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Ethylene oxide exposure

Biological monitoring by analysis of alveolar air and blood

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Summary. Occupational exposure to ethylene oxide (ETO) was studied in ten workers employed in a hospital sterilizer unit by testing environmental air, alveolar air and blood during and at the end of the workshift. Alveolar (Ca) and blood (Cb) ETO concentrations were correlated with each other ($r = 0.744$, $n = 36$, $P < 0.001$) and both with the environmental (Ci) concentrations ($r = 0.947$, $n = 144$, $P < 0.001$; $r = 0.827$, $n = 36$, $P < 0.001$). The alveolar retention of ETO (1-Ca/Ci) was equal to 75–80% of the inhaled ETO. In comparison with a blood/air partition coefficient equal in vitro to 90 (SD = 20), the mean Cb/Ca ratio found in the exposed workers was equal to 12–17. During work the blood ETO concentration was, on average, three times the environmental ETO concentration.

Key words: Ethylene oxide – Alveolar air – Blood biological monitoring

Introduction

Ethylene oxide (ETO) is a water soluble, colorless gas, which is widely used as an intermediate in the production of various chemicals. It is also used in small quantities for the sterilization of a number of medical devices in hospitals and industries. Numerous pathological aspects due to irritant, toxic, mutagenic and carcinogenic effects of ETO have been reported in the international literature (Calleman et al. 1978; Hogstedt et al. 1979; Wolfs et al. 1983; Yager et al. 1983; Currier et al. 1984; Laurent et al. 1984; Ohnishi et al. 1985; Sarto et al. 1985). However, there seems to be a paucity of publishied studies on the absorption of ETO in workers occupationally exposed in plants (Ehrenberg et al. 1974; Yager et al. 1983; Hines et al. 1984). In a previous paper

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(Brugnone et al. 1985) we reported our research on the monitoring of ETO in the alveolar air of workers. The purpose of the present investigation has been to shed more light on the biological monitoring of exposure by testing ethylene oxide in the alveolar air and blood of occupationally exposed workers.

Materials and methods

Exposure to ethylene oxide (ETO) was studied in a group of ten workers employed in a hospital sterilizer unit, by testing environmental air, alveolar air and blood.

Air samples

Eight alveolar air samples were taken at hourly intervals for each worker during the work shift. Eight out of the ten workers supplied the eight alveolar samples twice, on two different days. Air samples were collected in stoppered glass tubes with screw caps at both ends, and with an interior volume of 70 ml. Standing in the work place, and after a normal inspiration, workers forced expiration, keeping the glass tube between their lips. The tube was immediately sealed with the two caps after the end of the expiration. One instantaneous environmental air sample was collected at the same time as every alveolar air sampling. Instantaneous environmental air samples were taken from the breathing zone of each worker where he was during the alveolar air sampling. Instantaneous environmental samples were collected in glass tubes, similar to those used for alveolar air, by manual pump. A total of 144 alveolar air samples as well as environmental air samples were collected from the ten workers.

Venous blood samples

Blood samples were collected from nine workers. Each worker supplied four blood samples. Two of the four blood samples were taken at the end of the workshift on two different days. The other two blood samples were taken at the fourth and eighth hours of the same workshift on a third day. One instantaneous environmental air sample and one alveolar air sample were always collected together with each blood sample.

With regard to eight workers, continuous measurements of the environmental ETO were carried out (on the third day) when the two blood samples at the fourth and the eighth hours were collected. Continuous environmental samplings were carried out by means of portable pumps (Zambelli) and charcoal tubes. For each worker, four samplings, lasting 2 h each, were collected. Sampling flow was 100 ml/min. Blood samples, which were taken at the work place, were analysed by a head space technique. Three ml of heparinated venous blood were injected into a 12-ml glass vial provided with screw cap and perforable Teflon membrane. For the analysis, 1 ml of the vial head space was injected, with a heated gas-tight syringe, into the gas-chromatograph. The glass vials were put in a room heated at 37°C, for 2–3 h before analysis.

Gas-chromatography

The ETO concentrations in all the air and blood samples were measured with a C. Erba Fractovap 2350 gas-chromatograph, equipped with FID detector, stainless steel column (240 cm × 2 mm ID), packed with Carbowax 1500 0.2% on Carbopack C 80–100 mesh; temperature: oven 48°C, detector 150°C; carrier gas: nitrogen; flow rate: 12 ml/min. The detection limit of ETO was 0.05 μg/l.

