Ann. Occup. Hyg., Vol. 53, No. 2, pp. 161–165, 2009

doi:10.1093/annhyg/men077

Exposure to Hydrogen Peroxide and Eye and Nose Symptoms Among Workers in a Beverage Processing Plant

G. MASTRANGELO[1]*, R. ZANIBELLATO[2], E. FADDA[1], J. H. LANGE[3], L. SCOIZZATO[1] and R. RYLANDER[4]

[1]*Department of Environmental Medicine and Public Health, University of Padua, 35128 Padua, Italy;* [2]*Medical Surveillance Unit, Ditta San Benedetto SPA, 30037 Scorzè, Italy;* [3]*Envirosafe Training and Consultants, Pittsburgh, PA 15239, USA;* [4]*BioFact Environmental Health Research Center, 44391 Lerum, Sweden*

Received 24 January 2008; in final form 7 November 2008; published online 24 December 2008

Objectives: Two cross-sectional studies were undertaken on workers in a beverage processing plant to investigate the association between low H_2O_2 exposure and symptoms of irritation (2005 study) and to investigate the effect of wearing respiratory protection (2006 study). Methods: The study comprised 69 workers exposed to H_2O_2 in sterile chambers and 65 unexposed controls. The exposure was assessed from measurements and work task information from employment records. The severity of work-related symptoms was evaluated using questionnaires. Data were analyzed by the Student's *t*-test, multiple linear regression and analysis of variance for repeated measures of symptoms. Results: Symptoms of eye, nose and throat irritation were significantly ($P < 0.001$) more severe among exposed workers compared to controls. Exposure values were occasionally above American Conference of Governmental Industrial Hygienists (ACGIH) threshold limit value–time-weighted average (TLV-TWA) in the sterile chambers. The relationship between the severity of symptoms and the number of entrances in the chambers was significant ($P < 0.0001$) in 2005 but not in 2006, when respirators were used during work in the sterile chamber. No differences were found between exposed who entered a sterile chamber in 2005 but not in 2006 and exposed who entered a sterile chamber both in 2005 and 2006. This suggests that respirators provided an efficient protection and that the irritative effects of exposure to H_2O_2 in 2005 did not disappear after 1 year. Conclusions: The source of risk was exposure in the sterile chamber, even though the time of exposure was generally only ~30 min. To ensure complete worker protection, there is a need for a short-term exposure limit for H_2O_2 in addition to the existing ACGIH TLV–TWA value.

Keywords: eye inflammation; hydrogen peroxide; nose inflammation; respiratory protection; symptom questionnaire

Introduction

Hydrogen peroxide (H_2O_2) is a gas commonly used in industry for disinfection, bleaching and as a general oxidizing agent. A number of organizations have published values on maximum allowable concentrations. The American Conference of Governmental Industrial Hygienists (ACGIH) states a TLV–time-weighted average (TWA) of 1 p.p.m. (1.4 mg m^{-3}) (ACGIH, 2008). This value is based mainly on results from mice inhalation studies suggesting that 1 p.p.m. would be sufficient for protection against irritation (Gagnaire *et al.*, 2002). The German Research Foundation has proposed a maximum allowable concentration (MAK) value of 0.5 p.p.m. (0.74 mg m^{-3}) (DFG, 2008) and in UK, the value suggested by the Health and Safety Executive was 1 p.p.m. for a TWA 8-h exposure and 2 p.p.m. as a short-term exposure level (STEL) (HSE, 2005). The database for the values cited above is, however, rather meager and some data indicate that these threshold values do not provide optimal protection for the workers. Rough skin on the hands and decolorized hairs were observed in workers exposed to H_2O_2 concentrations <1 p.p.m. (Suenaka *et al.*, 1984).

