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Effects of Manchego-type cheese-making process on contents of mineral elements

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Variations in mineral content were determined throughout the process of making cheese, taking samples of natural, pasteurised milk, with additions of rennet, curd, whey, pressed curd, pressing whey and cheese. The mean contents in cheese were 7·59 mg/g for calcium, 0·309 mg/g for magnesium, 6·46 mg/g for sodium, 1·38 mg/g for potassium and 0·364 μg/g for manganese on a fresh weight basis. The contribution of the consumption of this type of product to the daily intake estimates in a Spanish diet are 23·6 mg/day for calcium, 1·0 mg/day for magnesium, 20·1 mg/day for sodium, 4·3 mg/day for potassium and 1·1 μg/day for manganese. By a variance analysis the existence of statistically significant differences ($P<0{\cdot}001$) was confirmed in the products from the cheese-making for the five minerals, expressed both by fresh weight and dry weight. Certain differences were observed in the groups formed by using a Scheffe homogeneity test ($P<0{\cdot}05$) depending on whether the mineral content was expressed on a fresh weight or a dry weight basis. Slight rises in the contents of the five minerals investigated on a dry weight basis were verified mainly due to the retention of the minerals by the curd and, secondly, by possible contamination occurring during the process

INTRODUCTION

The mineral content of cheese is highly variable and depends on numerous factors such as the differences in mineral content among the species producing the milk with which it is made (Franco *et al.*, 1981; Garcia Olmedo *et al.*, 1981*a*,*b*), the geographical area in which it is made (Coppini *et al.*, 1979), and also the characteristics of the making process (Feeley *et al.*, 1972).

During cheese manufacture physicochemical changes may take place; these changes may be designed to produce a particular taste, aroma or texture, but the changes may also affect the nutritional composition of the product (Scott, 1989).

Some cheeses are prepared by acid precipitation (starter only); other cheeses are prepared using rennet as well as starter. Factors such as pH, temperature and salt balance are also important in determining the characteristics of the curd and whey. In relation to these factors, the mineral-soluble fraction (part of whey components) may change and may be lost during the 'wheying-off' procedure.

The effects of cheese manufacturing on mineral levels have been shown by Scott (1989) for different types of cheese and they indicate that levels of calcium are generally up to 10 times higher in hard cheese and four to five times higher in mould cheese than in milk. Magnesium is of a similar order to calcium but only five times higher in hard cheese and two to three times in mould cheese. Levels of potassium are no higher in cheese than in milk and levels of sodium are dependent on the amount of salt added.

Manchego cheese is the most popular indigenous Spanish cheese, consumed both in Spain and outside its borders. Its average yearly production is estimated to be over 50 000 tonne/year (Fox, 1987). This product is made following a traditional process using different proportions of ewe's, goat's and/or cow's milk and there are also many other cheeses which, although they do not receive the denomination of Manchego, are also made in the same way.

In previous works we studied the variation of copper, iron and zinc levels in Manchego-type cheese during the traditional cheese-making process (Moreno-Rojas *et al.*, 1994*a*), and copper, iron, zinc, calcium, magnesium, manganese, sodium and potassium variations during ripening of Manchego-type cheese (Moreno-Rojas *et al.*, 1992, 1994*b*).

The aim of this study is to determine the influence of the traditional process of making Manchego-type cheese on contents of calcium, magnesium, manganese, sodium and potassium. This could be determined by two main factors: the relation of each one of the minerals studied with the curdling and soluble fractions of the milk (Renner *et al.*, 1989) and the possible contamination which is produced in the process (Pertoldi Marletta & Gabrielli Favretto, 1983).

MATERIALS AND METHODS

A routine production batch of Manchego-type cheese made in a commercial cheese-making factory in Spain was studied.

A mixture of milk (22% goat's milk and 78% cow's milk) was pasteurized and subsequently placed in the curdling vat, where 3350 litres of milk with a 3·9% fat content and 15·5° Dornic acidity was processed. Addition to the milk of 70 g ferment (Bioferment Dac Homo), 280 g discolouring agent, 175 cm^3 lysozyme, 670 g $CaCl_2$ and 800 cm^3 rennet (Proinalsa) was made. After the formation of curds these were pressed into plastic moulds. The pressed curd was placed in brine with a 20% salt content, pH 5·3 and 13° Dornic in which it remained for 40 h.

