FARINDOLA SHEEP CHEESE[1]

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Abstract

Samples of sheep cheese produced with pig rennet in the Farindola area were compared with samples of sheep cheese produced with calf rennet which were produced using the same methods of processing and aging as the Farindola cheese.

To distinguish the samples 18 chemical parameters (among which, 9 amino acids) and 25 volatile substances were chosen. In addition a taste analysis was carried out by a Panel of tasting experts. All of the data thereby obtained was analysed statistically through discriminate analysis. Both the analytical data and the taste expert Panel results show a net differentiation between the cheeses produced with the two different types of rennet. The different procedures used to treat the surfaces of the two different types of cheese account to some extent for the differentiation but the rennet used is by far the more significant factor in distinguishing the cheeses.

Riassunto

Sono stati presi in esame campioni di formaggio pecorino prodotti con caglio di maiale nell'area tipica di Farindola in confronto con campioni

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prodotti con caglio di vitello nelle stesse condizioni di processo e stagionatura. Per la differenziazione dei campioni sono stati dosati 18 parametri chimici (fra cui 9 aminoacidi) e 25 sostanze volatili ed inoltre è stata eseguita un'analisi sensoriale con un Panel Test di esperti assaggiatori. Tutti i dati ottenuti sono stati analizzati statisticamente mediante l'analisi discriminante.

Sia il confronto dei dati analitici che quello del Panel test hanno mostrato una netta differenziazione fra i formaggi prodotti con due cagli diversi. Il trattamento di superficie, previsto dal Disciplinare di produzione, influisce sulla differenziazione dei campioni ma non tanto quanto il caglio utilizzato.

Keywords: Pig rennet, calf rennet, sheep cheese, Farindola sheep cheese.

Introduction

Farindola sheep cheese is a characteristic product of the Farindola area and is perhaps unique among Italian sheep cheeses, which typically use calf rennet, since Farindola cheese is made instead with pig rennet. There are historical records of the use of pig rennet in the production of sheep cheese in the eastern part of the Gran Sasso area dating back to Roman times; it is referred to as the "cheese of the Vestini."

The denomination "Farindola Sheep Cheese" appears in some texts on Italian cheese in the early 1900's. It must be noted that while the animals usually used for rennet (calves, lambs, kids) have not yet been weaned, the animals used for pig rennet are over one year of age[3].

A distinctive characteristic of pig rennet, which is emphasised by all of the experts, is that it gives the cheese produced from it a much less sharp flavour than cheese produced from calf rennet, even after lengthy aging (1).

[3] Liquid pig rennet is made by working the mucous membrane of pig stomachs, first washing it carefully and then cutting it into narrow strips which are then covered with salt for 2 or 3 days (dry salting). It is then left to marinate in a dark glass container in a mixture of white vinegar, white wine, hot peppers and, sometimes, pepper corns. The mucous membrane is left in the marinade for 3 to 4 months. The rennet is then filtered through a linen cloth at least 5 or 6 days before use and it is stored in a dark place

The territory in which this traditional production method is used includes nine towns located within the provinces of Pescara and Teramo (Figure 1). The towns located in the interior of the traditional production area for the province of Pescara are Farindola, Montebello di Bertona, Villa Celiera and Carpineto della Nora.

The town within the traditional production area of the Province of Teramo is Arista. There are other towns that are not completely within the borders of the traditional production areas, and they are Penne and Civitella Casanova in the province of Pescara and Bisenti and Castelli in the province of Teramo.

Fig. 1 – Production area for Farindola Sheep Cheese.

Traditionally the natural animal "rennet" obtained from the stomach of a mammal (calf, lamb, buffalo, pig, etc.) was used to make cheese. In this regard, Merker, in a conference held in 1918 observed that "it has been known for a number of years that pepsin, a substance produced from the mucous membrane of the pig's stomach, would give excellent results in curdling milk for the manufacture of cheese" (2). In the modern cheese manufacturing industry, however, the use of industrially produced bovine rennet is the preferred method for obtaining curd, because of its standardised production and enzymatic activity which allows the cheese manufacturer to accurately determine clotting and cutting times.

As a result of the preference for enzyme coagulants, cheeses produced with the traditional method directly from pig rennet have not been the object of a systematic study. In fact, a consultation of the major international data banks will show that there are no scientific works on cheeses obtained from pig rennet[4].

