Bull. Environ. Contam. Toxicol. (1990) 45:711–717

Environmental Contamination and Toxicology

Levels of Zinc, Copper, and Lead in Wines from the Area South of Seville

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The assay of inorganic ions in wines, both major elements (Mg, Fe, Na, K, Ca), and trace elements, has long been the concern of most enologiers (Ribereau-Gagon et al. 1972; Amerine et al. 1976), as they consider that the quality of wine depends, to a considerable degree, on the content of these elements. Some of them, such as copper and zinc, occur naturally in grapes, in certain enzyme complexes. Their presence in the vine depends on the type and pH of the soil, the ripeness of the grape and the general climatic conditions (Fernandez et al., 1987).

However, the presence of metals in wines can be of technological origen (Cordonnier 1965) due to residues of agrochemical products used as agricultural insecticides and fungicides, which contain copper and lead. It can also be due to contamination from deteriorated metallic receptacles; from environmental contamination where industrial complexes exist close to the vinyards; and, in the case of lead, from exhaust gases from motor vehicles, among other sources.

The purpose of our study is to find out if wine elaborated or consumed in the area immediately south of Seville, Spain, presents high metal content (Cu, Pb, Zn); if this is due to environmental contamination; and also if it would be possible to use wine for monitoring exposure "in situ". For this purpose the content of these 3 metals in 150 samples of wines has been studied. We used wines sold loose and consumed in large quantities in the southern area of the province of Seville.

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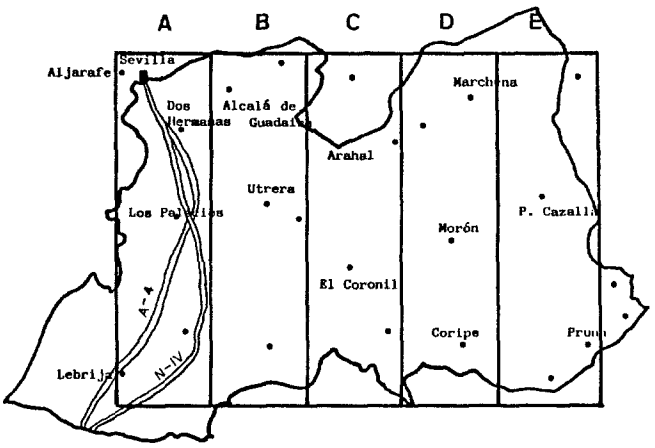

Figure 1. Map of the Southern Area of the province of Seville, divided into the five sampling areas.

MATERIALS AND METHODS

The vine growing and wine producing area in South Seville was divided into five areas (A, B, C, D and E) as shown in Fig.1. From each area, we took thirty samples of wine, of which 60% were "finos", 30% "olorosos", and 10% red wines.

The metals assayed were copper, zinc and lead. In the determination of the first two, the wines richer in carbohydrates were digested using the method recommended by Ough et al. (1982), which consists in the evaporation of 50 ml of wine to one third of its volume, followed by heating with concentrated nitric acid and hydrogen peroxide. The residue is made up to the original volume with deionized distilled water. Wines with low carbohydrate content were analysed directly. For the detection of lead the samples were diluted 1:3 with deionized distilled water.

When the samples needed digestion, the standard solutions were treated in the same way. In all other cases standard solutions were prepared to contain alcohol and carbohydrates in similar proportions to those found in the samples (Slavin et al. 1965, Strunk 1967 and Frey et al. 1969).

The final solutions were analysed by Atomic Absorption Spectrophotometry (AAS), with flame for copper and zinc, and flameless for lead, in a Perkin Elmer model 2380 Spectrophotometer equipped with an HGA-500 graphite furnace and AS-40 automatic sampler from the same company.