During the process, 10 statistical random samples were taken of each one of the following stages of the process: natural milk, pasteurised milk, the addition of ferment, curd, whey exuded from the curd and whey from pressed curd, and also 10 random samples were taken from each one of three portions situated at different depths in the pressed curd and in the fresh cheese.

For the mineralisation of the samples, the method of Moreno-Rojas *et al.* (1994*b*) was followed. The mass of the sample used for analysis in each case depended on its nature: for liquid samples 50 g were taken and, for the solid ones, 10 g.

The crucibles containing the dried samples were incinerated in a furnace at 460°C overnight. After cooling, 2 M nitric acid (2 ml) was added, and the solutions were dried on a thermostatic hotplate. They were subsequently placed once again in the furnace where they remained at 460°C for 1 h. The recovery of the ash was carried out with 2 M nitric acid (5 ml) and 0·1 M nitric acid (20 ml), in a 25-ml volumetric flask (subsequently stored in polypropylene flasks under refrigeration). For Ca, the solution was diluted 1:100 and lanthanum chloride ($LaCl_3.7H_2O$) was added to give 0·27% in the final solution.

Analyses were performed using a Perkin-Elmer Model 2380 atomic absorption spectrophotometer, using an air–acetylene flame. Single-element hollow cathode lamps were used for all elements except for Na and K which were determined by emission using the same instrument. For each element being determined, the analyses included duplicate analyses of samples, one spiked recovery analysis, and one standard reference material (non-fat milk powder, NBS 1549) from the National Institute of Standards and Technology (NIST). For calculation of the detection limit (3SD), the definition and criteria of the IUPAC were followed (Long & Winefordner, 1983; Analytical Methods Committee, 1987).

The sensitivities (μg/ml) obtained for Ca, Mg, Na, K and Mn, were 1·38, 0·089, 6·42, 6·70 and 0·034, respectively. The minimum concentrations detectable in cheese, on a fresh weight basis (μg/g), were 93·7, 12·2, 100, 50 and 0·023 for Ca, Mg, Na, K and Mn, respectively. Mean recoveries in 'non fat milk powder (NBS 1549)' were Ca, 102%;Mg, 101%; Na, 104%; K, 96%; and Mn, 98%.

Statistical analysis

Data obtained from the chemical analysis of the samples were evaluated statistically using a variance analysis with Scheffè multiple range test (Snedecor & Cochran, 1971)

RESULTS AND DISCUSSION

Table 1 shows the mean concentration of calcium, magnesium, sodium, potassium and manganese, and Figs 1–5 their evolutions at fresh (A) and dry (B) weight through the eight products analysed.

A one-factor variance analysis was used to determine if there were significant differences between the products studied and it was found that there were differences both in the moisture content and in the content of the five elements investigated (expressed both at fresh and dry weight) ($P < 0·001$) after which a Scheffè multiple range analysis ($P < 0·05$) was carried out and the groups observed in Table 1 were ascertained.

Table 1. Contents of moisture, calcium, magnesium, sodium, potassium and manganese at fresh weight (mean ± SD) in the different products formed in the cheese-making process. Homogeneous group from Scheffè multiple range test ($P < 0·05$) in fresh and dry weight[a]