Works are to be found, instead, regarding the characteristics of the enzymes (pepsin and chymosin) extracted from the mucous membrane of various mammals. In the case of "pig rennet" the enzyme which is extracted is pepsin. The literature on the subject reports that, of the available enzymes, pig pepsin is the only one which is potentially less proteolytic than chymosin, since it is readily denaturated in the cheese making process. Pig pepsin is unstable above the pH 6.0 compared to chymosin: pH 6.7 (3). Clotting activity of pig pepsin was extremely pH-dependent around pH 6.6 and coagulation did not occur above pH 6.68 (4). Other works deal with the activity of enzymes and their coagulatory and proteolytic effect on curd (4-6). In one work published in 1972 pig pepsin was used in the experimentation on cheese but only "as pig rennet substitutes" (7).

In order to distinguish animal rennet and other enzymes the characteristics of the chymosin gene, isolated from tissue extracts of buffalo, camel, cow and pig, were studied through SDS PAGE Polyacrylamide gel electrophoresis and PCR amplification techniques. Two prominent proteins were found in cow and buffalo rennet, while only one protein was observed in camel and pig (8).

It is known that the type of rennet used affects the rates of proteolysis and lipolysis and consequently flavour development in cheese (9-11). In light of these findings, further research was deemed to be called for into the differences in composition and organoleptic qualities of two samples of

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[4] The Scopus data bank, which reviews more than 18,000 journals, contains no works on this argument.

sheep cheese obtained under the same conditions, but using different rennet; one sample was produced with pig rennet and the other with calf rennet. In order to identify any possible effects from the treatment of the surface of the cheese with oil and vinegar, as required by Production Regulations, two of each sample were produced, one treated and one untreated.

Material and Methods

Freshly milked sheep milk, coming from sheep that produce less than 1 litre in about 100 milking days, was kept cool (10-12 °C) but not refrigerated. The milk was curdled at 32-34 °C with pig rennet obtained using the method described in footnote 2 above. The setting time varied from 40 to 60 minutes; after the curdle was broken into granules of 0.5 to 2 cm, it was placed into straw forms to harden; it was then dry salted with coarse salt on both sides. The salt was then washed off. The surface of the cheeses were periodically treated with extra virgin olive oil and vinegar (or tomato juice)[5]. The period of aging varied from a minimum of three months to a maximum of a year and each whole cheese weighed between 1 and 2 kilos.

Samples and Parameters: 1) 1 kg forms have been used that are generally introduced into commerce after 3-6 months, but even up to 1 year, ripened to a temperature between 10 and 14 °C; 2) time 3, 6 and 9 months; 3) treatment with oil and vinegar and without.

Using the parameters mentioned above, the time evolution of 9 amino acids (histidine, tyrosine, ornithine, lysine, phenylalanine, tryptophan, methionine, cystein and citrullin), and of 9 other variables such as water, cholesterol, unsatured fats, satured fats, linoleic acid isomers, Vitamine A and E, polyphenols, have been analyzed. A further 25 volatile substances have been determined qualitatively as a function of time evolution.

Apparatus: HPLC Dionex with electrochemical Detector (AA direct gold electrode) (12) for amino acids (13-14).

GasChromatograph Agilent Technologies 6890N with mass spectrometry Agilent 5973 inert, column HP 5MS for fatty acid, vitamin and polyphenol analyses, and Phenomenex ZB WAX PLUS for aromatic substance analysis.

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5 From this point on, for purposes of simplicity, the words “treated” or “non-treated” will be used.

Sample preparation: for the amino acid analysis the samples have been homogenized in methane sulfonic acid 4 M, evaporated in nitrogen current and taken back with bi-distillated water.

For the fatty acid analysis a cold trans-esterification with methanolic solution of potassium hydroxide has been carried out.

For vitamine and polyphenol analysis an extraction with hexane/acetone 1:1 has been carried out. The extracted substance is dried and is made to react with 50 μL di N-methyl-N-trimethylsilyl-trifluoroacetamide (MTSF) for 15 min at 75 °C.

For volatile substance qualitative analysis, about 5 g of the sample is placed in a 20 mL holding vial. The sealed vial is then inserted into the sampler for headspace at a temperature of 90 °C for 30 min. After 30 min. the sampler automatically extracts 500 μL of headspace injecting it into GC/MS.

Sensorial analysis: the Panel Test was constituted by 5 experienced tasters that have taken into consideration the following parameters: sweetness, acidity, saltiness, bitterness, spiciness, humidity, solubility and friability. The evaluation scheme used is that suggested by the National Cheese Taster organization (ONAF) in Cuneo.

Statistical Analysis: Linear Discriminant Analysis (LDA) was applied to the separation of the analysed cheese samples according to different aging temperatures and to the Panel Test judgement. As the group-membership of each sample was already known, LDA was applied to this variable set in order to evaluate the sample differentiation and classification of the data expressed as discriminant scores. LDA has been extensively discussed by several authors (15-17).

Results and Discussion

The aminoacid and all the other component evolution is reported in Table 1a and 1b.

The volatile substances present in the sheep cheese samples analysed are reported in Table 2.

