



75th EAAP ANNUAL MEETING – FLORENCE 2024

THE ROLE OF COLOUR INDEXES FOR THE ASSESSMENT OF SPECIES-SPECIFIC DIFFERENCES IN QUALITY TRAITS OF MILK FROM DOMESTIC RUMINANTS



JAVIER CABALLERO VILLALOBOS | Assistant Professor MVB · MSc · PhD · CertDHH



Meet the **research team**

A quick glance at some of the people involved in this research line



Javier Caballero-Villalobos
UNIVERSIDAD DE CÓRDOBA



Ana Garzón
UNIVERSIDAD DE
CÓRDOBA



José Perea
UNIVERSIDAD DE
CÓRDOBA



Eoin Ryan
UNIVERSITY COLLEGE
DUBLIN



Orla Keane
TEAGASC
GRANGE



Elena Angón
UNIVERSIDAD DE
CÓRDOBA



Miguel Cantarero
UNIVERSIDAD DE
CÓRDOBA



Ramón Arias
CERSYRA

CIENCIA ANIMAL
AGR-195



INTRODUCTION





Technological aptitude of milk

The cheese sector demands the development of rapid and low-cost methods to determine milk efficiency.



GAPS OF KNOWLEDGE
TECHNOLOGICAL
PERFORMANCE OF MILK



INDUSTRY CHALLENGES
ECONOMIC
VALUE OF MILK



RESEARCH OPPORTUNITIES
RAPID AND INEXPENSIVE
ANALYTIC METHODS





Colour in the food industry

Colour has been traditionally used to characterize the composition and quality of food products.

The colour of milk can vary greatly and depends to a large extent on its physical structure, the fat content and the pigmentation due to the presence of carotenoids and riboflavins.



- 1 Variation of **colloidal particles**.
- 2 Effect of **nutrition** in grazing herds.
- 3 Herd **health and management** practices.

OUR PROPOSED LINE OF RESEARCH

- To explore if colour can be adopted as a tool to predict technological quality of ruminant milk intended for cheesemaking.
- To assess if colour can be used as a discriminator, not only at a species level, but also at a breed level.





Background investigation

What have we achieved so far with this research line?



J. Dairy Sci. 103:4951–4957
<https://doi.org/10.3168/jds.2019-17201>
© American Dairy Science Association®, 2020.

Using multivariate analysis to explore the relationships between color, composition, hygienic quality, and coagulation of milk from Manchega sheep

A. Figueroa,¹ J. Caballero-Villalobos,^{1*} E. Angón,¹ R. Arias,² A. Garzón,¹ and J. M. Perea¹

¹Departamento de Producción Animal, Universidad de Córdoba, Córdoba 14071, Spain

²Centro Regional de Selección y Reproducción Animal de Castilla-La Mancha, Valdepeñas, Ciudad Real 13300, Spain



31
WBC
WORLD BUIATRICS CONGRESS
SEPTEMBER 4TH TO 8TH
MADRID 2022

VERY GOOD PREDICTOR FOR COAGULATION

NON-COAGULATING MILK → FALSE NEGATIVES (23-33%)



METHODOLOGY





Sample collection

Milk samples ($n = 2400$) were collected from dairy farms located in Spain and Ireland in the last 10 years.





Milk composition and SCC

Milk composition was performed by direct measurement of the percentage of its major components using a Milkoscan™ FT-120, and Somatic Cell Count was measured with a Fossomatic™ Minor Cell Counter.

