

Urinary 8-oxo-7, 8-dihydro-2'-deoxyguanosine and Biopyrrins Levels among Construction Workers with Asbestos Exposure History

Rie YOSHIDA^{1*}, Yasutaka OGAWA¹, Izuru SHIOJI², Xiaozhong YU¹, Eiji SHIBATA³,
Ippei MORI¹, Hitoshi KUBOTA¹, Akiko KISHIDA⁴ and Naomi HISANAGA¹

¹National Institute of Industrial Health, Kawasaki, Kanagawa, 6–21–1, Nagao, Tama-ku, Kawasaki, Kanagawa 214-8585, Japan

²Shino-Test Corporation, Sagamihara, Kanagawa, Japan

³Department of Medical Technology, Nagoya University School of Health Sciences, Nagoya, Aichi, Japan

⁴Kitasato University, Sagamihara, Kanagawa, Japan

Received December 18, 2000 and accepted February 23, 2001

Abstract: It has been suggested that oxidative stress is associated with the cancers caused by asbestos. Since construction workers are sometimes exposed to low levels of asbestos, we investigated whether oxidative stress was elevated in construction workers who had been exposed to low levels of asbestos. The subjects were 48 Japanese construction workers. The defined asbestos-exposed group consisted of subjects who had the history of suspected exposure to asbestos and were diagnosed to have irregular opacities or pleural plaques. We measured the amount of 8-oxo-7, 8-dihydro-2'-deoxyguanosine (8-oxodG) and biopyrrins in the urine of the subjects. The results showed that 8-oxodG and biopyrrins levels in the defined asbestos-exposed group were higher, although they were not statistically significant, than those in the control group. In addition, the urinary 8-oxodG levels tended to correlate positively with the duration of suspected exposure to asbestos. These results suggest that even low-level asbestos exposure may induce oxidative stress and that the resulting oxidative stress might be related to lung cancer in construction workers.

Key words: Asbestos, Construction workers, Oxidative stress, 8-oxo7, 8-dihydro-2'-deoxyguanosine, Biopyrrins

It has been suggested that oxidative stress is associated with cancers resulting from exposure to asbestos because asbestos induce oxidative DNA damage in both culture cells^{1–5)} and rats⁶⁾. It has also been demonstrated that 8-oxo-7, 8-dihydro-2'-deoxyguanosine (8-oxodG), one of the markers of oxidative DNA damage⁷⁾, increased in the urine or blood samples of workers exposed to asbestos at asbestos plants^{8–10)}. Construction workers have a high incidence of exposure to asbestos from construction materials¹¹⁾. Although the average total asbestos fiber concentration in the lungs of construction workers (6×10^6 fibers/g dry lung tissue)¹¹⁾

was shown to be lower than those of asbestos plants workers (7×10^8 fibers/g dry lung tissue)¹¹⁾, it was considered to be a high enough level to cause an excess incidence of lung cancer¹¹⁾. Anttila *et al.*¹²⁾ also reported that 69% of lung cancer patients with a history of occupational exposure to asbestos were construction workers.

Since construction workers are considered to be exposed to lower levels of asbestos compared to asbestos plant workers¹¹⁾, our research subjects, who are construction workers, are also suspected to have been exposed to lower levels of asbestos than those workers indicated in previous reports^{8–10)}. In this study, we investigated whether the level of oxidative stress was elevated in construction workers who

*To whom correspondence should be addressed.

Table 1. Characteristics of subjects

	Group C	Group A	
The number of subject	41	7	
Age	59.4 (\pm 6.6)	60.1 (\pm 7.1)	N.S.
Smoker (%)	87.7	100	N.S.
The number of cigarette smoked per day	23.3	26.7	N.S.
Subjects drunk alcohol more than twice/week (%)	72.3	67	N.S.

N.S. No significant difference between the control (Group C) and the defined asbestos-exposed (Group A) groups.

had been exposed to low levels of asbestos.

The subjects were 48 male Japanese construction workers, age from 43 to 72 (mean 59.5). All subjects were interviewed by an experienced occupational health doctor concerning their occupational histories, drinking and smoking habits, medical history, and other lifestyle factors. The duration of suspected exposure to asbestos for each subject was evaluated based on these data. Another doctor who had a specialty in pneumoconiosis diagnosed irregular opacities and pleural plaques by using chest x-rays films. The defined asbestos-exposed group (Group A, n=7, mean age 60.1) consisted of subjects who were evaluated to have the history of suspected exposure to asbestos, and were diagnosed to have irregular opacities or pleural plaques. The rest of subjects were classified as the control group (Group C, n=41, mean age 59.4).

