

spectrum of these salts somewhat resemble that of anomalous water, the spectra contain many additional lines.

Curve A of Fig. 1 reproduces the reported (2) infrared spectrum of anomalous water and is equivalent to the spectrum reported by other investigators (5, 6). Curve B is the infrared spectrum of sodium acetate. The spectrum of anomalous water is strikingly similar to that of sodium acetate. In particular, strong absorption lines for sodium acetate occur at 1595 and 1410 cm^{-1} , exactly as in the spectrum of anomalous water. In addition, in the sodium acetate spectrum there are weak lines around 1046, 1014, and 925 cm^{-1} , which can also be observed in the anomalous water spectrum. The spectral line for anomalous water that sometimes appears at 1120 cm^{-1} varies from preparation to preparation and has been assigned as a sulfate impurity (3, 5). Finally, the additional line at 1365 cm^{-1} probably results from a nitrate impurity, for example, sodium nitrate, which has a single strong line at that frequency (7). Thus all the unique characteristics of the infrared spectrum of anomalous water appear to originate from simple impurities, a major component being sodium acetate.

Two processes are suggested to account for the production of sodium acetate. In many laboratories it is common practice to wash tubing, capillaries, dishes, and desiccators with ethyl alcohol. The oxidation of ethyl alcohol produces acetic acid which reacts with the sodium-rich, highly absorptive glass surface to produce sodium acetate. Sodium acetate is also produced in the vicinity of acetylene-oxygen flames, which are used to draw capillaries and to flame tools and containers. Acetic acid is produced from acetylene in the presence of water and oxygen, both of which are readily available.

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References

1. B. V. Deryagin and N. V. Churayev, *Priroda (Moskva) No. 4* (1968), p. 16; translated in *Joint Publ. Res. Serv. No. 45* (1968), p. 989.
2. E. R. Lippincott, R. R. Stromberg, W. H. Grant, G. L. Cessac, *Science* 164, 1482 (1969).
3. D. L. Rousseau and S. P. S. Porto, *ibid.* 167, 1715 (1970).
4. S. W. Rabideau and A. E. Florin, *ibid.* 169, 48 (1970).
5. T. F. Page, Jr., R. J. Jakobsen, E. R. Lippincott, *ibid.* 167, 51 (1970).
6. D. L. Rousseau, *Phys. Today* 23, 17 (Oct. 1970).
7. F. Miller and C. Wilkins, *Anal. Chem.* 24, 1253 (1952).

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Carbon Monoxide: Association of Community Air Pollution with Mortality

Abstract. Regression analysis of daily mortality in Los Angeles County shows that there is a significant association between community carbon monoxide concentrations and mortality. Cyclic variation and maximum temperature were the main contributors. No association was demonstrated between oxidant and mortality.

Acute carbon monoxide poisoning claims more than 1000 lives each year in the United States. Exposures to lower concentrations, on the order of 50 parts per million (ppm), may result in temporary impairment of judgment and motor performance but are not known to produce any serious or long-lasting impairment (1). Cohen *et al.* have reported elsewhere a possible effect of exposures to community carbon monoxide on case fatality rates for myocardial infarction (2). We report here statistical evidence that carbon monoxide, when present in the ambient atmosphere at concentrations of the order of those that occur in Los Angeles, is associated with increased mortality.

Our data consist of the total number of deaths occurring in Los Angeles County for each day from 1 January 1962 through 31 December 1965, together with temperature, carbon monoxide, and total oxidant measurements for each day from 26 December 1961 through 31 December 1965. Temperature measurements are the maximum temperatures for downtown Los Angeles as published by the U.S. Weather Bureau. Carbon monoxide and oxidant concentrations are basin averages (arithmetic mean of all measurements at all monitoring stations in the Los Angeles Air Pollution Control District during each 24-hour period).

An arbitrary numbering sequence was established, with each day numbered consecutively starting with day 1 for 1 January 1962 to day 1461 for 31 December 1965. This gave us a total of five variables for input: total number of deaths, day number ("day of occurrence"), maximum temperature, basin averages for carbon monoxide concentration, and basin averages for oxidant concentration, for each day in the study period, with environmental data for the week preceding the study period.

A number of factors entered into our choice of analytic method. Mortality follows a cyclic pattern, with mortality maxima occurring in winter (3, 4) (Fig. 1). Many environmental factors, such as temperature and levels of air pollution, also occur in a cyclic pattern

with similar periodicity. Temperature is an important factor and would tend to mask the relatively small contribution expected for air pollutants (5). In Los Angeles, there is a secular trend both for total mortality and for carbon monoxide concentration. Substantial correlations exist among environmental variables. The effect of some environmental variables might be delayed by hours or days, so that the possibility of lag effects must be considered. A further consideration is that we wanted a model that could be adapted for surveillance on a real-time basis. For these reasons we chose multiple regression, because this one formulation will simultaneously identify significant variables, estimate the contribution and relative importance of each, and provide an estimate of the expected number of deaths under specified conditions.