Panel test for all samples is referred in Table 3.

An example of the results of sensorial analysis is reported in Figure 2; it shows a graph related to a sample obtained with pig rennet “treated” and a graph of a sample obtained with calf rennet “treated”.

TABLE 1a

AMINOACID CONTENT (mg/kg) IN FIVE SHEES CHEESE SAMPLES AS A FUNCTION OF TIME OF AGING

Sample	histidine	tyrosine	ornithine	lysine	phenylalanine	tryptophane	methionine	cystein	citrullin
Pig rennet "Treated"									
3 months	6,130	12,130	1,480	11,970	11,520	2,560	6,180	2,300	2,890
6 months	7,121	13,100	1,860	13,860	12,920	2,710	6,320	2,180	2,960
9 months	7,110	14,020	1,620	12,610	12,620	2,630	6,910	2,430	3,160
Pig rennet "Untreated"									
3 months	7,080	12,620	1,590	12,530	12,130	2,720	6,530	2,910	2,630
6 months	6,610	13,350	1,730	14,100	12,160	2,620	6,260	3,190	2,810
9 months	7,830	13,650	1,830	13,950	13,510	3,060	7,260	2,610	3,320
Calf rennet "Treated"									
3 months	6,950	11,460	1,680	12,620	11,890	2,850	6,980	2,580	2,750
6 months	7,520	12,520	1,820	12,650	12,380	3,180	6,830	2,560	3,280
9 months	7,650	13,280	1,920	13,160	12,960	3,210	7,110	2,530	3,160
Calf rennet "Untreated"									
3 months	6,420	11,980	1,540	12,090	11,310	2,360	6,880	2,460	3,090
6 months	7,950	13,910	1,980	13,980	13,620	3,050	7,950	3,420	3,760
9 months	8,130	13,910	1,850	13,820	13,190	2,410	7,610	2,420	3,160
Castel del Monte									
3 months	5,850	10,820	1,390	11,420	10,680	1,910	5,910	2,050	2,010
6 months	6,110	11,830	1,610	12,120	11,760	2,190	5,980	2,750	2,810
9 months	7,085	12,980	1,720	13,180	12,080	2,820	5,910	1,940	2,720

TABLE 1b

OTHER VARIABLE CONTENT IN FIVE SHEEP CHEESE SAMPLES AS A FUNCTION OF TIME OF AGING

Sample	Water %	Cholesterol (mg/kg)	Unsatured fats %	Satured ats %	Linoleic acid isomers %	Vitamin A (µg/kg)	Vitamin E (mg/kg)	Polyphenols (mg/kg)
Pig rennet "Treated"								
3 months	34.91	770	6.93	18.63	0.54	3,180	3.60	41.60
6 months	29.88	790	7.71	20.63	0.80	3,710	3.90	41.20
9 months	27.06	710	9.71	21.73	0.83	4,026	4.20	48.30
Pig rennet "Untreated"								
3 months	33.53	700	7.18	20.04	0.65	3,610	4.20	37.30
6 months	30.12	720	7.53	20.55	0.72	3,520	4.10	38.80
9 months	25.28	840	10.51	23.09	0.91	4,220	4.90	52.60
Calf rennet "Treated"								
3 months	32.29	630	6.98	20.79	0.76	3,870	4.50	29.70
6 months	29.38	670	7.65	21.53	0.75	4,130	4.80	35.20
9 months	25.04	810	10.30	22.94	0.90	4,113	4.60	46.90
Calf rennet "Untreated"								
3 months	33.31	740	6.40	19.39	0.66	3,490	3.40	40.30
6 months	27.57	750	8.10	22.54	0.71	3,910	3.90	44.70
9 months	22.49	950	10.78	24.83	1.03	4,125	5.10	55.10
Castel del Monte								
3 months	37.62	540	5.93	14.49	0.51	2,820	2.90	33.50
6 months	30.36	610	6.53	16.80	0.60	3,106	3.30	36.30
9 months	30.20	690	7.95	18.06	0.58	3,620	3.80	38.90

