Continuous Sorbent Preconcentration for the Electrothermal Atomic Absorption Spectrometric Determination of Ultratrace Amounts of Cobalt in Milled Wheat Fractions

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A method for the rapid determination of cobalt at ultratrace levels was applied in flour and flour byproducts (shorts and bran) obtained from various types of wheat that includes on-line preconcentration and electrothermal atomic absorption spectrometry is proposed. Solutions in 0.1 mol/L HNO_3 of milled wheat fractions subjected to wet ashing are preconcentrated in a straightforward flow-injection system by sorption on a RP-C_{18} column following chelation. The eluent, ethanol, is carried by an air stream, and the chelate is eluted and collected in a 500 μL PTFE autosampler cup. The determination of cobalt features a precision (RSD) of 6% for a concentration of 0.2 ng/mL and a sensitivity (slope of the calibration graph) of 0.38 ± 0.03 A s × ng/mL. The cobalt content in each type of wheat was found to be influenced by its geographical origin and texture. Also, it was found to depend on the amount of bran present in the milled wheat fraction. On the other hand, it is not significantly affected by the technological processes involved in wheat milling at a flour-producing factory.

Keywords: *Cobalt; sorption; wheat products; electrothermal atomic absorption spectrometry*

INTRODUCTION

The analysis of trace metals in foods has become a subject of considerable interest because trace metals play important roles as enzyme activators or inhibitors in biological processes (WHO, 1996). Deficiencies in some essential trace metals have occasionaly been detected; however, the area of metal toxicity from accidental or technological contamination is of special interest in the nutritional field (Stringari et al., 1998). Cereals are important staple foods in most countries and an essential source of mineral elements for humans. The mineral content of wheat flour is highly variable and depends on a number of factors such as the type of soil used to grow the wheat and the technological processes involving its milling to obtain flour, which can generate contamination through contact with stainless steel materials (Mahoney, 1982). Cobalt is essential for living species as it is required to synthesize vitamin B_{12} (Underwood, 1977).

Minerals in foods are the residual constituents remaining following ashing of plant and animal tissues; they fall into two categories: major elements and trace elements. The use of atomic spectrometry for the direct determination of trace elements in food is as old as the technique itself. A number of sample digestion and preconcentration methods for the determination of trace metals in foods by atomic absorption spectrometry (AAS) have been reported (Taylor et al., 1999). The metals in foods most frequently determined using the slurry technique are Zn, Pb, Fe, Cu, Mn, Cd, Ca, Mg, and K, followed by Cr, Na, and Hg, among others; cobalt, whose content in foods is pivotal in nutritional and toxicological terms, has only occasionally been deter-

mined in this manner, however (Ebdon et al., 1990; Gervais and Salin, 1991; Mochizuki et al., 1991). The ETAAS determination of cobalt in foods generally entails dry or wet digestion (Wu and Wu, 1988); occasionally, however, extraction methods have been used for this purpose (Borggaard et al., 1984; Blanchflower et al., 1990) to improve the sensitivity (cobalt is present at very low levels in foods). Cobalt in cereals has generally been determined by photometry (Wang, 1991) in addition to miscellaneous other techniques; however, few ETAAS methods for this purpose have so far been reported (Borggaard et al., 1984; Jorhem and Sundstrom, 1993; Zhang and Pan, 1997).

On-line separation and preconcentration methods have contributed to increasing sample throughput and decreasing sample contamination, which are critical in trace analysis. A monograph on a continuous flow system with atomic spectrometric detectors was recently published (Sanz-Medel, 1999) where the potential flow analysis for on-line sample pretreatment, separation and preconcentration, and the applications of this combination are explained in detail. Automatic preconcentration methods for cobalt determination have usually been applied to water samples (Yan et al., 1997; Balaji et al., 1998); however, only one preconcentration method for the determination of this metal in wheat flour by ETAAS has to our knowledge been developed (González et al., 1999). Such a method uses fullerene as sorbent material because it exhibits better sorbent properties than conventional solid materials when using isobutyl methyl ketone as the eluent (Gallego et al., 1994). However, RP-C_{18} performs in the same way as fullerene when using ethanol as the eluent in the ETAAS technique. In this paper, the solid phase extraction system proposed elsewhere (González et al., 1999) for cobalt determination in wheat flour using fullerene as the sorbent was initially adopted, but using silica RP-

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C_{18} as sorbent. In addition, the influence of wheat texture and geographical origin on the cobalt content and the effect of technological processes on potential cobalt contamination or losses from milled wheat fractions were also studied.

