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# 우리나라 일부 농업 종사자의 개인보호구 착용, 작업위생행위에 따른 소변 중 MDA, 8-OHdG 농도 변화

# Changes in Urinary MDA and 8-OHdG Concentrations due to Wearing Personal Protective Equipment and Performing Protective Behaviors among Agricultural Workers in Korea

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#### **ABSTRACT**

**Objectives:** Oxidative stress and DNA damage have been proposed as mechanisms linking pesticide exposure to health effects such as cancer and neurological diseases. We investigated whether protective measures could significantly reduce the levels of biomarkers for oxidative stress and DNA damage in agricultural workers.

**Methods:** In the present study, the levels of malondialdehyde (MDA) and 8-hydroxy-2'-deoxyguanosine (8-OHdG), biomarkers related to oxidative stress and DNA damage, respectively, were analyzed in urine samples collected from agricultural workers in two provinces of Korea (n=60). The influence of wearing personal protective equipment (PPE) and performing protective behaviors on the levels of these two biomarkers was also evaluated.

**Results:** The median urinary levels of MDA and 8-OHdG were 10.45 nmol/mg creatinine and 14.42 ng/mg creatinine in subjects living in region A, while they were 6.25 nmol/mg creatinine and 24.77 ng/mg creatinine in subjects living in region B, respectively. The levels of MDA and 8-OHdG were higher in male farmers. Farmers wearing greater numbers of PPE and performing more protective behaviors had significantly lower levels of MDA. Greater numbers of protective behaviors was significantly associated with lower levels of 8-OHdG.

**Conclusion:** The results of the present study indicate that pesticide exposure could induce oxidative stress and DNA damage in agricultural workers, and that protective measures are important for mitigating pesticide exposure.

**Keywords:** Malondialdehyde, 8-hydroxy-2'-deoxyguanosine, personal protective equipment, pesticide, protective behavior

## I. Introduction

Pesticides increase the yield and quality of agricultural products by protecting crop against pests.<sup>1)</sup> However, these chemicals are biologically active and can adversely affect the health when

humans are exposed for extended periods.<sup>2)</sup> The potential health effects of pesticide exposure in agricultural workers are of interest as these workers are exposed to extensive amounts of pesticides with high frequency.<sup>3)</sup> In agricultural workers, pesticide toxicity is generally associated with the class of the

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pesticide compound. Routine exposure to pesticides can result in long-term health effects, including cancer and adverse effects on reproduction.4) Previous study showed that long-term pesticide exposure is significantly related to cancer incidence, such as leukemia, lung, pancreatic, colon, rectal, bladder, prostate, and skin cancer.5)

Measuring pesticide metabolites in urine samples collected before and after pesticide exposure has been frequently used to evaluate pesticide exposure.<sup>6)</sup> However, this method does not represent the impact on health. Oxidative stress biomarkers have been used to link pesticide exposure with the risk of chronic diseases such as cancer, inflammatory disease, and neurodegenerative diseases.7-9) Two such oxidative stress biomarkers, malondialdehyde (MDA) and 8-hydroxy-2'deoxyguanosine (8-OHdG) have been commonly measured in human biological samples. MDA is the end products of lipid peroxidation<sup>10)</sup> and 8-OHdG is one of the predominant forms of free radicalinduced oxidative lesions, and has therefore been widely used as a biomarker for oxidative stress and carcinogenesis.<sup>11)</sup> For example, increased levels of lipid peroxides in the serum and urine<sup>7,9)</sup> as well as DNA damage<sup>8,9,12,13)</sup> have been detected following pesticide exposure. However, studies on the level of oxidative stress in agricultural workers in Korea are lacking.

