## Carboxyhemoglobin Concentrations in Blood from Donors in Chicago, Milwaukee, New York, and Los Angeles

Abstract. To determine the carbon monoxide exposure experienced by the residents of Chicago, Los Angeles, Milwaukee, and New York, venous blood samples were obtained from adults at arbitrarily chosen blood bank collection sites in the four cities and analyzed for circulating carbon monoxide, carboxyhemoglobin. For comparative purposes, blood was obtained from volunteers breathing carbon monoxide–free air and was found to contain 0.45 percent carboxyhemoglobin. By contrast a high percentage of all the nonsmoking blood donors breathing city air had carboxyhemoglobin saturations greater than 1.5 percent, which indicated that exposure to carbon monoxide in excess of that permitted by the quality standards of the Clean Air Act of 1971 was widespread and occurring regularly.

To protect the public from excessive exposure to carbon monoxide (CO), the quality standards of the Clean Air Act. of 1971 limit CO exposure. Compliance with the 8-hour standard would keep the CO saturation in blood, termed carboxyhemoglobin (COHb), below 1.5 percent in active nonsmokers. If we assume that exposure to CO concentrations in excess of the standards could be detrimental to human health. the immediate question is, "Are Chicago, New York, and Los Angeles, cities which have had problems with air pollution, able to comply with the CO standards?"

To answer this question would require the analysis of the CO in the breathing zone of each of the persons placed under surveillance by the Clean Air Act. None of these cities currently has the capability to do this. Instead they rely upon CO exposure data supplied by a system of air monitoring stations. This permits the estimation of CO encountered by the citizenry but has obvious limitations (1). A Los Angeles monitoring station located 40 feet (1 foot = 0.3 m) above an expressway entrance cannot measure the concentration of CO four blocks away in the breathing zone of a mother and her child crossing a busy intersection, nor that of a clerk on the 20th floor of an office building adjacent to the intersection.

Since the inspired CO passes through the lungs and circulates in the blood as COHb, the measurement of COHb provides an accurate method for assessing individual CO exposure occurring in the previous 15-hour interval (2). To answer the question regarding the magnitude of CO exposure, venous blood samples were obtained from blood donors at arbitrarily chosen blood bank collection sites in the three cities and analyzed for COHb. Sampling sites were chosen so that the population would include persons from urban, suburban, rural, and airport terminal locations. For comparative purposes, COHb measurements were also made on samples from blood donors in Milwaukee and from four adults breathing CO-free air.

Two research associates were trained in interviewing techniques. At the time of blood collection they interviewed the donors and completed detailed questionnaires about them. These allowed us to assess the influence of age, vreight, height, sex, race, smoking habits, place of residence, occupation, place of work, and meteorological conditions on the COHb measurements. From every tenth donor, an alveolar breath sample was obtained for CO analysis which served as an independent check on the stability of the COHb until time of analysis (3, 4).

The venous blood samples were collected in 5-ml Vacutainer tubes containing ethylenediaminetetraacetate (EDTA) as the anticoagulant, and alveolar breath samples were collected





in 35-ml glass pipettes (4). The samples were coded and air mailed to the Department of Environmental Medicine laboratory, Milwaukee, Wisconsin, for analysis. Blood samples were analyzed by the automated differential spectrophotometric procedure (CO-oximeter) based on the method of Malenfant (5) and which was calibrated by the gas chromatographic procedure based on the method of Collison (6) (Fig. 1). Breath samples were analyzed by gas chromatography by the method of Porter and Volman (7).

To determine the stability of the COHb in venous blood collected in 5ml Vacutainer tubes containing EDTA, samples were stored at 22°C and at 4°C. Tubes from each group were sequentially analyzed and showed stable COHb in the refrigerated samples for periods up to 6 weeks and in unrefrigerated samples for 2 weeks (8).

To check on the ability of the two COHb analytical methods to accurately measure low concentrations of COHb, four adults, three males and one female, breathed 100 percent oxygen delivered through a Scott Aviation Demand Oxygen face mask for a period of 3.5 hours; after this they breathed air for 1.5 hours which had been passed through Hopcalite to remove CO. The mean COHb saturation of the four adults prior to breathing 100 percent oxygen was 0.65 percent with a range of 0.55 to 0.75 percent. After 3.5 hours of oxygen inhalation, the mean COHb was 0.38 percent with a range of 0.30 to 0.40 percent. After breathing CO-free air for 1.5 hours, the mean COHb saturation was 0.45 percent with a range of 0.35 to 0.5 percent. This COHb saturation is in agreement with that due to endogenous CO production (9), and demonstrated the sensitivity of the analytical procedures in detecting low concentrations of COHb.