MATERIALS AND METHODS

Apparatus. A Perkin-Elmer model 1100-B atomic absorption spectrometer (Überlingen, Germany) and an HGA-700 graphite furnace atomizer were used throughout. A hollow cathode lamp operating at 30 mA and a spectral bandwidth of 0.2 nm were used for the determination of cobalt at its primary resonance line of 240.7 nm. Samples were injected with a Perkin-Elmer model AS-70 autosampler. Argon at a flow rate of 300 mL/min was used as the inert gas except during the atomization step, where the flow was stopped. The injected volume was 20 μL, and pyrolytic graphite-coated tubes and pyrolytic graphite L'vov platforms (Perkin-Elmer) were used. Background-corrected integrated absorbance (integration time, 6 s) was used as the analytical signal. The furnace program was as follows: ramp-dry 1 to 80 °C in 5 s, hold for 15 s, ramp-dry 2 to 110 °C in 1 s, hold for 15 s, ramp-pyrolyses to 1500 °C in 6 s, hold for 20 s, atomize by heating to 2600 °C in 0 s, hold for 6 s, clean up by heating at the highest rate to 2650 °C in 1 s, and hold for 4 s. The flow manifold consisted of a Gilson Minipuls-2 peristaltic pump (Villiers-le-Bel, France) furnished with poly(vinyl chloride) tubes, two Rheodyne 5041 injection valves (Cotati, CA), and a laboratory-built sorption minicolumn packed with 80 mg of silica RP-C_{18} (1.6 cm × 3 mm i.d.). The column (with a usable life of at least 6 months) was made from a PTFE capillary with an i.d. of 3 mm that was sealed at both ends with small cotton-wool plugs to prevent material losses. It was conditioned prior to each use by flushing 1 mL of 0.1 mol/L HNO_3 and cleaned occasionally with 2 mL of ethanol.

Reagents and Standard Solutions. A stock solution of cobalt (1000 mg/L) was prepared by dissolving 1.000 g of the metal in a small volume of concentrated HNO_3 and diluting to 1 L with 1% v/v HNO_3. A 0.1% w/v aqueous solution of ammonium pyrrolidine dithiocarbamate, APDC (Aldrich, Madrid, Spain), which remained stable for at least 3 days, was prepared. A 1% w/v solution of neocuproine (Merck, Darmstadt, Germany) in ethanol (Merck) was also prepared. Hydroxylammonium chloride and polygosyl-bonded silica reversed phase sorbent with octadecyl functional groups (RP-C_{18}), 60–100 μm particle size (Millipore, Madrid, Spain), was employed as the sorbent material. Standard solutions (25 mL) containing 0.04–1 ng/mL cobalt were all freshly prepared by appropriate dilution of a stock solution (1000 mg/L) in 0.1 mol/L HNO_3. An organic stock solution of Co-PDC in ethanol was prepared by mixing 1 mL of the 1000 mg/L cobalt stock solution with 10 mL of the 0.1% w/v APDC solution, prepared in ethanol, and diluting to 50 mL with more ethanol. Working-strength solutions were prepared on a daily basis by diluting the 20 μg/mL stock solution with ethanol. Solutions of potentially interfering ions were prepared by dissolving the amount of each metal or salt needed to obtain a 100 μg/mL concentration of each ion.