Gas-chromatographic determination of ETO in charcoal tubes was carried out after desorption with carbon disulphide, which was injected into a Perkin-Elmer Sigma 3B gas-chromatograph equipped with FID detector, stainless steel column (200 cm × 2 mm ID), packed with Porapak Q 50–80 mesh; temperature: oven 200°C, detector 200°C; carrier gas: nitrogen; flow rate: 40 ml/min.

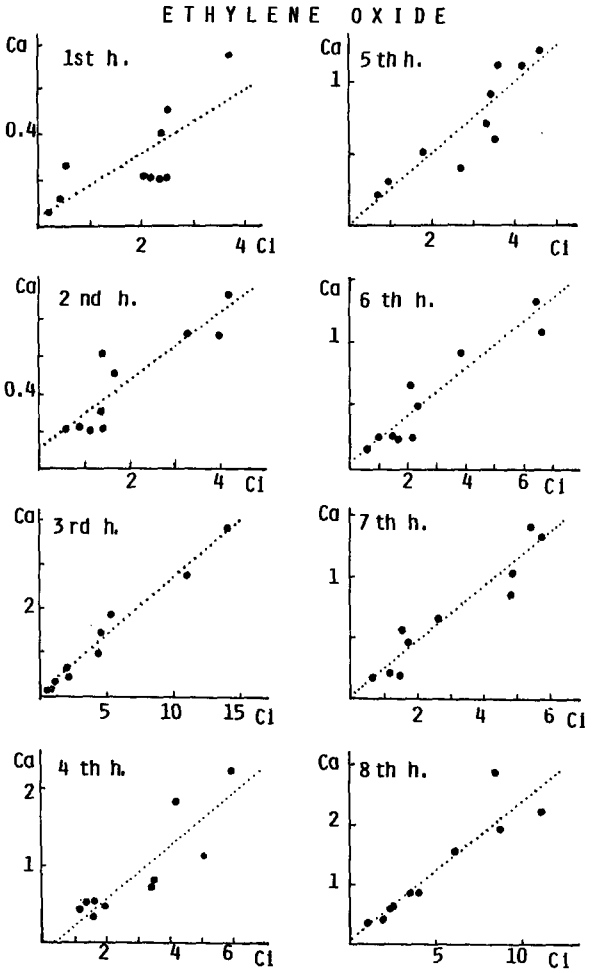

Fig. 1. Correlations between alveolar (Ca) and environmental (Ci) concentrations of ethylene oxide at hourly intervals: 1st h: Ca = 0.136 Ci + 0.02; $r = 0.7509$; $n = 10$; $P < 0.05$; 2nd h: Ca = 0.172 Ci + 0.09; $r = 0.8858$; $n = 10$; $P < 0.001$; 3rd h: Ca = 0.243 Ci + 0.02; $r = 0.9873$; $n = 10$; $P < 0.001$; 4th h: Ca = 0.340 Ci − 0.10; $r = 0.8776$; $n = 10$; $P < 0.001$; 5th h: Ca = 0.250 Ci + 0.002; $r = 0.9029$; $n = 10$; $P < 0.001$; 6th h: Ca = 0.221 Ci + 0.06; $r = 0.9367$; n 10; $P < 0.001$; 7th h: Ca = 0.229 Ci + 0.02; $r = 0.9511$; $n = 10$; $P < 0.001$; 8th h: Ca = 0.232 Ci + 0.02; $r = 0.9307$; $n = 10$; $P < 0.001$

Results

The results of the alveolar (Ca) and environmental (Ci) ethylene oxide (ETO) determinations carried out at hourly intervals in the ten workers during the work shift are reported in Fig. 1. This figure shows that a very high correlation ($P < 0.001$) exists between the alveolar and environmental concentrations from the second to the eighth hours of the workshift (r between 0.9873 at the third hour and 0.8776 at the fourth hour). Only at the first hour was the correlation lower: $r = 0.7509$ with $P < 0.05$. The slopes (Fig. 1) ranged between 0.136 at the first hour and 0.340 at the fourth hour, suggesting that the alveolar ETO concentration ranged between 14 and 34% of the environmental concentration.

