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Effect of perchloroethylene, smoking, and race on oxidative DNA damage in female dry cleaners

Mark Toraason [a,*], Mary Ann Butler [a], Avima Ruder [a], Christy Forrester [a], Lauralynn Taylor [a], David L. Ashley [b], Patty Mathias [a], Kate L. Marlow [a], Kenneth L. Cheever [a], Edward Krieg [a], Howard Wey [c]

[a] *The National Institute for Occupational Safety and Health, 4676 Columbia Parkway, Cincinnati, OH 45226, USA*
[b] *The National Center for Environmental Health, Atlanta, GA, USA*
[c] *South Dakota State University, Brookings, SD, USA*

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Abstract

Perchloroethylene (PERC) is used widely as an industrial dry cleaning solvent and metal degreaser. PERC is an animal carcinogen that produces increased incidence of renal adenomas, adenocarcinomas, mononuclear cell leukemia, and hepatocellular tumors. Oxidative DNA damage and lipid peroxidation were assessed in 38 women with (dry cleaners) or without (launderers) occupational exposure to PERC. PERC exposure was assessed by collecting breathing zone samples on two consecutive days of a typical work week. PERC levels were measured in blood drawn on the morning of the second day of breathing zone sample collection in dry cleaners and before a typical workday in launderers. Blood PERC levels were two orders of magnitude higher in dry cleaners compared to launderers. A significant correlation was noted between time weighted average (TWA) PERC and blood PERC in dry cleaners ($r = 0.7355$, $P < 0.002$). 8-Hydroxydeoxyguanosine (8-OHdG), ng/mg deoxyguanosine (dG) in leukocyte nuclear DNA was used as an index of steady-state oxidative DNA damage. Urinary 8-OHdG, μg/g creatinine was used as an index of oxidative DNA damage repair. Urinary 8-epi-prostaglandin $F_{2\alpha}$ (8-epi-PGF), ng/g creatinine was used as an index of lipid peroxidation. The mean ± S.D. leukocyte 8-OHdG in launderers was 16.0 ± 7.3 and was significantly greater than the 8.1 ± 3.6 value for dry cleaners. Urinary 8-OHdG and 8-epi-PGF were not significantly different between dry cleaners and launderers. Unadjusted Pearson correlation analysis of log transformed PERC exposure indices and biomarkers of oxidative stress indicated a significant association in launderers between blood PERC and day 1 urinary 8-OHdG ($r = 0.4661$, $P < 0.044$). No significant associations between exposure indices and biomarkers were evident in linear models adjusted for age, body mass index, race, smoking (urinary cotinine, mg/g creatinine) and blood levels of the antioxidants Vitamin E and β-carotene. The mean ± S.D. leukocyte 8-OHdG value in control white women was 17.8 ± 7.4 and was significantly greater than the 11.8 ± 5.9 in control black women. No significant differences by race were evident for the other biomarkers. Smoking status was not significantly associated with any of the oxidative damage indices. Results indicate a reduction in oxidative DNA damage in PERC exposed dry cleaners relative to launderers, but PERC could not clearly be defined as the source of the effect. Published by Elsevier B.V.

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1. Introduction

Perchloroethylene (PERC) (CAS No. 127-18-4) is used widely as an industrial dry cleaning solvent and

* Corresponding author. Tel.: +1-513-533-8207; fax: +1-513-533-8138.
E-mail address: mtoraason@cdc.gov (M. Toraason).

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metal degreaser. The National Institute for Occupational Safety and Health Administration (OSHA) [1] estimates that over 1.5 million industrial workers in the USA are potentially exposed to PERC.

PERC is an animal carcinogen associated with increased incidence of renal adenomas or adenocarcinomas in male rats, increased incidence of mononuclear cell leukemia in rats, and a dose related increase in hepatocellular tumors in mice [2–4].

Human studies have consistently demonstrated increased risk among dry cleaning workers for urinary tract and other cancers [5–8]. Case control population-based studies also have shown increased cancer risk for those exposed to dry cleaning solvents [9,10]. Although PERC exposure has been associated with increased risk for a wide range of cancers [11–14], the human data for PERC-only exposures are equivocal. Therefore, it has not been possible to determine whether the excess cancers are due to PERC.