Wheat Materials. All samples of whole grain and their fractions in different degrees of milling and from different geographical locations were obtained from an Andalusian flour-producing factory with a production of 570 tons/day. A 5 kg portion of sample was taken from their respective horizontal stores (hugh in extension but not very high, to avoid an excess of moisture in the grain) by introducing an aluminum lancet through the roof at various points of the store. Subsamples (0.5 kg) were then milled. Wheat fractions were obtained using the "split-run milling" technique at a pilot plant as depicted in Figure 1A. Conditioning of the wheat to prepare it for the milling process involved the addition of variable amounts of water depending on the wheat texture up to a moisture content about 17%. Mills were all stainless steel. To study the influence of the wheat texture and geographical origin, and also the
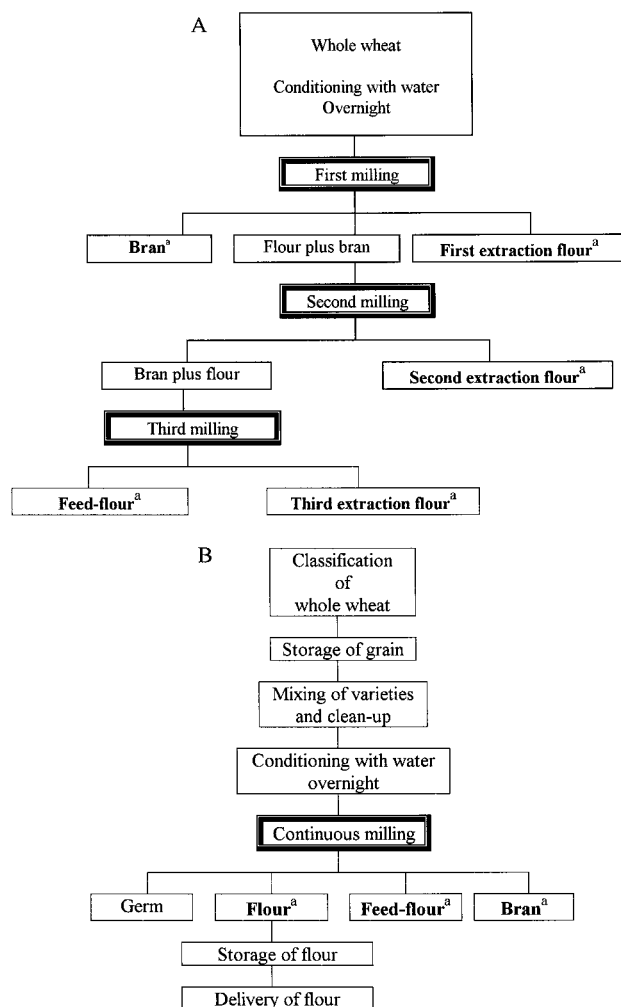

Figure 1. Flowchart of the milling process at the pilot plant (A) and the factory (B). [a] Samples taken for analysis.

distribution of cobalt in the different milled wheat fractions, the samples that were analyzed included the different milled fractions obtained as shown in Figure 1A and a mixture of the three flours (the flour type supplied to bakeries).

Samples obtained directly from the factory were used to study the effect of the manufacturing process on the cobalt content. Wheat grains from various areas corresponding to several wheat varieties were selected, taking into account quality parameters based on end-use, and milled using the "straight-run milling" technique; the flour was thus the result of the different extractions obtained by continuous milling at the factory. Stainless steel mills (with plain and striated rollers) were used for milling. As can be seen in Figure 1B, four different fractions were obtained, and subsamples of 1–5 kg of flour, shorts, and bran were taken as laboratory samples.

Sample Preparation. Each type of sample, both at the pilot plant and at the factory, was screened through a 132 μm sieve. Flour and byproduct samples (50–100 g) were dried to constant mass in an oven at 130 °C. An accurately weighed amount of 0.3–0.5 g was digested with 4 mL of 65% HNO_3 and a few drops of 30% H_2O_2 in a glass beaker by heating the mixture to near dryness on a hot plate at about 200 °C for ~10 min. The cool residue was diluted with 0.1 mol/L HNO_3 and transferred quantitatively into a 25 mL calibrated flask to which 1 g of hydroxylammonium chloride and 0.5 mL of 1% neocuproine (to avoid the interference of iron and copper, respectively) had been added before the volume had been made up with the same nitric acid solution. A reagent blank was prepared in parallel. The diluted sample (25 mL) was analyzed immediately after preparation by inserting it into the manifold of Figure 2.

(A) Sorption

(B) Elution
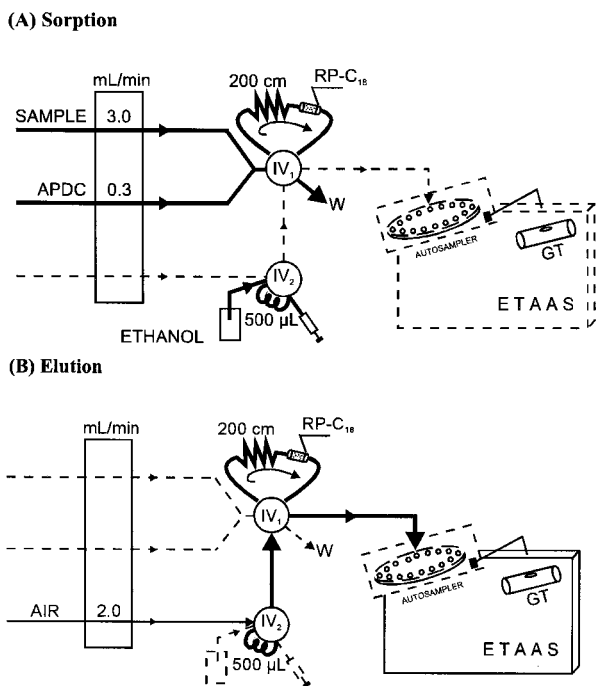
Figure 2. FI manifold for the on-line preconcentration of cobalt and its off-line determination by ETAAS. IV, injection valve; W, waste; GT, graphite tube.