Figure 2 reports the results of all 144 determinations carried out in the instantaneous alveolar and environmental air samples collected from the ten workers. The figure shows that alveolar and environmental concentrations were highly correlated ($r = 0.947$; $P < 0.001$). According to the slope of the regression line the alveolar concentration corresponded to 21.3% of the environmen-

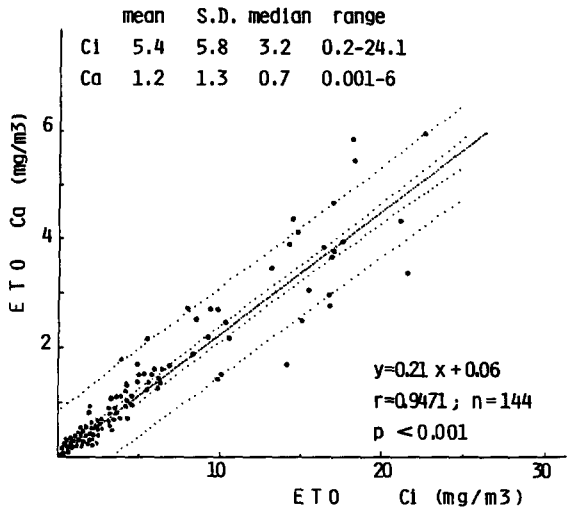

Fig. 2. Correlation between all the environmental (Ci) and alveolar (Ca) concentrations of ethylene oxide measured during the work-shift with 95% confidence and tolerance limits

Table 1. Blood ethylene oxide concentrations (Cb) determined 4 times in 9 occupationally exposed workers simultaneously with the environmental (Ci) and alveolar (Ca) concentrations

	Determinations											
	1st			2nd			3rd			4th		
Worker	Cb	Ci	Ca	Cb	Ci	Ca	Cb	Ci	Ca	Cb	Ci	Ca
(1) MS	4.6	4.0	0.8	nd*	2.4	0.4	nd	5.2	1.4	80.9	19.5	5.5
(2) FI	14.8	3.5	0.8	4.9	1.8	0.5	16.0	1.7	0.4	51.1	15.1	1.7
(3) GE	12.3	2.5	0.6	4.9	2.4	0.2	11.2	5.7	1.5	46.5	18.0	3.7
(4) MA	nd	2.0	0.4	7.3	2.0	1.5	6.2	9.0	2.5	53.4	18.1	3.8
(5) MI	19.5	8.8	1.9	7.3	0.4	0.2	10.4	4.6	1.0	72.2	17.8	3.0
(6) OR	4.0	2.2	0.2	41.8	6.2	1.5	6.2	6.8	1.6	42.7	22.5	4.4
(7) PI	nd	0.5	nd	54.2	13.1	7.0	nd	2.0	0.4	51.1	16.0	2.5
(8) RI	31.6	8.5	2.8	27.0	4.3	0.9	41.5	4.7	0.7	103.7	17.5	4.0
(9) FR	4.0	2.5	0.6	41.8	5.5	1.5	14.7	1.1	0.3	46.5	11.2	2.2
Mean	10.1	3.8	0.9	21.0	4.2	1.5	11.8	4.5	1.1	60.9	17.3	3.4
SD	10.5	3.0	0.8	20.4	3.8	2.1	12.5	2.6	0.7	20.5	3.1	1.2

*nd: not detectable

tal concentration. The mean of the individual Ca/Ci ratios was 0.239 (SD = 0.08). Table 1 shows the results of the four blood determinations of ETO compared with those in the instantaneous environmental and alveolar air samples simultaneously collected. With regard to the data in Table 1, statistical analysis showed that blood ETO concentrations correlated significantly (Table 2) with alveolar ($r = 0.914$ and 0.739) and environmental ($r = 0.866$ and 0.886) concentrations at the first and second determinations, while no correlation existed at the third and fourth determinations (Table 2).

Figure 3 shows the correlation between the environmental and blood concentrations of ETO with regard to all 36 determinations performed on the nine

Table 2. Parameters of the correlations of blood (Cb) ETO concentrations with instantaneous environmental (Ci) and alveolar (Ca) ETO concentrations in 9 workers tested four times

	Determinations				
	1st	2nd	3rd	4th	All data
Number	9	9	9	9	36
Cb = Y; Ci = X					
Slope	2.9	4.7	−0.6	0.9	3.3
Intercept	−0.9	0.9	15	45	1.2
$r =$	0.866	0.886	−0.129	0.137	0.827
$P <$	0.01	0.01	ns*	ns	0.001
Cb = Y; Ca = X					
Slope	11.2	7.1	−5.3	7.1	12
Intercept	0.6	10	18	36	5
$r =$	0.914	0.739	−0.315	0.413	0.744
$P <$	0.001	0.05	ns	ns	0.001