PERC is oxidized by cytochrome P450 to produce trichloroacetyl chloride, trichloroethanol, and trichloroacetic acid (TCA) [15]. TCA is a metabolite of PERC in both man and experimental animals, and is also a rodent hepatocarcinogen [16–20]. TCA induces lipid peroxidation and oxidative DNA damage, which have been implicated in its carcinogenesis [21–24]. Although rats did not exhibit elevated oxidative DNA damage following a single exposure to PERC [25], the ability of Vitamin E to protect against PERC toxicity in mice [26] suggests that oxidative stress may occur to some degree following PERC exposure.

DNA is subject to continuous oxidative damage from oxygen radicals generated during normal cellular respiration [27]. Unrepaired DNA damage can lead to mutation and possibly cancer [28–33]. Environmental exposures that increase the level of damage above background levels increase the potential for unrepaired lesions to become permanent mutations [34–36]. This concept has led to the use of oxidative DNA damage in the assessment of exposures to chemicals that are capable of inducing oxidative stress [37]. Oxygen-based radicals can produce a variety of DNA lesions [38], but the 8-hydroxydeoxyguanosine (8-OHdG) adduct has been used most extensively as a biomarker of exposure [39–42].

Presently, biomonitoring for PERC exposure in the workplace is accomplished by breath, blood, or urine analysis of the parent compound or metabolites [4,43]. These represent biomarkers of exposure, but provide little information on potential activation of these compounds or interactions with target tissues. If PERC induces oxidative stress, then biomarkers of oxidative damage could serve as biomarkers of exposure for workers in the dry cleaning industry. Therefore, the objective of the present study was to determine if increased oxidative damage, including the formation of oxidative DNA adducts, could be detected in female dry cleaners with PERC exposure.

Ideally, an assessment of chemically induced oxidative DNA damage would measure 8-OHdG in DNA of target tissue [32,41,44,45]. In the present study, peripheral leukocytes were assessed as a potential target tissue and as a surrogate for liver and kidney. Urinary 8-OHdG was also measured as an index of DNA repair of oxidative DNA adducts [46,47]. An additional assessment of oxidative stress associated with PERC exposure was made by measuring excretion of the isoprostane, 8-epi-prostagladin $F_{2\alpha}$ (8-epi-PGF). 8-Epi-PGF is formed by the peroxidation of lipid membranes [48,49] and is emerging as a sensitive, specific, and reliable marker of in vivo lipid peroxidation and oxidative damage [50].

2. Materials and methods

2.1. Subjects

This study was approved by the NIOSH Human Subjects Review Board. The study population is described in Table 1. Eighteen dry cleaning workers and 20 laundry workers (controls) were recruited from seven shops in and around Cincinnati, OH, USA. All participants were women, under the age of 70 who had worked in the dry cleaning or laundry industry for at least 1 year. Dry cleaning workers were machine operators or pressers who worked in the area of the dry cleaning machines. All facilities used third generation dry-to-dry machines. Laundry workers had not previously worked in a facility with exposure to PERC. Laundry workers were matched by race, smoking status, and age (±5 years when possible) to already selected dry cleaners. Ethnic differences, noted as white (Caucasian of western European descent) or black (African–American), were of similar distribution.