MILKOSCAN™ FT-120



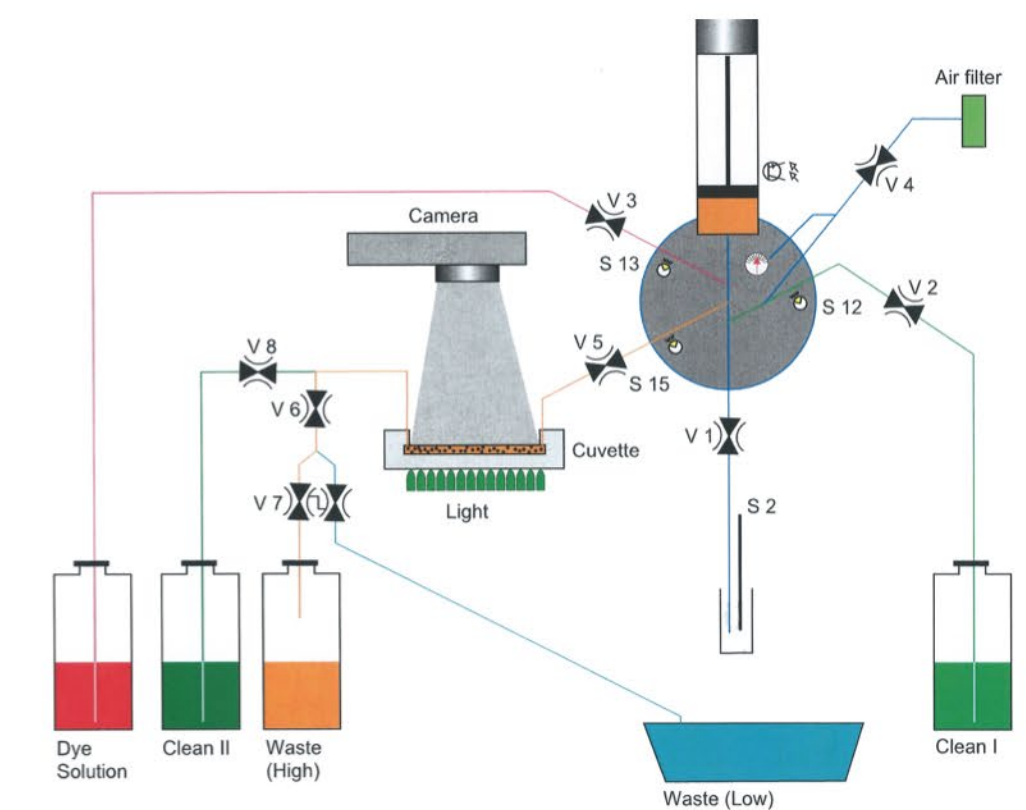
FAT

PROTEIN

LACTOSE

TOTAL
SOLIDS

FOSSOMATIC™ MINOR CELL COUNTER