TABLE 2

VOLATILE SUBSTANCES PRESENT IN FIVE SHEEP CHEESE SAMPLES AS A FUNCTION OF TIME OF AGING*

Volatile substances **	**Pig rennet "treated"**			**Pig rennet "untreated"**			**Calf rennet "treated"**			**Calf rennet "untreated"**			**Castel del Monte**		
	months			**months**			**months**			**months**			**months**		
	3	6	9	3	6	9	3	6	9	3	6	9	3	6	9
Ethyl alcohol	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Butyric acid, ethyl ester	x	x	x	x	x		x	x	x	x	x	x	x	x	x
Methyl propyl ketone	x	x	x	x	x	x	x	x	x	x	x	x		x	
Acetic acid, ethyl ester	x	x	x	x	x	x	x	x	x	x	x	x		x	
2-Propanol	x	x	x	x	x	x	x	x	x	x	x			x	x
1-Pentanol	x	x	x	x	x	x	x	x	x	x	x	x		x	
2-Pentanol	x	x	x	x	x	x	x	x	x	x	x	x		x	
2-Butanol	x	x		x		x	x			x		x	x	x	x
Acetone		x	x			x		x	x		x	x	x	x	x
Capric acid, ethyl ester		x			x			x	x		x			x	
1-Butanol		x			x			x			x		x	x	
Volatile substances ***															
2-Butanone, 3-hydroxy					x			x		x	x				
Methyl amyl ketone						x		x	x						
2-methyl-valeraldehyde						x			x			x			
Methyl ethyl ketone						x							x		x
1-Propanol								x						x	x
Dimethyl diketone						x			x						
Isopropyl aldeide								x			x				
2-Butanone, 4-hydroxy			x												
Methylethylacetaldehyde						x									
Dimethyl disulfide						x									
1,3-Propanedioll, 2-amino-2-ethyl						x									
Acetic acid, iso-propyl-ester									x						
Propanoic acid, ethyl ester											x				
Formic acid, butyl ester													x		

* The order in which the substances appear in the table is in function of their decreasing presence in the cheeses studied.

** Substances present in all of the varieties of cheese.

*** Substances present in only some of the types of cheese studied.

TABLE 3

PANEL TEST FOR FIVE SHEEP CHEESE SAMPLES AS A FUNCTION OF TIME OF AGING

Sample	Sweetness	Acidity	Saltiness	Bitterness	Spiciness	Humidity	Solubility	Friability
Pig rennet "Treated"								
3 months	4	2	1.7	0	0	3	5	3
6 months	2	0	2.7	0	0	2.7	5.7	5
9 months	4	1	4	0	2	3	4	3
Pig rennet "Untreated"								
3 months	3	3	2.7	0	0	3	5	3
6 months	2	0	3	1	0	3	5	3
9 months	5	0	3	0	1	2	3	2
Calf rennet "Treated"								
3 months	4	4	2	0	0	4.5	5.5	4
6 months	2	1.7	3	0	0	2.7	5.7	5
9 months	4	2	3	1	1	1.7	3	4
Calf rennet "Untreated"								
3 months	3	4.5	2	0	0	4.5	5	4
6 months	1.7	1	2.7	0	0	1.7	4.5	5
9 months	4	1	3	2.7	1	3	4	2
Castel del Monte								
3 months	1	0	2	2	1	5.5	5	3
6 months	1	0	3.7	4	2	3	6	4.5
9 months	2	1	5	3	1	4	4	2