Urine was sampled from each subject in the morning and was frozen immediately after sampling. 8-OxodG was measured using anti-8-oxodG monoclonal antibody, N45.1 (Japan Institute for the Control of Aging, Fukuroi, Shizuoka, Japan). Urinary biopyrrins, which is the oxidative metabolite of the antioxidant bilirubin¹³⁾ was selected as an indicator to estimate the level of systemic oxidative stress and measured using anti-bilirubin monoclonal antibody 24G7 (Shino-Test corporation, Sagamiara, Kanagawa, Japan).

Table 1 shows characteristics of Group A and C. All workers of Group A were smokers who smoked 26.7 cigarette/day. Sixty seven percent of them drunk alcohol three or more times a week. In Group C, 87.7% workers were smokers who smoked 23.3 cigarette/day and 72.3% of them drunk alcohol three or more times a week. There was no significant difference in age, the frequency of alcohol consumed, the number of smokers and the number of cigarette smoked/day between Groups A and C.

The urinary 8-oxodG level of Group A ($8.0 \pm 0.9 \mu\text{g/g}$ creatinine) was a little higher than that of Group C ($6.4 \pm 2.1 \mu\text{g/g}$ creatinine) (Fig. 1). The difference was near the border of confidence interval but was not statistically

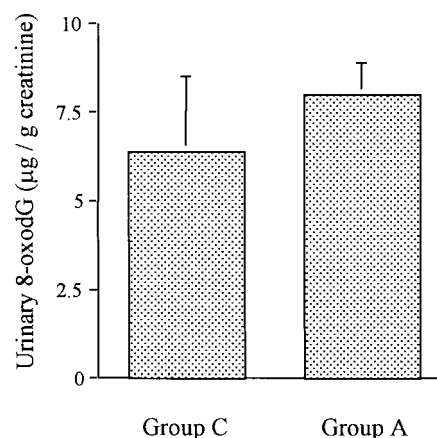


Fig. 1. Comparison of the amount of urinary 8-oxodG between the defined asbestos-exposed group (group A) and the control group (group C). The amount of urinary 8-oxodG was measured by ELISA and the value was corrected by creatinine.

significant ($p = 0.057$). We also investigated the relationship between the duration of suspected exposure to asbestos and the urinary 8-oxodG levels of the Group A. The 8-oxodG levels of Group A tended to correlate positively with the duration of suspected exposure to asbestos ($r=0.7$, $p=0.1$) (Fig. 2). We further investigated whether the 8-oxodG levels were associated with age. There were no correlation between age and the urinary 8-oxodG levels of Group C ($p=0.71$). The urinary biopyrrins level of the Group A ($2.3 \pm 1.5 \mu\text{mol/g}$ creatinine) was slightly higher than that of the Group C ($1.9 \pm 1.1 \mu\text{mol/g}$ creatinine), but the difference was not statistically significant ($p=0.35$).

Although the results were not statistically significant, urinary 8-oxodG and biopyrrins levels of the Group A were higher than those of the Group C. In addition, urinary 8-oxodG levels of the Group A increased with the duration of suspected exposure to asbestos. Since the urinary 8-oxodG levels were not correlated with age, increased urinary 8-oxodG levels of Group A might depend on the suspected

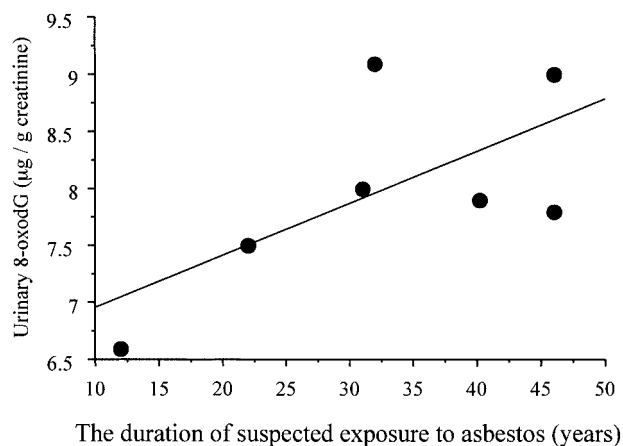


Fig. 2. A relationship between the duration of suspected exposure to asbestos and the amount of urinary 8-oxodG.

exposure to asbestos. Considering that asbestos had induced oxidative stress in cells¹⁻⁵, rats⁶, and asbestos plant workers who were exposed to high levels of asbestos⁸⁻¹⁰, our results suggest that exposure to even low levels of asbestos also increase oxidative stress level. Since 8-oxodG induces genetic mutation¹⁴, the increased oxidative stress induced by asbestos may be associated with lung cancer in construction workers. The measurement of urinary 8-oxodG might be a reliable supplemental way to assess the cancer risk of construction workers who have been exposed to asbestos in addition to routine chest x-ray diagnosis. Due to the limitation of the sample size in this study, further studies are needed to determine the validity of our recommendation.

