriptions of spraying materials, mixing in synthetic leather production, dipping or assembling of PC board, printing, researching and developing, preparing textiles in synthetic leather production, and mixing materials in PC board production. Low DMF exposure was defined by job descriptions of maintenance and quality control in PC board production, operators,

Table I. Characteristics of workers by categories of DMF exposure level.

	High exposure ($n=44$)	Low exposure ($n=22$)	Total ($n=66$)
Age (years)	34.25* ± 5.58 (23–45)	31.32 ± 4.52 (25–42)	33.27 ± 5.4 (23–45)
Employment (years)	8.43* ± 4.78 (1–21)	5.5 ± 2.3 (2–9)	7.46 ± 4.33 (1–21)
GOT	25.18** ± 9.64 (13–60)	22.23 ± 3.82 (16–30)	24.2 ± 8.26 (13–60)
GPT	30.16** ± 21.77 (11–120)	22.96 ± 9.69 (6–47)	27.76 ± 18.86 (6–120)
DMF concentration (ppm)	23.87 ± 19.74 (5.2–86.6)	2.41 ± 0.76 (0.9–4.3)	16.71 ± 19.02 (0.9–86.6)
HBV (+)	8 (18.2%)	7 (31.8%)	15 (22.7%)
Drink (+)	10 (22.7%)	9 (40.9%)	19 (28.8%)
BMI (>27)	24 (54.6%)	12 (54.6%)	36 (54.6%)
GSTT1 (null type)	20 (45.5%)	14 (63.6%)	34 (51.5%)
GSTM1 (null type)	18 (40.9%)	9 (40.9%)	27 (40.9%)

Data are mean ± standard deviation, range (minimum–maximum).
High exposure: ≥5 ppm; low exposure <5 ppm.
*$p<0.05$, **$p<0.1$.
GOT, glutamate oxaloacetate transaminase; GPT, glutamate pyruvate transaminase; HBV (+), positivity of HbsAg; BMI, body mass index.

epoxy resin unloading, research and development, and maintenance in epoxy resin production.

Informed consent was obtained from all subjects. Demographic data, work history and alcohol consumption were determined by standard self-administered questionnaire. Habitual drinking was defined as alcohol drinking greater than once each week. Calculation of body mass index (BMI) was determined by dividing body weight by the square of height. LFTs including were determined with a clinical chemistry analyser (Hitachi 7450; Hitachi Co. Ltd., Tokyo, Japan). HBsAg and anti-hepatitis C virus (HCV) assays were determined by ELISA. The positivity of HBV was defined as the presence of HBsAg, and the positivity of HCV was defined as the presence of anti-HCV. The abnormality of LFTs was defined as oxaloacetate transaminase (GOT) > 34 or glutamate pyruvate transaminase (GPT) >36, respectively.

Using the high-resolution real-time machine Aloka 650 (Aloka Co., Ltd., Tokyo, Japan), certified physicians performed abdominal ultra-sonograms to detect structured clinical liver abnormalities on 15 subjects with abnormal LFTs. A multi-parameter scoring system was applied in the evaluation of real-time ultra-sonograms for the detection of liver abnormalities (Yang et al. 1988). The parameters used for the assessment of chronic liver diseases (parenchymal liver diseases or liver cirrhosis other than fatty liver) were changes of liver surface, inferior edge, echotexture, echogenicity, hepatic vein, inner diameters of portal veins as well as splenic veins, and the size of the liver and spleen. As defined by the scoring system, parenchymal liver diseases were given at scores in the range of 6–7 and liver cirrhosis as a total score ≥8. Signs and symptoms that indicated fatty liver included the presence of the increased echogenicity (brightness), masking of walls of portal veins and gallbladder, blurring of hepatic veins and far-gain attenuation.

A total of 10 ml venous blood were drawn into heparinized tubes (vacutainer) and stored at 4°C. The whole blood was separated into plasma, buffy coat and red blood cells by centrifugation within 18 h of obtaining the blood, then stored in a −70°C