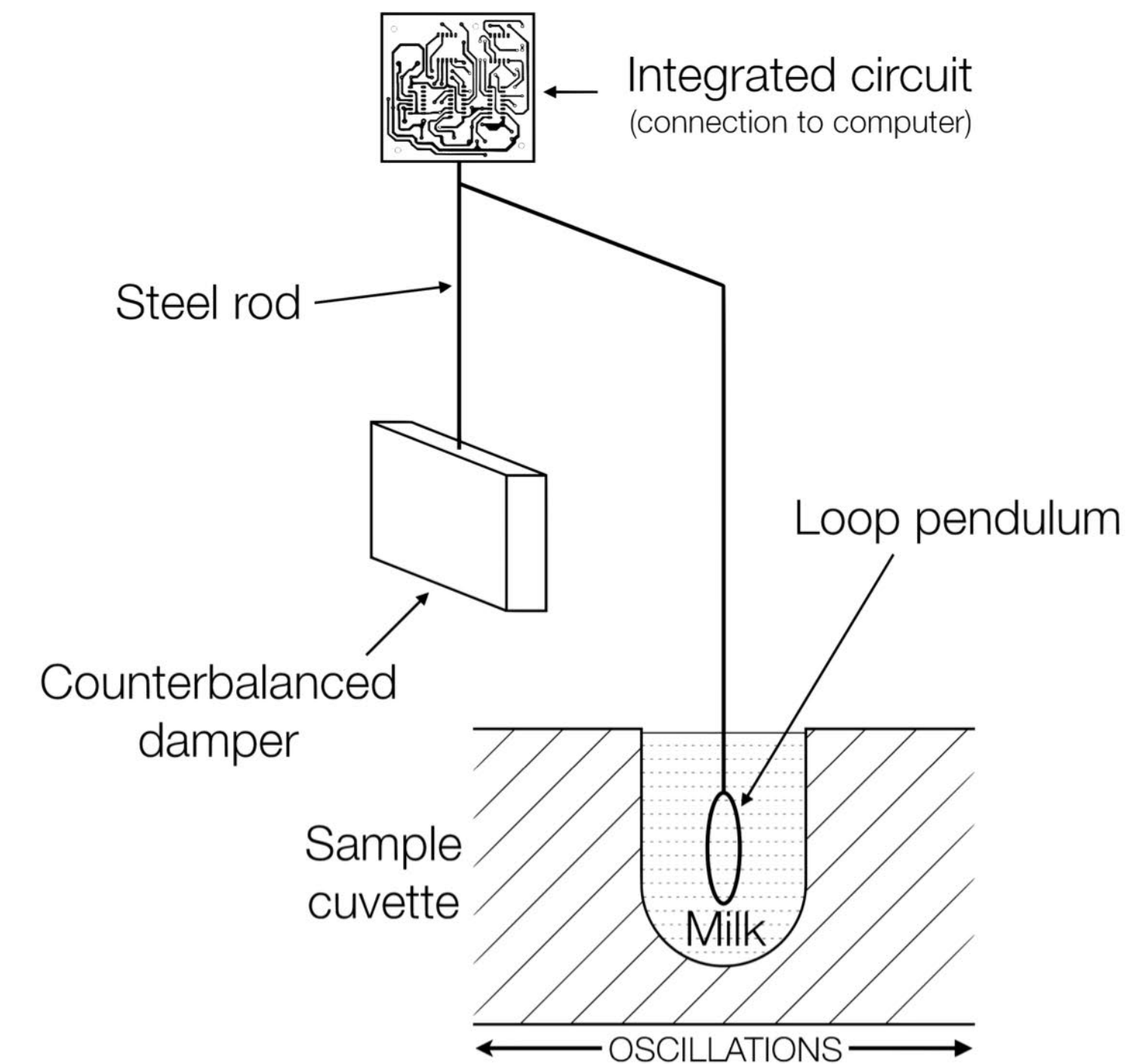
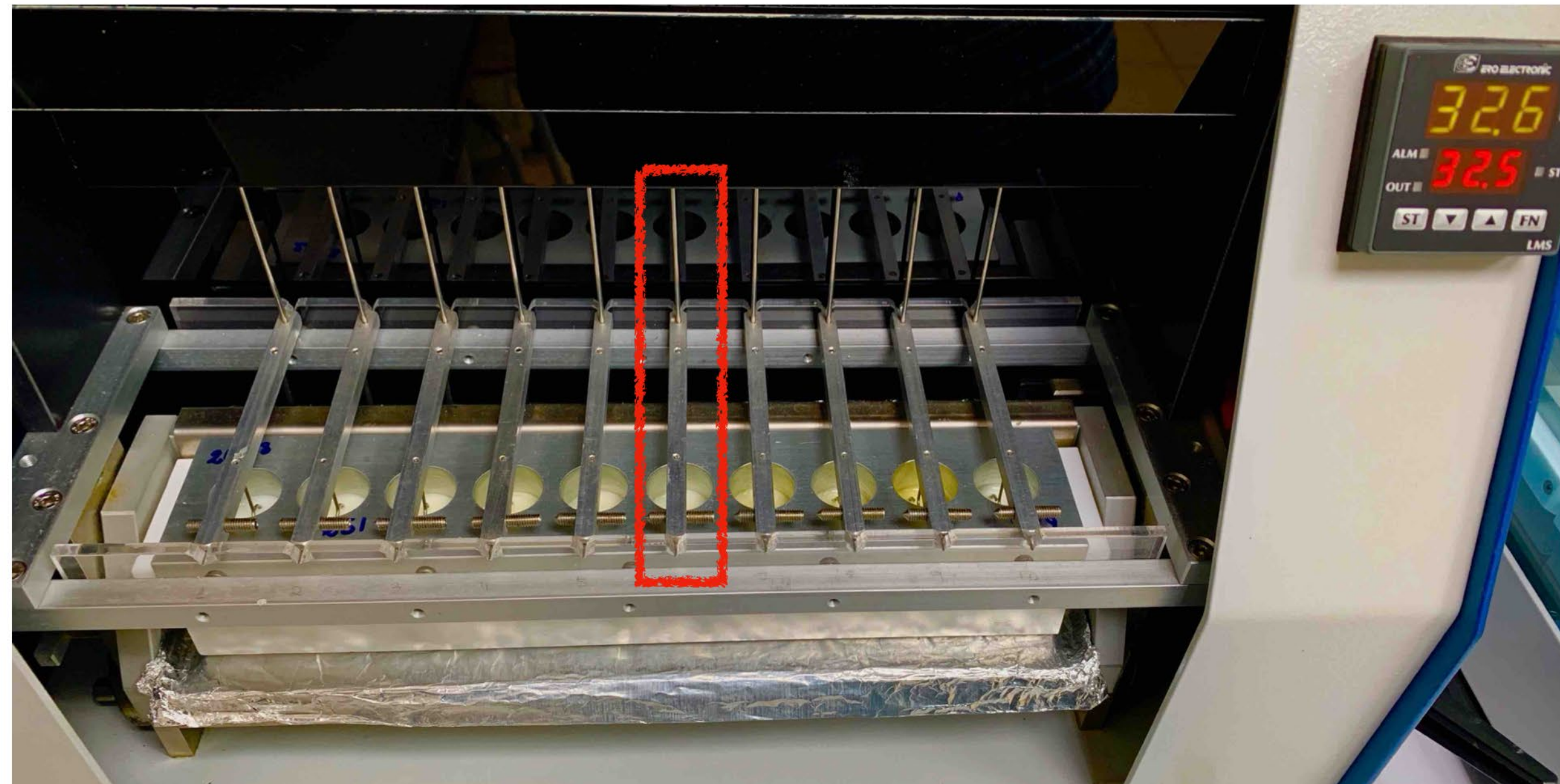
SCC normalized to
SOMATIC CELL SCORE