Table 1
Data summary

	Laundry	Dry cleaner
Demographic		
Facilities	3[a]	4
Workers	20	18
Race (white/black)	14/6	13/5
Current smokers	10	10
Age	39 ± 9[b]	40 ± 13
Body mass index (kg/m^2)	29 ± 4	28 ± 5
Exposure indices		
Day 1 PERC TWA (ppm)	<0.02[c]	2.4 ± 3.4
Day 2 PERC TWA (ppm)	<0.02	3.8 ± 5.3
Biomarkers		
Blood PERC (μg/l)[e]	0.19 ± 0.44	74.81 ± 104.27*
Cotinine (mg/g creatinine)[d]	1504 ± 879	1642 ± 1216
β-Carotene (μg/dl blood)	8.7 ± 5.6	19.9 ± 24.0
Vitamin E alpha (mg/l blood)	8.4 ± 3.1	9.0 ± 2.2
Vitamin E beta (mg/l blood)	2.4 ± 1.5	1.9 ± 0.9
Leukocyte 8-OHdG (ng/mg dG)[e]	16.0 ± 7.3	8.1 ± 3.6*
Pre-shift 8-epi-PGF (ng/g creatinine)[f]	535 ± 258	604 ± 314
Post-shift 8-epi-PGF (ng/g creatinine)[f]	433 ± 141	417 ± 174
Pre-shift 8-OHdG (μg/g creatinine)[f]	2.8 ± 1.7	2.8 ± 1.2
Post-shift 8-OHdG (μg/g creatinine)[f]	3.5 ± 1.3	3.1 ± 1.2

[a] Values are number of facilities, workers who are white or black, and number of smokers.
[b] All other values are mean ± S.D.
[c] PERC TWA in laundries was below the limit of detection (0.023 ppm).
[d] Continine means are from smokers as continine was below the limit of detection in nonsmokers.
[e] Blood was drawn on day 2 for PERC and leukocyte 8-OHdG measurement.
[f] Pre- and post-shift urines were collected on day 2 for measurement of 8-epi-PGF or 8-OHdG.
* Mean is significantly different than corresponding mean for laundry workers (t-test, $P < 0.05$).

2.2. PERC time weighted average (TWA) air concentration sampling

Personal breathing zone samples were collected from women on Wednesday (day 1) and Thursday (day 2) of a typical work week. Sampling analysis was performed to determine the concentration of PERC in air according to NIOSH method 1003 [51] described as follows. Each woman wore a personal sampling pump used to draw a known volume of air through a charcoal tube attached to clothing near her breathing zone. Personal samples collected PERC on the 100 mg/50 mg coconut-shell charcoal tubes. Each charcoal sample was collected throughout a typical work shift for a 120 min interval with an air flow rate of 0.1 l/min and a volume of 12 l. Samples were analyzed for PERC using carbon disulfide desorption with analysis by gas chromatography with flame ionization detector. It was assumed that exposures were constant over a workday for calculation of 8 h TWA. The 8 h TWAs for days 1 and 2 were averaged for correlation analysis.

2.3. Blood and urine samples

Venous blood was collected from dry cleaners before work following three consecutive days of PERC exposure, and from launderers on a typical work day. On the same day, urine was collected pre- and post-shift. Following aliquoting, urine samples were flash frozen and stored at −70 °C until analyzed. Blood was obtained via venipuncture into tubes containing EDTA at the same time the pre-shift urine samples were collected. Blood samples were fractionated into various components the same day they were collected and fractions or extraction products were frozen and stored at −70 °C until analyzed. Blood samples for measuring PERC were collected in gray-top vacutainers that had been previously processed to remove volatile contaminants. These whole blood samples were kept at refrigerator temperature until analysis.

2.4. Blood PERC

PERC was measured by the method of Ashley et al. [52]. Volatile compounds were separated from the blood matrix by purge and trap concentration. Detection, identification, and quantification of PERC was done by GC/MS. Quantification was achieved by isotope dilution and reference to commercially available standard compounds [52].

2.5. 8-OHdG in leukocyte DNA and urine

Peripheral blood leukocyte DNA was extracted from 2 ml whole blood on the day it was drawn using a WAKO DNA extractor WB kit (WAKO Chemicals

USA Inc., Richmond, VA, USA). DNA extracted from leukocytes was hydrolyzed by incubation with nuclease P1 and alkaline phosphatase. 8-OHdG was measured in urine and hydrolyzed leukocyte DNA using HPLC and electrochemical detection by ESA Laboratories Inc. (Chelmsford, MA, USA). DNA extracted from leukocytes is expressed as the ratio of 8-OHdG, ng to deoxyguanosine (dG), mg. Urinary 8-OHdG is expressed as μg/g creatinine. Creatinine was determined by ESA Laboratories Inc.

