

Trace-element concentrations in blood samples from welders of stainless steel or aluminium and a reference group

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ULFVARSON, U. and WOLD, S. Trace-element concentrations in blood samples from welders of stainless steel or aluminium and a reference group. *Scand. j. work environ. & health* 3 (1977) 183—191. The concentrations of 17 trace elements (e.g., copper, cobalt, iron, manganese, chromium, silicon and magnesium) were determined in whole blood samples of 81 persons working with different welding methods on stainless steel or aluminium and 68 nonwelders. Inorganic spark source mass spectrometry was used for the chemical analyses. The data were analyzed by the SIMCA method for pattern recognition (discriminant analysis). No differences were found between the five groups, either in the average levels of the trace elements or in the correlation structures between the trace elements. Thus no blood concentration data on the analyzed elements and collected from a single person contained any information with respect to exposure to the welding fumes investigated.

Key words: aluminium, blood concentrations, pattern recognition, stainless steel, welders.

The concentration levels of a few trace elements are routinely measured in blood or urine samples of workers subjected to exposure to metals. For lead, extensive data have been collected over a long period (14). This information has made it possible to set a biological standard limit for the lead concentration in whole blood (21). Exposure to mercury may be controlled from the determination of the mercury content in blood or urine. For alkyl-mercury compounds urinary determinations are not valid, but the concentration in blood correlates well with exposure (15). As re-

gards cadmium, there are difficulties in judging exposure from data on blood and urine concentrations of this metal (6, 7). The same is true for manganese (14). Approximate recommendations concerning the allowed limits for urinary concentrations have been discussed for nickel, chromium, arsenic, and selenium. For other metals, experience regarding what concentrations can be tolerated is largely lacking, especially for blood concentrations (14). The situation may partly be due to the earlier lack of sensitive methods for determining various elements in small concentrations.

The literature on the concentrations of various pertinent metals in the blood and urine of welders is rather scanty. Barborik and Sehnalová (2) found higher concentrations of manganese in blood samples from 20 welders who did shielded arc welding (arc welding with coated electrodes) with electrodes containing this metal. Jahr and Johnsen (10) have determined nickel in urine samples from welders and found a

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fairly high correlation between exposure and weekly mean values for nickel concentration. Hewitt and Hicks (9) exposed rats to welding fumes for periods of 4 h. High concentrations of cobalt in blood were observed, while the blood concentrations of chromium, antimony, zinc and iron were not significantly different from those in the unexposed control animals. Gylseth et al. (8) found a good correlation between exposure and the urinary chromium excretion of welders.

Welders are exposed to many elements. The composition of welding fumes depends on the material handled, and the intensity of the exposure varies greatly. The exposure situation is thus very complicated. Through the use of multielement analysis, e.g., mass spectrometry, many elements can be determined at the same time in the same sample. The present study is part of an epidemiologic investigation on welders (17) that was undertaken to ascertain whether the multielement analysis of blood samples from welders could give any information about the work environment. To extract as much information as

possible from the multivariate data, we have used the methodology of pattern recognition (3, 11, 12).

Pattern recognition is a methodology which extracts the regular behavior of the data from each group and describes the regularities in terms of parameter values in a simple mathematical model. Specifically, the methodology permits the testing of the hypothesis that the variables vary independently of each other and reveals whether variations within each group are correlated. In the present data the variables showed rather weak, but significant, correlation patterns, which motivates the use of the more complex data analysis.

MEASURED DATA

The concentrations of 17 trace elements (table 1) were measured with mass spectrometry in whole blood samples of the following groups of employees: (a) 68 individuals not working with welding. This reference group was selected from the

Table 1. Properties of the data. All variables were transformed by $y \rightarrow \log_e (1 + ay)$. Normal range of each element based on the 95 % confidence interval of $\log (1 + ay)$.