The use of protective behaviors may reduce exposure to agricultural pesticides and potentially minimizes adverse health outcomes. 14) The United States Environmental Protection Agency (US EPA) has established agricultural worker protection standard (WPS) to inform workers and handlers about pesticide safety, to provide protections from potential exposure to pesticides, and to mitigate exposures that do occur.<sup>15)</sup> The protective behaviors include washing hands before eating or using the restroom; wearing protective clothing to minimize skin contact with pesticide residue at work; showering and changing clothes immediately after

work; and washing work clothes separately from other laundry.16 Unlike the US, where the proportion of agricultural workers exposed to pesticides is low by introducing aviation control systems on large scale farms, there are many selfemployed farmers who spray pesticides directly in Korea.<sup>17)</sup> The Rural Development Administration is conducting safety education for farmers, and the Korea Occupational Safety and Health Agency (KOSHA) provide tips on safe pesticide use and first aid treatment for pesticide poisoning accidents. However, they are not offer differentiated protection method according to crop and pesticide application, and the specification of personal protective equipment are not standardized. 17) Studies on Korean farmers have shown that the actual personal protective equipment (PPE) wearing rate is poor because it is too hot and uncomfortable.<sup>17)</sup> Furthermore, there is a lack of awareness on the toxicity of pesticides and the necessity of wearing protective clothing. [18,19]

In the present study, we determined the levels of MDA and 8-OHdG in urine samples collected from agricultural workers in Korea and evaluated the contribution of protective measures on such exposure. Although the study population may not be representative of all agricultural workers in Korea owing to regional specificity, the results of this study will be useful in developing management plans for pesticides in the agricultural worker population.

#### II. Materials and Methods

# 1. Study population and description of the study area

A study population was recruited from Gyeonggi (site A) and Jincheon (site B) provinces in the Korean peninsula between July and August 2014. Cultivation in enclosed space production facilities (e.g., greenhouses) and rice farming are the representative agricultures in Korea, and therefore

Table 1. Demographic characteristics and the details of protective measures of study population

		Site A number (%) Site B number (%)				
	Variables	Male	Female	Male	Female	
All samples		15 (100%)	15 (100%)	15 (100%)	15 (100%)	
	≤ 49 years	3 (20.0%)	3 (20.0%)	0 (0.0%)	1 (6.6%)	
A	50 ~ 59 years	7 (46.7%)	8 (53.4%)	3 (20.0%)	2 (13.3%)	
Age	60 ~ 69 years	2 (13.3%)	2 (13.3%)	2 (13.3%)	5 (33.4%)	
	≥ 70 years	3 (20.0%)	2 (13.3%)	10 (66.7%)	7 (46.7%)	
X7 C	≤ 10	2 (13.3%)	1 (6.6%)	1 (6.6%)	4 (26.7%)	
Years of farming	11 ~ 20	2 (13.3%)	7 (46.7%)	4 (26.7%)	1 (6.6%)	
	≥ 21	11 (73.4%)	7 (46.7%)	10 (66.7%)	10 (66.7%)	
History of	Non-smoker	3 (20.0%)	13 (86.7%)	2 (13.3%)	14(93.4%)	
smoking	Past or present smoker	12 (80.0%)	2 (13.3%)	13 (86.7%)	1 (6.6%)	
Alcohol	Non-drinker	5 (33.3%)	13 (86.7%)	7 (46.7%)	7 (46.7%)	
consumption	Past or present drinker	10 (66.7%)	2 (13.3%)	8 (53.3%)	8 (53.3%)	
	Chemical-resistant hats	8 (53.4%)	10 (66.7%)	12 (80.0%)	12 (80.0%)	
	Protective eyewear	2 (13.3%)	0 (0.0%)	1 (6.6%)	6 (40.0%)	
Wearing	Respirator	3 (20.0%)	2 (13.3%)	1 (6.6%)	7 (46.7%)	
personal protective	Long-sleeved shirts	5 (33.4%)	6 (40.0%)	9 (60.0%)	10 (66.7%)	
equipment	Long-pants	3 (20.0%)	7 (46.7%)	3 (20.0%)	10 (66.7%)	
	Chemical-resistant gloves	3 (20.0%)	2 (13.3%)	5 (33.3%)	9 (60.0%)	
	Chemical-resistant footwear	10 (66.7%)	12 (80.0%)	14 (93.4%)	14 (93.4%)	
	Compliance with safe use standards of pesticides	10 (66.7%)	7 (46.7%)	13 (86.7%)	10 (66.7%)	
Protective behavior	Changing clothes after spraying pesticides	9 (60.0%)	9 (60.0%)	14 (93.4%)	12 (80.0%)	
	Showering immediately after spraying pesticides	10 (66.7%)	9 (60.0%)	14 (93.4%)	12 (80.0%)	
	Spraying pesticides in back wind	10 (66.7%)	8 (53.3%)	13 (86.7%)	12 (80.0%)	
	Having frequent breaks	12 (80.0%)	9 (60.0%)	10 (66.7%)	13 (86.7%)	

two regions were chosen based on considerations. Site A is close to the city, and approximately 80-90% grow cucumbers tomatoes in greenhouses. Site B is a rural area and serves as a cropland for rice. A total of 78 subjects were recruited; however, 18 individuals were excluded from the analysis because of missing relevant information (n=60, 30 males and 30 females) (Table 1). Written statements of informed consent were obtained from all subjects before they participated in the study.