* ns = not significant

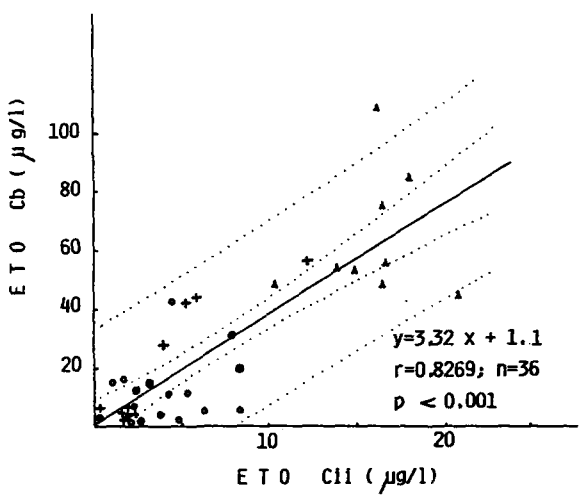

Fig. 3. Correlation between environmental (Ci) and blood (Cb) concentrations of ethylene oxide relating to 9 workers who were tested 4 times (First determinations: ●; second determinations: ○; third determinations: +; fourth determinations: ▲) with 95% confidence and tolerance limits

workers (9 × 4). As can be seen, a very high correlation was found ($r = 0.8269$; $P < 0.001$). The slope suggests that the blood ETO concentration was, on average, 3.3 times higher than the environmental concentration.

Figure 4 reports the data of all 36 determinations of ETO in the alveolar and blood samples collected from the nine workers. The plot shows that a highly significant correlation existed between alveolar and blood ETO concentrations ($r = 0.7443$; $P < 0.001$). The slope suggests that blood concentrations corresponded, on average, to 12 times the alveolar concentrations. According to the mean of the individual values the Cb/Ca ratio corresponded to 17.0 (SD = 14.2).

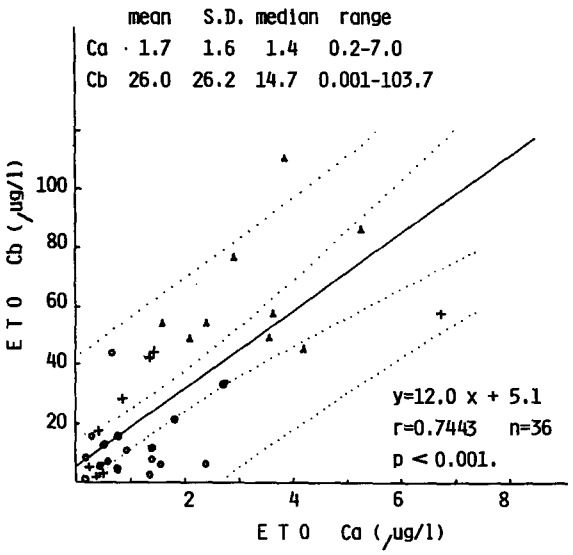

Fig. 4. Correlation between alveolar (Ca) and blood (Cb) concentrations of ethylene oxide relating to 9 workers who were tested 4 times (First determinations: ●; second determinations: ○; third determinations: +; fourth determinations: ▲) with 95% confidence and tolerance limits

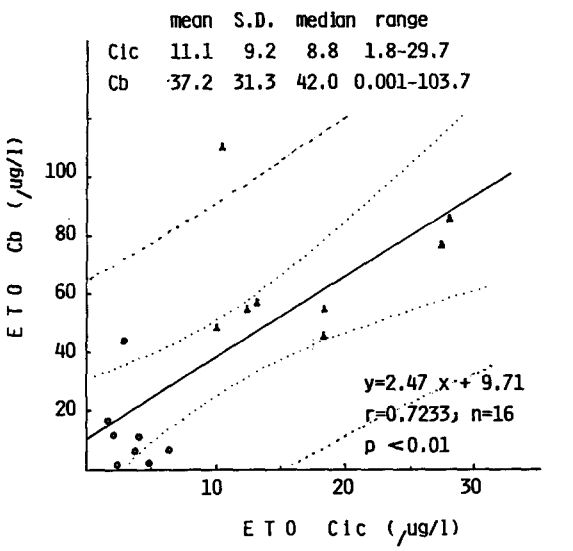

Fig. 5. Correlation between blood ethylene oxide concentrations measured at the fourth (○) and eighth (▲) hours of exposure, and the environmental exposure measured continuously (Cic) during the first and the second halves of the workshift with 95% confidence and tolerance limits