Rennet coagulation

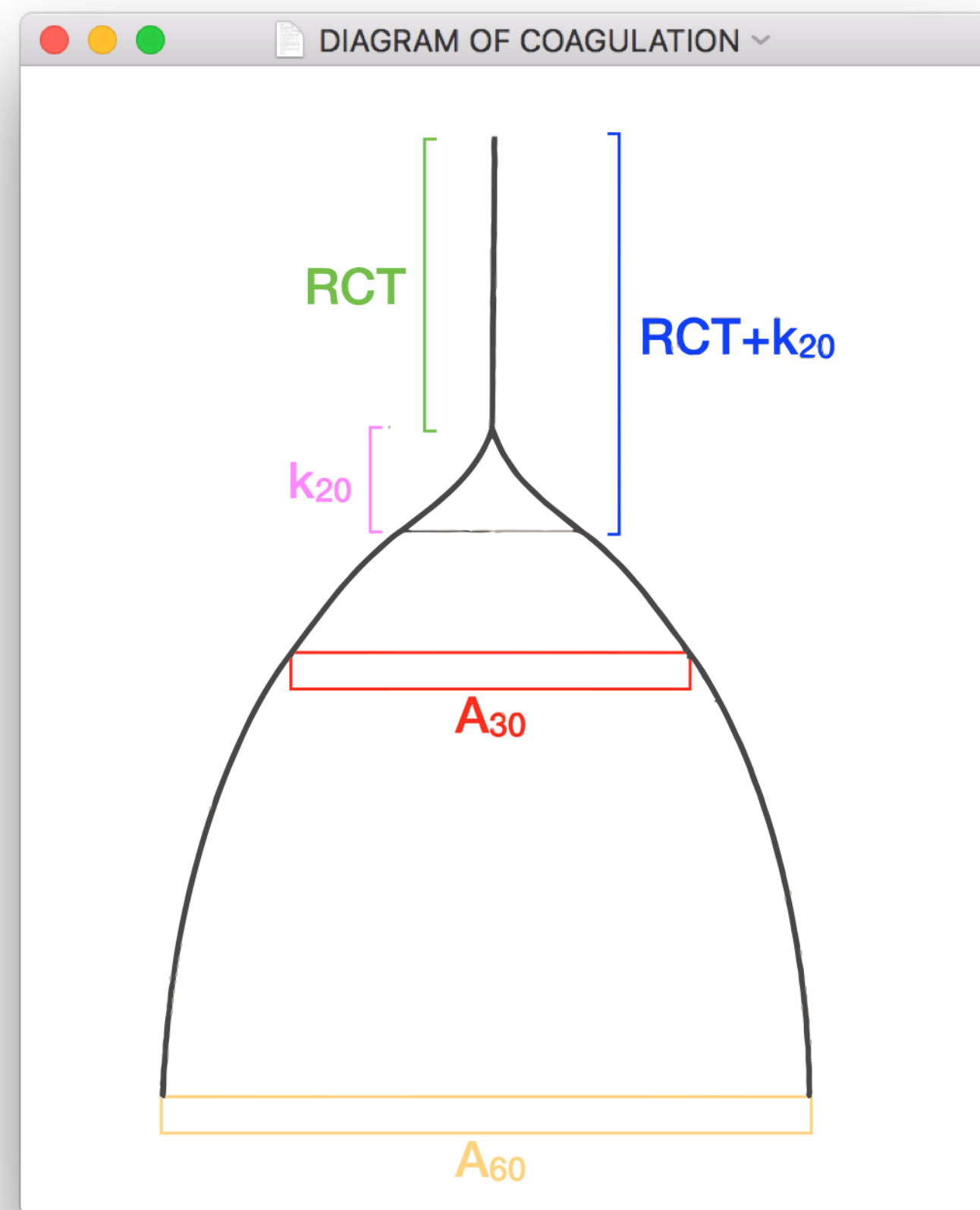
Renneting parameters were monitored using a Formagraph™ viscometer, based on the oscillatory motion of circular pendula immersed in milk during coagulation.





Rennet coagulation

Renneting parameters were monitored using a Formagraph™ viscometer, based on the oscillatory motion of circular pendula immersed in milk during coagulation.



- **RCT**: time elapsed (in minutes) until the formation of the curd.
- **k₂₀**: time (in minutes) from the start of gel development until a width of 20 mm is reached on the chart.
- **A₃₀**: width of the diagram (mm) at 30 minutes.
- **A₆₀**: width of the diagram (mm) at 60 minutes.