2.6. Urinary 8-epi-PGF

Urinary 8-epi-prostaglandin $F_{2\alpha}$ was evaluated using commercially available enzyme immunoassay (EIA) kits from Cayman Chemical (Ann Arbor, MI, USA). Prior to 8-epi-PGF immunoassay analysis, a BenchMate® II robotic workstation was used to automate sample preparation. Bond Elute® 500 mg C18 SPE columns were conditioned with MeOH and H_2O pH 3. Samples were filtered using a Whatman GD/X 0.2 μm nylon syringe filter and 10 ml loaded onto columns. Following this the columns were washed sequentially with 10 ml of H_2O and heptane prior to elution with 10 ml ethyl acetate:heptane (1:1). The ethyl acetate:heptane fraction was dried using Na_2SO_4 prior to normal phase cleanup. Bond Elute® 500 mg silica gel SPE columns were conditioned with 5 ml MeOH and ethyl acetate prior to loading the dried ethyl acetate:heptane fraction onto the column. A 5 ml ethyl acetate:MeOH (1:1) fraction containing >90% of 8-epi-PGF was collected, reduced to dryness under N_2 and stored at $-20\,^{\circ}C$ until ELISA analysis. All samples were assayed in triplicate. Urinary 8-epi-PGF is expressed as ng/g creatinine.

2.7. Urinary cotinine

Urinary cotinine was measured by ELISA method performed by LabCorp of America (Hudson, OH, USA).

2.8. Vitamin E and β-carotene

Alpha-, and beta–gamma-tocopherols, and β-carotene were measured in serum by ethanolic protein denaturation followed by hexane extraction and HPLC analysis. The method used an isocratic mobile phase C-18 reversed-phase columns and diode ray detection of carotenoids and fluorescent detection of tocopherols. Analysis was performed by Quest Diagnostics (Wood Dale, IL, USA).

2.9. Statistical analysis

SAS® (Version 8, SAS Institute, Cary, NC, USA) was used to analyze the data. Pearson correlation coefficients were used to determine the unadjusted relationships between the PERC exposure indices and the biomarkers of oxidative damage. Differences in oxidative damage between smokers and nonsmokers, exposed and unexposed workers, and races were determined with *t*-tests. Multiple linear regression models were used to determine the adjusted relationships between the PERC exposure indices and the biomarkers of oxidative damage. There was a separate model for each combination of exposure index and biomarker. The covariates included in the models were alpha-, and beta–gamma-tocopherols, β-carotene, body mass index, urinary cotinine, age and race. The exposure measures of PERC TWA and blood PERC were transformed using the base 10 logarithm for the Pearson correlation analysis and in the multiple linear regression models. Relationships were considered statistically significant if $P < 0.05$.

3. Results

Table 1 presents the demographics of the workers investigated for an association between PERC exposure and oxidative DNA damage. Twenty female laundry workers without prior occupational exposure to PERC were compared to 18 female workers in dry cleaning facilities that used PERC. Laundry workers were matched by race, smoking status, and age (±5 years) to already selected dry cleaners. There was no difference between the two groups for these variables. Smoking status was confirmed by measuring urinary cotinine, which was below the level of detection in women reporting they were presently nonsmokers. To assess antioxidant status of the participants, serum levels of β-carotene, and Vitamin E alpha and beta were measured. There were no significant differences between launderers and dry cleaners

Table 2
Oxidative damage in PERC exposed and control workers stratified by smoking status

PERC exposure (*n*)	Urinary 8-epi-PGF (ng/g creatinine)	Urinary 8-OHdG (μg/g creatinine)	Leukocyte 8-OHdG/dG (ng/mg dG)
Control, nonsmoker (10)	476 ± 199	3.1 ± 1.5	17.1 ± 7.4 (a)
Control, smoker (10)	479 ± 141	3.1 ± 1.3	14.8 ± 7.5 (a)
Control, smoker >20 cigarettes per day (5)	594 ± 97	3.2 ± 1.2	14.4 ± 7.5 (a)
Exposed, nonsmoker (8)	439 ± 177	2.8 ± 1.3	7.8 ± 2.6 (b)
Exposed, smoker (10)	539 ± 256	3.1 ± 1.1	8.4 ± 4.4 (b)
Exposed, smoker >20 cigarettes per day (4)	676 ± 319	3.0 ± 0.8	11.2 ± 6.0 (a and b)