Procedure. The complete preconcentration cycle for the Co-PDC chelate on the RP-C_{18} material, which consists of two steps, and the flow manifold are shown in Figure 2. In the sorption step, the 0.1% APDC solution (0.3 mL/min) was mixed on-line with the standard or treated sample (25 mL), containing 0.04–1 ng/mL cobalt in 0.1 mol/L HNO_3, and circulated at a rate of 3.0 mL/min. The cobalt chelate was adsorbed on the column and the sample matrix sent to waste. The residual aqueous solution inside the column and the FI connectors were flushed by passing an air stream through the carrier line of the second valve (IV_2) at a rate of 2.0 mL/min for 2 min; simultaneously, the loop of IV_2 was filled with 500 μL of eluent (ethanol) by aspirating the solvent with a syringe fitted to the end of the loop. In the elution step, both valves were switched and the loop contents were carried by the air stream and passed through the sorbent column. The extract was collected in the capped PTFE cup (500 μL capacity) of the instrument's autosampler. A volume of 20 μL of ethanol was used as a blank (0.003 A s).

RESULTS AND DISCUSSION

Graphite Furnace Conditions. Organometallic compounds often respond differently from inorganic salts to ETAAS (Sturgeon et al., 1980); this calls for the use of organometallic standards and the same solvent in ETAAS determinations. Also, the volatility and thermal stability of analyte–PDC complexes in ethanol solutions are important because many organometallic compounds may sublime undecomposed and give rise to analyte losses during pyrolysis (Sperling et al., 1991). In this work, the effect of the drying and pyrolysis temperatures on the integrated absorbance of the ethanol Co-PDC extracts was examined by studying the Co-PDC chelate simultaneously in ethanol and aqueous standards of cobalt in 0.2% HNO_3. Pyrolytic graphite-coated graphite tubes with platforms were used for this purpose as they provide improved sensitivity and decrease the absorbance difference between aqueous and organic solvents (Tserovsky and Arpadjan, 1991). Aqueous and organic standards of 25 ng/mL cobalt were

employed in all instances. Two dry steps, at 80 and 110 °C, for 20 μL of organic solution are recommended. A pyrolysis temperature of 1500 °C held for 20 s ensured that the APDC reagent was pyrolyzed and fully removed, so it was adopted for subsequent experiments as the thermal stability of cobalt in the chelate–ethanol solution was high enough to allow the analyte element to be isolated from more volatile concomitants present in the matrix sample, thereby increasing specificity and reducing background signals. At a pyrolysis temperature of 1500 °C, the highest integrated absorbance for cobalt was obtained at 2600 °C with both aqueous and ethanol standards. The use of magnesium nitrate as the chemical modifier (15 μL volume of standard and 5 μL of a 10 g/L chemical modifier) provided no advantages with aqueous and organic standards. At a pyrolysis temperature of 1500 °C and an atomization temperature of 2600 °C, the integrated absorbance was ca. 5% higher with Co-PDC in ethanol than with identical amounts of cobalt in 0.2% HNO_3.

Background absorption was found not to depend on the nature of the cobalt solution medium; thus, the signals were similar for aqueous and ethanol standards, and ranged from 0.006 to 0.014 A s. Results were similar for a blank of ethanol and a blank of 0.1% APDC in ethanol (ca. 0.003 A s), so 20 μL of ethanol was selected as a blank.

Solid Phase Extraction System. To investigate the potential of silica RP-C_{18} as a sorbent for cobalt determination, the above-mentioned system (González et al., 1999) was selected because of its simplicity. The effect of sample pH was examined by circulating 25 mL of a solution containing 0.2 ng/mL cobalt into the system depicted in Figure 2 for ca. 8 min. The optimal pH range was shorter, 1–1.5, so samples were prepared in 0.1 mol/L HNO_3. An APDC level of 0.1% was chosen to ensure an excess of chelating reagent (to avoid the interference of metals in real samples). Various organic solvents were tested for elution of adsorbed Co-PDC chelate from the column; the best results (difference between sample and blank) were provided by ethanol. Complete desorption of the chelate was accomplished above 400 μL; however, because the capacity of the autosampler cup was 500 μL, this was also the eluent volume that was selected. To minimize losses and contamination, the cup was fitted with a pierced anti-evaporation stopper to allow insertion of the flow line. Flow variables such as flow rates and the length of the preconcentration coil (200 cm, located before the sorbent column) were all set at the optimum valves found elsewhere (González et al., 1999).

