

Ion Chromatographic Determination of Chloride in Mustard Sauces

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A new, simple, precise, and rapid ion chromatography (IC) method has been developed to determine chloride in mustard sauces using a mixture of phthalic acid, acetone, and water adjusted to pH 5.0 as eluent. Conductometric detection was carried out. The retention time for chloride was 1.5 min. Linearity was obtained up to a concentration level of 100 mg/L NaCl. The method was statistically evaluated for accuracy and precision after being used to assay the chloride from mustard sauces. Within the same samples, the chloride levels obtained by IC were compared with the sodium concentrations quantified by atomic absorption spectrophotometry.

Keywords: *Ion chromatography; atomic absorption spectrophotometry; chloride; sodium; mustard sauces*

INTRODUCTION

The use of mustard sauces is becoming more and more popular among the Spanish people. Although Spanish legislation has established the limits of several of their components, at the moment there are no official methods to determine them.

Regarding the use of chloride, this anion can be used up to a maximum level of 5% w/w expressed as NaCl. This anion should be monitored to ensure that the legislation and regulatory limits in a wide variety of mustard sauces are observed.

Thiocyanide ion seriously interferes with the measurement of chloride when the volumetric method recommended by the AOAC (1990) is used. Consequently, it was important to find a method simple and rapid enough to be used in food quality control laboratories.

In recent years, an enormous number of papers about ion chromatographic (IC) inorganic anion determinations in a large variety of samples having very different natures have been published (Salas-Auvert et al., 1995; Achilli et al., 1995; Gros and Gorenc, 1997; Umile and Huber, 1993; Nesterenko et al., 1994; Ivask and Pentchuk, 1997). Among these papers, we emphasize those aimed at determining the anions and/or cations in food samples (Bosch Bosch et al., 1995; Miyahara and Sito, 1994; Ruiz et al., 1994; Trifiro et al., 1996; Swiader and Freiji, 1996; Ding et al., 1995; Ruiz et al., 1995; Jun et al., 1996; Buckee, 1995), as well as those aimed at determining the chlorides (Blackwell et al., 1997; Buldini et al., 1994). IC seems to be the most useful analytic tool for a small anion simultaneous determination.

This present paper proposes an IC method with conductivity detection combined with a surface ami-

nated resin-based anion exchange column and electronic suppression. We propose this method because of its rapidity and simplicity, because no interferences have been observed between chloride and the rest of the anions, and because no sample pretreatment is required.

The aim of the present study is to propose a suitable analytical method for chloride determination in mustard sauces. A statistical study on the linearity, accuracy, and precision of the method and the results of the chloride levels were compared with sodium levels obtained in the same samples by atomic absorption spectrophotometry (AAS), which was used as a reference method with regard to their chloride and sodium contents.

MATERIALS AND METHODS

Chemicals. All chemicals were of analytical reagent or HPLC grade (Merck, Darmstadt, Germany). Working standard solutions were prepared by appropriate dilutions of stock standard solutions. The pH was adjusted with aqueous sodium hydroxide. The eluents, standards, and all solutions were prepared using ultrapure 18 M Ω -cm water obtained by passing doubly distilled water through a Milli-Q system (Millipore). Phthalic acid was dissolved in the eluent reservoirs to 2 mM in a mixture on acetone/water (1:9), and the pH was adjusted to 5.0. Both eluents and standard solutions were thoroughly degassed with nitrogen (SEO N-48, 99.98% N₂).

Apparatus. The IC equipment components were the following: a Kontrom Model 320 high-pressure pump system, a Rheodyne injector with a 20 μ L loop, an IC anion PRP-X100 column (5 μ m particle size, 125 mm \times 4 mm i.d.) (Hamilton), and an IC Metrohm Model 690 conductometric detector. All tubing in the chromatography path was PEEK [0.005 in. (0.125 mm) i.d.]. Data collection was performed with a personal computer and the Kontron PC Integration Pack software.

The conditions for analysis were the following: eluent, 2 mM phthalic acid/10% acetone (pH 5.0); flow rate, 1.0 mL/min; injection volume, 20 μ L. All measurements were performed at room temperature. Under these conditions, the net retention time was 1.48 \pm 0.05 min.