In a previous study in a beverage processing plant, lung function was examined in workers exposed to

*Author to whom correspondence should be addressed.
Tel: +39-049-821-2543; fax: +39-049-821-2542;
e-mail: giuseppe.mastrangelo@unipd.it

airborne H_2O_2 at levels in compliance with the ACGIH TLV–TWA value (Mastrangelo *et al.*, 2005). The findings suggested that exposure did not induce lung function changes and thus supported the TLV–TWA value. In spite of this, the workers at the plant reported widespread work-related symptoms from the eyes and upper airways, suggesting that the value was not sufficient for complete worker protection.

In view of this, new studies were undertaken on the workers in the same bottling plant to (i) assess the presence of work-related symptoms related to H_2O_2 exposure and (ii) investigate the possible beneficial effect of wearing respiratory protection.

Methods

Plant characteristics and study subjects

There were two major sections in the plant. One included several aseptic bottling departments each having a built-in sterile chamber, in which 65 workers were regularly exposed to H_2O_2. The other consisted of several lines of automatic bottling of mineral water, where 69 workers unexposed to H_2O_2 acted as controls. The work task descriptions for each worker were provided by the plant management.

Measurements of H_2O_2

The measurements of H_2O_2 were performed once a year in 2005 and 2006 by means of stationary air samplers located 150 cm above the floor in several areas of the beverage bottling lines (sterile chamber, dressing room used to change for personal protective equipment, cap room and production lines). In addition, personal samplers (Escort Elf Pump, MSA Company, Pittsburgh, PA, USA) were used in the sterile chamber during the short interventions of maintenance, lasting in average 30 min.

The samples were analyzed for their content of H_2O_2 using an occupational hygiene standard method (INRS, 2008). Using concentrations obtained from stationary air samplers, a TWA level was calculated on the basis of the time spent in the corresponding areas of the plant, according to information provided by the unions and plant managers. The concentration values from personal samplers were reported as task-length average.

Symptom studies

In the first study, the 65 workers exposed to H_2O_2 were compared with the 69 unexposed workers. The presence of symptoms was determined using a standardized questionnaire on irritation in the eyes, nose, throat and skin. The symptoms experienced during the last week were graded for severity using a numerical score from 1 to 10 (1 = none and 10 = unbearably severe symptoms). The 10-point visual analog scale was reported in the questionnaire beside each symptom. Smoking habits were explored. The subjects completed the questionnaires themselves in September 2005 independently from the H_2O_2 measurements.

In the second study, to evaluate the effectiveness of a prevention measure, workers were encouraged to wear respirators (full-face mask, equipped with filter A1B1E1K1 suitable for organic gases and vapors, inorganic gases, acid gases and ammonia; Willson respiratory protectors, Santa Ana, CA, USA) when working in the sterile chambers. Workers for this study were recruited from the same group of workers who participated in the symptoms study in 2005, and of these, 60 exposed and 58 unexposed participated. The same questionnaire as in 2005 was used to assess the extent of symptoms. A second questionnaire investigating the actual use of respirators was completed by workers who worked in the sterile chambers in 2006. Both questionnaires were self-administered after a brief presentation in December 2006.

Statistical analysis

The statistical analyses were performed using the SPSS system and comprised the calculations of

(i) Multiple linear regression models (one for each symptom), in which the severity score was the dependent variable, while the independent variables were age, gender, smoking, hours spent in the bottling line and number of occasions having entered a sterile chamber from July 2004 to June 2005 (unexposed workers were scored as 0). With 24 symptoms and 11 risk factors, the possible associations were 264 (= 24 × 11). In order to avoid random significances, the Bonferroni method was used to decrease the alpha level of each individual statistical test. A *P*-value of 0.0001 was chosen for significance to ensure that the overall risk for a number of tests remained ∼0.05.

(ii) Analysis of variance for repeated measures, which simultaneously analyzed the repeated measures of severity scores of symptoms collected in 2005 and 2006 in four groups of workers: 58 unexposed workers (group 1), 9 workers exposed to H_2O_2 along the bottling lines but never entered a sterile chamber (group 2), 10 exposed who entered a sterile chamber in 2005 but not in 2006 (group 3) and 40 exposed who entered a sterile chamber both in 2005 and 2006 (group 4). To avoid false-positive results, a *P*-value of 0.001 was chosen for significance. Whenever a *F*-test reached the statistical significance, a pairwise comparison of the four groups was made using the post-hoc test of Bonferroni.