Product	*n*	Moisture (%)	Calcium (mg/g)	Magnesium (mg/g)	Sodium (mg/g)	Potassium (mg/g)	Manganese (μg/g)
Natural milk	10	89·2 ± 0·1D	1·43 ± 0·10Bc	0·118 ± 0·010Bb	0·55 ± 0·03Ac	1·71 ± 0·06Dc	0·023 ± 0·004Aa
Pasteurised milk	10	89·3 ± 0·1D	1·41 ± 0·07Bc	0·111 ± 0·002Bb	0·53 ± 0·03Ac	1·69 ± 0·05Dc	0·029 ± 0·003Aa
Milk + rennet	10	89·2 ± 0·1D	1·40 ± 0·05Bc	0·113 ± 0·005Bb	0·49 ± 0·02Ac	1·68 ± 0·09Dc	0·037 ± 0·006Aa
Curd	10	54·7 ± 0·4C	6·95 ± 0·34Ce	0·261 ± 0·017Ca	0·33 ± 0·02Ab	1·20 ± 0·05Bb	0·255 ± 0·023Bb
Curdling whey	10	94·2 ± 0·2E	0·58 ± 0·06Aa	0·086 ± 0·005Ac	0·50 ± 0·02Ad	1·81 ± 0·06Ed	0·014 ± 0·002Aa
Pressing whey	10	95·0 ± 0·1F	0·59 ± 0·05Ab	0·104 ± 0·005ABd	0·48 ± 0·01Ad	1·71 ± 0·06De	0·033 ± 0·005Ac
Pressed curd	30	47·8 ± 0·2A	7·67 ± 0·41Dd	0·322 ± 0·033Da	0·28 ± 0·01Aa	1·10 ± 0·06Aa	0·247 ± 0·044Bb
Cheese	30	49·2 ± 0·5B	7·59 ± 0·64Dde	0·309 ± 0·042Da	6·47 ± 5·23Be	1·38 ± 0·05Cb	0·364 ± 0·136Cd

[a]A–E Scheffè homogeneous groups ($P < 0·05$) at fresh weight. a–e Scheffè homogeneous groups ($P < 0·05$) at dry weight.

Each Scheffé homogeneous group is formed by a group of means that do not present any statistically significant differences from each other. Products marked with the same letter for an element or moisture had similar concentrations.

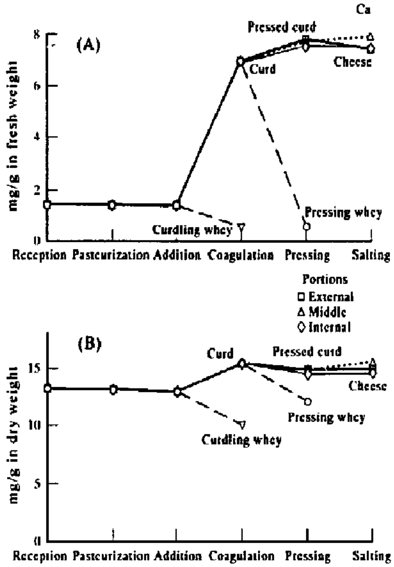

Fig. 1. Calcium evolution during the cheese-making process.

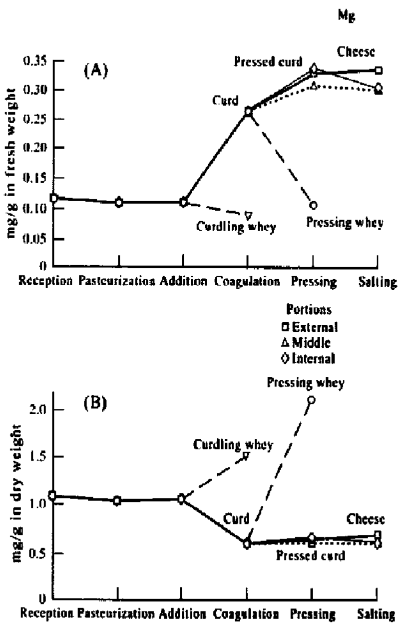

Fig. 2. Magnesium evolution during the cheese-making process.

For moisture and the five elements investigated, the homogenous groups formed with Scheffé's test were different and also differed if expressed at fresh or at dry weight (Table 1). An exception for all the elements and moisture were the liquid milks which belonged to the same homogenous group, both at fresh and dry weight.

For calcium four homogeneous groups were formed

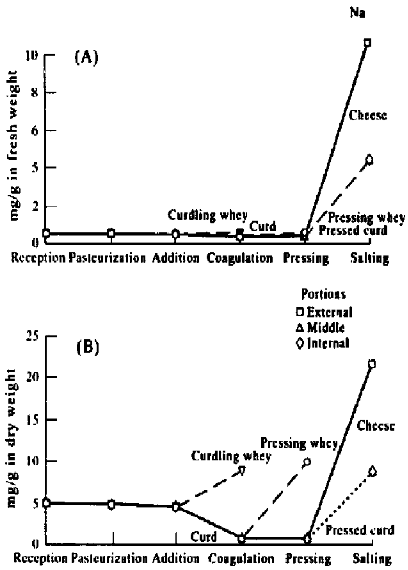

Fig. 3. Sodium evolution during the cheese-making process.