Sodium atomic absorption measurements were carried out with a Perkin-Elmer Model 2280 atomic absorption spectrophotometer equipped with an Na-hollow cathode lamp as primary source. An air/acetylene flame was applied, with the

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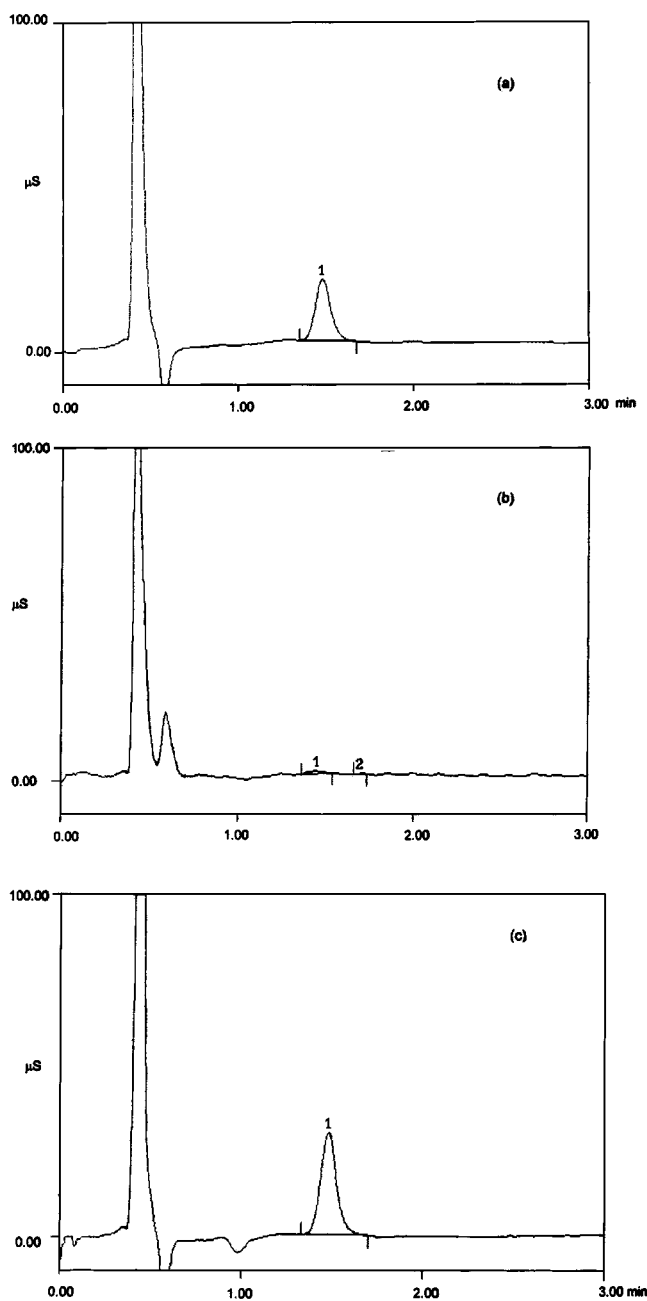


Figure 1. Chromatogram of (a) standard solution of chloride in mobile phase (5 µg/mL NaCl), (b) a mustard solid (1:100 in mobile phase), and (c) a fine sauce of mustard (1:100 in mobile phase). Chromatographic conditions: IC anion PRP-X100 column (5 µm particle size 125 mm × 4 mm i.d.); mobile phase, 2 mM phthalic acid/10% acetone (pH 5.0); flow rate, 1.0 mL/min; injection volume, 20 µL; conductometric detection.

automatic gas control unit set at 28 L/min air and 10 L/min acetylene. A solution uptake rate of 5 mL/min and three replicates of 1 s of aspiration time were used for all samples.