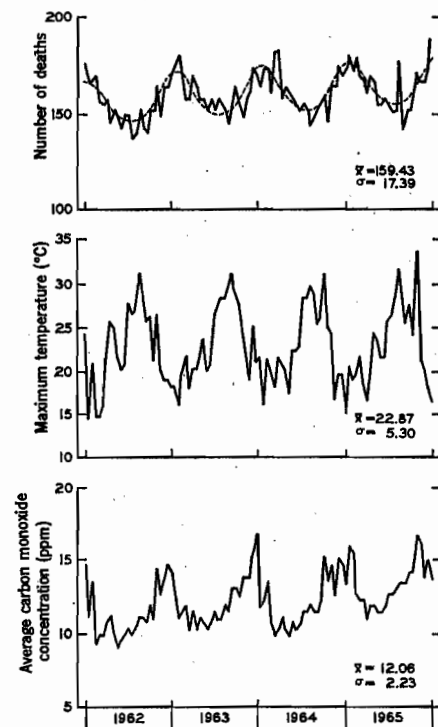


Fig. 1. Mortality, temperature, and carbon monoxide concentrations in Los Angeles, 1962-1965. Points plotted are the medians of 15-day intervals. The dashed line is the mortality predicted by the model with trend and cyclic variation terms. σ , Standard deviation; \bar{X} , mean.

Table 1. Coefficients for regressions of daily mortality in Los Angeles County, 1962-1965. The mortality is the total number of deaths from all causes on the day of occurrence, by the place of occurrence. The temperature is the maximum temperature in downtown Los Angeles. The carbon monoxide concentration is the 24-hour basin average (in parts per million). These figures are rounded; the original calculations were carried out to five or more significant figures. S.E., standard error; $\theta = 2\pi D/365.24$; D , day number, for day of occurrence; T , maximum temperature on the day of occurrence (in degrees Celsius); T_{-1} , maximum temperature 1 day before the day of occurrence (T_{-2} and T_{-3} are similarly defined); R^2 , the multiple coefficient of determination, is the proportion of the total variation about the mean "explained" by the regression.

Variable	Regression on trend, cyclic variation, temperature, and carbon monoxide concentration		Regression on trend, cyclic variation, and temperature		Regression on trend and cyclic variation	
	Coefficient	S.E.	Coefficient	S.E.	Coefficient	S.E.
Constant	192.48		209.36		151.63	
D	0.017	0.003	0.018	0.003	0.015	0.004
D^2	-0.000007	0.000002	-0.000006	0.000002	-0.000004	0.000002
$\sin \theta$	8.12	0.67	7.74	0.66	5.08	0.56
$\cos \theta$	10.44	0.87	12.22	0.65	10.76	0.55
$\sin 2\theta$	-0.41	0.55	-0.81	0.54	1.17	0.55
$\cos 2\theta$	2.55	0.54	2.89	0.53	1.60	0.55
T	-2.49	0.72	-2.10	0.71		
T^2	0.064	0.015	0.059	0.015		
T_{-1}	-1.61	0.88	-1.43	0.88		
T_{-1}^2	0.040	0.019	0.037	0.019		
T_{-2}	-0.45	0.88	-0.47	0.88		
T_{-2}^2	0.009	0.019	0.009	0.019		
T_{-3}	-2.06	0.70	-2.01	0.70		
T_{-3}^2	0.039	0.015	0.037	0.015		
Log CO	24.94	8.08				
Mean square for regression		10,926.4		11,576.1		19,564.5
Mean square for residuals		192.0		193.1		222.8
R^2		0.371		0.367		0.266

Our basic model is a linear regression in the usual form, with the total number of deaths from all causes as the dependent variable (6). Our original model included day number and its square for trend, five pairs of harmonic terms for cyclic variation (the leading terms of a Fourier series based on 1 year), and various representations for the environmental variables with lags of up to 6 days (that is, observations made 6 days before the day of occurrence). We deleted variables through backward elimination with restraints, with minimization of the mean square for residuals (MSE) as our major criterion; our final choice, from among several models with small differences in MSE, was the model with the fewest variables. The restraints on our elimination procedure specified that both variables of a pair in a quadratic or harmonic formulation must be taken together and that only variables at the beginning or at the end of a sequence could be deleted. Thus we did not permit any models in which the temperature variable, for example, could take the form: $T + T^2 + T_{-2}$, omitting T_{-1} , T_{-1}^2 , and T_{-2}^2 from the sequence (where T_{-1} denotes the maximum temperature 1 day before the day of occurrence and T_{-2} is similarly defined).