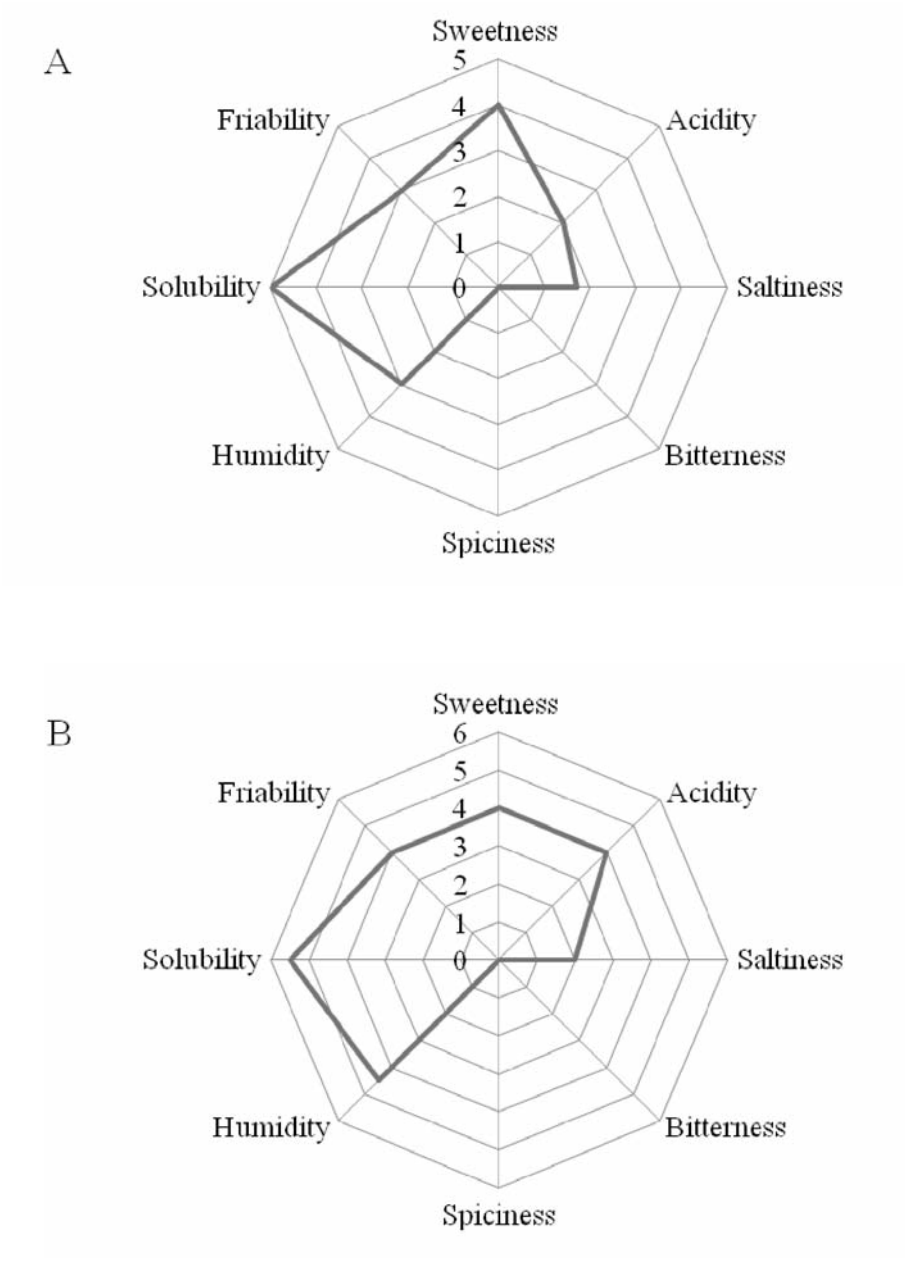

Fig. 2 – Graph A relates to a sample obtained with pig rennet "treated" with a time of aging of 3 months and graph B relates to a sample obtained with calf rennet "treated" with a time of aging of 3 months.

The presence of 25 volatile substances was ascertained. Although at this point of our paper we are simply noting the findings as to their presence or absence, some observations can be made here.

As can be seen from Table 2, 11 of these substances are present in all of the types of cheese studied; clearly, they are normal products of fermentation. The other 14 substances are present in only some of the types of cheese studied. It is of interest to note here that all 14 of these substances are absent in the treated cheese made from pig rennet, with the exception of 2-butanone-4-hydroxy, which gives a pleasing aroma with a note of butter. Some of the substances detected , for example, the last seven substances listed in the Table, were present only once.

In addition, in the sample of treated cheese made from pig rennet, the unpleasant smelling 2-methyl-valeraldehyde does not form, as it does in the other samples.

Methyl ethyl acetaldeide, dimethyl sulphide, 1,3-propanedioll,2-amino-2-ethyl, present exclusively in untreated cheese made from pig rennet, are generically unpleasant substances, and the treatment required by the Production Regulations would seem to serve in impeding the formation of just these substances.

In contrast, in products made with calf rennet the substances formed - 1-propanol, isopropyl aldeide, propanoic acid ethyl ester, formic acid butyl ester - are all substances that are generally considered unpleasant. One exception, however, is acetic acid iso-propyl ester which has an delicately fruited scent.

All data obtained were analysed statistically using the multivariate statistical approach, in particular Linear Discriminant Analysis.

This methodology was applied to separate the cheese samples based on the presence of amino acid and the other compound dosed and furthermore on the sensorial analysis, using rennet and treatment, as class identity. The aim of this procedure was to evaluate sample differentiation and classification of data expressed as discriminant scores.

Results show that the cheese samples are classified correctly; the overall classification success was 100.0% as a function of aging time.
Therefore, depending on the number of groups, one or two discriminant functions were extracted. To determine the number of linear discriminant functions to retain, Bartlett's classical test was applied,

$$b = -[N - (p + g)/2 - 1] \ln \Lambda$$

where N stands for the number of observations, p for the number of variables, g for the number of groups and Λ represents the ratio of the within-group sum of squares to the total sum of squares. Wilks' Λ value provides information pertaining to how much of the total variability is due to the differences between the group means or to the within-group variability. The value of Λ can range between 0 and 1: $\Lambda = 1$ when the two group means are equal, while $\Lambda = 0$ if they differ.

Once a set of q variables has been selected, the classification rule (also known as Fisher's linear Discriminant functions) can be computed using:

$$b_{ij} = (n-g)\sum_{l=1}^{q} w_{il}^{*}\overline{X_{lj}}$$

$i = 1, 2, \ldots, q;\ j = 1, 2, \ldots, g$

for the coefficient, and

$$a_j = \log p_j - \frac{1}{2}\sum_{i=1}^{q} b_{ij}\overline{X_{ij}}$$

$j = 1, 2, \ldots, q$

for the constant, where p_j is the prior probability of group j.