Sensitivity and Selectivity of the Method. Several calibration graphs were run by using the preconcentration flow system depicted in Figure 2. The sensitivity (average slope of the calibration graphs) was 0.38 ± 0.03 A s × ng/mL, and the linear range for cobalt was 0.04–1 ng/mL (for a sample volume of 25 mL). The detection limit, calculated as 3 times the standard deviation of the peak area for 15 injections of 20 μL of ethanol, was 20 ng/L. The precision, as the relative standard deviation, was checked on 11 standard solutions containing 0.2 ng/mL cobalt and was found to be 6%. To validate the proposed method, an SRM was used. When the available materials were taken into account, the most appropriate reference material for the intended purpose was SRM wholemeal flour no. 189 (from the European Commission), where the concentration of Co,

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however, is uncertified. Several samples were prepared, and the average content as determined in five individual determinations ($n = 5$) was 8 ± 1 ng/g. The wholemeal flour spiked from 5 to 20 ng of cobalt per gram of sample provided recoveries of $\sim$100%. In addition, the results obtained in this SRM by the proposed method and by the conventional dry ashing were quite consistent. Therefore, the proposed solid phase extraction method is fairly accurate and can be used to determine cobalt in similar samples.

Interference by various ions present in wheat that form complexes with APDC was examined by using standards containing 0.2 ng/mL cobalt at pH 1. The maximum concentration of potential interferents tested was 10 000 times that of cobalt. The study revealed that Al^{3+}, Mn^{2+}, and Sn^{2+} at concentrations up to 2 μg/mL, Hg^{2+} up to 1.6 μg/mL, Zn^{2+} up to 0.8 μg/mL, Bi^{3+} and Ni^{2+} up to 80 ng/mL, Cu^{2+} up to 60 ng/mL, Cd^{2+} and Fe^{3+} up to 20 ng/mL, and Pb^{2+} up to 16 ng/mL did not interfere with the determination of cobalt. Unless the wheat samples are highly contaminated, only Cu^{2+} and Fe^{3+} are normally encountered at concentrations of ca. 6 and 20 μg/g, respectively, in wheat samples (Belitz and Grosch, 1992); such contents interfere with the proposed method. Masking agents are thus required to overcome the interference of large amounts of Cu^{2+} and Fe^{3+} in analyzing wheat flour. The interference of Cu^{2+} was avoided by using 0.02% w/v neocuproine (in the presence of a reductant such as hydroxylammonium chloride); in this way, copper exhibited no disturbance of the determination of cobalt contents at least up to 1000 times higher (copper normally occurs at concentrations that are ca. 600 times those of cobalt in flour). Since Fe^{2+} has a lower affinity for APDC than Fe^{3+}, using 4% w/v hydroxylammonium chloride increased the tolerated level of this ion to 0.8 μg/mL to a 4000:1 Fe:Co ratio.

Influence of the Texture, Geographical Origin, and Milled Fraction of the Wheat on the Cobalt Content. Wheat grains can be classified as soft and hard according to their capacity to absorb water. The effect of the wheat texture was examined using various wheat samples, including three of the soft and five of the hard type (one of them, called durum, was an extra-hard type); samples were subjected to three consecutive milling operations (first, second, and third extraction) at a labotatory mill as shown in Figure 1A. Amounts of ca. 0.5 kg of whole wheat were subjected to successive milling steps that provided flour proportions with respect to whole wheat of 30, 15, and 15% w/w for soft and 20, 15, and 15% w/w for hard wheat in the first, second, and third extractions, respectively. The remaining material, 40 and 50% w/w, consisted of byproducts (bran plus shorts) from soft and hard wheat, respectively. The influence of texture and geographical origin on the cobalt content in the flours was examined by analyzing flour samples obtained by milling eight soft and hard types of wheat from various geographical areas (Spain, Denmark, England, Sweden, Canada, and Hungary). The samples were mixtures of the three extraction flours obtained in the three consecutive millings shown in Figure 1A. For analysis, samples were treated as described in Materials and Methods and processed using the flow system depicted in Figure 2. Table 1 gives the average concentration for each sample as obtained from three replicates and three injections per replicate ($n = 3$). In all instances, the highest cobalt