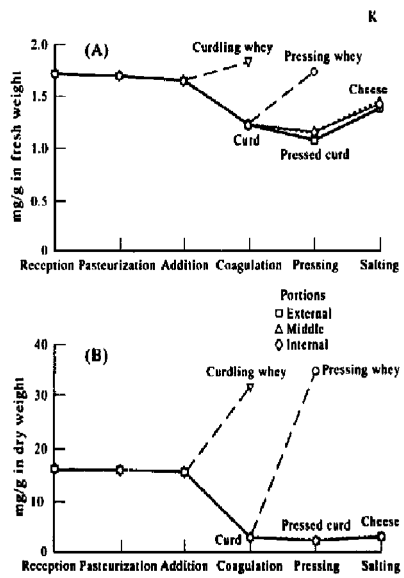

Fig. 4. Potassium evolution during the cheese-making process.

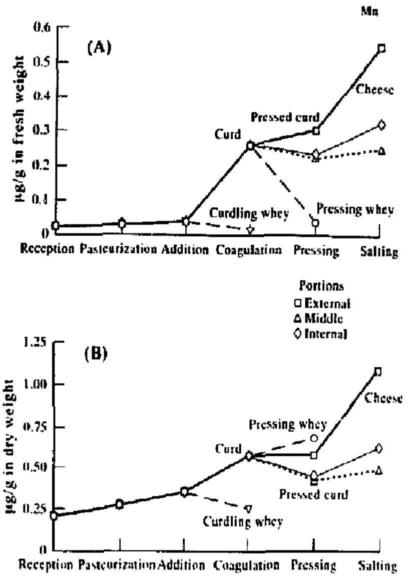

Fig. 5. Manganese evolution during the cheese-making process.

at fresh weight (Table 1): curdling and pressing whey formed the lowest concentration group, liquid milk formed the next group, curd constituted a group in itself and pressed curd and cheese formed the highest concentration group. At dry weight the trend of concentrations was that they were similar to fresh weight (Fig. 1(A) and (B)), but a higher number of groups were formed: curdling and pressing whey formed different groups and cheese formed a group between pressed curd with lower concentration and curd.

This distribution of concentrations indicated a higher association of calcium with solid fractions than with the whey fraction. The percentage of calcium associated with the curd which remained in the cheese, is calculated from the cheese produced and the mean concentrations of minerals in the different products of the cheese-making. In this work about 10·8% of the mass of milk was transformed into pressed curd, 87·3% into curdling whey and 1·9% into pressing whey. Allowing for the mean concentration for each of these, 62% of calcium remained in the pressed curd.

The observation coincides with the association of this element with the specific fractions of milk made by Renner *et al.* (1989) which indicated about 60% of calcium associated with the colloidal suspension of casein micelles. It also coincides with the degrees of filtration of this element in milk indicated by Fischbach-Green and Potter (1986): only 30% of calcium in milk is in a soluble form.

The homogeneous groups formed for magnesium by the Scheffè test at fresh weight were similar to calcium, but at dry weight magnesium distribution (Fig. 2(B)) in the different products, and the groups formed, was entirely different for that of calcium at dry weight, and also the reverse of the distribution of magnesium at fresh weight (Fig. 2(A)). The lower concentration group was formed by solid products (curd, pressed curd and cheese), the next group by liquid milks and, finally, curdling whey and pressing whey each formed a separate group with the highest concentrations (Table 1).

This reverse distribution at fresh and at dry weight of magnesium is due to the association of magnesium with specific fractions of milk. The percentage of magnesium associated with the curd remaining in the cheese now calculated is 31% and coincides with Renner *et al.* (1989) who indicated that about 70% of magnesium is in true solution, and also coincides with Fischbach-Green and Potter (1986) who indicated that 66% of magnesium in milk is in soluble form.