A significant Wilks Λ value was obtained when the cheese samples were classified as a function of the rennets. In this case, one discriminant function was estimated, since the number of groups in this sample was 2, and 2–1 is the maximum allowable number of eigenvalues for the matrix $W^{-1}B$. The first discriminant eigenvalue (41.013) had a Wilks Λ value close to zero (0.024).

The distribution of data expressed as discriminant scores along the first eigenvector is presented in Figure 3. In this representation of all data, the two sample classes, corresponding to cheese samples with pig rennet and cheese samples with calf rennet, respectively, were clearly distinct.

Based on the values for the two linear discriminant functions for each sample, the group membership could be predicted using a classification rule. Table 4 summarises the results of the classification for the cheese samples, where the actual and predicted group membership and, on the diagonal, the number of the samples classified correctly, are shown.

In this case, all cheese samples were correctly assigned to the group they belong to. Furthermore the overall classification success was 100.0%.

According to Wilks Λ value another distribution was quite significant. In fact, if the whole data set is analysed as a function of treatments, the results obtained are the following. In this case, one discriminant function was estimated since the number of groups in this sample was 2, and 2–1 is the maximum allowable number of eigenvalues for the matrix $W^{-1}B$. The first discriminant eigenvalue (7.864) had a Wilks Λ value close to zero (0.113).

The distribution of data expressed as discriminant scores along the first eigenvector is presented in Figure 4. In this representation of all data, the two sample classes, corresponding to cheese samples treated and untreated, respectively, were distinct.

Based on the values for the two linear discriminant functions for each sample, the group membership could be predicted using a classification rule. Table 5 summarises the results of the classification for the cheese samples, where the actual and predicted group membership and, on the diagonal, the number of the samples classified correctly, are shown. In this case, all cheese samples were correctly assigned to the group they belong to. The overall classification success was 100%.

By comparing the two elaborations above described, the variable "rennet" has a bigger discriminatory significance than variable "treatment".

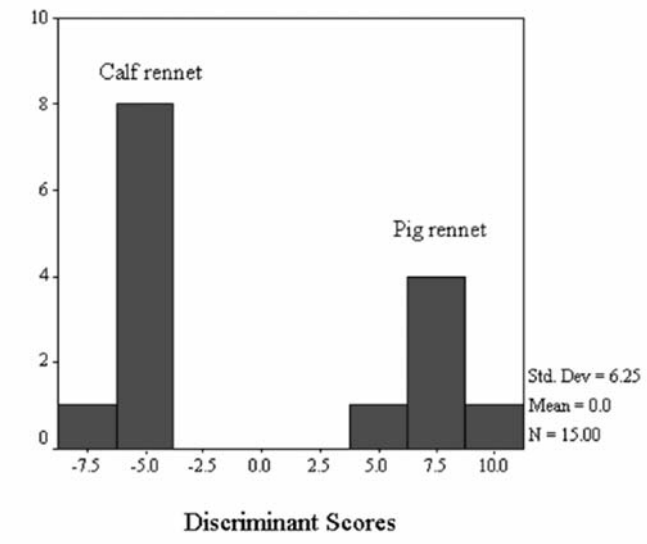

Fig. 3 - The distribution of data, related to cheese samples as function of the two rennets, expressed as discriminant scores along the first eigenvector.

TABLE 4

CLASSIFICATION TABLE FOR 2 GROUPS OF CHEESE SAMPLES AS A FUNCTION OF TWO RENNETS (G1 = CHEESES WITH PIG RENNET; G2 = CHEESES WITH CALF RENNET).

	Predicted Group		
Actual Group	G1	G2	Total
G1	6	0	6
G2	0	9	9
Total	6	9	15
Accuracy of prediction, %	**100.0**	**100.0**	**100.0**

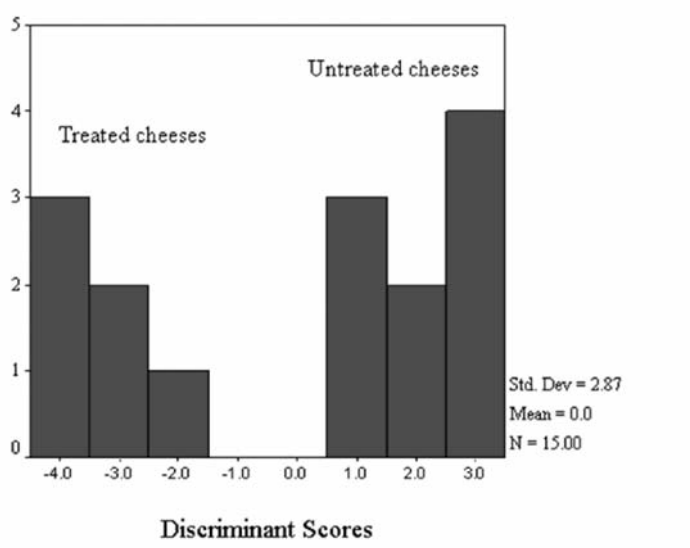

Fig. 4 - The distribution of data, related to cheese samples as function of the treatments, expressed as discriminant scores along the first eigenvector.