CURD YIELD
30 min 2800×g, 37 °C

DRY CURD YIELD
100 °C, 24 h

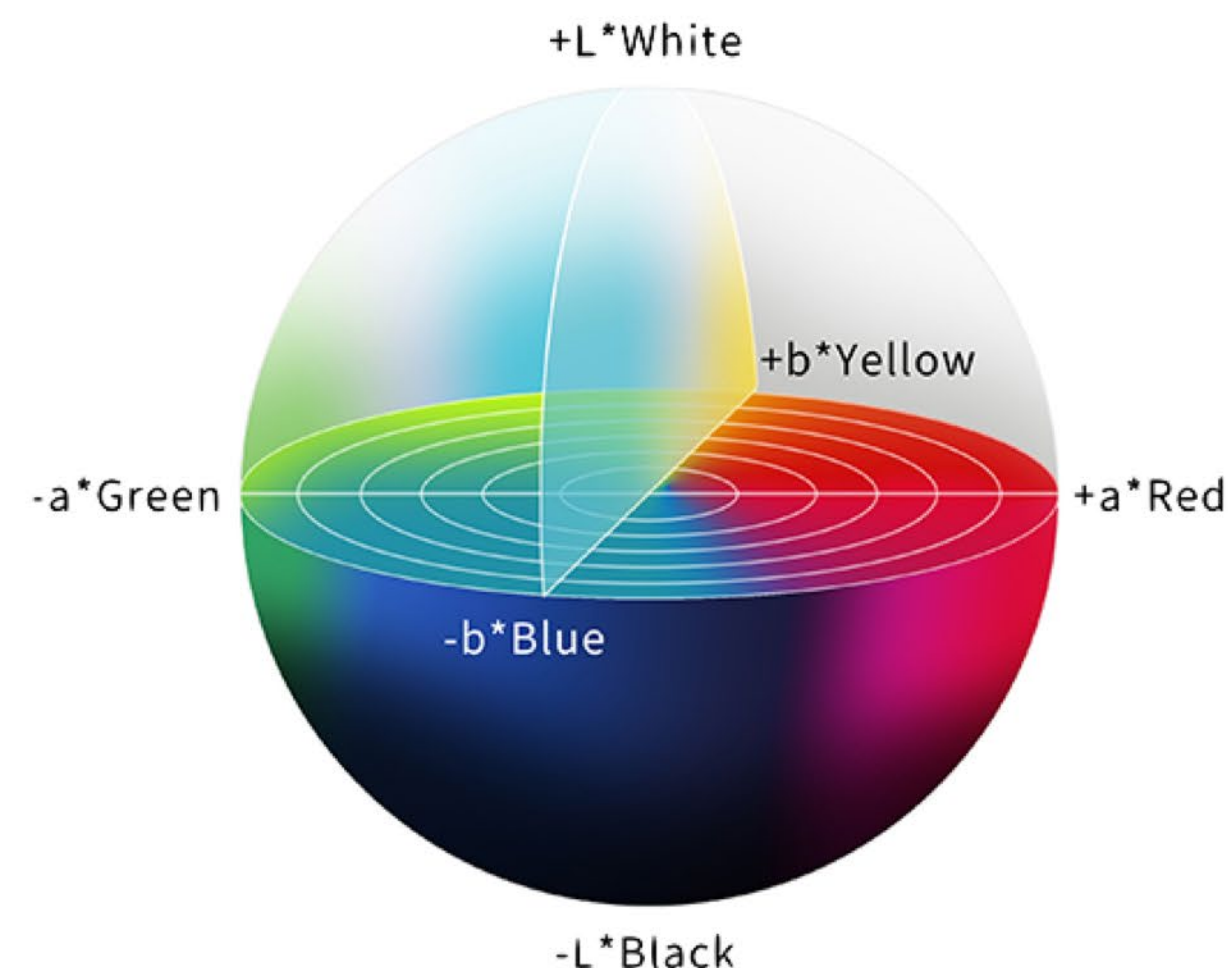




Colour in raw milk

For the measurement of colour values in milk we used a colour meter, which provided the values for brightness (L^*) and chromaticity (a^* and b^*).

- **Brightness values (L^*)** range from 0 (white) to 100 (black).
- **Chromaticity values** range from -60 to +60. Positive values for a^* are associated to red and negative values to green. Positive values for b^* are associated to yellow and negative values to blue. C^* (chroma) and h^* (hue) were then obtained using mathematical formulas (Daszkiewicz et al., 2012).





Statistical analysis

Multivariate analysis techniques were used to analyze differences and similarities in colour, milk composition, and milk coagulation.

CANONICAL DISCRIMINANT ANALYSIS

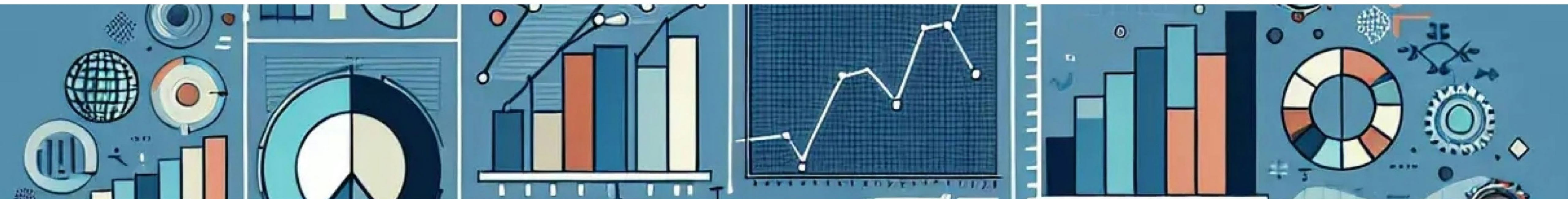
THE MOST DISCRIMINANT VARIABLES OBTAINED WERE IDENTIFIED

STEPWISE DISCRIMINANT ANALYSIS

PERFORMED ON 4 SETS OF VARIABLES: COMPOSITION VARIABLES; COAGULATION VARIABLES; COLOR VARIABLES; AND THE WHOLE SET OF VARIABLES.

DISCRIMINANT ANALYSIS

THE PREDICTIVE ABILITY OF EACH MODEL IS TESTED USING THE ABSOLUTE ASSIGNMENT OF SAMPLES TO THE PREASSIGNED GROUP



RESULTS & DISCUSSION





Canonical discriminant analysis

The table emphasises the variables selected that showed higher discriminatory ability among species

Variable	Cow	Goat	Sheep	Wilks' λ	F-Value	p-Value	R ²	CAN1*	CAN2*
FAT	2.44 ± 1.12^c	5.56 ± 1.23^b	6.54 ± 1.81^a	0.429	1518.51	<0.001	0.867	0.835	0.072
CP	4.18 ± 0.47^b	3.98 ± 0.54^c	5.59 ± 0.79^a	0.474	1264.47	<0.001	0.775	0.632	0.603
LAC	4.62 ± 0.26 ^c	4.86 ± 0.45 ^b	4.95 ± 0.36 ^a	0.850	201.44	<0.001	0.283	0.427	0.052
pH	6.70 ± 0.08 ^a	6.66 ± 0.13 ^b	6.61 ± 0.14 ^c	0.898	130.12	<0.001	0.423	-0.343	-0.111
RCT	20.48 ± 6.00	20.07 ± 7.38	19.88 ± 10.03	0.999	1.07	0.343	0.473	-0.034	-0.005
k ₂₀	9.39 ± 6.01 ^a	6.06 ± 3.95 ^b	3.53 ± 3.09 ^c	0.730	420.56	<0.001	0.518	-0.562	-0.145
A ₆₀	30.94 ± 9.44	25.09 ± 9.65 ^c	38.69 ± 10.99 ^a	0.817	255.28	<0.001	0.384	0.280	0.464
CY	16.64 ± 3.72^c	20.59 ± 4.78^b	26.76 ± 5.77^a	0.558	903.79	<0.001	0.879	0.695	0.295
DCY	35.02 ± 4.09	42.17 ± 6.08 ^a	42.37 ± 5.43 ^a	0.694	502.89	<0.001	0.536	0.611	-0.059
L*	78.27 ± 2.87^c	83.47 ± 1.28^b	83.61 ± 2.21^a	0.483	1221.35	<0.001	0.736	0.794	-0.077
a*	-4.13 ± 1.34	-1.12 ± 0.51^a	-2.46 ± 0.71^b	0.483	1217.35	<0.001	0.707	0.679	-0.505
b*	2.52 ± 3.17^c	3.29 ± 1.22^b	4.49 ± 1.93^a	0.870	169.63	<0.001	0.572	0.377	0.159