References

- 1) Okayasu R, Takahashi S, Yamada S, Hei TK, Ullrich RL (1999) Asbestos and DNA double strand breaks. *Cancer Res* **59**, 298–300.
- 2) Murata-Kamiya N, Tsutsui T, Fujino A, Kasai H, Kaji H (1997) Determination of carcinogenic potential of mineral fibers by 8-hydroxydeoxyguanosine as a marker of oxidative DNA damage in mammalian cells, *Int Arch Occup Environ Health* **70**, 321–6.
- 3) Fung H, Kow YW, Van Houten B, Mossman BT (1997) Patterns of 8-hydroxydeoxyguanosine formation in DNA and indications of oxidative stress in rat and human pleural mesothelial cells after exposure to crocidolite asbestos, *Carcinogenesis* **18**, 825–32.
- 4) Kamp DW, Israbian VA, Preusen SE, Zhang CX, Weitzman SA (1995) Asbestos causes DNA strand breaks in cultured pulmonary epithelial cells: role of iron-catalyzed free radicals, *Am J Physiol* **268** (3 Pt 1), L471–80.
- 5) Takeuchi T, Morimoto K (1994) Crocidolite asbestos increased 8-hydroxydeoxyguanosine levels in cellular DNA of a human promyelocytic leukemia cell line, HL60. *Carcinogenesis* **15**, 635–9.
- 6) Yamaguchi R, Hirano T, Ootsuyama Y, Asami S, Tsurudome Y, Fukada S, Yamato H, Tsuda T, Tanaka I, Kasai H (1999) Increased 8-hydroxyguanine in DNA and its repair activity in hamster and rat lung after intratracheal instillation of crocidolite asbestos, *Jpn J Cancer Res* **90**, 505–9.
- 7) Shigenaga MK, Gimeno CJ, Ames BN (1989) Urinary 8-hydroxy-2'-deoxyguanosine as a biological marker of in vivo oxidative DNA damage. *Proc Natl Acad Sci U S A* **86**, 9697–701.
- 8) Tagesson C, Chabiuk D, Axelson O, Baranski B, Palus J, Wyszynska K (1993) Increased urinary excretion of the oxidative DNA adduct, 8- hydroxydeoxyguanosine, as a possible early indicator of occupational cancer hazards in the asbestos, rubber, and azo-dye industries. *Pol J Occup Med Environ Health* **6**, 357–68.
- 9) Takahashi K, Case BW, Dufresne A, Fraser R, Higashi T, Siemiatycki J (1994) Relation between lung asbestos fibre burden and exposure indices based on job history. *Occup Environ Med* **51**, 461–9.
- 10) Marczyński B, Rozynek P, Kraus T, Schlosser S, Raithel HJ, Baur X (2000) Levels of 8-hydroxy-2'-deoxyguanosine in DNA of white blood cells from workers highly exposed to asbestos in Germany. *Mutat Res* **468**, 195–202.
- 11) Ebihara I, Hirata M, Hisanaga N, Shibata E, Sakai K (1997) Respiratory findings of construction workers exposed to asbestos. In: *Health and toxicology* ed. by Cheremisinoff PN, 93–126, Gulf publishing company, Houston.
- 12) Anttila S, Karjalainen A, Taikina-aho O, Kyyronen P, Vainio H (1993) Lung cancer in the lower lobe is associated with pulmonary asbestos fiber count and fiber size. *Environ Health Perspect* **101**, 166–70.
- 13) Yamaguchi T, Shioji I, Sugimoto A, Komoda Y, Nakajima H (1994) Chemical structure of a new family of bile pigments from human urine. *J Biochem (Tokyo)* **116**, 298–303.
- 14) Kuchino Y, Mori F, Kasai H, Inoue H, Iwai S, Miura K, Ohtsuka E, Nishimura S (1987) Misreading of DNA templates containing 8-hydroxydeoxyguanosine at the modified base and at adjacent residues. *Nature* **327**, 77–9.