TABLE 5

CLASSIFICATION TABLE FOR 2 GROUPS OF CHEESE SAMPLES AS A FUNCTION OF TRATTAMENTI (G1 = TREATED CHEESES; G2 = UNTREATED CHEESES).

	Predicted Group		
Actual Group	G1	G2	Total
G1	6	0	6
G2	0	9	9
Total	6	9	15
Accuracy of prediction, %	**100.0**	**100.0**	**100.0**

A significant Wilks Λ value was obtained when the cheese samples were classified as a function of the rennets and of the treatment. In this case, 3 discriminant functions were estimated, since the number of groups in this sample was 4, and 4–1 is the maximum allowable number of eigenvalues for the matrix $W^{-1}B$.

The first discriminant eigenvalue (38.552) had a Wilks Λ value close to zero (0.001).

The distribution of data expressed as discriminant scores along the first eigenvector is presented in Figure 5.

In this representation of all data, the four sample classes, corresponding to cheese samples with pig rennet “treated” (G1), pig rennet “untreated” (G2), calf rennet “treated” (G3) and calf rennet “untreated” (G4), respectively, were clearly distinct.

Based on the values for the four linear discriminant functions for each sample, the group membership could be predicted using a classification rule.

Table 6 summarises the results of the classification for the cheese samples, where the actual and predicted group membership and, on the diagonal, the number of the samples classified correctly, are shown. In this case, all cheese samples were correctly assigned to the group they belong to. Furthermore the overall classification success was 100.0%.

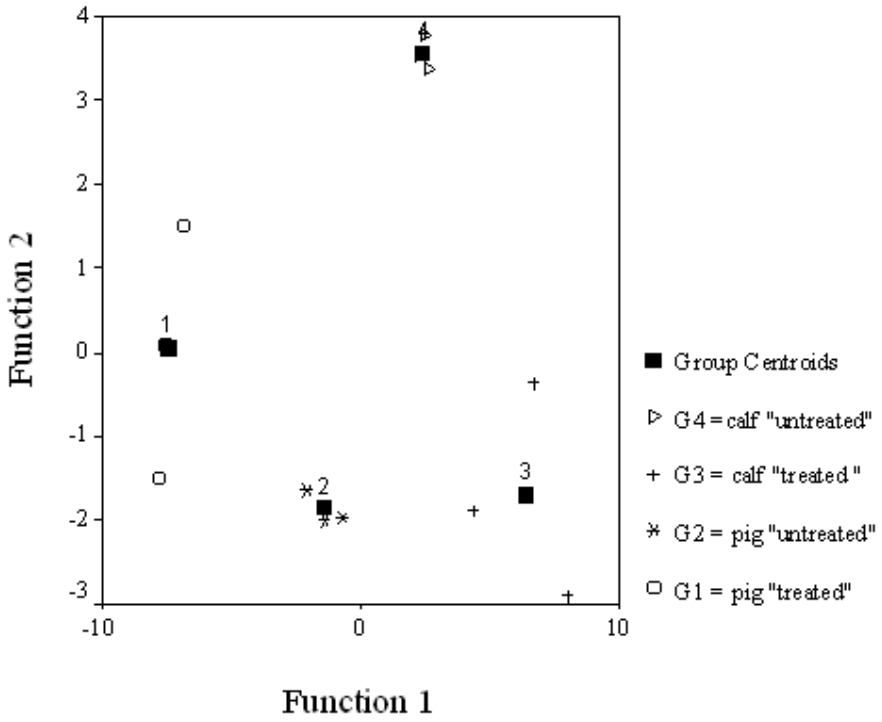

Fig. 5 - Discrimination of cheeses as a function of rennets and treatments, expressed as discriminant scores along the first two eigenvectors.

TABLE 6

CLASSIFICATION TABLE FOR 4 GROUPS OF CHEESE SAMPLES AS A FUNCTION OF RENNETS AND TREATMENTS (G1 = PIG RENNET "TREATED"; G2 = PIG RENNET "UNTREATED"; G3 = CALF RENNET "TREATED"; G4 = CALF RENNET "UNTREATED").