* Correlation of each variable with the canonical function





Canonical discriminant analysis

The table emphasises the variables selected that showed higher discriminatory ability among species

Variable	Cow	Goat	Sheep	Wilks' λ	F-Value	p-Value	R ²	CAN1*	CAN2*
<i>Composition</i>									
FAT	2.44 ± 1.12^c	5.56 ± 1.23^b	6.54 ± 1.81^a	0.429	1518.51	<0.001	0.491	0.855	-0.266
CP	4.18 ± 0.47^b	3.98 ± 0.54^c	5.59 ± 0.79^a	0.474	1264.47	<0.001	0.482	0.770	0.512
LAC	4.62 ± 0.26 ^c	4.86 ± 0.45 ^b	4.95 ± 0.36 ^a	0.850	201.44	<0.001	0.021	0.441	-0.116
pH	6.70 ± 0.08 ^a	6.66 ± 0.13 ^b	6.61 ± 0.14 ^c	0.898	130.12	<0.001	0.077	-0.370	0.004
<i>Coagulation</i>									
RCT	20.48 ± 6.00	20.07 ± 7.38	19.88 ± 10.03	0.999	1.07	0.343	0.286	-0.038	0.004
k ₂₀	9.39 ± 6.01 ^a	6.06 ± 3.95 ^b	3.53 ± 3.09 ^c	0.730	420.56	<0.001	0.467	-0.647	-0.048
A ₆₀	30.94 ± 9.44	25.09 ± 9.65 ^c	38.69 ± 10.99 ^a	0.817	255.28	<0.001	0.223	0.385	0.797
CY	16.64 ± 3.72^c	20.59 ± 4.78^b	26.76 ± 5.77^a	0.558	903.79	<0.001	0.307	0.819	0.289
DCY	35.02 ± 4.09	42.17 ± 6.08 ^a	42.37 ± 5.43 ^a	0.694	502.89	<0.001	0.132	0.668	-0.374
<i>Colour</i>									
L*	78.27 ± 2.87^c	83.47 ± 1.28^b	83.61 ± 2.21^a	0.483	1221.35	<0.001	0.605	0.821	0.564
a*	-4.13 ± 1.34	-1.12 ± 0.51^a	-2.46 ± 0.71^b	0.483	1217.35	<0.001	0.597	0.903	-0.047
b*	2.52 ± 3.17 ^c	3.29 ± 1.22 ^b	4.49 ± 1.93 ^a	0.870	169.63	<0.001	0.409	0.300	0.504

* Correlation of each variable with the canonical function





Stepwise discriminant analysis

Discriminant canonical models for composition, coagulation, colour, and the whole set of variables.



Model	No. Variables in model	Wilks' λ	F-Value	p-Value
Whole set	12	0.083	468.30	<0.001
Composition	4	0.172	802.49	<0.001
Coagulation	5	0.308	364.78	<0.001
Colour	3	0.261	725.89	<0.001