Values are mean ± S.D. The values shown are means of the average of pre- and post-shift values. No significant differences were noted for urinary 8-epi-PGF or 8-OHdG whether pre- and post-shift values were evaluated individually or combined. Values without a letter in common are significantly different.

for cotinine, β-carotene, or Vitamin E. Although not statistically significant, the mean value for β-carotene in dry cleaners was more than double the mean value in launderers. This is due to a serum β-carotene level of 106 μg/dl measured in one dry cleaner. The mean ± S.D. without outlier was 14.88 ± 10.94. As the outcome of data analysis was the same with or without this value it was included in the final analysis. Leukocyte 8-OHdG was significantly reduced in dry cleaners compared to laundry workers. In contrast, urinary levels of 8-epi-PGF or 8-OHdG did not differ between launderers and dry cleaners.

Table 2 presents the effects of smoking status on the biomarkers of oxidative damage. No significant differences between smokers and nonsmokers were detected for leukocyte 8-OHdG, urinary 8-epi-PGF or urinary 8-OHdG.

Table 3 presents a comparison of oxidative damage in women stratified by exposure and race. In the present study, black launderers had significantly lower levels of leukocyte 8-OHdG than white launderers. Black dry cleaners also appear to have had lower levels of leukocyte 8-OHdG than white dry cleaners, but the difference is not statistically significant. No differences among the groups were detected for urinary 8-epi-PGF, or urinary 8-OHdG.

PERC TWAs for employees in two of three laundry facilities were below the limit of detection (0.023 ppm). In the third facility, the average for the employees was below the limit of detection, but some workers had detectable levels (<0.1 ppm) of PERC. Because PERC TWAs for launderers were near or below the limit of detection, the TWAs from launderers were not included in the correlation analysis or the linear models. Dry cleaners had average blood PERC levels that were more than two orders of magnitude greater than those in launderers. This occurred despite employees in dry cleaning facilities having PERC TWAs that were below 5 ppm. The PERC blood concentrations were measured in samples taken pre-shift on day 2. In dry cleaners, there was a significant association between log transformed

Table 3
Oxidative damage in PERC exposed and control workers stratified by race

PERC exposure (*n*)	Urinary 8-epi-PGF (ng/g creatinine)	Urinary 8-OHdG (μg/g creatinine)	Leukocyte 8-OHdG (ng/mg dG)
Control, black (6)	411 ± 90	2.8 ± 1.1	11.8 ± 5.9 (a)
Control, white (14)	506 ± 188	3.2 ± 1.5	17.8 ± 7.4 (b)
Exposed, black (5)	457 ± 171	3.1 ± 1.4	5.9 ± 1.2 (a)
Exposed, white (13)	509 ± 246	2.9 ± 1.1	9.0 ± 3.9 (a)

Values are mean ± S.D. The values shown are means of the average of pre- and post-shift values. No significant differences were noted for urinary 8-epi-PGF or 8-OHdG whether pre- and post-shift values were evaluated individually or combined. Values without a letter in common are significantly different.

Table 4
Unadjusted Pearson correlation coefficients for PERC exposure indices and biomarkers of oxidative damage

	PERC TWA	Blood PERC launderers	Blood PERC dry cleaners
Pre-shift			
Urine 8-epi-PGF (ng/g creatinine)	0.3587 (a)	−0.3199	0.0932
	0.157 (b)	0.211	0.741
	17 (c)	17	15
Urine 8-OHdG (μg/g creatinine)	−0.1199	0.4661	0.3079
	0.635	0.044	0.246
	18	19	16
Leukocyte 8-OHdG (ng/mg dG)	0.3296	−0.3032	0.2016
	0.196	0.237	0.471
	17	17	15
Post-shift			
Urine 8-epi-PGF (ng/g creatinine)	0.4688	0.0797	0.1843
	0.058	0.761	0.511
	17	17	15
Urine 8-OHdG (μg/g creatinine)	−0.3577	0.4296	0.0878
	0.145	0.075	0.747
	18	18	16

Values are (a) correlation coefficient, (b) *P*-value, and (c) sample size.