	Predicted Group				
Actual Group	G1	G2	G3	G4	Total
G1	3	0	0	0	3
G2	0	3	0	0	3
G3	0	0	3	0	3
G4	0	0	0	3	3
Total	3	3	3	3	12
Accuracy of prediction, %	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**

According to Wilks Λ value another distribution was quite significant. In fact, if the whole data set is analysed as a function of Panel Test, the results obtained are the following.

In this case, 3 discriminant functions were estimated, since the number of groups in this sample was 4, and 4–1 is the maximum allowable number of eigenvalues for the matrix $W^{-1}B$.

The first discriminant eigenvalue (13.090) had a Wilks Λ value close to zero (0.018).

The distribution of data expressed as discriminant scores along the first eigenvector is presented in Figure 6. In this representation of all data, the four sample classes, corresponding to cheese samples with pig rennet "treated" (G1), pig rennet "untreated" (G2), calf rennet "treated" (G3) and calf rennet "untreated" (G4), respectively, were distinct.

Based on the values for the four linear discriminant functions for each sample, the group membership could be predicted using a classification rule.

Table 7 summarises the results of the classification for the cheese samples, where the actual and predicted group membership and, on the diagonal, the number of the samples classified correctly, are shown. In this case, all cheese samples were correctly assigned to the group they belong to. The overall classification success was 100%.

The organoleptic differences between Farindola sheep cheese and that produced in industrial cheese factories with calf rennet are to be confirmed. In fact, the Panel Test found that the "industrial" sheep cheese is systematically more spicy than the Farindola one, whereas this latter is always more sweet and never bitter. As it is known, a positive correlation was found between β-casein degradation and the bitter taste (18) and the main peptides responsible for the bitter taste in cheese appear to be those corresponding to the C-terminal portion of β-casein (19-20).

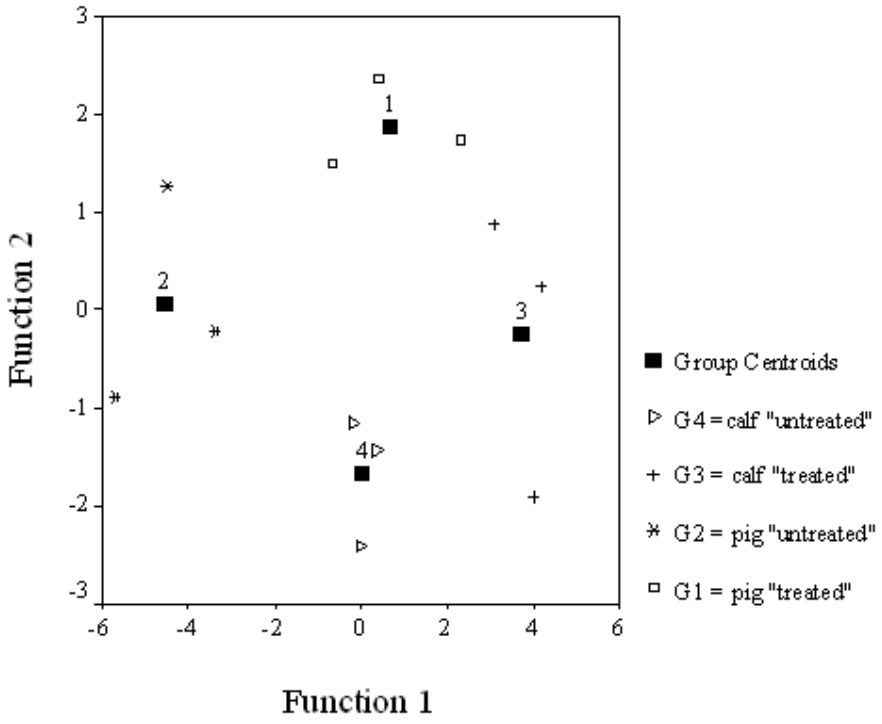

Fig. 6 - Discrimination of cheeses obtained with different rennets, as a function of Panel Test, expressed as discriminant scores along the first two eigenvectors.

TABLE 7

CLASSIFICATION TABLE FOR 4 GROUPS OF CHEESE SAMPLES AS A FUNCTION OF PANEL TEST (G1 = PIG RENNET "TREATED"; G2 = PIG RENNET "UNTREATED"; G3 = CALF RENNET "TREATED"; G4 = CALF RENNET "UNTREATED").

	Predicted Group				
Actual Group	G1	G2	G3	G4	Total
G1	3	0	0	0	3
G2	0	3	0	0	3
G3	0	0	3	0	3
G4	0	0	0	3	3
Total	3	3	3	3	12
Accuracy of prediction, %	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**

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