#### 2. Questionnaire survey and urine sampling

An extensive questionnaire was developed to gather information on demographic characteristics of the participants (sex, age, years of farming, history of smoking, and alcohol consumption) and the details of pesticide use (wearing PPE, and protective behaviors following pesticide use). The wearing PPE and protective behaviors were investigated for 7 items (chemical-resistant hats, protective eyewear, respirator, long-sleeved shirts, long-pants, chemicalresistant gloves, and chemical-resistant footwear) and 5 items (compliance with safe use standards of pesticides, changing clothes after spraying pesticides, showering immediately after spraying pesticides, spraying pesticides with back wind, and having frequent breaks), respectively, based on the WPS. Details of the PPE characteristics<sup>20)</sup> are shown in Table 2. One-on-one interviews by a trained

Table 2. Description of personal protective equipment for agricultural working condition

Terminology	Description						
Chemical-resistant hats	- Chemical resistant hat with a wide brim						
Protective eyewear	<ul> <li>Goggles</li> <li>Face shield</li> <li>Safety glasses with front, brow, and temple protection</li> <li>Full-face respirator</li> </ul>						
Respirator	<ul> <li>Self-contained breathing apparatus</li> <li>Full-facepiece air-purifying respirator</li> <li>Half-mask air-purifying respirator</li> <li>Filtering facepiece</li> </ul>						
Long-sleeved shirts	- Loose-fitting, one- or two-piece chemical-resistant garment that covers, at a minimum, the						
Long-pants	entire body except head, hands, and feet						
Chemical-resistant gloves	- Protective gloves specified on the pesticide product labeling (gloves made of leather, cotton, or other absorbent materials may not be worn while performing handler activities unless gloves made of these materials are listed as acceptable for such use on the pesticide product labeling)						
Chemical-resistant footwear	<ul> <li>Chemical resistant shoes</li> <li>Chemical resistant boots</li> <li>Chemical resistant shoe coverings worn over shoes or boots</li> </ul>						

interviewer were conducted at the time of urine sampling. The type of crop treated, the commercial names of the pesticides they had used, and the frequency of pesticides application were also recorded. Participants were instructed on the proper self-collection of the first-void urine (i.e., filling the first 20 mL in a marked cup), and sent to be laboratory within 4 hours. Urine samples were stored at -80°C prior to analysis.

# 3. Analysis of urinary MDA and 8-OHdG concentrations

Urinary levels of MDA were measured using OxiSelect<sup>TM</sup> thiobarbituric acid reactive substances (TBARS) assay kit (STA-330; Cell Biolabs Inc., San Diego, CA, USA) following the manufacturer's instructions. Briefly, aliquots of urine samples (100 μL) or standards were added to the TBA reagent and heated in a water bath at 95°C for 60 min. Samples were cooled in ice water and centrifuged at 3,000 rpm for 15 min. Absorbance was measured at 532 nm using a spectrophotometric plate reader (TECAN infinite® M 200, TECAN Group ltd., Männedorf, Switzerland).

Urinary concentrations of 8-OHdG were measured using OxiSelect<sup>TM</sup> oxidative DNA damage ELISA kit (STA-320; Cell Biolabs Inc., San Diego, CA, USA) following the manufacturer's instructions. Briefly, urine samples (50 µL) or standards were added to the wells of the 8-OHdG conjugate-coated plate and incubated at 25°C for 10 min. Anti-8-OHdG antibody, secondary antibody-enzyme conjugate, and substrate solution were added to each well and incubated at room temperature. Finally, a stop solution was added into each well, and absorbance was measured at 450 nm using a spectrophotometric plate reader (TECAN infinite® M 200). The urinary concentrations of MDA and 8-OHdG were adjusted for creatinine levels in the urine.