PERC blood and log transformed TWA ($r = 0.7355$, $P = 0.0012$).

Table 4 presents assessment by Pearson correlation of the association between indices of PERC exposure and biomarkers of oxidative damage. There were no statistically significant relationships between PERC TWA and biomarkers of oxidative damage. Blood PERC was significantly associated with urinary 8-OHdG, but only within the launderer group.

Linear models that included adjustment for age, body mass index, race, urinary cotinine, serum Vitamin E, and serum β-carotene were used to assess the relationships between variables shown in Table 4. The significant direct association identified by Pearson correlation analysis between urinary 8-OHdG and blood PERC among launderers was not statistically significant in the linear model.

In the 20 linear models, Vitamin E had a significant interaction in five models, race in four models, and cotinine in one model. These findings and the consistency between non-adjusted and adjusted analyses indicates that the terms used to adjust the linear models had limited effect on the relationship between the PERC exposure indices and the oxidative damage biomarkers.

4. Discussion

4.1. PERC exposure

The PERC TWA for 17 of 18 dry cleaner participants was below 5 ppm. NIOSH classifies PERC as an occupational carcinogen [53], and recommends exposure concentrations be minimized and that the number of workers exposed should be limited. More specific guidelines are provided by the Occupational Safety and Health Administration which regulates PERC exposure with an 8 h TWA permissible exposure limit (PEL) of 100 ppm. The American Conference of Governmental Industrial Hygienists (ACGIH) recommends a threshold limit value (TLV) of 25 ppm over an 8 h exposure [43]. ACGIH [43] also recommends a biological exposure index for PERC in blood of 0.5 mg/l. The mean value for PERC in blood of dry cleaners in the present study was 0.078 mg/l. Therefore, mean PERC exposures in the individuals examined were below levels recommended to protect workers. Personal exposures were also lower than those of other assessments of dry cleaning operations reporting geometric mean levels of 3.3, 10 and 13 ppm in dry cleaning operations [54–56]. Although the PERC TWA mean in this study was lower

than these published values, elevated levels of PERC were still evident in blood. Therefore, in the present study, oxidative DNA damage was compared to log transformed personal PERC levels and blood PERC concentration.

4.2. Oxidative DNA damage

The hypothesis at the onset of this investigation was that exposure to PERC in dry cleaning facilities would elevate oxidative stress. It was expected that this would be manifested as increased oxidative DNA damage in peripheral leukocytes and elevated excretion of byproducts of DNA repair products in urine. Confirmation of increased oxidative stress would be evident by the presence of increased levels of lipid peroxidation products (8-epi-PGF) in urine. With the present small population, the coefficient of variation for the oxidative stress biomakers for the two populations (Table 1) averaged 42%. Nonetheless, results demonstrate that dry cleaners with PERC exposure have decreased oxidative DNA damage in leukocytes relative to launderers. This result was not evident within the two exposure groups using Pearson correlation analysis or adjusted linear model analysis. The absence of associations within these two analyses leaves open the possibility that differences in leukocyte 8-OHdG may be attributable to unidentified differences between dry cleaners and launderers other than blood PERC.

While the leukocyte 8-OHdG assessment suggests that PERC exposure resulted in reduced damage from oxidative stress or increased repair of oxidative DNA damage, the other measures of oxidative stress do not support this conclusion. The assessment of urinary 8-epi-PGF is consistent with the conclusion that there is no association between PERC exposure in the study population and oxidative stress.