The distribution of sodium in the different products of the cheese-making process at fresh weight (Fig. 3(A)) and the homogeneous groups formed (Table 1) indicated only one factor which supposedly caused significant changes in the concentration of this element, the salting. Only two groups were formed by the Scheffè multiple range test, the lowest concentration group was formed by all the products except cheese which constituted in itself the other group. At dry weight (Fig. 3(B)), the groups formed indicated the association of sodium with the soluble fraction which produced lower concentration groups of solid products (except cheese). Actually the association of sodium with the soluble fraction should produce a similar distribution to that expressed at dry and fresh weight, but the greatest effects of salt addition were most notoriously expressed at fresh weight. The percentage of sodium associated with the curd remaining in the cheese now calculated is only 6%. Renner *et al.* (1989) indicated that sodium in milk is entirely soluble, and Fischbach-Green and Potter (1986) showed 98% soluble sodium. This 6% of sodium in pressed curd must be associated logically with the moisture of the product (47·8%).

A Scheffè range multiple test for potassium showed five homogeneous groups at fresh weight (Table 1); lower concentration groups were formed for solid products, pressed curd, curd, and cheese which formed three different groups of increasing concentration, respectively; liquid milk and pressing whey formed another group and curdling whey itself constituted the highest concentration group. At dry weight (Table 1), five groups were also formed and were similar to those formed at fresh weight with slight differences. The distribution of potassium at fresh and dry weight (Fig. 4) and the grouping mentioned above indicated the relation of potassium with the soluble fraction of milk, and the percentage calculated by us was 7% of the potassium remaining in cheese, probably associated with moisture (49·2%). Fischbach-Green and Potter (1986) indicated 98% of soluble potassium and Renner *et al.* (1989) reported that potassium in milk is entirely soluble.

The distribution of manganese in the products of the cheese-making process at fresh (Fig. 5(A)) and dry weight (Fig. 5(B)) was similar to the calcium distribution, except for the pressing whey at dry weight.

Only three homogeneous groups were formed by the Scheffé test for manganese at fresh weight (Table 1), the lowest being formed by all the liquid products, another formed by the curds and the highest formed by the cheese itself. At dry weight the groups formed were similar except for the pressing whey which itself constituted an intermediate group between curds and cheese groups. This abnormal concentration in pressing whey at dry weight could be due to contamination by manganese in the manufacturing process (pressing and perhaps salting), a fact previously noted by Pertoldi Marletta and Gabrielli Favretto (1983) in the making of Parmesan cheese, when contamination by iron was observed, and by Moreno-Rojas *et al.* (1994*a*) who observed contamination by iron, copper and zinc in Manchego-type cheese.

If the sources of contamination are removed, it can be seen that the degree of association of manganese with the precipitable fraction is similar to that of calcium. The calculated percentage of manganese in pressed curd is 67%; according to Fischbach-Green and Potter (1986) it was 64% and Renner *et al.* (1989) found 68% of manganese associated with casein and fat.

In order to verify the hypothesis of a possible contamination and the effect of salt added, three portions situated at different depths in the pressed curd and in the cheese were studied and analysis of variance showed statistically significant differences ($P<0{\cdot}001$) between portions for manganese in the two products, both at fresh and dry weight and for sodium in cheese also at fresh and dry weight (obviously because of salting). For instance, the highest concentration of manganese in both products was in the external portion and, of course, sodium in cheese was also in the external portion. Calcium, magnesium and potassium did not show statistically significant differences ($P>0{\cdot}05$) between portions.

The estimate made in Spain of the consumption of this type of product is approximately 3·11 g per person per day in a total diet of 1387 g (Instituto Nacional de Estadística, 1985). On considering the mean concentrations in fresh cheese for the five elements analysed, the daily intake estimated is 23·6 mg/day for calcium, 1·0 mg/day for magnesium, 20·1 mg/day for sodium, 4·3 mg/day for potassium and 1·1 μg/day for manganese. The density of nutrients presented by the minerals studied in fresh cheese for a male adult, with an energy contribution of 1800 kcal/day (NRC, 1989) is 1082% for calcium, 110% for magnesium, 45% for potassium, 8–21% for manganese and 307% for the highest and 1475% for the lowest recommended allowances for sodium, which indicates that fresh Manchego-type cheese is a poor source of manganese and potassium, a good source of magnesium, an excellent source of calcium and a dangerous source of sodium for people with a cardiovascular risk. The consumption of this type of cheese may be a problem for persons with kidney disease (litiasis).