The agreement between the alveolar breath CO and the matched blood COHb collected in the field did not approach the accuracy of the relationship obtained experimentally (4, 10); however, the agreement was good enough to validate that COHb was stable until time of analysis.

In the analysis of the data, the median and 90 percent range were used to describe the distribution of COHb saturation in the various geographical locations. Mean COHb saturation was used to investigate the effect of the other variables such as smoking and occupation. The statistical methods used to assess the observed differences between mean COHb saturations were the *t*-test and analysis of variance for two or more means. Multiple regression techniques were used to investigate the influence of meteorological variables on mean COHb saturation. In all cases, a significance level of  $P \leq .01$  was used to declare statistical significance.

The median COHb saturations of the blood donors and the range in which 90 percent of them fell are listed in Table 1. Carboxyhemoglobin saturation was an order of magnitude greater in cigarette smokers than in nonsmokers. Among the nonsmokers, 76 percent in Los Angeles, 74 percent in Chicago, 35 percent in New York, and 26 percent in Milwaukee had COHb saturations in excess of 1.5 percent, indicating CO exposure in excess of that permitted by air quality standards (2).

Persons sampled in urban areas with high automobile density consistently had COHb saturations greater than those in areas with low automobile density. This was dramatically evident in the comparison of individuals working on Governor's Island with persons working in adjacent New York City areas. The average COHb saturation of individuals on Governor's Island was half that of person working in adjacent New York City.

Both the median and the mean COHb saturation of blood donors from the urban areas of Los Angeles and Chicago were higher than those observed in donors in urban New York and Milwaukee. This is not proof that the ambient CO levels in Los Angeles and Chicago were higher than in the other two cities, but that the specific locales sampled had higher average ambient CO concentrations than the average of the locales visited in New York City and Milwaukee.

One observation did, however, suggest that the ambient CO concentrations in Los Angeles and Chicago were higher than those in New York City. During the sampling period at the East Blood Center a weather inversion occurred in New York City, which was associated with COHb saturations comparable to those observed in the other two cities during fair weather periods.

Barometric pressure, temperature, visibility, and wind speed data were correlated with COHb saturations. Of these, only wind speed had a positive but relatively minor influence on COHb saturation (10).

There were significant differences between the COHb saturations of the oc-28 DECEMBER 1973 cupational groups studied (10). Students and housewives had the lowest COHb concentrations. Other low COHb groups included those associated with mental health, education, library science, religion, art, road paving, and entertainment. The vehicle-related occupational groups had higher COHb saturations than most groups. Other high COHb groups included those that were associated with metal processing, chemical processing, stone and glass processing, printing, welding, electrical assembly and repair, and graphic art.

Taxicab drivers generally showed the highest COHb saturations observed in any occupational group. In New York City, 14 nonsmokers were sampled on 18 October 1970. Eight cab drivers coming from work showed a mean COHb saturation of 2.5 percent with a range from 1.3 to 5.8 percent. Six off-duty cab drivers coming from home showed COHb saturations ranging from 1.0 to 1.5 percent with a mean of 1.2 percent. Twelve cigarette-smoking cab

Table 1. Median COHb and 90 percent range for nonsmokers and cigarette smokers at various locations. For sample sizes less than 20, the 90 percent COHb range is not computed.