freezer. Genomic DNA was extracted and purified from the buffy coat. The GSTM1 and GSTT1 genotypes were determined by co-amplification of both genes (Comstock et al. 1990, Pemble et al. 1994) with polymerase chain reaction (PCR). Briefly, PCR was performed in a 25-μl mixture containing the buffer supplied by Promega (Madison, WI, USA), 250 ng genomic DNA, Taq DNA polymerase (1 U), four bases (dNTP) and 200 μg of each primer. The primers used for the GST M1 gene were 5′-CTGCCCTACTTGATTGATGGG-3′ and 5′-CTGGATTGTAGCA-GATCATGC-3′. The primers used for the GSTT1 gene were 5′-TTCCTTAC TGGTCCTCACATCT C-3′ and 5′-TCACC-GGATCATGGCCA GCA-3′. The human B-globin gene (110 bp) was also amplified in each reaction as a positive control to confirm the presence of amplifiable DNA in the samples. The primers used for B-globin were 5′-ACACAACT GT GTTCACTAG-C-3′ and 5′-CAACTCATC-CACGTTCACC-3′. The amplification was carried out in 35 cycles with denaturation at 94°C for 1 min 30 s, annealing at 52°C for 1 min, and extension at 65°C for 1 min. The PCR products were then resolved by electrophoresis in 2% agarose gels, stained with ethidium bromide and photographed under ultraviolet light. Individuals with one or more GSTM1 alleles had a 273-bp fragment, and individuals with one or more GSTT1 alleles had a 480-bp fragment.

The data from the 66 workers were encoded, entered and analysed with the assistance of the Statistical Analysis System (SAS) PC software Package (SAS Institute 1999–2001) and Database III Plus (1986). By using SAS PC programs, Student's *t*-test and chi-square were performed to test significant differences among groups. Odds ratios were calculated to test the magnitude and the significance of differences in prevalence of abnormal LFTs among exposure and control groups or GST status. Multivariate logistic regression analysis was used to confirm the relationship between abnormal liver function, GST status and DMF exposure after adjusting for other factors.

Results

Table I shows a summary of the basic characteristics of the study cohort. The DMF exposure concentration mean was 16.71 ppm (range 0.9–86.6 ppm). A ratio of 44 of 66 (66.6%) workers had high ($\geq$5 ppm) DMF exposure in comparison with a ratio of 22 of 66 (33.4%) of workers exposed to low ($<$5 ppm) DMF concentrations. The low DMF exposure workers were significantly younger than the high exposure group. Low DMF exposure workers also had a significantly shorter duration of employment. There were borderline significant higher liver enzyme (GOT, GPT) level in high DMF exposure workers than low DMF exposure workers. The overall prevalence of GSTT1 null genotype and GSTM1 null genotype were 51.5 and 40.9%, respectively.

The prevalence for abnormal LFTs by DMF exposure status is shown in Table II. High DMF exposure workers had a significantly higher prevalence of abnormal LFTs and chronic liver disease than the low DMF exposure workers (29.6 versus 9.1%, 15.9 versus 0%, respectively). Compared with the low DMF exposure workers, the odds ratio (adjusted for duration of employment, HBV status and DMF exposure) abnormal LFT was 6.78, 95% CI = (0.94–48.7), and for chronic liver disease was 3.11, 95% CI = (0.3–32) for the high DMF exposure workers.

The prevalence of abnormal LFTs by GSTM1 status and GSTT1 status are shown in Tables III and IV, respectively. GSTT1 null genotype workers had a significantly

Table II. Prevalence of abnormal liver function tests by DMF exposure status.

	High exposure ($n=44$)	Low exposure ($n=22$)	Total ($n=66$)
GOT (+)	7* (15.9%)	0 (0%)	7 (10.6%)
OR[a] (95% CI)	9* (0.49–165.1)	1	
AOR[b] (95% CI)	3.2* (0.54–18.8)	1	
GPT (+)	11 (25%)	2 (9.1%)	13 (19.7%)
OR[a] (95% CI)	3.3 (0.67–16.6)	1	
AOR[b] (95% CI)	3.5 (0.62–20)	1	
LFT (+)	13* (29.6%)	2 (9.1%)	19 (28.8%)
OR[a] (95% CI)	4.19** (0.85–20.6)	1	
AOR[b] (95% CI)	6.78* (0.94–48.7)	1	
Chronic liver disease	7* (15.9%)	0 (0%)	7 (10.6%)
OR[a] (95% CI)	9* (0.49–165.1)	1	
AOR[b] (95% CI)	3.11 (0.3–32)	1	

[a]Crude odds ratio.
[b]Odds ratio adjusted for duration of employment, HBV, GSTT1 status.
*$p<0.05$, **$p<0.1$.
GOT (+) is defined as GOT >34; GPT (+) is defined as GPT >36; LFT (+), abnormality of liver function tests, is defined as GOT >34 or GPT >36.

higher prevalence of abnormal LFTs and chronic liver disease than the GSTT1-positive genotype workers (32.4 versus 12.5%, 14.7 versus 0%, respectively) (Table III). Compared with the GSTT1-positive genotype workers, the odds ratio (adjusted for duration of employment, HBV status and DMF exposure) abnormal LFT was 4.41, 95% CI = (1.15–16.9) and for chronic liver disease was 4.41 95% CI = (0.63–31) for the GSTT1 null genotype workers. Compared with the GSTM1-positive genotype workers, the odds ratio (adjusted for duration of employment, HBV status and DMF exposure) for abnormal LFT was 1.11, 95% CI = (0.25–4.83), and for chronic liver disease was 2.63, 95% CI = (0.39–17.8) for the GSTM1 null genotype workers (Table IV).