(iii) Geometric mean, geometric standard deviation (GSD) and an upper limit of 95% confidence interval were calculated for the airborne levels of H_2O_2 in the different areas of the bottling lines. The Shapiro–Wilk *W*-test was used to assess the log-normal distribution of airborne H_2O_2 values and the Kruskal–Wallis test (chi-square) to determine group homogeneity.

Results

Table 1 reports the results for airborne levels of H_2O_2 in different areas of the aseptic departments: bottling lines and sterile rooms in 2005 and 2006. The average value in the sterile room was lower in 2006 than in 2005, but the GSD was larger, yielding a wider range of values. The airborne levels were log-normally distributed (W equal to 0.92, $P = 0.49$ and 0.99 and $P = 0.99$, for the log-transformed values measured along bottling lines and sterile chambers, respectively) and homogenous among workers (chi-square 4.90, $P = 0.42$).

Table 2 reports a breakdown of the workers examined in relation to background variables. Most of the variables were similar in the two groups except the proportion of smokers which was higher in the exposed group.

Table 3 shows that workers exposed to H_2O_2 reported more severe symptoms compared to controls. Statistically significant ($P < 0.001$) differences were found for teary eyes, sore eyes, redness of eyes, throat irritation, nasal secretion, nasal obstruction, nasal itching, sneezes and cough.

Table 4 shows that there was a significant ($P = 0.0001$) exposure–effect relationship between the number of entries in a sterile chamber in the past 12 months and the severity of sore eyes, teary eyes, redness of eyes, nasal secretion and throat irritation.

Regarding the second study, where the workers were encouraged to wear respirators before entering sterile chambers, adherence was accomplished by 38 of 40 workers ever entering sterile chambers. There were no significant exposure–effect relationships between the number of entries in the sterile chamber and severity of symptoms (data not shown).

Figure 1 illustrates the severity score in relation to the year of study (2005 and 2006).

In groups 1 and 2, the average severity was always just ∼2 (mild or occasional symptoms). In the groups 3 and 4, it was ∼4 (steady but easily tolerable) for nasal symptoms and roughly 6 (moderately bothersome—hard to tolerate) for eye symptoms. The four lines represent the time trend of a particular symptom (sore eyes, redness of eyes, teary eyes, nasal secretion, sneezes and nasal itching) in each group of workers. The trend lines are overlapping or very close for groups 1 and 2, as well as for groups 3 and 4. For workers wearing respirators during work in sterile chambers in 2006 (group 4), the scores were lower in 2006 than in 2005 for eye symptoms resulting in downward time trend lines. In the same group, the upward time trend for nasal symptoms could not be related to exposure since a parallel trend was observed in all groups, including unexposed workers.

At analysis of variance for repeated measures (the F_2-test with 3 and 113 degrees of freedom), values were 14.46, 13.35, 15.93, 13.17, 8.14, 7.81 and 6.51 for sore eyes, redness of eyes, teary eyes, nasal secretion, throat irritation, sneezes and nasal itching, respectively (always $P < 0.001$).

Table 5 shows the difference between the means of symptom severity as well as the P-values (the post-hoc test of Bonferroni) at the pairwise comparisons of the four groups of workers. Significant differences in the severity of symptoms were found between control workers and those wearing respirators during work in sterile chambers. This could not be explained by the H_2O_2 exposure (Fig. 1). There were no significant differences between groups 1 and 2, suggesting that exposure along the bottling lines was most likely acceptable. Likewise there were no differences between groups 3

Table 1. Summary statistics for airborne levels of hydrogen peroxide (mg m^{-3}) in different areas of the aseptic plants in 2005 and 2006: geometric mean, GSD and upper limit of 95% confidence interval (CI)

	Bottling lines[a]		Sterile chambers[b]	
	2005	2006	2005	2006
Geometric mean	0.13	0.07	0.82	0.47
GSD	2.11	1.88	1.37	2.24
Upper limit of CI	0.58	0.25	1.53	2.28
No. of samples	6	6	6	6

[a]Stationary air samplers. Sampling time = 330 min.
[b]Personal samplers. Sampling time = task length.