Focusing only on the leukocyte 8-OHdG, assessment of oxidative DNA damage appears to have revealed a biological response to PERC that would generally be considered beneficial. The decreased oxidative DNA damage could be the result of increased removal of hydroxylated guanosine by specific base and nucleotide excision repair pathways [57]. Excretion of excised 8-OHdG in the urine would provide an index of oxidative DNA damage repair. However, urinary 8-OHdG can arise from a variety of sources including bacteria [58]. Few studies assessing 8-OHdG as a biomarker of oxidative DNA damage in humans, have measured 8-OHdG in both urine and leukocyte DNA. Gedik et al. [58] reported a statistically significant direct association between urinary and lymphocyte 8-OHdG. In contrast, two other recent studies [59,60] reported non-statistically significant inverse associations between urinary excretion of 8-OHdG and leukocyte 8-OHdG. The discrepancy among these studies may be attributed to the fact that the former study was in a control population, and latter studies were investigating exposure related responses. In the present study, no statistically significant association was found between urinary and leukocyte 8-OHdG using Pearson correlation analysis. If increased repair was responsible for decreased leukocyte 8-OHdG, a corresponding increase in urinary 8-OHdG would be expected. This is not the case. This suggests that decreased leukocyte 8-OHdG is the result of increased antioxidant capacity. It may be that PERC exposure up regulates genes that result in a reduction of endogenous oxidative DNA damage. The effect is such that it is detectable in DNA, but undetectable in the byproducts of oxidative damage excreted in the urine.

4.3. Tobacco smoking

The three biomarkers of effect used in the present study have been reported to be associated with smoking. Results in Table 2 indicate that none of the biomarkers of oxidative stress were effective in demonstrating a statistically significant difference between smokers and nonsmokers. In well-controlled studies, 8-epi-PGF has been consistently elevated in smokers relative to nonsmokers [48,49], and elevation of 8-epi-PGF is dependent upon the number of cigarettes smoked [61]. Smokers in the present study would not be considered heavy smokers. Nine of the 20 smokers reported that they smoked 10 or fewer cigarettes per day. While 8-epi-PGF was not statistically significantly elevated in smokers, data in Table 2 demonstrates an apparent dose-dependent response that may not be achieving statistical significance because of small sample size.

Although several studies have demonstrated a direct association between smoking and 8-OHdG [42], occupational studies have rarely demonstrated a positive association [37,62]. Recently, two studies [60,63] reported significantly decreased leukocyte 8-OHdG in

smokers relative to nonsmokers. van Zeeland et al. [63] attributed the effect to enhanced rapid repair. Nai et al. [60] reported that 21 healthy smokers in their study had reduced levels of lymphocyte 8-OHdG and decreased urinary excretion of 8-OHdG relative to healthy nonsmokers. They [60] concluded that oxidative stress imposed by cigarette smoke had a low impact on pathways involved in DNA damage, which is consistent with present results.

4.4. Race

The finding of increased leukocyte 8-OHdG in black women relative to white women is consistent with several studies demonstrating reduced levels of oxidative damage in individuals of African descent relative to Caucasians. In examining the effect of vitamin supplementation on 184 healthy nonsmoking adults, Haung et al. [64] reported that urinary excretion of 8-OHdG was decreased in African–Americans relative to Caucasians. Simon et al. [65] examined another oxidative DNA adduct, 5-hydroxymethyl-2′-deoxyuridine, and found levels were reduced in populations of African descent relative to Caucasians. Lahiri et al. [66] reported increase resistance to oxidative stress in Africans relative Caucasians, and Collins et al. [67] examined 8-OHdG in leukocyte DNA in five western European countries and found significant differences among countries, and a north-to-south declining gradient. While present results demonstrate a marked difference in leukocyte 8-OHdG between black and white women, reduced leukocyte 8-OHdG also was associated with PERC exposure in both races.

4.5. Summary

The primary objective of the present study was to assess the association between PERC and oxidative damage. Contrary to the original hypothesis, women exposed to PERC while working in dry cleaning shops had decreased levels of oxidative damage relative to launderers without PERC exposure. It is important to note that PERC exposures were well below recommended exposure limits. To what extent an absence of hypothesized response can be attributed to low exposures is not evident. Additional assessment of alternative measures of oxidation products and antioxidants did not provide an insight into the mechanism of the inverse association. It may be that a factor(s) not assessed in the present study is responsible for the difference in oxidative DNA damage between launderers and dry cleaners. The present study also revealed a race-dependent difference in leukocyte 8-OHdG in the worker population that was independent of PERC exposure.

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