	СОНЬ					
Location	Nonsmokers			Cigarette smokers		
	No.	Median	90% range	No.	Median	90% range
Chicag	o-Samp	led Novem	ber 1970			
Loyola Lakeshore	148	1.5	1.0-2.2	83	3.6	1.2-7.8
Michael Reese Hospital	41	2.1	1.0-3.7	163	5.9	2.3-9.9
A.B. Dick, Niles	80	1.7	1.2–2.7	81	6.1	2.5-9.6
O'Hare International Airport	32	2.5	1.8-3.0	16	6.6	5.2-11.1
Imperial Eastman, Niles	34	2.0	1.5-2.7	47	6.8	3.2-10.4
Montgomery Ward, downtown	30	2.7	2.2–3.7	34	6.9	3.2-9.3
Palatine, Ill.	41	1.4	0.8-4.4	16	4.8	1.5-7.7
Los Angeles-Sample	ed Januar	y, Februar	y, May, an	d June 1	972	
Burbank	419	1.8	1.0-3.4	295	6.5	2.5 - 10.5
Torrance	58	1.9	1.5-2.3	46	6.8	2.0-10.5
Long Beach	105	1.6	1.2-5.4	59	5.2	1.5-9.4
El Segundo	172	1.8	1.2-2.5	66	5.9	2.0-9.9
Hollywood	148	2.2	1.4-3.0	130	6.2	2.5-10.0
Van Nuys	361	1.8	1.0-2.7	208	6.8	2.0-10.7
El loro	31	1.8	1.5-6.9	62	5.3	1.5-7.8
El Monte	119	2.0	1.2-3.2	108	5.9	2.5-10.2
Blood Center	15	2.7		13	7.5	
Reseda	28	1.8	1.5-2.2	8	6.2	
Duarte	51	1.7	1.5-4.5	39	6.3	3.4-9.4
Westwood	127	2.0	1.2-2.8	62	6.3	2.2-10.7
Lagie Rock	117	2.0	1.5-2.5	17	4.3	
Ananeim	525	1.7	0.6-2.3	212	6.4	3.2-9.7
Downtown	166	2.7	1.0-3.2	108	6.0	2.0-9.4
Hawthorne Death	12	2.2	1.7-2.7	39	7.0	2.5-8.7
Clandala	12	1.6	1.2-2.3	37	6.1	2:0-9.0
Aliment	01	1.7	1.4-2.0	27	6.1	2.0-9.1
Airport Contury City	213	1.4	1.0-2.1	75	5.6	1.2-9.6
Century City	30	1.4	1.0-1.8	21	6.3	2.0-8.3
Milwaukee-Sampled April-Dec	cember 19	969, Januar	y–Decembe	er 1970, i	and April	1971
Milwaukee County General Hospital	1831	1.2	0.4-3.4	1117	3.7	0.8–9.1
Milwaukee Children's Hospital	193	1.0	0.5-2.5	2	1.2	
Peawaukee	120	1.0	0.6-2.5	61	5.3	0.9-8.6
Milwaukee Blood Center	209	1.4	0.4-3.0	180	5.3	1.0-11.9
Allen Bradley	102	1.4	0.6-2.7	72	5.2	1.2-9.3
Brookfield	72	0.8	0.4-3.2	31	5.8	0.9–9.5
Brown Deer	92	1.4	0.6-3.5	59	7.0	1.4–9.8
Cedarburg	5 121	0.7	0 5-2 1	2	6.9 5 5	10.05
New Yet City C	1.5	1.0	0.5-2.1	. 00	5.5	1.0-9.5
Islin Long Island	a Decemi	per 1970 an	id October-	-Decemb	per 1971	
Met Life And Madison Ave	110	1.4	0.9-2.3	68	3.9	1.4-7.2
Long Island	04	1.2	0.5-2.2	63	4.3	1.0-7.6
Brony	440	1.0	0.5-3.4	2.5	3.5	1.0-8.8
Manhattan	13	1.4	0.8-2.0	75	4.7	1.6-7.6
IEK International Airport	041	1.4	0.8-2.3	8.3	5.2	1.4-9.2
Brooklyn	38	2.1	1.5-2.8	46	6.9	2.3 - 10.9
Hightstown N I	8	2.1	0015	16	7.3	
Governor's Island	40	1.2	0.8-1.7	31	4.4	1.4-10.4
Fast Blood Center	38	0.8	0.4-1.4	45	2.8	0.7-4.7
Flizabeth N I	21	2.0	1.0-2.7	24	5.4	1.8-7.0
Holmdel NI	04 114	1.3	0.3-3.3	67	4.6	1.2-7.6
East View NY	140	1.0	0.0-1.0	20	5.1	1.6-8.3
Croton NY	107	1.4	0.9-2.0	69	5.8	2.1-8.6
New Jersey	130	1.0	0.4-3.7	113	4.1	1.2-8.1
-	100		0.7-2.0	1.00	4.7	1.4-0.3

drivers coming from work showed a mean COHb saturation of 6.9 percent with a range from 3.0 to 13.0 percent.