Table 1. Cobalt Content[a] (ng/g) in Flour Made from Wheat of Variable Texture and Geographical Origin Using Straight-Run Milling Flour

wheat texture	geographical origin	Co found
soft	Spain	5.6 (0.4)
soft	Denmark	6.4 (0.4)
soft	England	7.5 (0.5)
hard	Sweden	13.0 (0.9)
hard	Canada	13.6 (1.0)
hard	Hungary	10.6 (0.7)
hard	Spain	9.0 (0.6)
durum	Spain	10.4 (0.7)

[a] Standard deviations ($n = 3$) are given in parentheses.

contents were obtained for hard wheats (average of 11.3 ± 1.9 ng of cobalt/g) and the lowest for soft wheats (average of 6.5 ± 0.9 ng of cobalt/g); variability was also higher in hard wheats (17% RSD). We can thus conclude that the cobalt content is influenced by the texture of the wheat. Such a content also depends on the type of agricultural soil (geographical origin) used to grow the wheat since the average values obtained in flours of different origin (9.5 ± 2.9 ng of cobalt/g) exhibit a high variability (30% RSD).

Flour extraction may be defined as the proportion of the wheat recovered as flour by the process of milling. The chemical composition of the flour depends on the extraction rate because the proportion of starch decreases and that of the constituents from the grain's cover increases with an increasing extraction rate. The effect of the different wheat fractions extracted after three consecutive millings on the cobalt concentration was studied at a laboratory mill like that depicted in Figure 1A. For this purpose, eight varieties of wheat grains were subjected to milling and sieving, and the five fractions obtained from each (first-, second-, and third-extraction flour, shorts, and bran) were individually analyzed as described above. Table 2 lists the average fraction concentrations for the samples ($n = 3$), from which several conclusions can be drawn. In all cases, the cobalt content increases as the number of extractions increases. Thus, the content in byproducts (shorts and bran) is higher than that in the flours; this is the result of the mineral content increasing from the inner to the outer cells of the endosperm. Because the proportion of constituents from the cover increases with an increasing extraction rate (the proportion of wheat recovered as flour by the milling process), the variation of cobalt content (nanograms per gram) with the extraction rate (percentage) for the three extraction flours of each type of wheat studied is a stright line ($r = 0.912 - 0.998$) which confirms that the cobalt content increases with an increasing extraction rate. Figure 3 shows the variation of the cobalt content (nanogram per gram) in each type of wheat (the Spanish durum type excluded) with the intercepts of the stright lines obtained previously by the representation of cobalt content versus extraction rate. As can be seen, the variation is nonlinear; therefore, a graphical method was used to study data (Haswell, 1992). The data were displayed as points in a two-measurement space where the coordinate axes were defined by the measured variables, i.e., Co content and the intercepts of the stright lines obtained previously by plotting cobalt content (nanograms per gram) versus extraction rate (percentage). By examining the graph, searching for similarities and dissimilarities among the samples, we found that flour contents form natural clusters that span the intercept range from 2.6

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Table 2. Cobalt Content[a] (ng/g) in Milling Products Obtained at a Pilot Plant

	flour			byproducts	
geographical origin	first extraction[b]	second extraction[b]	third extraction[b]	shorts[b]	bran[b]
Spain	5.5 (0.4)	6.5 (0.4)	7.3 (0.6)	8.6 (0.7)	12.1 (0.9)
Spain	5.7 (0.4)	19.5 (1.2)	17.1 (1.3)	22.4 (1.6)	28.3 (2.0)
Denmark	5.5 (0.3)	7.8 (0.6)	8.6 (0.7)	9.9 (0.8)	10.5 (0.9)
Hungary	7.7 (0.5)	9.7 (0.7)	12.0 (1.0)	20.2 (1.4)	24.9 (1.8)
England	8.8 (0.6)	12.6 (0.9)	14.3 (1.0)	21.0 (1.3)	29.2 (2.3)
Sweden	6.9 (0.5)	9.8 (0.7)	10.8 (0.9)	17.1 (1.2)	21.9 (1.8)
Canada	9.6 (0.7)	12.7 (0.9)	18.3 (1.3)	20.6 (1.4)	24.9 (1.9)