#### 4. Statistical analysis

Statistical analyses were performed using SAS, version 9.3 (SAS Institute Inc., Cary, NC, USA). The concentrations of MDA and 8-OHdG were found to be skewed and were log-transformed before the analysis. Independent t-test and Dunnett's analysis of variance (ANOVA) were used to evaluate the influence of variables (site, sex, age, years of farming, history of smoking, alcohol consumption, wearing PPE, and protective behavior) on urinary concentrations of MDA and 8-OHdG. For this analysis, the number of PPE adopted by a farmer is simply categorized into 3 groups without considering their efficiency of protection of exposure to pesticide as follows; less than 2, 2~3, and more than 4. The protective behavior was also categorized by 2 or less, 3~4, and 5.

The relationship between protective measures (number of wearing PPE and performing protective behavior) and urinary concentrations of oxidative stress biomarkers was evaluated using regression analysis. Two variables (number of wearing PPE and performing protective behavior) that were found significant determinant of urinary MDA and 8-OHdG concentrations based on simple regression analysis (p<0.05) were included in the multiple regression analysis. We adjusted models for sex, age, site, history of smoking, and alcohol consumption to consider the possibility for differences in MDA and 8-OHdG concentrations by these variables. In the regression analysis, original values of the number of wearing PPE (1 to 7 items) and performing protective behavior (1 to 5 items) were included in the model. P values less than 0.05 were considered significant.

#### III. Results

The demographic and lifestyle information (i.e., age, history of smoking, and alcohol consumption), and pesticide characteristics obtained from the questionnaire (i.e., years of farming, wearing PPE and protective behaviors) was summarized in Table 1. Site A was composed of the subjects under the age of 59 (70%), while site B mainly had subjects over 60 years of age (80%). Most of the participants answered that they have been engaged in agriculture for more than 21 years (64%). The average number of pesticide application days per year was 26 days in site A and 18 days in site B. The most frequently used PPE were chemicalresistant footwear (83%), chemical-resistant hats (70%), and long-sleeved shirts (50%). Fewer participants reported regular use of long-pants (38%), chemical-resistant gloves (32%), respirator (22%), and protective eyewear (15%). Regarding protective behaviors, the majority mentioned that they normally showered immediately after direct contact with a pesticide (75%), changed their clothes after spraying pesticides (73%), took frequent breaks (73%), and sprayed pesticides with back wind (72%).

The urinary MDA and 8-OHdG concentrations of the study participants are summarized in Table 3. The creatinine-adjusted median urinary levels were 9.37 nmol/mg creatinine for MDA and 19.06 ng/ mg creatinine for 8-OHdG. Significant differences in urinary MDA levels were found among distinct groups based on the sites, sexes, history of smoking, number of PPE worn, and number of protective behavior performed. In significant differences in urinary 8-OHdG levels were observed among different sites and alcohol consumption. Urinary levels of MDA were higher in male farmers living in site A, while those of 8-OHdG were higher in male farmers living in site B.

The creatinine-adjusted concentrations of MDA and 8-OHdG in participants by the number of PPE worn and the number of protective behaviors performed are shown in Fig. 1. **Participants** wearing less number of PPE and performing less number of protective behaviors tended to have greater concentrations of MDA. The wearing of PPE and protective behavior were found to be significant determinants of MDA and 8-OHdG in urine after the adjustment for age, sex, site, history of smoking, and alcohol consumption in multiple regression analysis (Table 4).

**Table 3.** Urinary malondialdehyde (MDA) and 8-hydroxy-2'-deoxyguanosine (8-OHdG) in the participating adult population