Large international airports, such as O'Hare and J.F.K., had surprisingly high ambient CO concentrations attributable in part to jet engine CO production and in part to the high automobile density near airport terminal entrances. Persons with advanced heart or lung disease planning to travel in aircraft pressurized for 6000 feet could, as a result of prolonged CO exposure in airport terminals, unknowingly subject themselves to an additional anoxic stress.

The major purpose of our investigation was to establish the range of CO exposure experienced by the American population in these four cities in 1969 to 1972. In so doing we have established that a significant percentage of their populations was continuously exposed to CO concentrations in excess of those permitted by the air quality standards. These worrisome baseline data should stimuate a reexamination of the scientific basis for the air quality standards and should provide a means to measure the effectiveness of the antipollution measures which we as a nation develop and use to control CO exposure.

RICHARD D. STEWART EDWARD D. BARETTA LEIGH R. PLATTE ELIZABETH B. STEWART JOHN H. KALBFLEISCH BARBARA VAN YSERLOO Alfred A. Rimm

Department of Environmental Medicine, Medical College of Wisconsin, Milwaukee 53226

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## Linoleic Acid Hydroperoxide: Impaired Bacterial Uptake by Alveolar Macrophages, a Mechanism of Oxidant Lung Injury

Abstract. Exogenous linoleic acid hydroperoxide causes in vitro impairment of both bacterial uptake and the phagocytic stimulation of  ${}^{14}CO_{2}$  production from [1-14C] glucose in rabbit alveolar macrophages by an undefined effect on the cell membrane. This effect may be one mechanism for the defective pulmonary bacterial clearance characteristic of oxidant lung injury.

Exposure of lungs to oxidants (NO<sub>2</sub>,  $O_3$ , and excess  $O_2$ ) has many effects, including impaired pulmonary bactericidal activity (1) and the formation of lipid hydroperoxides (2). Although pulmonary bacterial clearance is a complex process, one important factor is bacterial ingestion by alveolar macrophages (AM). While lipid hydroperoxides are highly cytotoxic (3) and affect a number of membrane functions, notably in erythrocytes (4), their effect on phagocytosis has not been studied. We therefore studied the effects of exogenous linoleic acid hydroperoxide (LPO) on bacterial uptake and glucose metabolism in the AM.

The LPO was prepared by aerobic oxidation of linoleic acid and purified by silicic acid column chromatography.



Fig. 1 (left). Effects of LPO on bacterial uptake. Alveolar macrophages  $(1 \times 10^7)$ were suspended in 2 ml of GRPS (Ringer-phosphate solution containing 5.5 mM glucose, pH 7.4) and the appropriate amount of LPO. Control flasks contained no AM. After incubation at 37°C, for 1 hour, 0.25 ml of homologous serum and  $2.5 \times 10^8$ live Staphylococcus aureus 502 A were added to both control and experimental flasks, and the incubation was continued for another hour. The AM were removed by centrifugation at 2500g for 15 minutes, and the supernatants containing bacteria were quantitatively cultured by a pour plate technique. Bacterial uptake was calculated from the difference between the colony counts in the absence and presence of AM. Light microscopy indicated that the majority of the bacteria are ingested by the AM, although some surface adherence cannot be excluded. Bacterial uptake in control flasks (no LPO) was taken as 100 percent. Data from seven experiments are expressed as percentage of control  $\pm 1$  standard error (S.E.). Fig. 2 (right). Effect of LPO on <sup>14</sup>CO<sub>2</sub> production from [1-<sup>14</sup>C]glucose. Alveolar macrophages ( $2 \times 10^8$ ) were suspended in 2 ml of GRPS in the presence of the appropriate amount of LPO. After 1 hour of incubation, 12.5 percent homologous serum, 0.1  $\mu c$  of [1-14C]glucose, and, where appropriate,  $2 \times 10^{16}$  heat-killed Staphylococcus epidermidis were added and the incubation continued for another hour. Production of <sup>14</sup>CO<sub>2</sub> was measured as described (8). Data are mean  $\pm 1$  S.E. for five experiments performed in triplicate.

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