Table III. Prevalence of abnormal Liver function tests by GSTT1 status.

	GSTT1 (null type) ($n=34$)	GSTT1 (positive type) ($n=32$)	Total ($n=66$)
GOT (+)	4 (11.8%)	3 (9.4%)	7 (10.6%)
OR[a] (95% CI)	1.3 (0.27–6.27)	1	
AOR[b] (95% CI)	1.8 (0.34–9.41)	1	
GPT (+)	9 (26.5%)	4 (12.5%)	13 (19.7%)
OR[a] (95% CI)	2.52 (0.69–9.2)	1	
AOR[b] (95% CI)	3.01** (0.79–11.6)	1	
LFT (+)	11** (32.4%)	4 (12.5%)	15 (22.7%)
OR[a] (95% CI)	3.35** (0.94–11.9)	1	
AOR[b] (95% CI)	4.41* (1.15–16.9)	1	
Chronic liver disease	5* (14.7%)	2 (6.3%)	7 (10.6%)
OR[a] (95% CI)	2.59 (0.46–14.4)	1	
AOR[b] (95% CI)	4.41* (0.63–31)	1	

[a]Crude odds ratio.
[b]Odds ratio adjusted for duration of employment, HBV, DMF status.
*$p<0.05$, **$p<0.1$.

Table IV. Prevalence of abnormal liver function tests by GSTM1 status.

	GSTM1 (null type) ($n=27$)	GSTM1 (positive type) ($n=39$)	Total ($n=66$)
GOT (+)	5** (18.5%)	2 (5.13%)	7 (10.6%)
OR[a] (95% CI)	4.21** (0.75–23.5)	1	
AOR[b] (95% CI)	2.63 (0.39–17.8)	1	
GPT (+)	7 (25.9%)	6 (15.4%)	13 (19.7%)
OR[a] (95% CI)	1.93 (0.57–6.6)	1	
AOR[b] (95% CI)	1.27 (0.29–5.6)	1	
LFT (+)	8 (29.6%)	7 (18%)	15 (22.7%)
OR[a] (95% CI)	1.93 (0.6–6.2)	1	
AOR[b] (95% CI)	1.11 (0.25–4.83)	1	
Chronic liver disease	5 ** (18.5%)	2 (5.13%)	7 (10.6%)
OR[a] (95% CI)	4.21** (0.75–23.5)	1	
AOR[b] (95% CI)	2.63 (0.39–17.8)	1	

[a]Crude odds ratio.
[b]Odds ratio adjusted for duration of employment, HBV, GSTT1 status.
*$p<0.05$, **$p<0.1$.

In multivariate logistic regression analysis, HBV status was significantly associated with GOT (+), GPT (+), LFT (+) and CLD (+) ($p=0.062$, 0.011, 0.033 and 0.024, respectively) after adjusting for other factors (Table V). GSTT1 null genotype status was significantly associated with GPT (+), LFT (+) and CLD (+) ($p=0.047$, 0.011 and 0.079, respectively) after adjusting for other factors. High DMF exposure status was significantly associated with GPT (+) and LFT (+) ($p=0.043$ and 0.0014, respectively) after adjusting for other factors.

Analysis of risk of abnormal LFTs stratified by GSTT1 status and categories of DMF exposure is shown in Table VI. Compared with the low DMF group with GSTT1-positive genotype workers, the odds ratio (adjusted for HBV status) of abnormal LFT was 2.55, 95% CI = (0.22–29.54) for the low DMF group with GSTT1 null genotype workers. Compared with the low DMF group with GSTT1-positive genotype workers, the odds ratio (adjusted for HBV status) of abnormal LFT was 3.06, 95% CI = (0.29–32.73) for the high DMF group with GSTT1-positive genotype workers. Compared with the low DMF group with GSTT1-positive genotype workers, the odds ratio (adjusted for HBV status) of abnormal LFT was 12.38, 95% CI = (1.04–146.9) for the high DMF group with GSTT1 null genotype workers. The high DMF group with GSTT1 null genotype workers had a synergy index of 1.587 departure from expected adjusted odds ratio, and indicated more than a multiplicative interaction effects between DMF exposure and GSTT1 null genotype.