Variables N			MDA (nmol/mg creatinine)					8-OHdG (ng/mg creatinine)						
		N (%)	25%	50%	75%	90%	95%	<i>p</i> -value	25%	50%	75%	90%	95%	<i>p</i> -value
All samples		60 (100%)	5.44	9.37	14.44	20.93	22.58	-	13.30	19.06	25.07	29.35	33.34	-
Site	A	30 (50%)	8.44	10.45	14.95	19.33	23.17	0.03*	11.22	14.42	20.58	24.35	28.76	-0.00*
	В	30 (50%)	4.19	6.25	12.68	20.93	22.00		15.75	24.77	26.94	33.34	35.15	
Sex -	Male	30 (50%)	8.44	10.91	16.86	21.74	28.38	0.04*	13.32	19.50	25.06	28.11	33.88	- 0.95
Sex	Female	30 (50%)	4.98	6.94	12.59	16.50	20.80	0.04	12.39	17.85	25.09	29.63	32.80	
	≤49	7 (11%)	6.79	8.44	8.93	10.37	10.37		12.59	15.71	24.55	36.66	36.66	- 0.13
A ~~	50~59	20 (34%)	7.08	10.28	15.36	19.33	36.12	0.52	10.93	13.85	20.69	25.82	29.46	
Age -	60~69	11 (18%)	5.06	9.87	14.60	17.22	20.80	- 0.52 -	13.32	15.92	24.84	27.73	29.84	
<del>-</del>	≥70	22 (37%)	4.66	9.12	14.61	22.00	23.17		15.94	24.46	26.94	29.42	33.88	
	≤10	8 (13%)	6.46	8.46	14.86	50.76	50.76	_	15.26	25.33	31.32	36.66	36.66	0.13
Years of farming	11~20	14 (23%)	5.83	10.28	13.15	14.29	14.95		11.22	15.02	19.85	25.06	25.09	
iaining -	≥21	38 (64%)	4.92	9.74	15.78	21.49	23.17	•	13.34	20.10	25.31	29.28	33.88	
History of	Past or present smoker	24 (40%)	6.31	10.45	14.60	22.00	28.38	0.04*	13.34	20.32	25.59	29.42	33.88	0.37
smoking -	Non-smoker	36 (60%)	5.02	8.51	13.22	17.18	20.80	•	12.35	16.33	24.90	28.76	32.80	,
Alcohol consumption	Past or present drinker	28 (47%)	6.12	9.32	14.45	22.00	28.38	0.46	13.34	20.32	25.59	29.42	33.88	0.01*
	Non-drinker	32 (53%)	4.95	9.45	13.94	20.80	21.49	•'	11.58	15.81	24.09	28.76	32.80	
N. 1 C	<2	16 (27%)	14.16	17.11	21.74	28.38	50.76		15.93	20.93	25.41	29.28	36.66	
Number of wearing PPE-	2~3	21 (35%)	8.07	9.87	12.59	14.60	14.95	0.00*	12.59	15.71	24.70	26.94	33.88	0.54
	≥4	23 (38%)	2.88	5.06	7.07	9.04	10.37	•	12.31	16.73	25.31	29.42	29.84	•
Number of protective behavior	≤2	16 (27%)	11.74	16.41	21.27	28.38	50.76		16.46	24.09	29.02	35.15	36.66	
	3~4	13 (21%)	6.82	8.53	10.37	17.22	22.00	0.00*	15.94	19.59	24.70	25.52	26.88	0.10
	5	31 (52%)	3.41	6.07	11.28	13.15	14.60		12.59	14.60	25.06	27.73	32.80	

Asterisk (\*) indicates significant difference between groups (p < 0.05) using independent t-test or ANOVA. -: Not available.

#### IV. Discussion

In general, compared to Korea's general population, pesticide sprayers had higher levels of MDA and 8-OHdG. Although our study does not have a control group, the median concentrations of MDA (13.19  $\mu$ mol/L in males and 7.52  $\mu$ mol/L in females) and 8-OHdG (22.58 ng/mL) obtained in the present study were higher than those reported in the general population of Korea (8.14  $\mu$ mol/L in males and 5.52  $\mu$ mol/L in females for MDA<sup>21)</sup> and 4.46 ng/mL for 8-OHdG<sup>22)</sup>). Previous studies have

also shown that the levels of MDA<sup>23-24)</sup> and 8-OHdG<sup>8,9,13,25)</sup> in the urine or plasma samples collected from pesticide sprayers are greater than those of the control participants. A pilot study of pesticide applicators and farm workers working in fruit orchards (e.g., apples and pears) in Oregon showed that serum MDA and urinary 8-OHdG levels were higher than those in the controls.<sup>10)</sup> These data suggest that agricultural workers are frequently exposed to higher concentrations of chemicals that induce oxidative stress and DNA damage.