Variable no.	Name	Typical value ^a μg/g wet tissue	Min	Max	Skewness of log (1 + ay)	a	Mean ^b log (1 + ay)	SD ^b log (1 + ay)	
1	Pb	Lead	0.04	0	1.870	—1.1	1,000	3.664	1.935
2	Sr	Strontium	0.04	0.007	0.27	—1.8	1,000	3.825	0.896
3	Rb	Rubidium	10	3.5	25	0.6	1	2.375	0.439
4	Br	Bromine	0.2	0.06	0.75	—0.3	1,000	5.349	0.636
5	Ga	Gallium	0.025	0.0004	0.51	—1.2	1,000	3.270	1.482
6	Zn	Zinc	1.7	0.63	4.2	0.6	10	2.882	0.444
7	Cu	Copper	0.7	0.35	1.4	—0.4	10	2.113	0.301
8	Co	Cobalt	0.09	0.016	0.49	—1.9	1,000	4.520	0.835
9	Fe	Iron	520	270	980	—0.4	1	6.247	0.320
10	Mn	Manganese	0.06	0.009	0.37	—2.5	1,000	4.118	0.901
11	Cr	Chromium	0.03	0.002	0.34	—1	1,000	3.443	1.197
12	Ca	Calcium	47	27	83	0.0	1	3.873	0.275
13	K	Potassium	2,500	1,500	4,000	—1	1	7.811	0.247
14	S	Sulfur	390	200	760	0.1	1	5.965	0.333
15	P	Phosphorus	540	290	1,010	0.2	1	6.295	0.314
16	Si	Silicon	6	1.4	18	0.2	1	1.913	0.524
17	Mg	Magnesium	22	8	60	—2	1	3.135	0.483

^a Typical values [from mean of $\log (1 + ay)$].

^b Data scaled by subtraction of mean and subsequent division by the standard deviation. Hence, the original data are obtained from the transformed data as

$$Y_{\text{orig}} = \{ \exp[Y_{\text{tr}} \cdot \text{SD} + \text{Mean}] - 1 \} / a.$$

same workplaces as the welders. Some of them were exposed to dust from the same general atmosphere as the welders. (b) 23 welding operators working with shielded metal arc welding (arc welding with coated electrodes) on different types of stainless steel containing between 10 and 20 % nickel and chromium, together with much smaller percentages of some other metals. (c) 7 welders working with gas shielded welding (TIG) on stainless steel of the same general composition as that on which group b worked. (d) 28 welders working with gas shielded welding (MIG) on aluminium and aluminium alloys, containing 1–5 % magnesium and/or silicon and/or zinc. (e) 23 welders working with gas shielded welding (TIG) on aluminium and aluminium alloys with the same general composition as that on which group d worked.

All welders were working between 40 and 100 % of their shifts on the operation in question. The day the blood samples were taken the welders working with stainless steel were exposed to welding fumes with an average concentration of 5 mg/m³ (range 0.8–21 mg/m³). The median of the chromium exposure, calculated as CrO₃, was 0.3 mg/m³ (range 0.01–1.4 mg/m³). The median of the nickel exposure was 0.03 mg/m³ (range 0.0007–0.16 mg/m³).

The welders working with aluminium and its alloys were exposed to welding fumes with an average concentration of 12 mg/m³ (range 0.3–150 mg/m³). All concentrations were measured in the breathing zone inside the welding mask or helmet. The general atmosphere in the workroom, to which some of the referents were exposed, had an average dust concentration of 2 mg/m³. The spread of concentrations in general workroom atmospheres was much less than that of concentrations of welding fumes inside the mask or helmet. No dust concentration in the general atmosphere exceeded the occupational health standard limit of 5 mg/m³ for fine particles of inert dust.

In view of the negative result of this investigation, no further details of the exposure situation were considered necessary for this report.