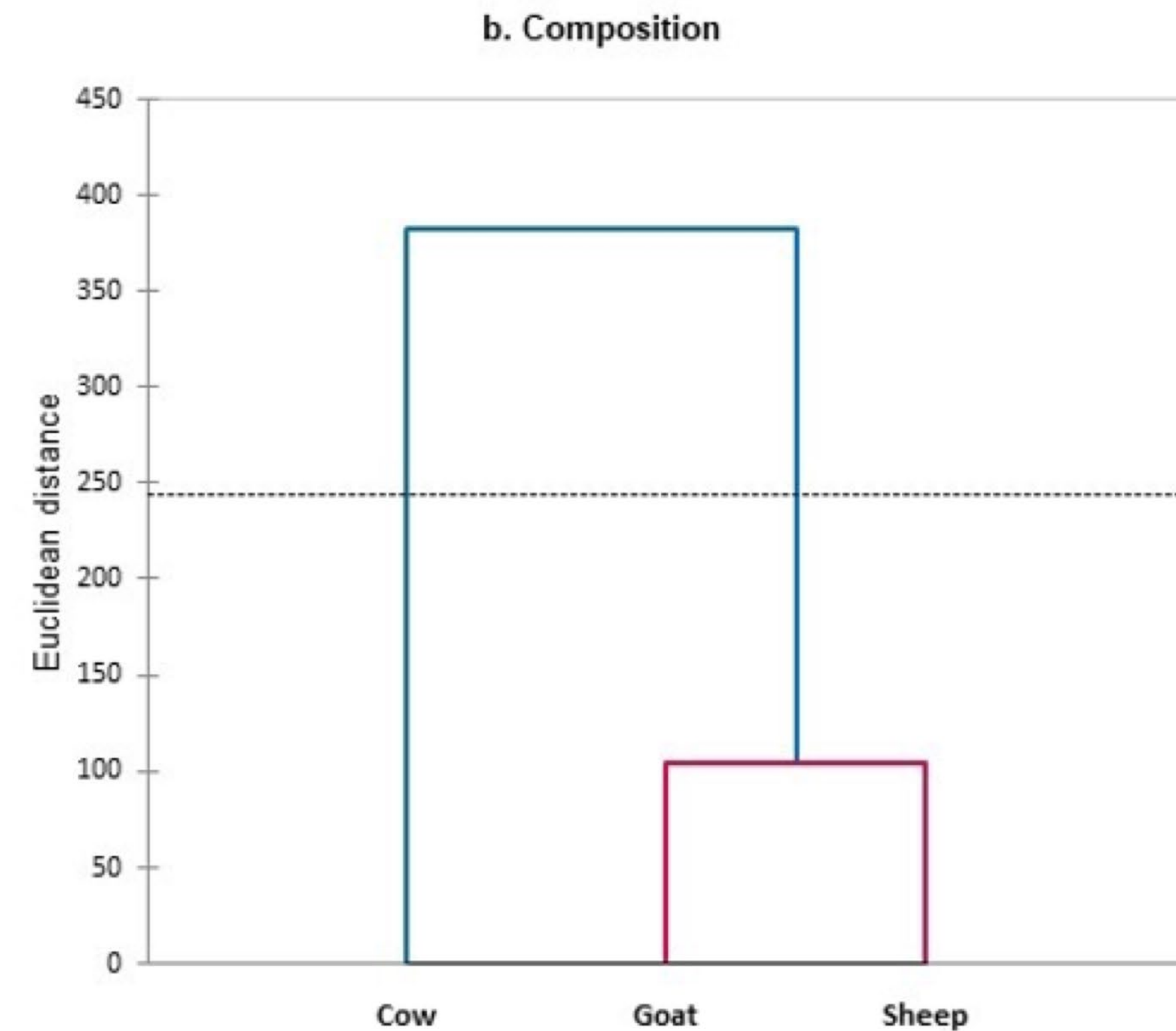
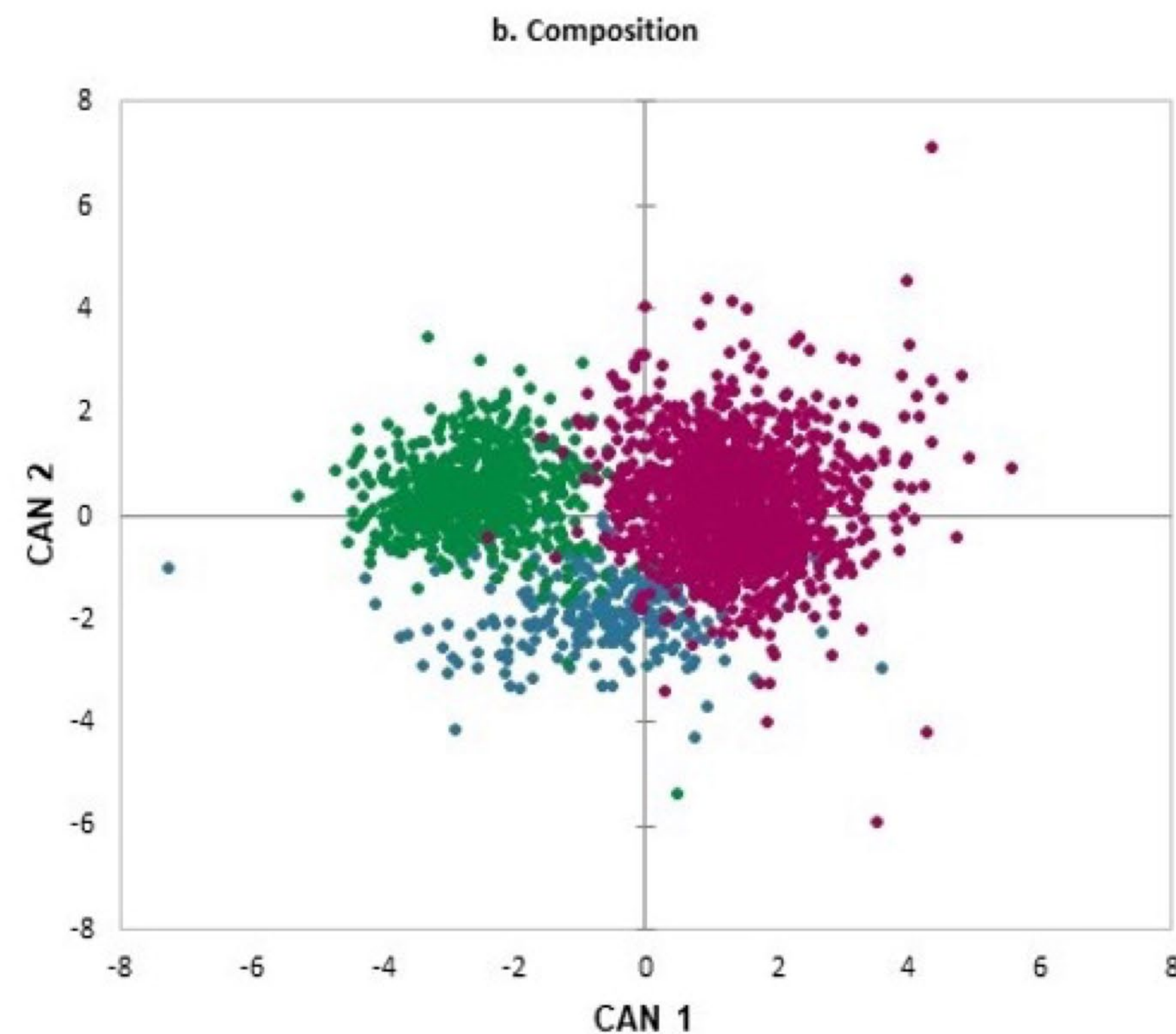
In all cases, the extracted canonical functions significantly discriminated among the three species. The F-statistics revealed a higher discriminating ability for the variables related to milk composition and colour.