Table 1 shows the results of regressions for three separate models: one model with trend and cyclic variation only, one with temperature added, and one with both temperature and carbon

monoxide concentration added. The models selected as providing the "best" representation include two terms for trend, two pairs of harmonic terms for cyclic variation, quadratic representations for temperature through 3 days of lag, and the logarithm of the carbon monoxide concentration. The MSE for the model with carbon monoxide is 192.0, a reduction of 36 percent from the variance of the original observations (7).

The coefficient for carbon monoxide is highly significant ($P < .002$). We believe that this result constitutes evidence that carbon monoxide, in the concentrations encountered as a constituent of community air pollution, is associated with excess mortality.

Previous work with daily mortality has suggested that a substantial autocorrelation would exist, which might invalidate a regression of this form (4). Fortunately, analysis of residuals for the two models with temperature terms indicated that no substantial autocorrelation exists (8). However, because of our concern for a possible autocorrelation or a possible day-of-the-week effect, which might be confounded with the carbon monoxide concentration, we divided our data into 7 (or in some cases 28) interpenetrating subsamples, each subsample with observations spaced 7 (or 28) days apart, and performed parallel analyses for each subsample. Using the models shown in Table 1, we obtained virtually the same findings

with respect to the proportion of explained variance and the magnitude of the carbon monoxide coefficient (9).

We also applied the models shown in Table 1 to that fraction of total mortality attributed to arteriosclerotic heart disease with similar results. This is consistent with the findings of Cohen *et al.* (2).

The logarithm of the carbon monoxide concentration, used in our model, does not provide a direct measure of the contribution of carbon monoxide. However, comparisons between concentrations are readily obtained. The estimated contribution to mortality for Los Angeles County for an average carbon monoxide concentration of 20.2 ppm (the highest concentration observed during the 4-year period), as compared with an average carbon monoxide concentration of 7.3 ppm (the lowest concentration observed), is 11 deaths for that day, all other factors being equal.

Similar regression models were formulated with oxidant as an environmental variable. No model with oxidant gave an MSE as small as the corresponding model without oxidant. We take this as evidence that the association of oxidant with mortality, if there is any such association, is substantially less than that of carbon monoxide with mortality.

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References and Notes

1. Committee on Effects of Atmospheric Contaminants on Human Health and Welfare, "Effects of Chronic Exposure to Low Levels of Carbon Monoxide on Human Health, Behavior, and Performance" (National Academy of Sciences, Washington, D.C., 1969); J. R. Goldsmith and S. I. Cohen, *J. Air Pollut. Control Ass.* 19, 704 (1969).
2. S. I. Cohen, M. Deane, J. R. Goldsmith, *Arch. Environ. Health* 19, 510 (1969).
3. J. R. Goldsmith and N. M. Perkins, in *International Biometeorological Congress, 3rd, Pau, France, S. W. Tromp et al., Eds.* (Pergamon, New York, 1967), vol. 2, p. 97.
4. H. H. Hechter and J. R. Goldsmith, *Amer. J. Med. Sci.* 241, 581 (1961).
5. R. K. MacPherson, F. Ofner, J. A. Welch, *Brit. J. Prev. Soc. Med.* 21, 17 (1967); C. A. Bridger and L. A. Helfand, *Int. J. Biometeorol.* 12, 51 (1968).
6. For a general discussion of linear regression and associated statistical techniques, see N. R. Draper and H. Smith [*Applied Regression Analysis* (Wiley, New York, 1966)].
7. An MSE of 190.4 was obtained with a 26-variable model, which included two terms for trend, four pairs of terms for cyclic varia-

tion, terms for temperature through 6 days of lag, a three-way interaction term for temperature, and a carbon monoxide term. The coefficient for the logarithm of carbon monoxide was 22.8 ($P < .003$).

8. First-order autocorrelation coefficients were less than 0.05 ($P > .05$) for both models with temperature terms. For the model with trend and cyclic variation terms only, the autocorrelation coefficient was 0.155 ($P < .001$).
9. If the subsamples are regarded as statistically independent, and if the day-of-the-week effect, if any, is a simple displacement of the mean for total mortality, then each of the 28 subsamples provides an independent and unbiased estimate of the same set of regression coefficients. For the model shown in Table 1, the mean coefficient for the logarithm of carbon monoxide is 26.28, t (the ratio of the coefficient to its standard error) = 2.67 ($P < .01$).
10. We thank Dr. W. R. Gaffey and H. K. Ury for many helpful discussions and suggestions during the course of this study. Supported in part by contract PH 86-68-35 with the National Air Pollution Control Administration.

