Determination of calcium, iron and phosphorus in meat and meat products.

K. KOLAR and R. NILSSON

Swedish Meat Research Centre
Kävlinge, Sweden.

In our work on the chemical composition of meat and meat products we found it necessary to develop an appropriate system for serial analyses of calcium, iron and phosphorus. Many methods exist for the determination of these elements, but they had to be modified in different ways to serve our purpose. The aim of the work was to arrive at a simple analytical technique that allowed calcium, iron and phosphorus to be determined on the same pretreated sample.

METHODS

Preparing the sample

The sample was ground twice. 10 g was dried in a porcelain crucible at 135° for 2 h and then ashed at 550° for 5—6 h. The ash was dissolved in 5 ml of 6-n hydrochloric acid. The solution was then heated to dryness on a boiling water bath. The residue was dissolved in 15 ml of 0,5-n hydrochloric acid and filtered into a 50 ml volume flask. The crucible and the filter were washed with 3 × 10 ml of destilled water. The volume was adjusted to 50 ml.

Determination of calcium

Two methods were used for determination of calcium, one for the samples containing less than 0,1 % of calcium and one for samples containing more than 0,1 %.

A. Samples containing less than 0,1 % calcium

The calcium content of meat is about 10 mg per 100 g. This is too low a concentration to be determined accurately by titration with e.g. erichrom black as an indicator, as magnesium and phosphorus interfere. Neither was it possible to precipitate the calcium ions in this low concentration as sulphate.

But accurate determination are obtained by titrating 20 ml of the hydrochloric acid as solution with 0,001-n EDTA-solution with calcein as indicator

(1). Interfering phosphatates are removed by adding zirkonyloxichloride as described by INKLAAR and SANDIFORT (2).

The titration is carried out at a pH exceeding 12 at which pH magnesium-hydroxide is practically insoluble.

The use of calcein as an indicator makes it necessary to titrate in ultra violet light as calcium and calcein form a complex which has a strong green fluorescens when illuminated with light of a wavelength of about 360 nm. The end point of the titration is very sharp and is indicated by the extinction of the fluorescens.

B. Samples containing more than 0,1 % calcium

Meat products which had been enriched with calcium by adding bone, bone meal etc. were analysed by the MORGAN (3) method. In this case interfering substances were not removed but the calcium ions were precipitated from 20 ml of the hydrochloric acid acid ash solution as calcium sulphate and then determined by titration with 0,01—n EDTA. Eriochromblack T was used as an indicator.

Determination of phosphorus

The colorimetric method developed by ALLEN (4) was used. Phosphoric acid forms in perchloric acid solution a complex with molybdenic acid and this complex is reduced to a blue compound which is measured by absorption at 660 nm. For determination 0,3 ml of the hydrocloric acid ash solution is used.

Determination of iron

A method worked out by *Nordisk Metodikkommitté* (5). was used. It is a colorimetric method in which the absorption at 505 nm of a red ortho-phenontroline-iron complex is determined. 20 ml of the hydrochloric acid ash solution is used.

RESULTS AND DISCUSSION

To test the accuracy of the method we analysed lean meat to which phosphate, calcium salt and iron salt were added. Two levels of calcium content were analysed, one at about 30 mg per 100 g meat and one at about 100 mg per 100 g meat. The results are shown in table 1.

The maximum deviation from the mean values was for the calcium content according to method A ± 3,1 %, according to method B ± 1,8 %, for the phosphorus content ± 2,5 % and for the iron content ± 2,6 %. The accuracy of the methods seemed to be satisfactory for the intended purpose.

The recoveries were also satisfactory as 99 %, 98 %, 96 % and 100 % respectively of added calcium, phosphorus and iron was recovered.

The determination of both the calcium and the iron content requires a limited pH range. This determined the choice of method for ashing. Wet ashing is usually simpler and faster to perform than dry ashing. But in wet ashing strong sulphuric acid or perchloric acid is used, which makes a subsequent pH-adjustment difficult. We therefore preferred the dry ashing as it is then possible to dissolve the ash in a known volume of hydrochlorid. It is also possible to remove the hydrochloric acid by evaporation if necessary. Dry ashing is generally thought to give lower values for calcium than wet ashing, but the error is small and can be disregarded for our purposes.

The determination of calcium in meat and meat products is complicatep partly by the small amounts present in meat and partly the presence of interfering substances such as phosphorus, magnesium and iron. These problems were discussed above. It is of course possible to use method A even for determination of calcium in samples containing more than 0,1 % calcium, but as titration in ultra violet light is trying to the eyes we used it only when necessary.

The methods are now in use at our institute for the determination of minerals in meat from different species and in different meat products. The results of these investigations will be reported elsewhere.

Table 1. *Determination of calcium, iron and phosphorus in lean meat to which phosphate, calcium salt and iron salt were added.*

	Ca		*Ca*		*P*		*Fe*	
	Method A		*Method B*					
Sample	*mg/100 g*		*mg/100 g*		*mg/100 g*		*mg/100 g*	
Lean meat	10.9	11.2			203	207	2,84	2,96
	10,4	10,7			204	211	3,02	2,87
	11,5	11,3			206	208	2,87	2,82
	10,7	11,1			205	207	2,90	2,94
	11,2	11,3			202	209	2,85	2,89
	11,3	11,3			201	208	2,90	2,94
	MV	11,1			206		2,9	
Lean meat +	30,4	30,8						
20 mg Ca/100 g	30,5	31,5						
	30,6	30,9						
	30,7	30,7						
	30,6	31,5						
	30,8	31,3						
	MV	30,9						
Lean meat +			108	111	300	306	4,80	4,86
100 mg Ca/100 g +			106	109	299	303	4,83	4,86
100 mg P/100 g +			110	113	306	297	4,87	4,84
2 mg Fe/100 g			107	109	304	305	4,85	4,89
			107	111	300	301	4,86	4,85
			109	110	296	304	4,87	4,83
			MV	109	302		4,9	
Recovery	19,8		98		96		2,0	

REFERENCES

1. Diehl, H. and Ellingboe, J. L.: Indikator for Titration of Calcium in Presence of Magnesium using EDTA. Anal. Chem. 28, 882, 1956.
2. Inklaar, P. and Sandifort, J.: Determination of Calcium and Magnesium in Meat. J. of Food Sci. 32, 622, 1967.
3. Montag, A.: Bestimmung von Calcium und Magnesium in phosphatreichen Lebensmittelashen. Z. f. Lebensm. Unt. u. Forsch. 134, 287, 1967.
4. Allen R. J. L.: The estimation of Phosphorus. Biochem. J. 34, 858, 1940.
5. Bestemmelse af Jern i Cerealier. Nordic Committee on Food Analysis, 22, 1955.