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REFERENCES

Analytical Methods Committee (1987). Recommendations for definition, estimation and use of detection limit. *Analyst*, **112**, 199–204.

Coppini, D., Monzani, A., Parenti, C. & Plessi, M. (1979). Indagine spettrofotometrica di assobimento atomico sul contenuto di alcuni elementi metallici nel formaggio parmigiano reggiano. *Scienza e Tecnica Lattiero-Casearia*, **30**, 263–79.

Feeley, R. M., Criner, P. E., Murphy, E. W. & Toepfer, E. W. (1972). Major mineral elements in dairy products. *Research*, **61**, 505–10.

Fischbach-Green, L. & Potter, N. N. (1986). Effects of ultrafiltration on retention of minerals and other components of milk. *J. Food Sci.*, **51**, 345–7.

Fox, P. F. (1987). *Cheese: Chemistry, Physics and Microbiology*, (Vols I and II). Elsevier Applied Science, London, UK.

Franco, M. A., Balestrieri, F., Sabbatini, M. & Serra, A. (1981). Valutazione della concentrazione di ioni metallici presenti nel latte prodotto in Sardegna. *La Riv. della Soc. Ital. Sci. Aliment.*, **10**, 35–40.

Garcia Olmedo, R., Diez Marqués, M. C., Coll Hellin, L. & Barrera Vázquez, C. (1981*a*). Estudio de elementos minerales en leches de cabra y oveja. I Macroelementos. *Anales de Bromatología*, **33**, 1–10.

Garcia Olmedo, R., Diez Marqués, M. C., Coll Hellin, L. & Barrera Vázquez, C. (1981*b*). Estudio de elementos minerales en leches de cabra y oveja. II Microelementos. *Anales de Bromatología*, **33**, 77–84.

Instituto Nacional de Estadística (1985). *Encuesta de Presupuestos Familiares, Estudio sobre Nutrición*(5). DL M-32466-85, Spain.

Long, G. L. & Winefordner, J. D. (1983). Limit of detection a closer look at the IUPAC definition. *Anal. Chem.*, **55**, 712A–24A.

Moreno-Rojas, R., Amaro-López, M. A. & Zurera-Cosano, G. (1992). Mineral content modifications in Manchego-type cheese during ripening. *Food Chem.*, **45**, 319–22.

Moreno-Rojas, R., Amaro-López, M. A. & Zurera-Cosano, G. (1994*a*). Copper, iron and zinc variations in Manchego-type cheese during the traditional cheese-making process. *Food Chem.*, **49**, 67–72.

Moreno-Rojas, R., Pozo-Lora, R., Zurera-Cosano, G. & Amaro-López, M. A. (1994*b*). Calcium, magnesium, manganese, sodium and potassium variations in Manchego-type cheese during ripening. *Food Chem.*, **50**, 373–8.

NRC (1989). *Recommended Dietary Allowances (RDA)*. National Research Council, National Academy Press, Washington, DC, USA.

Pertoldi Marletta, G. & Gabrielli Favretto, L. (1983). Effects of processing on the occurrence of heavy metals in milk products. Proceedings of *Euro Food Chem II*, pp. 305–14.

Renner, E., Schaafsma, G. & Scott, K. J. (1989). Micronutrients in milk. In *Micronutrients in Milk and Milk-Based Food Products*, ed. E. Renner. Elsevier Applied Science, London, UK.

Scott, K. J. (1989) Micronutrients in milk products. In *Micronutrients in Milk and Milk-Based Food Products*, ed. E. Renner. Elsevier Applied Science, London, UK.

Snedecor, G. W. & Cochran, W. G. (1971). *Metodos Estadísticos*. Compañia Editorial Continental, Mexico.