It has not been possible to analyze all the elements of primary interest, since some elements were present in concentra-

tions below the detection limit of the method. Some of the elements that were possible to detect are of less immediate interest. The 17 elements treated in this investigation are those detectable in the blood samples. Among the 17 elements only zinc, manganese, chromium, silicon and magnesium were assumed to be important constituents of the welding fumes in the operations studied. Gas shielded welding (TIG and MIG) on aluminium and its alloys produced, among others, zinc, silicon and magnesium in the fumes. The most important component, aluminium, could not be determined in the blood (see the section on sampling method and chemical analysis). Shielded metal arc welding on stainless steel produces chromium, manganese, and silicon (from the electrode coating). Nickel could not be determined in the blood (see the section on sampling method and chemical analysis). Iron is an important component of welding fumes in this case, but since this metal is also an important natural constituent of blood, it is scarcely likely that data on blood concentrations of this metal contain information concerning the exposure situation. Other elements determined in the blood samples were included in the data analysis to see if the data reflected the exposure situation, although the concentration of these elements in the breathing zone was considered to be insignificant. There is also the possibility that the exposure may have effects on the blood level of other trace elements than those found in welding fumes.

SAMPLING METHOD AND CHEMICAL ANALYSIS

Venous whole blood samples were taken at the end of the shift in plastic flasks with heparin added. The samples were deep frozen and stored until analysis.

About 0.5 g of the blood samples was mineralized in a low temperature oven, type 500 A, Uppsala Elektronik, operating with activated oxygen. The temperature never exceeded 180°C. The procedure was repeated several times after hydrochloric acid had been added. The ash content of the whole blood was about 1.5 % after the completed mineralization.

The mineralized blood sample was analyzed with inorganic spark source mass spectrometry at the Department of Analytical Chemistry, Royal Institute of Technology, Stockholm. All elements with an atomic mass of 7 (Li) or more can be analyzed with a precision of about 30 %. The detection limit varies according to the details of the procedure between 0.01 and 0.1 ppm in the sample (ash). When the total analytical procedure is considered, this means a detection of at least about 0.002 $\mu\text{g/g}$ wet tissue. The detection limit is approximately the same for all elements. The determination of trace elements is sometimes disturbed by the presence of other elements, e.g., nickel and aluminium in the presence of iron.

According to the published data on element concentrations in human organs (1), all the elements determined in the blood samples in this investigation have concentrations in blood (in some cases plasma) of healthy people well above the detection limit. This is one indication that the reported concentrations are not due to artifacts. The concentration levels of the elements represented in table 2 agree approximately with published data.

Values below the detection limit were treated as zero. Since such values were rare among the elements selected for statistical treatment, the data analysis was not affected.

DATA ANALYSIS

For the data analysis, the computer program ARTHUR (4) was used for the pre-processing and plotting of the data, while the program SIMCA-1B (18) was used for the SIMCA analysis.

Preprocessing

SIMCA and most other data analytical methods are most efficient for fairly centrally distributed data. Since several of the variables showed appreciable skewness, the data were transformed from y to $\log(1 + ay)$, which removed most of the skewness (table 1). So that each variable would have the same weight in the analysis, the data were then regularized (auto-scaled) by the subtraction of the variable means and subsequent division by the variable standard deviations (table 1).

Table 2. Modeling power and resulting α_i (q) values.

Variable no.	Modeling power ^a	Parameters α_i (q) ^b				
		Class 1	Class 2	Class 3	Class 4	Class 5
1	0.0	0.093	0.096	0.474	-0.524	0.124
2	0.08	-0.026	0.043	0.541	-0.093	-0.016
3	0.21	-0.186	0.139	0.571	-0.111	0.373
4	0.17	-0.004	-0.048	-0.302	0.241	-0.153
5	0.13	0.026	-0.119	-0.148	-0.150	0.267
6	0.24	-0.059	0.296	-0.033	-0.144	0.065
7	0.20	-0.127	0.141	0.286	-0.133	0.310
8	0.25	0.030	-0.048	-0.103	-0.194	0.227
9	0.17	-0.141	-0.062	0.611	0.082	0.192
10	0.14	0.088	0.040	-0.275	-0.057	-0.148
11	0.29	-0.144	0.155	0.231	0.055	0.134
12	0.34	-0.117	-0.144	-0.106	0.299	0.157
13	0.02	0.096	0.104	-0.727	-0.055	-0.100
14	0.31	-0.044	-0.235	0.207	0.071	0.217
15	0.16	-0.035	-0.096	0.121	-0.003	0.167
16	0.05	-0.140	0.256	0.484	0.010	0.000
17	0.23	0.001	0.095	0.228	-0.119	-0.024

^a Calculated as $1 - \sum S_{qi}$, where S_{qi} is the residual standard deviation of variable i in class q after the extraction of the average α_i and one term $\beta_i\beta_n$ ($A = 1$ in equation 1).