Table 2. Background characteristics for investigated workers in the beverage processing plant in the two repeated cross-sectional studies

	First study (of 2005)				Second study (of 2006)			
	64 Exposed		69 Controls		59 Exposed		58 Controls	
	%	Mean	%	Mean	%	Mean	%	Mean
Females	9		13		10		12	
Age (years)		34.3		36.1		35.8		36.7
Years smoked		11.3		10.1		11.3		10.1
Cigarettes per day		11.6		11.5		5.1		3.7
Present smokers	55		26		48		31	

Table 3. Differences between exposed and unexposed workers in relation to severity scores of symptoms (significant differences only) reported in 2005

Symptoms	Difference between means	Standard error	*T*-test	*P*-value
Teary eyes	3.77	0.54	7.04	<0.001
Sore eyes	3.30	0.56	5.93	<0.001
Redness of eyes	3.11	0.54	5.80	<0.001
Throat irritation	2.30	0.47	4.89	<0.001
Nasal secretion	1.92	0.48	3.97	<0.001
Nasal obstruction	1.81	0.51	3.56	<0.001
Nasal itching	1.79	0.45	3.94	<0.001
Sneezes	1.57	0.44	3.59	<0.001
Cough	1.51	0.40	3.81	<0.001

Table 4. Different models (one for each symptom) of multiple regression analysis: coefficient of regression (*b*), its standard error [SE(*b*)], test of significance [$T = b/\text{SE}(b)$] and two-tail error probability (*P*-value)

	b	SE(*b*)	*T*-test	*P*-value
Sore eyes	0.229	0.041	5.550	0.0001
Teary eyes	0.226	0.046	4.903	0.0001
Redness of eyes	0.194	0.041	4.790	0.0001
Nasal secretion	0.151	0.035	4.340	0.0001
Throat irritation	0.133	0.034	3.885	0.0001

Dependent variable: severity score of symptom reported in 2005. Independent variable: number of entries in the sterile chamber (the only independent variable entered in the model).

and 4, which suggests that respirators provided an effective protection, but also that the irritative effects of exposure to H_2O_2 in 2005 did not disappear after 1 year.

Discussion

The study has certain shortcomings. The number of workers is relatively small. Because of the strict criteria for significance, some of the differences between the groups could have been real although they did not meet the criteria for the *P*-value used.

The workers were aware of the aims of the study and had themselves initiated the study on symptoms. There is thus a possibility that the extent of and severity of symptoms could be biased. Even if some bias were present, the symptom profile corresponds to what could be expected after exposure to a highly reactive gas in relatively small amounts. This suggests that the reporting was relevant. Furthermore, previous studies in other environments have shown that a standardized questionnaire to record work-related symptoms will yield valid data (Rylander *et al.*, 1990). We thus feel that the symptoms reported in the questionnaires to a large extent represent a real effect among the workers.

Pulmonary effects by H_2O_2 have been described previously in a 41-year-old dairy worker, exposed chronically to an aerosol of hydrogen peroxide ($41\ \text{mg m}^{-3}$) (Kaelin *et al.*, 1988). The absence of such