[a] Standard deviations ($n = 3$) are given in parentheses. [b] Fractions extracted as per the flowchart of Figure 1A.
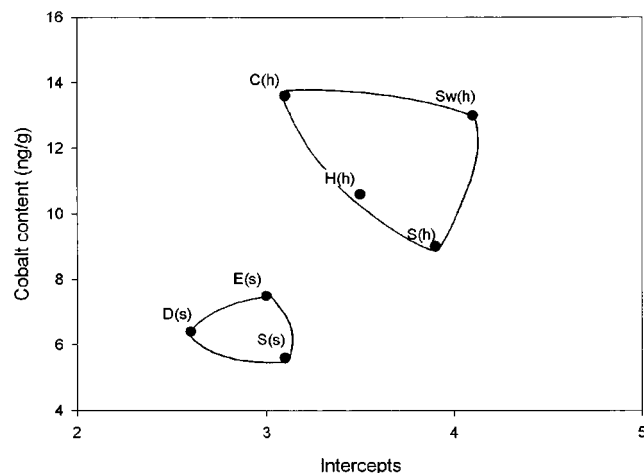
Figure 3. Cluster analysis of the three flours extracted from wheat from various countries: Canadian hard (C), Swedish hard (Sw), Hungarian hard (H), Spanish hard (S), English soft (E), Denish soft (D), and Spanish soft (S) wheat. For details, see Table 2.

to 3.1 and from 3 to 4.1 and the cobalt content ranges from 5.6 to 7.5 ng of cobalt/g and from 9 to 14 ng of cobalt/g, respectively. The clusters consist of flours from the soft and hard wheat types with the lowest and highest cobalt contents, respectively. This type of plot may thus be useful with a view of classifying wheat according to texture.

Influence of the Manufacturing Process on the Cobalt Content at a Flour-Producing Factory. The mineral (ash) content of flour is considered to be a measure of its quality, although it is not as such related to its final performance; although some wheats naturally possess a higher endosperm ash content because of the prevailing soil conditions, genetic factors, and others, it provides some indication of the miller's skills and the degree of refinement in the milling process. To study the potential increase or decrease in the cobalt content in flour obtained by milling the wheat grains at a flour-producing factory, nine wheat samples (and their respective flours, shorts, and brans) obtained at an Andalusian factory during daily routine production were analyzed. Figure 1B (the flowchart of the milling process of the flour-producing factory) shows the three milled wheat fractions (straight-run flour, shorts, and bran) that were obtained. Overall, 27 milled wheat fractions were analyzed with the proposed method. Thus, samples were dissolved as described in Materials and Methods, and cobalt in the resulting solutions was preconcentrated using the flow system depicted in Figure 2. The results obtained for flour, shorts, and bran were 9.2−11.5, 15.2−22.9, and 19.8−27.2 ng/g, respectively. A comparison with the results of Table 1 (the

total flour fraction) and Table 2 (shorts and bran), obtained at the laboratory mill, with those obtained at the factory revealed the following facts. The average cobalt content was the same in the flours (9.5 ± 2.9 and 10.0 ± 0.9 ng/g) and similar in the shorts (16.6 ± 5.4 and 19.0 ± 2.4 ng/g) and the brans (21.0 ± 7.2 and 23.8 ± 2.3 ng/g), for samples from the laboratory mill and the factory, respectively. The results obtained for the different types of flour, shorts, and bran were ca. 10% higher in the samples milled at the flour-producing factory than in those obtained at the laboratory mill; these slightly increased cobalt levels in the wheat products may have resulted from differences in the characteristics of the wheat used in the two types of milling as the cobalt content in wheat (and hence that in its fractions) depends not only on its texture and origin but also on other factors that are considered in the selection process at the flour-producing factory. On the other hand, the high variability among samples from the laboratory mill (ca. 30%) relative to those from the factory (ca. 10%) can be explained as described above, taking into account the fact that the wheat used to obtain flour at the factory is possibly more uniform with regard to physicochemical characteristics related to metal composition, whereas the test samples used in the laboratory mill milling process spanned a wide range of wheat types of different origin and texture and with differential physicochemical properties.

LITERATURE CITED

Balaji, T.; Chiranjeevi, P.; Naidu, G. R. K. Simultaneous determination of trace amounts of chromium, cobalt and lead in waste water and plant materials by extraction-atomic absorption spectrometry. *Anal. Lett.* **1998**, *110*, 1081−1094.

Belitz, H. D.; Grosch, W. *Food Chemistry*; Springer-Verlag: Berlin and Heidelberg, 1992; Chapter 7.

Blanchflower, W. J.; Cannavan, A.; Kennedy, D. G. Determination of trace amounts of cobalt in feed grains and forages by solvent extraction and graphite furnace atomic absorption spectrometry. *Analyst* **1990**, *115*, 1323−1325.

Borggaard, O. K.; Christensen, H. E. M.; Lund, S. P. Determination of cobalt in feedingstuffs by solvent extraction and graphite furnace atomic-absorption spectrometry. *Analyst* **1984**, *109*, 1179−1182.