^b Averages of variable i for class q of scaled data (see table 1).

Plot

Fig. 1 shows an eigenvector projector (13) of the preprocessed data. No apparent patterns or differences between the five groups can be seen.

SIMCA analysis

For the data analysis we have used the SIMCA method (19, 20), which is a method of pattern recognition (discriminant analysis) specifically designed for chemical data analysis. The method is based on the fact that multivariate data (M variables) measured for a group of similar objects (in this case blood samples) can be closely approximated by the principal components model in equation 1.

In the present application the objects correspond to the collected blood samples and the M variables measured for each object are the 17 trace-element concentrations. Thus the data element $y_{ik}^{(q)}$ denotes the (transformed) concentration of the i th trace element measured on the k th blood sample in group q . The number of groups, Q , is 5, the reference group and four groups of welders.

$$y_{ik}^{(q)} = \alpha_i^{(q)} + \sum_{a=1}^{A_q} \beta_{ia}^{(q)} \theta_{ak}^{(q)} + \varepsilon_{ik}^{(q)} \quad (1)$$

The parameters in equation 1 have a simple interpretation. Thus the parameters $\alpha_i^{(q)}$ are the averages of the variables (index i) in group q . The parameters $\beta_{ia}^{(q)}$ and $\theta_{ak}^{(q)}$ express the correlation structure of the group, the former being specific for the variables and the latter being specific for the objects (index k). The number of product terms in equation 1, A_q , determines the complexity of this correlation structure. Finally, the residuals $\varepsilon_{ik}^{(q)}$ describe the nonsystematic part of the data in group q , the "random noise."

Equation 1 has a simple geometrical interpretation. In the M -dimensional space, defined by giving each of the M variables one coordinate axis, the systematic part of equation 1 is an A_q -dimensional hyperplane. Each data point deviates from this hyperplane by the amount of

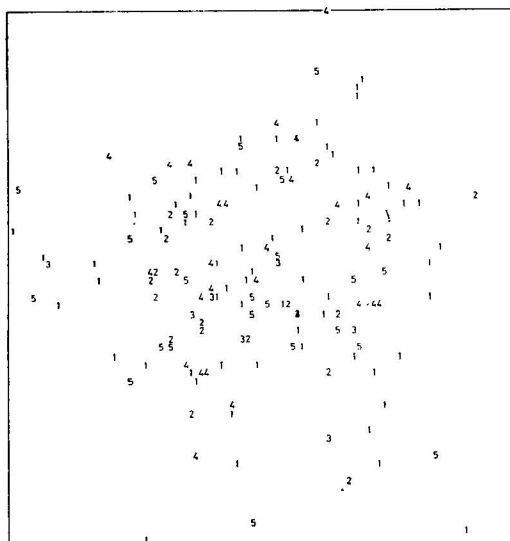


Fig. 1. Eigenvalue plot of the data. Each number in the plot corresponds to one individual in that class. The eigenvalue plot preserves as much of the variance in the original 17-dimensional space in the projection down to the 2-dimensional space of the plot.

$\varepsilon_{ik}^{(q)}$ in each dimension i . With $A_q = 1$, the model is particularly simple, being a straight line in M -space.

In the data analysis equation 1 is fit separately to the data of each group q by nonlinear least squares. The resulting parameters can then be interpreted and, which is equally important in the present analysis, may be used for the determination of how well objects in the other groups fit each of the Q class models.