Table 1. Copper, lead and zinc content in wines of the area (µg/mL)

Type of Wine	N		Zinc	Copper	Lead
Fino	90	Range	0.01-37	0.01-24	0.005-03
		Average	3.90	0.96	0.05
		Standard Dev.	±7.60	±3.2	±0.05
Oloroso	45	Range	0.01-26	0.01-13.84	0.012-0.25
		Average	5.40	1.49	0.08
		Standard Dev.	±6.90	±2.66	±0.06
Tinto	15	Range	0.01-0.85	0.01-0.71	0.012-0.19
		Average	0.3	0.18	0.05
		Standard Dev.	±0.2	±0.22	±0.05

N= number of samples

In air-acetylene flame atomization, a mixture of 23 and 40 flow units respectively, was used. The resonance lines measured were 324.7 nm for copper and 213.9 nm for zinc. In flameless AAS the solutions were subjected to a sequence of steps of drying (120°C), mineralization (900°C) and atomization (1900°C) using an 0.2% solution of Diammonium hydrogen phosphate as matrix modifier. For each measurement, 20 µL of sample and 20 µL of modifier were injected. The resonance line for the measurement of lead was at 283.3 nm.

All the water used was purified in a Milli-Q system (Millipore Corporation) at a resistance of 18 megaohms/ /cm. The reagents were Suprapur (Merck) quality and the standards Titrisol (Merck) quality.

The laboratory material to be used was kept previously in a 5% nitric acid solution all night and subsequently washed with deionized water and dried in a dust free atmosphere.

Six measurements per sample were made for the assay of zinc and of copper, obtaining an average standard eviation of 0.01 µg/mL for zinc, and of 0.02 µg/mL for copper.

For lead, four measurements were made per sample with a standard deviation of 1.1 µg/mL.

For each type of wine and metal an analysis of variance with one single factor (area) was carried out. A Duncan test was applied for the multiple comparisons.

Table 2. Lead, copper and zinc in "Fino" wines from the areas studied. Results of the comparative statistical study.

Area		Zn	Cu	Pb
A	Average	10.40	6.30	0.13
	Standard Dev.	±11.00	±4.30	±0.08
B	Average	5.20	0.52	0.03
	Standard Dev.	±5.10	±0.60	±0.02
C	Average	2.20	0.41	0.04
	Standard Dev.	±1.70	±0.38	±0.02
D	Average	0.50	0.30	0.04
	Standard Dev.	±0.43	±0.30	±0.02
E	Average	1.20	0.30	0.04
	Standard Dev.	±1.05	±0.27	±0.01

* P<0.01

\+ P<0.05

	D	E	C	B	A
D					
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A	*	*	*	*	*

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RESULTS AND DISCUSSION

The rates of recovery from three standard solutions of copper (II) (treated by wet digestion in open flasks in the same way as the samples) were the following: 1 µg/mL, 98.3%; 2 µg/mL, 102.1%; 3 µg/mL, 101.5%.

For zinc (II) they were: 0.1 µg/mL, 99.8%; 0.5 µg/mL, 99.5%; 1 µg/mL, 100.9%. These results indicated that there is no loss of these elements during the process of digestion.
In the case of lead, to reveal the influence of the matrix, both the direct method and that of standard addition were applied. No significant differences between the two calibration techniques were observed, consequently the direct method was used.

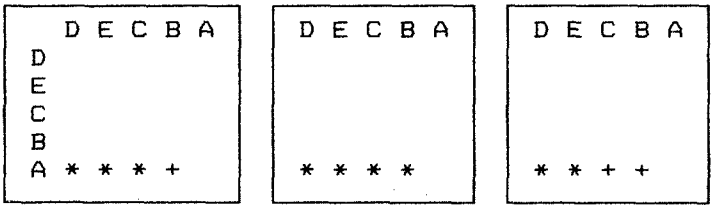
Table 1 shows the copper, zinc and lead content in the wines studied. It can be seen that the zinc concentration is higher in the "olorosos" ($x=5.40\pm6.90$) than in the "finos" ($x=3.90\pm7.60$) ($p<0.005$) and even

Table 3. Lead, copper and zinc in "Oloroso" wines from the areas studied. Results of the comparative statistical study.