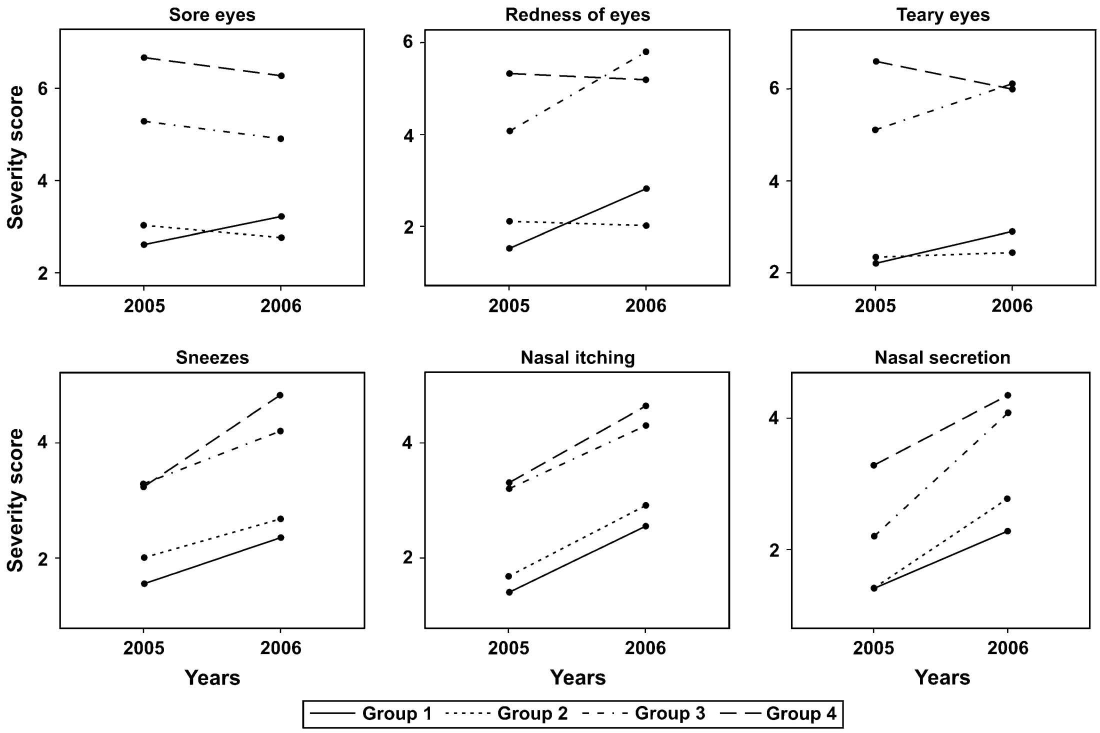

Fig. 1. Severity score of symptoms recorded in 2005 and 2006 in group 1 (controls), group 2 (bottling line), group 3 (sterile chamber in 2005) and group 4 (sterile chamber in 2005 and 2006).

Table 5. Analysis of variance for repeated measures of severity scores of symptoms collected in 2005 and 2006.

Symptoms	Group (i)	Group (j)	Difference between means (i − j)	Standard error	*P*-value
Sore eyes	1	2	0.2	0.980	1.000
		3	−2.19	0.937	0.128
		4	−3.56	0.562	0.000
	3	4	−1.38	0.968	0.948
Redness of eyes	1	2	0.02	0.980	1.000
		3	−2.19	0.937	0.128
		4	−3.56	0.562	0.000
	3	4	−1.38	0.968	0.948
Teary eyes	1	2	0.17	1.007	1.000
		3	−3.04	0.963	0.012
		4	−3.73	0.578	0.000
	3	4	−0.69	0.994	1.000
Nasal secretion	1	2	0.21	0.779	1.000
		3	−1.32	0.745	0.476
		4	−2.69	0.447	0.000
	3	4	−1.38	0.769	0.459
Throat irritation	1	2	−0.05	0.841	1.000
		3	−1.78	0.804	0.173
		4	−2.25	0.483	0.000
	3	4	−0.48	0.830	1.000
Sneezes	1	2	−0.39	0.786	1.000
		3	−1.80	0.752	0.109
		4	−2.08	0.451	0.000
	3	4	−0.28	0.776	1.000
Nasal itching	1	2	−0.33	0.846	1.000
		3	−1.80	0.808	0.167
		4	−2.03	0.485	0.000
	3	4	0.23	0.834	1.000

Post-hoc test of Bonferroni (*P*-value) for the pairwise comparisons of symptoms (means of severity) in the four groups of workers. Group 1, unexposed workers; group 2, workers exposed to H_2O_2 along the bottling lines but never entered a sterile chamber; group 3, exposed who entered a sterile chamber in 2005 but not in 2006 and group 4, exposed who entered a sterile chamber both in 2005 and 2006.

effects in workers exposed to H_2O_2 in the bottling plant examined here is probably due to the lower exposure level (Mastrangelo *et al.*, 2005).