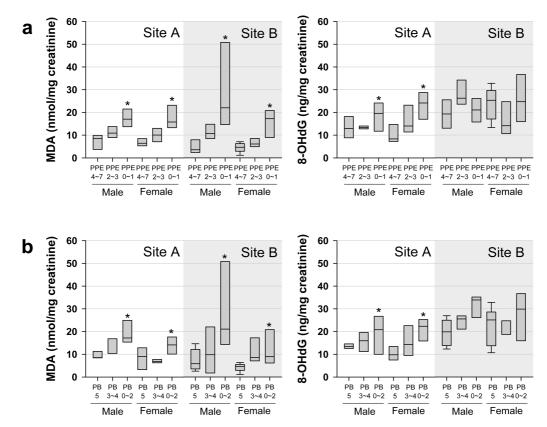


Fig. 1. Urinary malondialdehyde (MDA) and 8-hydroxy-2'-deoxyguanosine (8-OHdG) concentrations in participants by number of wearing personal protective equipment (a) and protective behavior (b). Box indicates interquartile range showing the 25th and 75th percentile values. The black line in the box denotes median value, and black circle denotes  $5^{\text{th}}$  and  $95^{\text{th}}$  percentiles. Asterisk indicates significant difference between groups (p<0.05). PPE: personal protective equipment, PB: protective behavior.

We compared the levels of oxidative stress biomarkers in two different agricultural sites. We found that MDA concentrations were higher in site A, while 8-OHdG concentrations were higher in site B. Since the period of pesticide use varies depending on the cultivated crops and harvesting times, the levels of effective biomarkers can vary. The subjects living in site A mainly produce cucumber and tomatoes, and have been used chlorothalonil fungicide (trade name, Dakonil) and emamectin benzoate insecticide (trade name. APEM). The study participants living in site B cultivated rice and used carbofuran (product name, Furadan) validamycin-a (product and name.

It has been reported that (TBARS) was increased concentration- and time-dependent manner in rat hepatocytes exposed to chlorothalonil. 26-27) addition, MDA was significantly increased in male rats exposed to 2.5 mg emamectin benzoate/kg body weight for 28 days.<sup>28)</sup> These results support those of our study that showed significantly greater MDA concentrations in the urine collected from participants in site A.

The MDA and 8-OHdG levels in the agricultural population of the US, 9,10,13) Greece, 8) Egypt, 29) and Korea<sup>30-31)</sup> from the literature are summarized in Table 5. The urinary concentrations of 8-OHdG

Table 4. Relationships between oxidative biomarker and protective measures based on multiple regression analyses

Oxidative biomarkers	Variables	β	95% confidence interval	p
MDA	Wearing PPE	-4.06	(-0.66, -0.22)	0.0002
(nmol/mg creatinine)	Protective behavior	-2.41	(-0.45, -0.04)	0.0196
8-OHdG	Wearing PPE	-0.19	(-0.15, 0.12)	0.8523
(ng/mg creatinine)	Protective behavior	-3.05	(-0.32, -0.06)	0.0036

Adjusted for sex, age, site, history of smoking, and alcohol consumption. PPE: personal protective equipment.

Table 5. Concentrations of malondialdehyde (MDA) and 8-hydroxy-2'-deoxyguanosine (8-OHdG) in agricultural farmers in worldwide