Ebdon, L.; Fisher, A. S.; Parry, H. G. M.; Brown, A. A. Direct atomic spectrometric analysis by slurry atomization. Part 10. Use of an air-ashing stage in electrothermal atomic absorption spectrometry. *J. Anal. At. Spectrom.* **1990**, *5*, 321−324.

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J. Agric. Food Chem., Vol. 48, No. 10, 2000 **4519**

Gallego, M.; Petit de Peña, Y.; Valcárcel, M. Fullerenes as sorbent materials for metal preconcentration. *Anal. Chem.* **1994**, *66*, 4074−4078.

Gervais, L. S.; Salin, E. D. Heated sample introduction system for the analysis of slurries by inductively coupled plasma atomic-emission spectrometry. *J. Anal. At. Spectrom.* **1991**, *6*, 41−47.

González, M. M.; Gallego, M.; Valcárcel, M. Effectiveness of fullerene as a sorbent for the determination of trace amounts of cobalt in wheat flour by electrothermal atomic absorption spectrometry. *J. Anal. At. Spectrom.* **1999**, *14*, 711−716.

Haswell, S. J., Ed. *Practical Guide to Chemometrics*; Marcel Dekker: New York, 1992; Chapter 7.

Jorhem, L.; Sundstrom, B. Levels of lead, cadmium, zinc, copper, nickel, chromium, manganese and cobalt in foods on the Swedish market, 1983−1990. *J. Food Compos. Anal.* **1993**, *6*, 223−241.

Mahoney, A. W. Mineral contents of selected cereals and baked products. *Cereal Foods World.* **1982**, *27*, 147−150.

Mochizuki, T.; Sakashita, A.; Iwata, H.; Ishibashi, Y.; Gunji, N. Application of slurry nebulization to trace elementary analysis of some biological samples by inductively coupled plasma mass spectrometry. *Fresenius J. Anal. Chem.* **1991**, *339*, 889−894.

Sanz-Medel, A. *Flow Analysis with Atomic Spectrometric Detectors*; Elsevier: Amsterdam, 1999.

Sperling, M.; Yin, X.; Welz, B. Flow injection on-line separation and preconcentration for electrothermal atomic absorption spectrometry. Part 2. Determination of ultra-trace amounts of cobalt in water. *J. Anal. At. Spectrom.* **1991**, *6*, 615−621.

Stringari, G.; Pancheri, I.; Möller, F.; Failla, O. Influence of two grinding methods on the uncertainty of determinations of heavy metals in atomic absorption spectrometry/electrothermal atomisation of plant samples. *Accredit. Qual. Assur.* **1998**, *3*, 122−126.

Sturgeon, R. E.; Berman, S. S.; Desaulniers, A.; Russell, D. S. Pre-concentration of trace metals from sea-water for deter-mination by graphite-furnace atomic-absorption spectrom-etry. *Talanta* **1980**, *27*, 85−94.

Taylor, A.; Branch, S.; Halls, D. J.; Owen, L. M. W.; White, M. Atomic spectrometry update. Clinical and biological materials, foods and beverages. *J. Anal. At. Spectrom.* **1999**, *14*, 737−772.

Tserovsky, E.; Arpadjan, S. Behaviour of various organic solvents and analytes in electrothermal atomic absorption spectrometry. *J. Anal. At. Spectrom.* **1991**, *6*, 487−491.

Underwood, E. J. *Trace Elements in Human and Animal Nutrition*, 4th ed.; Academic Press: New York, 1977.

Wang, G. Study on the catalytic-kinetic-photometric determi-nation of trace cobalt. *Lihua Jianyan Huaxue Fence* **1991**, *27*, 375−379.

World Health Organization. *Trace Elements in Human Nutri-tion and Health*; World Health Organization: Geneva, 1996; Chapter 1.

Wu, Z.; Wu, Z. Direct determination of trace cobalt in grains and forages by graphite-furnace atomic-absorption spec-trometry. *Fenxi Huaxue* **1988**, *16*, 710−712.

Yan, X. P.; Van Mol, W.; Adams, F. Flow injection on-line sorption preconcentration in a knotted reactor for electro-thermal atomic absorption spectrometric determination of ultratrace amounts of cobalt in natural waters. *Lab. Rob. Autom.* **1997**, *9*, 191−199.

Zhang, Y.; Pan, J. H. Determination of cadmium, cobalt and nickel in grain by graphite-furnace atomic-absorption spec-trometry. *Guangpuxue Yu Guangpu Fenxi* **1997**, *17*, 94−96.