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Chemical Communication and "Propaganda" in Slave-Maker Ants

Abstract. *Slave-maker ants of the Formica sanguinea group direct their raids by means of odor trails. Artificial trails made from whole-body extracts and extracts of Dufour's glands and hindguts can be used to guide columns of workers to selected target colonies and to initiate raids. In workers of F. pergandei and F. subintegra, members of the F. sanguinea group, the Dufour's glands are hypertrophied and contain large quantities of three acetates (decyl, dodecyl, and tetradecyl), which are discharged at defending workers during the slave raids. The acetates produce very efficient, long-lasting alarm signals that attract the slave-makers but disperse the defenders; in effect, therefore, they are "propaganda substances."*

Slavery is practiced by species belonging to at least six ant genera: *Leptothorax*, *Strongylognathus*, and *Harpagoxenus* in the subfamily Myrmicinae; and *Formica*, *Polyergus*, and *Rossomyrmex* in the subfamily Formicinae. Although most or all of these cases represent independent phylogenetic developments, the basic pattern is the same. The slave-maker workers raid nests of another species in the same or a closely related genus, where they repel or kill defending workers, penetrate their nest, and capture their worker pupae. When adults later eclose from the pupae, they accept the slave-makers as nestmates and readily assist in the domestic work of the slave-maker nest. Workers of the most specialized of the slave-maker species are capable only of conducting raids and are wholly dependent on their slaves for their day-to-day existence.

Previous experimental work indicates that the raids of *Harpagoxenus americanus* (1) and *Polyergus lucidus* (2) are both initiated and guided by odor trails laid down by scout workers from the target nest back to the home nest of the slave-makers. Neither the

glandular source nor the chemical identity of the trail pheromone has been identified in these species. We extended the result to the species of the *F. sanguinea* group in the following way. It was learned that raids could be initiated at the discretion of the investigator during late July and August by placing fragments of colonies belonging to slave species (*F. subsericea*) a short distance from the edge of the

slave-maker nests. When slave-maker scouts encountered the colony fragments, they returned to their own nests, apparently laying odor trails. Columns of workers, indistinguishable from those seen during natural raids, immediately emerged and began attacking the colony fragments. They subdued the workers of the slave species and carried their pupae back to the slave-maker nests. We were able to initiate and guide raids in the following way. Whole-body extracts of ten slave-maker workers (*F. rubicunda*) were made in ether and land down with watercolor brushes over the soil, from the nest entrance to selected points about 1 m away. The *F. rubicunda* workers, accompanied by a few adult slaves, followed the trails to the end and milled in confusion in the area beyond. When fresh fragments of slave-species colonies (*F. subsericea*) were now placed at the end of the trail, a full-scale raid ensued. Similar results were obtained with workers from a *F. subintegra* nest. *Formica subsericea* slaves did not accompany their *F. subintegra* mistresses on these artificially induced raids. It was further discovered that *F. subintegra* workers can be easily diverted for distances of up to 1 m or more from raided colony fragments with the use of artificial trails consisting of synthetic acetates in the mixtures found naturally in the Dufour's gland (see further discussion of the chemistry of the secretions below). However, it was not demonstrated that the natural trail substances originate in the Dufour's gland. When a trail made from three combined Dufour's glands was laid in competition with one laid from three combined hindguts, the latter had far greater attracting power. Only a single worker followed the trail made from the Dufour's gland during 5 minutes, whereas over 30 followed the trail made from the hindguts. In view of the fact that the hindgut is the source of recruitment odor trails in other kinds of formicine ants (3), we conclude that this organ also produces a trail pheromone in *F. subintegra*. It remains to be determined whether substances from the Dufour's gland, and, in particular, the acetates, also serve as trail substances.

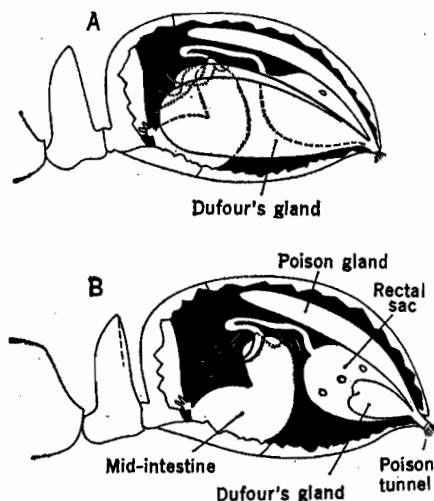


Fig. 1. The abdomens of workers of (A) a slave-maker ant species (*F. subintegra*) and (B) one of the ant species it utilizes as slaves (*F. subsericea*), showing the location of the gut and principal exocrine glands.