Country	Biomarker (sample)	Characteristics	N	Sampling year	Age (years)	Arithmetic mean (unit)	Median (unit)	Reference	
USA	) (D)	Farm workers	10	2000	$35.9 \pm 9.7$	-	1.4 (μmol/L)	<del></del>	
	MDA (serum)	Applicators	12		$44.0 \pm 6.9$	-	6.7 (μmol/L)		
		Control	9		$31.4 \pm 11.2$	-	0.29 (µmol/L)		
	MDA (serum)	Knapsack motor sprayer	80	_	20~60	9.50 (mmol/dL)	-	[28]	
Egypt		Conventional motor sprayer	70	2007	20~60	12.6 (mmol/dL)	-		
		Control	60		23~55	5.36 (mmol/dL)	-		
	MDA (urine)	Pesticide exposed farmer	84	2011	adult	0.13 (µmol/g creatinine)	-	[30]	
Korea	MDA (urine)	Farmers	60	2014	62.1 ± 11.3	11.6 (µmol/L) 10.8 (µmol/g creatinine)	10.7 (μmol/L) 9.3 (μmol/g creatinine)	This study	
	8-OHdG (urine)	Farm workers	63	2003	$57.0 \pm 2.9$	3.42 (ng/mL)	-	[13]	
		Control	48	- 2003	$46.0 \pm 1.6$	3.00 (ng/mL)	-		
USA	8-OHdG (urine)	Farm workers	10		$35.9 \pm 9.7$	-	190 (nmol/L)	[9, 10]	
		Applicators	12	2000	$44.0 \pm 6.9$	-	85 (nmol/L)		
	(urnic)	Control	9		$31.4 \pm 11.2$	-	23 (nmol/L)		
	8-OHdG (whole blood)	Sprayers	80	_	adult	-	17.31 (pg/mL)		
Greece		Rural and urban residents	206	2010	-	-	13.24 (pg/mL)	[8]	
Korea	8-OHdG (urine)	Pesticide exposed farmer	84	2011	adult	0.93 (ng/mg creatinine)	-	[30]	
	8-OHdG (urine) 8-OHdG (urine)	Farm workers (sunblock users)	48	2015	66.8 ± 10.9	7.76 (ng/g creatinine)	-	F2.13	
		Farm workers (non-sunblock users)	38	2015	68.3 ± 11.3	7.93 (ng/g creatinine)	-	[31]	
		Farmers	60	2014	62.1 ± 11.3	19.8 (ng/mL) 19.4 (ng/mg creatinine)	22.5 (ng/mL) 19.0 (ng/mg creatinine)	This study	

MDA: malondialdehyde, 8-OHdG: 8-hydroxy-2'-deoxyguanosine, -: Not available.

(median: 19.06 ng/mg creatinine) in the Korean population were higher than those in the US population (median: 3.42 ng/mL).<sup>13)</sup> These observations may suggest that the Korean agricultural population

is exposed to higher concentrations of pesticides than the US agricultural population. The greater use of pesticides by Korean farmers could partly explain this result: the use of pesticides per unit area in Korea (11.3 kg/ha in 2014) is more than that in the US.<sup>32)</sup> The observation that males significantly greater concentrations of MDA and 8-OHdG compared to females is comparable to the results of other study,8) who showed that the median 8-OHdG concentrations in males (15.39 pg/mL) were significantly greater than those in females (10.24 pg/mL). Although apparent reasons for sexdependent differences are not known, the greater use of pesticides by males could partly explain these observations.

Since pesticides can be exposed mainly through the dermal route, it is important to wear PPE when spraying pesticides and to perform protective behavior. In the present study, participants wearing a greater number of PPE and performing more protective behaviors had significantly less levels of MDA and 8-OHdG. Participants mostly wore chemical-resistant footwear and hats; however, the wearing rates of chemical-resistant respirator, and protective eyewear were the lowest. Consistent with our results, among protective clothing use, the wearing rate of protective hats and footwear was the highest in Gyeongju farmers.<sup>33)</sup> In the case of Carolina farmers in the US, the majority of participants wore hats (93%) and water-resistant outerwear (68%).16) It has been also reported that farmers in Chungnam area in Korea,34) as well as in Greece,8) frequently wore protection footwear, hats, and gloves. These wearing rates were lower than or similar to those of our study participants. Although the reduction of pesticide exposure should not rely solely on PPE use, safety training on PPE helps to encourage farmworkers to use PPE. In addition, 68% of the farmworkers did not wear gloves while working in the fields; therefore, it is necessary to solve this problem in Korean farmers.

#### V. Conclusion

In summary, it is important to reduce pesticide exposure and induction of oxidative stress by using appropriate protective measures, since pesticides are mostly absorbed through dermal contact. The results of this study showing that wearing more numbers of PPE and performing protective behaviors could significantly reduce the accumulation levels of MDA and 8-OHdG. However, there are some limitations that must be considered for interpreting the results of this study. First, the research subjects are limited to small sample size of farm workers in rural areas; therefore, it is difficult to generalize them as whole farmers. In addition, it was difficult to control the effective ingredients of pesticides, because we recruited subjects as voluntary participants. The lack of control groups that are not exposed to pesticides, absence of pesticide exposure assessment, one-time urine sampling, and self-report of behavior are also limitations. Given the importance of wearing PPE and performing protective behavior in pesticide users, further studies are needed to investigate the influence of protective measures to pesticide exposure with an appropriate control group and a larger study population.

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