Sample Preparation. *IC Determination.* Mustard seeds were milled in a grinder and sieved before dissolution in the eluent. Sauce samples were manually homogenized until becoming completely homogeneous. Then a known amount of an accurately weighed sample (≈1 g) was added to ~40 mL of the eluent and stirred for 2 h. The solution was transferred into a 50 mL volumetric flask and diluted to the mark with eluent. A 1:100 dilution of samples with eluent was performed to obtain an anion concentration falling within the linear range of the calibration curves. These final solutions were filtered with 0.45 µm filters and then injected for the analysis. The same method was used for all samples. Figure 1 shows the

chromatograms of a standard solution, a solid mustard, and a fine mustard sauce. The second one shows a very small peak corresponding to the chloride and another unidentified one near the unretained solute peak. In the chromatogram obtained with the fine sauce we can observe the chloride peak and the negative peak corresponding to the acetate that is found in these samples as a consequence of the vinegar added during the sauce elaboration.

AAS Determination. Mineral content was determined by ignition of an accurate amount of sample (0.5 g for solid mustards and 1 g for liquid mustards) in a crucible, which was cleaned with nitric acid and rinsed several times and then introduced into a muffle at 400 °C for 4 h before use. Previous ignition was performed in a Bunsen gas burner, and then ignition was finished in a muffle at 450 °C for 16 h. The ashes were completely soluble in 2 mL of 50% v/v HNO₃ and 2 mL of 50% v/v HCl, and a clear solution was obtained. Then the solution was filtered and placed in a 50 mL volumetric flask and diluted to the mark with water. These final solutions were analyzed in the spectrophotometer. The analytical wavelength was 330.2 nm with a slit corresponding to a 0.7 nm spectral bandwidth instead of 589.0 nm because of a high concentration of analyte in these samples that exceeds the linearity range.

RESULTS

Chromatographic Determination of Chloride.

Sensitivity. Because the concentrations of chloride in mustard sauces are generally significantly higher, the sensitivity of the method is not expected to restrict its application.

Linearity. A series of standard solutions with increasing values of chloride were used for the calibration graph for chloride determination. The plot of the peak area versus the chloride concentration was found to be linear up to 100 mg/L of NaCl, with the following linear regression equation:

$$y = 0.529 + 0.727x \quad (r^2 = 0.9999)$$

Precision. The analytic precision of real samples was assessed by choosing a mustard at random from a series of 10 successive injections of such a sample. The mean chloride value of these samples was 4.86%, with a minimum value of 4.63% and a maximum one of 5.09%. The relative standard deviation was 3.29%.

Recovery. Three mustard sauces were chosen at random to carry out recovery experiments; 1 was selected from the 6 granulated sauces analyzed and 2 were chosen from the 34 fine ones. The analytical recovery was determined by adding a known amount of chloride to a previously analyzed mustard sample and comparing the concentration measured with and without the amount added. The spike was added to the mustard sauce before any treatment was done to see how reproducible the whole procedure is from extraction to analysis. The chloride concentrations of the samples were between 3.99 and 4.86 g of NaCl/100 g; the chloride added, exactly measured, was ~5.00 g of NaCl/100 g. In these conditions, recovery ranged from 92.45 to 102.50%.

Interferences. To verify if the proposed method did not show interferences, different anion-containing solutions were analyzed and fluoride, bromide, acetate, nitrite, nitrate and sulfate were assayed. The elution order was acetate = F⁻ < Cl⁻ < NO₂⁻ < Br⁻ < NO₃⁻ < SO₄⁻². None of them interfered with chloride.

Chromatographic Analysis of Mustards. *Chloride Content.* The mustards analyzed were divided into three types according to their nature: solid mustards, mus-

Table 1. Comparison between IC Results for Chloride and AAS Results of Sodium in Mustard Samples

	solid mustards		granulated sauces		fine sauces	
	chloride ^a	sodium ^a	chloride ^a	sodium ^a	chloride ^a	sodium ^a
min	0.02	<0.04	3.99	3.60	1.39	0.72
max	0.35	0.86	7.68	5.35	8.51	5.83
mean	0.12	0.49	6.26	4.88	5.23	3.66
SD	0.11	0.31	1.27	0.66	1.74	1.43
RSD (%)	88.0	61.8	20.2	13.5	33.2	39.0

^a Grams of NaCl per 100 g of sample.