The absence of work-related symptoms among workers wearing respiratory protection could be due to a decrease of the inhaled dose, although it cannot be excluded that the lower average levels in 2006 could play a role. On the other hand, peak values according to the upper limit of confidence interval (Table 1) were higher in 2006 than in 2005. Whatever the case, the results obtained support recommendations to wear respiratory protection in the sterile chambers in the plant.

The symptoms reported by the workers appeared after a relatively brief exposure period (30 min in average). The effect is explainable from a physiological point of view as H_2O_2 is a highly reactive gas where the adverse effects develop quite quickly. The results from the study thus support the need for a short-term exposure limit for full protection of the workers.

The TLV–STEL is the concentration to which workers can be exposed continuously for a short period of time and a value of 2 p.p.m. for 30 min has been suggested in the UK (HSE, 2005) and by the European Union (SCOEL, 2008). STEL values may also be set as TLV–ceiling values. In view of the results from our exposure measurements (Table 1) and the very rapid effect induced by H_2O_2, it is prudent to suggest a STEL value at 2 p.p.m. for a 15 min period to provide a satisfactory protection of workers exposed to H_2O_2.

Conclusions

The source of risk was exposure in the sterile chamber, even though the time of exposure was generally only ~30 min. The presence of symptoms among the workers entering the sterile room even for short period suggests that there is a need for a short-term exposure limit for H_2O_2 exposure.

Funding

University of Padova and the bottling plant.

REFERENCES

ACGIH. (2008) Annual reports of the committees on TLVs and BEIs for year 2007. American Conference of Governmental Industrial Hygienists. publ 0108 A, pp.1–31.

DFG German Research Foundation. (2008) MAK und BAT Werte Liste. Wiley-VCH, Weinheim, Germany. http://www.dfg.de. Accessed October 2008.

Gagnaire F, Marignac B, Hecht G, *et al*. (2002) Sensory irritation of acetic acid, hydrogen peroxide, peroxyacetic acid and their mixture in mice. Ann Occup Hyg; 46: 97–102.

HSE. Health and Safety Executive. London, UK: EH40/2005 Workplace exposure limits.

Institut Nationale de la Recherche et Securité de Travail (INRS). (2008) http://www.inrs.fr—fiche 047. Accessed October 2008.

Kaelin RM, Kapanci Y, Tschopp JM. (1988) Diffuse interstitial lung disease associated with hydrogen peroxide inhalation in a dairy worker. Am Rev Respir Dis; 137: 1233–5.

Mastrangelo G, Zanibellato R, Fedeli U, *et al*. (2005) Exposure to hydrogen peroxide at TLV level does not induce lung function changes: a longitudinal study. Int J Environ Health Res; 15: 313–7.

Rylander R, Peterson Y, Donham K. (1990) Questionnaire evaluating organic dust exposure. Am J Ind Med; 17: 121–6.

SCOEL. (2008) Scientific Committee on Occupational Exposure Limits (SCEL) on hydrogen peroxide. http://www.ser.nl/~/media/Files/Internet/Grenswaarden/SCOEL/GSW354Prov134 Waterstofperoxide.ashx. Accessed October 2008.

Suenaka T, Akaska S, Hirata M. (1984) A survey of occupational exposure to hydrogen peroxide –H2O2 exposure level and its effects on lipid peroxide and its related enzyme activities. Proceedings of Osaka Prefecture Institute of Public Health, Edition of Industrial Health; 22: 9–13.