Discussion

The results are consistent with other reports (Potter 1973, Redlich et al. 1987a,b, 1988, 1990, Wang et al. 1991, Kawai et al. 1992) that hepatoxicity can occur in DMF-exposed workers. The results suggest that for workers exposed to DMF above 5 ppm, interventions need to be done to avoid further DMF exposure and liver damage.

The interaction between environmental factors and genetic susceptibility are important in determining disease risk (Ottman 1990, Ottman et al. 1991). The role

Table V. Multivariate logistic regression analyses between abnormal liver function tests, GSTT1 status, DMF exposure categories and related indicators.

	GOT (±)	GPT (±)	LFT (±)	Chronic liver disease
Intercept:				
PE±SE[a]	−14.99±237	−4.94±1.42	−5.68±1.63	−19.4±200
p	(0.93)	(0.0005)	(0.0005)	(0.92)
Duration:				
1 (>7.46)				
0 (≤7.46)				
PE±SE	−0.55±0.98	−0.23±0.8	−0.73±0.84	0.52±1.14
p	(0.58)	(0.77)	(0.39)	(0.65)
HBV status:				
1 (yes), 0 (no)	1.89±1.01	2.17±0.85	2.93±1	3.19±1.41
PE±SE	(0.062)	(0.011)	(0.0033)	(0.024)
p				
BMI status:				
1 (>27)				
0 (≤27)				
PE±SE	−0.104±0.99	0.73±0.8	0.64±0.82	1.86±1.47
p	(0.92)	(0.36)	(0.44)	(0.21)
Alcohol:				
1 (yes), 0 (no)	−0.91±1.24	0.1±0.82	−0.32±0.87	0.46±1.29
PE±SE	(0.46)	(0.9)	(0.71)	(0.72)
p				
GSTT1:				
1 (Null)				
0 (Positive)				
PE±SE	0.81±0.92	1.62±0.81	2.33±0.92	2.51±1.43
p	(0.38)	(0.047)	(0.011)	(0.079)
Exposure:				
Categories				
1 (High DMF)				
0 (Low DMF)				
PE±SE	12.9±237	2.11±1.04	2.90±1.18	13.53±200
p	(0.96)	(0.043)	(0.0014)	(0.95)

[a]PE±SE, parameter estimate±standard error.

of genetic susceptibility of metabolic enzymes might provide some explanations to the individual differences in sensitivity to the development of environmental disease. The mammalian GST super-gene family (composed of alpha, pi, mu and theta) catalyses the conjugation of GSH with electrophilic substrates. Some individuals carry a large deletion (null genotype) in the glutathione *S*-transferase μ (GSTM1) gene or the glutathione *S*-transferase θ (GSTT1) gene. Individuals with the GSTM1 or GSTT1 null genotype are expected to detoxify environmental toxicants less effectively, thus increasing the concentrations of toxic metabolites in plasma, affecting the clinical outcomes. An individual without an intact GSTM1 may have a higher risk of developing cancer or cytogenetic damage (Seidegard et al. 1986, 1990, Wiencke et al. 1990). Many chemicals are substrates for GSTT1, including ethylene oxide, dihaloalkalane and diepoxybutane (Fost et al. 1991, Their et al. 1991, Wiencke et al. 1995). DMF is conjugated with glutathione to detoxify. The prevalence of

Table VI. Analysis of risk of abnormal liver function tests by GSTT1 status, and DMF exposure categories.

DMF exposure	GSTT1 type	LFT (+)	LFT (−)	OR 95% CI	(AOR[+] 95% CI)
High	null	9	11	14.04[a]⋆	714–276.2 (12.38[a]⋆1.04–146.9)
High	positive	4	20	3.732[b]	18–77.170 (3.06[b]⋆⋆.29–32.73)
Low	null	2	12	3.4[c]	144–80.05 (2.55[c]0.22–29.54)
Low	positive	0	8	1	
Total		15	51		
	Expected OR[a] departure from expected (Expected AOR[a] departure from expected)				
Additive	3.732+3.4−1=6.132	14.04−6.132=7.098	(3.06+2.55−1=4.61	12.38−4.61=7.77)	
Multiplicative	3.732×3.4=12.69	14.04/12.69=106	(3.06×2.55=7.803	12.38/7.903=1.587)	

⋆$p<0.05$, ⋆⋆$p<0.1$.
[a]Odds ratio for LFT (+) among high DMF group with GSTT1 null type.
[b]Odds ratio for LFT (+) among high DMF group with GSTT1 positive type.
[c]Odds ratio for LFT (+) among low DMF group with GSTT1 null type.
[+]AOR, odds ratio adjusted for HBV status.