The fitting of an object not in class q to the q th class model is then made with the help of simple multiple regression. The parameters α and β are then fixed to the values calculated for the q th class, and only the parameters θ need be determined for this object. The resulting residuals, ε , indicate how well the object fits the class; specifically, the residual standard deviation (SD) $s_k^{(q)}$ measures the orthogonal distance between the point of the object k to the class model in M -space:

$$s_k^{(q)2} = \sum_{i=1}^M \varepsilon_{ik}^{(q)2} / (M - A_q) \quad (2)$$

The residuals $\varepsilon_{ik}^{(q)}$, obtained when fitting objects to their own classes, are used for the calculation of the measures of relevance of the variables. Thus, a variable i , having a large residual SD, s_i , over all groups, is considered to be less relevant than a variable having a smaller residual SD (one must remember that the variance of the original data is 1.0 for each variable due to the preprocessing of the data). The modeling power of a variable calculated as $1.0 - s_i$ is used in the present analysis to exclude irrelevant variables (see table 2).

$$s_i^2 = \frac{\sum_{q=1}^Q \sum_{k=1}^{n_q} \varepsilon_{ik}^{(q)2}}{\sum_{q=1}^Q (n_q - A_q - 1)} \quad (3)$$

Determination of the number of components, A_q , in equation 1

This process is critically important for the subsequent data analysis. The value of A_q , i.e., the number of product terms used in equation 1 to approximate the data of each class, corresponds to the amount of systematic structure in the data. A high value of A_q corresponds to the assumption that most of the variation in the data is systematic and just a little is "random noise," and vice versa. Hence the use of an A_q that is too large will correspond to an overestimation of the structure in the data and some results of the data analysis will be mere artifacts.

Too small an A_q leads to underutilization of the information inherent in the data, i.e., bad research economy.

In SIMCA, A_q is determined by cross-validation. This is a procedure whereby part of the data is left out of the analysis and predicted from the rest of the data and equation 1 with different values for A_q . The exclusion of part of the data is rotated over the whole data set and the value of A_q is finally selected to give the smallest differences between the predicted and the actual values for the excluded data.

In the present application the cross-validation procedure showed that all five groups are best described by equation 1 with $A_q = 1$. The same results were obtained in later stages of the analysis when some of the variables were excluded.

Analysis with 17 variables

After the determination of A_q to 1 in all five groups, equation 1 was fitted separately to each of the five data-sets of the five groups by the method of non-linear least squares. The resulting parameter values are not discussed in this report since some of the variables had a very low modeling power, i.e., they showed little regular behavior within each class (column 1 in table 2). The five variables with the lowest modeling power were therefore excluded from further analysis (no. 1, 2, 5, 13 and 16, that is, lead, strontium, gallium, potassium and silicon).

Table 3. Resulting $\beta_i^{(q)}$ values for classes 1 to 5.

Variable no.	β_{il}				
	Class 1	Class 2	Class 3	Class 4	Class 5
3	0.308	0.357	0.460	0.138	0.385
4	0.103	0.160	0.128	0.110	0.340
6	0.274	0.410	0.150	0.279	0.290
7	0.309	0.393	0.079	0.396	0.216
8	0.282	0.192	0.111	-0.016	0.136
9	0.309	0.175	0.155	0.262	0.397
10	0.134	0.063	0.680	0.307	0.350
11	0.387	0.200	0.161	0.240	0.176
12	0.288	0.359	0.328	0.238	0.313
14	0.347	0.374	0.309	0.465	0.294
15	0.307	0.191	0.095	0.474	0.221
17	0.292	0.328	0.071	0.129	0.204

Analysis with 12 variables

With the remaining 12 variables, the data were reanalyzed. Cross-validation still gave $A_q = 1$ for all groups. The parameter values resulting from the least squares fitting of equation 1 to each of the five groups are shown in tables 2 and 3. The variable averages in each class, α_i , were the same whether 17 or 12 variables were utilized.

The distances between classes p and q were calculated as the residual standard deviation (SD) of the data in group p when fitted to equation 1 with the parameters of group q . The distances between groups a to e , normalized by division with the residual SD of class q , are shown in table 4. It is seen that there is no appreciable difference between any of the classes; for significance a distance value of at least 2.0 is required.

Analysis with 6 variables

The six variables with the highest negative α values in class 1 (no. 3, 7, 9, 11, 12 and 16, that is, rubidium, copper, iron, chromium, calcium and silicon) were selected for a final analysis for the investigation of whether the variables showing on the average the largest increase from referents to welders contained any information. The resulting distance matrix calculated on the basis of these variables showed the same lack of difference between the groups as before and is therefore not given.