Area		Zn	Cu	Pb
A	Average	16.10	7.21	0.12
	Standard Dev.	±15.40	±6.11	±0.07
B	Average	7.70	0.62	0.05
	Standard Dev.	±6.90	±0.70	±0.03
C	Average	2.60	0.70	0.05
	Standard Dev.	±2.30	±0.65	±0.04
D	Average	0.70	0.80	0.07
	Standard Dev.	±0.62	±0.73	±0.03
E	Average	1.70	0.50	0.07
	Standard Dev.	±1.48	±0.48	±0.03

* P<0.01

+ P<0.05



higher than in the red wines (x=0.30±0.2) (p<0.005). Although in general normal concentrations of this element are close to 0.5 µg/mL most studies show high zinc content in andalusian wines than in wines from other regions of Spain. The type of wine is an important factor in the concentration, higher values always being observed in "olorosos" than in "finos" (Fernandez Pereira et al., 1987).

The copper content follows a similar pattern, being more abundant in "olorosos" (x=1.49±2.66) than in "finos" (x=0.96±3.27) (p<0.005) and red wines (x=0.18±0.22) (p<0.0005). Normal copper content ranges from 0.03 to 0.80 µg/mL. In andalusian wines high values usually appear; in "finos" from Montilla, the range is between 0.47 and 1.48 µg/mL. If we eliminate three samples of "fino" in which the copper content varies considerably from the average, the concentrations of our samples (0.96±3.27) are within the range found in other areas of Andalucia.

With regard to lead, although there are differences in the three types of wine, the results show that the lead content in most of the samples is below the level considered dangerous. The legal limit established for lead content varies from one country to another (Sharper et al. 1982) being 0.1 µg/mL in Spain. Only ten of the samples analysed exceeded this value. The values in the wines of this region are inferior to those described by other authors using scant samples of wines from other regions of Spain (Marks et al., 1987) Ribeiro 0.25, Jerez 0.36, Rioja 0.94 µg/cc. These differences are justified among other reasons by the fact that we used poorer quality wines, produced in this same year, and for the most part, sold loose. Old wines bottled with lead foil closures (Marks et al., 1957) (Wai et al., 1979) have a higher content of this metal than new wines (Vives et al., 1980) (Grandjean et al., 1981). The higher lead content observed in the "oloroso" wines is a fact both known and described (Fernandez Pereira et al., 1987).

Tables 2 and 3 show copper, zinc and lead levels in "fino" and "oloroso" wines respectively, from the different areas, and also statistical analysis comparing the various areas with one another, with indication of the limits of confidence. A similar statistic evaluation was not included for the red wines due to the low number of these studied.

A study of the values appearing in these tables shows one of the most interesting conclusions of our study, that is, the evidence of higher lead levels in new wines from vine-growing and wine-producing areas of Seville (Aljarafe, Los Palacios and Lebrija) included in area A, than in the others, further away.

Probably this fact is fundamentally due to the proximity of the city of Seville and of the two roads A-3 and N-4 which lie parallel along the area and carry a lot of traffic. The contamination would be due to the deposit of dust on the bunches of grapes, as the capacity of absorption of lead through the roots of the plants is almost nil according to Ganje et al. (1983) and Douglas et al. (1976) who have demonstrated correspondence between the lead content of plants, soil and air and the distance from large motorways.

Another of the conclusions of the study is the greater proportion of copper and zinc in the wines from area A. The presence of these elements can be considered to be due to the use on the vines of fungicides such as zinc ditiocarbamate (zineb) and copper oxychloride, widely used in Spain; the greater content in the wines from area A could be explained as due to application of the fungicides shortly before harvesting.

Acknowledgments. The authors received a research Grant from the Social Services Health Research Fund for this work, and also wish to thank J.A.Guerrero and Dra.P. Sanz for assistance with statistical analysis, S.A. Owens,M.R.Ph.S. for translation, and Miss A.M.Eguino for assistance in the preparation of the manuscript.

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Received October 16, 1989; accepted February 19, 1990.