GSTT1 null genotype in the present study was within the 95% CI of the prevalence of GSTT1 null genotype from a previous report on a Chinese population (Nelson et al. 1995). This study suggests that DMF workers with a susceptible genotype (GSTT1 null type) have an increased prevalence of abnormal LFTs. The GSTT1 null genotype was significantly associated with abnormal GPT value, abnormal LFTs and chronic liver diseases. In contrast, GSTM1 genotypes were not significantly associated with LFTs. The high DMF group with GSTT1 null genotype workers had a synergy index of 1.587 departure from expected adjusted odds ratio, and indicated more than a multiplicative interaction effects between DMF exposure and GSTT1 null genotype.

There were 15 (22.7%) DMF-exposed workers with HBV infection, and 0 (0%) workers with HCV infection. In this study there was a significant association between abnormal liver enzyme tests and HBV carrier status regardless of DMF exposure with $p = 0.0033$, and HBV carrier status had synergistic effects with DMF and GSTT1 null genotype in causing liver abnormalities. These findings were consistent with other reports that hepatitis status were strongly associated with increased liver enzyme activity (Reichling & Kaplan 1988), and also confirm the previous report by Wang et al. (1991) that HBV carriers might be more sensitive to DMF exposure, especially at concentrations greater than 5 ppm. The precise mechanism of interaction between HBV infection and chemicals is still unclear. Animal studies indicated that HBV might cause persistent liver damage, change metabolism and toxicity of chemicals, and induce liver cell regenerative hyperplasia (Chisari et al. 1989).

There were seven of 44 (15.9%) workers with chronic liver disease by sonogram in the high exposure group, and none of 22 (0%) CLD in the low exposure group. Compared with the low DMF exposure workers, the adjusted odds ratio (and 95% confidence intervals) for chronic liver disease was 3.11, 95% CI = (0.3–32) for the high DMF exposure workers. A significant association was also found between chronic liver diseases and HBV carrier status and GSTT1 null genotype. Previous pathological reports from others showed multiple zonal necrosis in one worker with acute DMF exposure (Wang et al. 1991), and micro- or macrovesicular steatosis, spotty necrosis, and regeneration in other workers with acute (from less than 2 weeks to 4 months) DMF exposure, and moderately severe micro- or macrovesicular steatosis with spotty necrosis, and regeneration in workers with 10 years of DMF exposure (Redlich et al. 1988, 1990). Hepatitis B and C virus infections have been reported to be the major causes of chronic liver diseases (including liver cirrhosis, liver cancer) in Taiwan (Beasley et al. 1982, Chen et al. 1991, Yu et al. 1991). The sonogram result in the present study indicated that HBV might interact with DMF and GSTT1 null genotype to cause CLD. All seven CLD cases were in the high DMF exposure group compared with none in the low DMF exposure group. The only liver cirrhosis case was a HBV carrier with 83.3 ppm DMF exposure for 14 years.

There were two of 44 (4.6%) workers with fatty liver change by sonogram in the high exposure group, and two of 22 (9.1%) workers with fatty liver change in the low exposure group. Compared with the low DMF exposure workers, the adjusted odds ratio for fatty liver was 0.51, 95% CI = (0.08–3.27) for the high DMF exposure workers. No association was also found between fatty liver change with DMF exposure, BMI, alcohol consumption, GSTT1 status and HBV exposure (data not shown). It is speculated that DMF workers in Taiwan with low DMF exposure only suffered from fatty liver, but with HBV infection and high DMF exposure they might have liver abnormalities progressing to chronic liver disease, even cirrhosis.

Sonography is a quick, non-invasive and accurate tool, according to previous reports that they can predict the presence of liver cirrhosis, fatty liver and chronic liver disease with high sensitivity, specificity and accuracy (Yang et al. 1988, Khan et al. 2000). Nevertheless, the current sonogram results are limited by the small sample size and that some workers with normal liver enzymes may have abnormal sonograms, and further pathologic diagnosis will be necessary to confirm or clarify any discrepancies.

This study is limited by its small sample size and results (with borderline significance and confidence intervals <1), which indicate that abnormal LFTs are associated with DMF exposure and there are more than multiplicative interaction effects on abnormal LFTs between the DMF exposure and the GSTT1 genotype. Further studies are needed to clarify better the nature of the observed association.

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