Analysis with 12 variables, combined groups

Since the welding methods of groups b and c were related, as well as those of groups d and e , the data were combined to form the following three groups: referents, users of methods $b + c$, and users of methods $d + e$. The data were reanalyzed as described in the foregoing, but they gave the same negative results, which are not given, however, as they were identical with earlier results.

Table 4. Distances between classes 1 to 5 (12 variables). D_{ij} is the residual standard deviation when the data of class i are fitted to the model of class j (equation 1 with $A = 1$ and parameters in table 1 and table 2). The elements D_{ij} are then normalized through division by the residual standard deviation obtained when the data for class j are fitted to the model of class j , i.e., "its own model." The sixth column shows the residual standard deviation of the classes.

	$j = 1$	2	3	4	5	SD
$i = 1$	1.00	1.10	1.41	1.04	1.05	0.862
2	0.94	1.00	1.28	0.98	0.97	0.771
3	1.01	1.09	1.00	1.05	0.96	0.753
4	1.03	1.13	1.37	1.00	1.06	0.839
5	1.01	1.12	1.30	1.05	1.00	0.822

Finally, the data were combined to form two groups, i.e., referents and welders. The data analysis still gave the same negative results, which accordingly are not given.

Linear discriminant analysis

To check the results of the SIMCA analysis, we also used linear discriminant analysis (5, 16) to analyze differences between the five groups. This analysis gave the same lack of significant differences between the groups and is therefore not presented in detail.

SUMMARY OF THE RESULTS OF THE DATA ANALYSIS

The main conclusion is that the data in the present study do not contain any information about differences in the pattern of trace-element concentrations in blood, either between referents and welders or between welders using different welding methods. Most of the variation in the data, about 80 % of the standard deviation, is nonsystematic, "random" (table 4).

The data structure differs slightly between the groups in the variable means α_i (table 2). These small differences are

completely masked by the regular variation within the groups described by $\beta_i^{(q)}$ $\theta_k^{(q)}$ (table 3) and the "random" variation $\varepsilon_{ik}^{(q)}$. The θ values in all classes range between ± 5 with a standard deviation of about 2. The differences between the classes are further diminished because of the similarity of the β values of all the groups (table 3). Thus neither the averages of the variables in each group nor the regular variation within the groups nor their combination can be used to distinguish the group of an incoming blood sample of unknown assignment.

The averages α_i , when transformed back to the original data space, give the "typical" values for each element shown in table 1. A 95 % confidence interval based on the standard deviation of the logarithmated data is also given in table 1. One must remember that there is a weak correlation between the variables, so that if a variable is large for a given individual, there is also a tendency for the other variables to be large, a probability of about 0.6 that any other variable is above its typical value.

DISCUSSION

The absence of statistical differences between the five groups may be due to a large biological variation in blood pattern within each group that obscured any smaller but systematic differences between the groups. The low precision in the analyses, about 30 %, is not considered to affect seriously the result, since the number of samples in most groups was high. It is obvious from the exposure data that the exposure varied considerably within each group. The differences in the type of exposure between the two main groups, stainless steel welders and aluminium and aluminium alloy welders, is, however, very high. The exposure to chromium in the former group should be reflected in the blood concentration of this metal, if blood concentrations are significantly affected by chromium exposure at the level in question.

The most important conclusion of the investigation is that the chromium exposure of the welders could not be controlled by blood concentrations of this metal. This

finding is consistent with others (14) and it also indicates that it has little to do with the analytical method.

The concentration of zinc and magnesium in the breathing zone of welders working on aluminium alloys is low, and the blood level was hardly expected to be affected in this case. The same is true for manganese in the exposure picture for stainless steel welders. The manganese concentration in the welding fumes in this case is low.

Since no significant differences were found in the blood concentrations of the investigated groups, the concentrations reported as typical values in table 2 may be considered to be normal background concentrations of the elements in question in healthy employees in the workshop industry. As has already been mentioned, the concentrations found for all of these 17 elements agree approximately with published data (1).

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