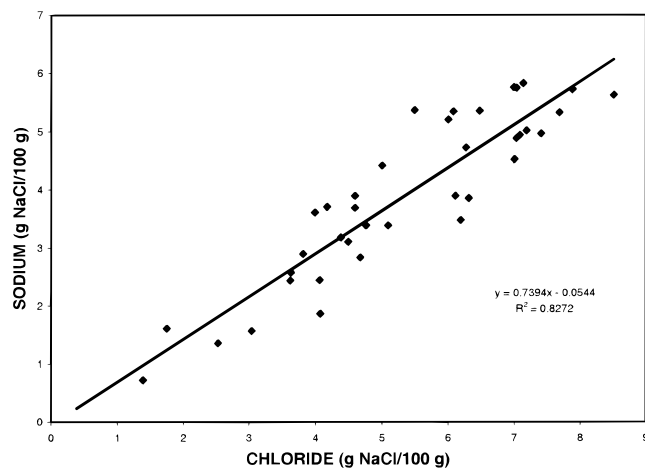


Figure 2. Correlation graph between the chloride concentration of mustard (grams of NaCl/100 g) obtained by IC and sodium concentration of mustard (milligrams/100 g) obtained by AAS.

tard seeds, and mustard sauces, which in turn were divided into granulated and fine sauces, according to their aspect. Figure 1 shows the three chromatograms corresponding to a standard solution of chloride (5 mg/L NaCl), a sample of solid mustard, and a sample of fine mustard sauce. In contrast to the mustard sauces, it can be seen how the chloride content in the solid mustard is close to the detection limit of the method. The results obtained in these analyses are shown in Table 1.

Sodium Content. Although the accuracy and precision of the proposed method have been verified, it would be interesting to have a reference method to compare the results obtained. That is why we proposed studying the sodium content, because the origin of most of the sodium and chlorides would be the same—the salt added in the sauces elaboration. The sodium concentration was determined by AAS, and the results obtained are shown in Table 1.

Correlation between the Chloride and Sodium Levels. Finally, the relationship between the sodium and chloride contents of the samples analyzed was studied (Figure 2). As could be expected, a reasonable correlation between them is observed, with a correlation coefficient of 0.91.

DISCUSSION AND CONCLUSIONS

The mustard seeds present a very low level of chlorides as is indicated by the contents obtained in the six samples analyzed. The mean content of these samples was 0.12 g of NaCl/100 g of sample (Table 1). The relative standard deviation (RSD) was very high due to the low levels of chloride found, ~0.1 mg/L in the solution injected in the chromatograph, and this concentration is difficult to quantify accurately with the proposed method.

As Table 1 shows, the chloride contents of the granulated mustard sauces were variable, ranging from 3.99 to 7.68 g of NaCl/100 g. The heterogeneity of the results obtained in the fine mustard sauces gives us an idea about the different proportions of common salt incorporated in its elaboration. The chloride levels varied from 1.39 to 8.51 g of NaCl/100 g. Regarding the maximum value permitted by law (5 g/100 g), only 1 sample belonging to the granulated sauce group and 18 of the 40 fine sauce samples fulfill the Spanish Guideline regarding the sodium chloride content. The remaining sauces studied exceeded this value.

A simple, rapid, and low-cost method such as that proposed in this paper could help to carry out the routine measurements of chlorides in mustard sauces, with the consequent importance that this would have to be able to comply with the present legislation.

The mean sodium content of the granulated sauces is slightly greater than that of the fine sauces: 4.88 g of NaCl/100 g compared to 3.66 g of NaCl/100 g (Table 1). The values provided by Elmadfa et al. (1989), by Jimenez et al. (1994), and by Mataix et al. (1995) in processed mustards were, respectively, 1307, 1252, and 1232 mg/100 g, which are similar to those found in this study with the fine mustard sauces, 1441.7 mg/100 g. The label of one of the samples, corresponding to fine sauces, indicated, as nutritional information, a sodium content of 1.1 g/100 g. In the analysis of this sample we obtained a sodium level of 0.96 g/100 g. The sodium levels maintain a proportional relationship to those of the chlorides, as the reasonable coefficient of the correlation obtained between these two concentrations shows.

This IC method is proposed for the determination of chloride in all kinds of mustard sauces because of its accuracy, precision, rapidity, and simplicity. Hence, it can be easily and conveniently adopted for routine quality control analyses.

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