In Fig. 5 blood ETO concentrations measured at the end of the first and second halves of the workshift (third and fourth determinations in Table 1) are plotted against the environmental ETO concentrations measured throughout the first and the second 4 h of the work shift. As shown in Fig. 5, the blood ETO concentrations and the environmental exposure were, during the first half of the shift, lower than during the second half. Moreover, the blood ETO concentrations measured at the fourth and eighth hours of the workshift were found to correlate significantly ($r = 0.7233$; $P < 0.01$) with the corresponding environmental exposure which was being continuously tested. According to the slope of the regression line (Fig. 5), the blood concentrations corresponded, on average, to 2.5 times the environmental concentrations.

Discussion

We tested the alveolar concentration of ETO eight times at hourly intervals in ten workers, eight of whom supplied the alveolar air samples twice. The data in Fig. 1 show that at each hour throughout the workshift the alveolar and environmental ETO concentrations were highly correlated in the ten workers studied. Figure 2 summarises all alveolar and environmental ETO determinations (n = 144) carried out in the ten workers. According to the slope in Fig. 2, the alveolar ETO concentration is found to correspond, on average, to about 21% of the environmental concentration, while it ranged between 14 and 34% according to the slopes in Fig. 1. In a previous report (Brugnone et al. 1985) with only 80 determinations, the alveolar ETO concentration corresponded to about 24%. According to the mean of individual Ca/Ci ratios resulting from all 144 determinations, the alveolar ETO concentration corresponds to 23.9% of the environmental concentration. It can be argued from these data that alveolar retention of ETO [1-(Ca/Ci)] corresponds to about 75–80% of the inhaled ETO. With the present TLV of 2 mg/m^3 (ACGIH) and an alveolar ventilation of 10 l/min, this means an absorption of 7.2–7.7 mg of ETO in 8 h of exposure. We refer to the previous paper (Brugnone et al. 1985) for other comments on the ETO concentration in the alveolar air of workers during occupational exposure.

When we compared the four blood ETO determinations performed on each worker (Table 1) with the determinations of ETO in the instantaneous environmental air samples collected simultaneously, we found that a correlation existed at the first and second determinations, but not at the third and fourth determinations (Table 2).

The same result, i.e. a correlation at the first and second determinations but not at the third and fourth determinations, was also found between blood and alveolar ETO concentrations. It could be interesting to bear in mind that the last two blood ETO determinations related to the fourth and eighth hours of the exposure (on the third day), when the environmental ETO concentration was monitored continuously throughout the workshift. The plot in Fig. 5 shows that the blood ETO concentrations measured at the fourth and eighth hours of the workshift correlated with the environmental exposure measured throughout the corresponding first and second half of the workshift. Moreover, Fig. 5 shows that the environmental exposures during the first half of the workshift were lower than during the second half.

When we considered the four determinations of the blood ETO concentrations together, we found that a significant correlation existed between blood and instantaneous alveolar ETO concentrations as well as between blood and instantaneous environmental ETO concentrations (Fig. 3 and 4). Moreover, we found that the blood concentration was higher than environmental concentration by a mean ratio of about 3 (slopes in Figs. 3 and 5) and higher than alveolar concentration by the mean ratio of 12 (slope in Fig. 4). We found a mean value of 17 for the individual Cb/Ca ratios.

The blood/air partition coefficient of ETO studied "in vitro" was found to be 90 (SD = 20). The difference between our values "in vivo" 12–17 and "in vitro" 90, can be explained by the fact that we dealt with venous blood and not arterial

blood. Appelgren et al. (1977) reported that ETO concentrations in the liver, kidney and pancreas were 3 to 4 times higher than in blood. This means that until the tissues are saturated and as long as the lung uptake of ETO is lower than its metabolism rate, the ETO concentration in venous blood cannot approach the level present in arterial blood. This is the reason why the ratio between venous and alveolar ETO concentrations is generally lower than the blood/air partition coefficient measured "in vitro". If the values we found are exploited by substitution according to the following formula: $Ca/Ci \times Cb/Ca = Cb/Ci$, we can suppose that, at the exposure level of the TLV (2 μg/l), the alveolar and blood ETO concentrations in occupationally exposed workers should not exceed, as a mean of the 8-h exposure, the following figures:

Ca: 0.4–0.5 μg/l and Cb: 6.6–8.5 μg/l according to the Cb/Ci and Cb/Ca ratios.

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