Canonical discriminant analysis (composition)

Graphical representation of the results obtained by canonical discriminant analysis for milk composition variables, defined by the axes of the first 2 canonical variables.



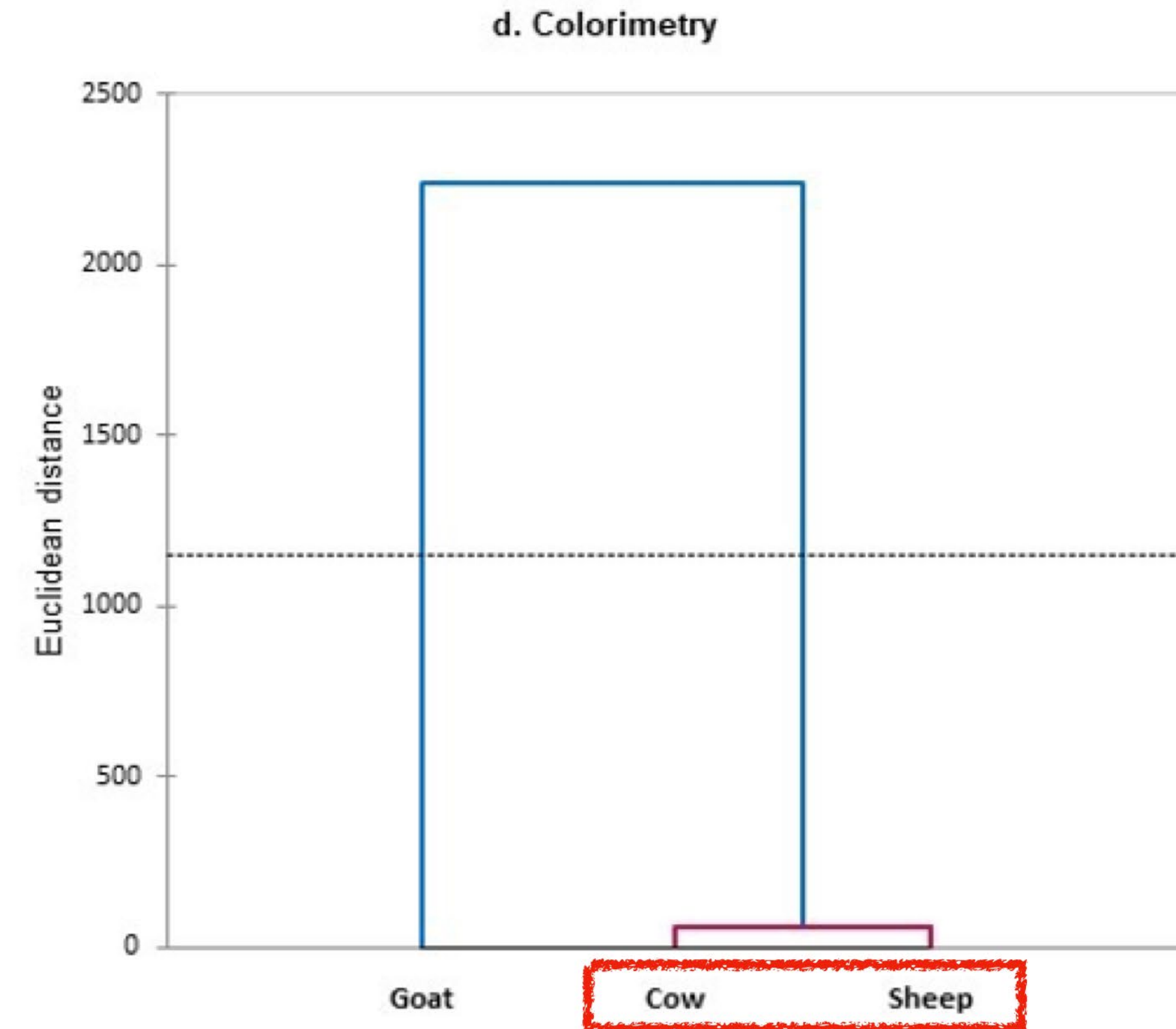
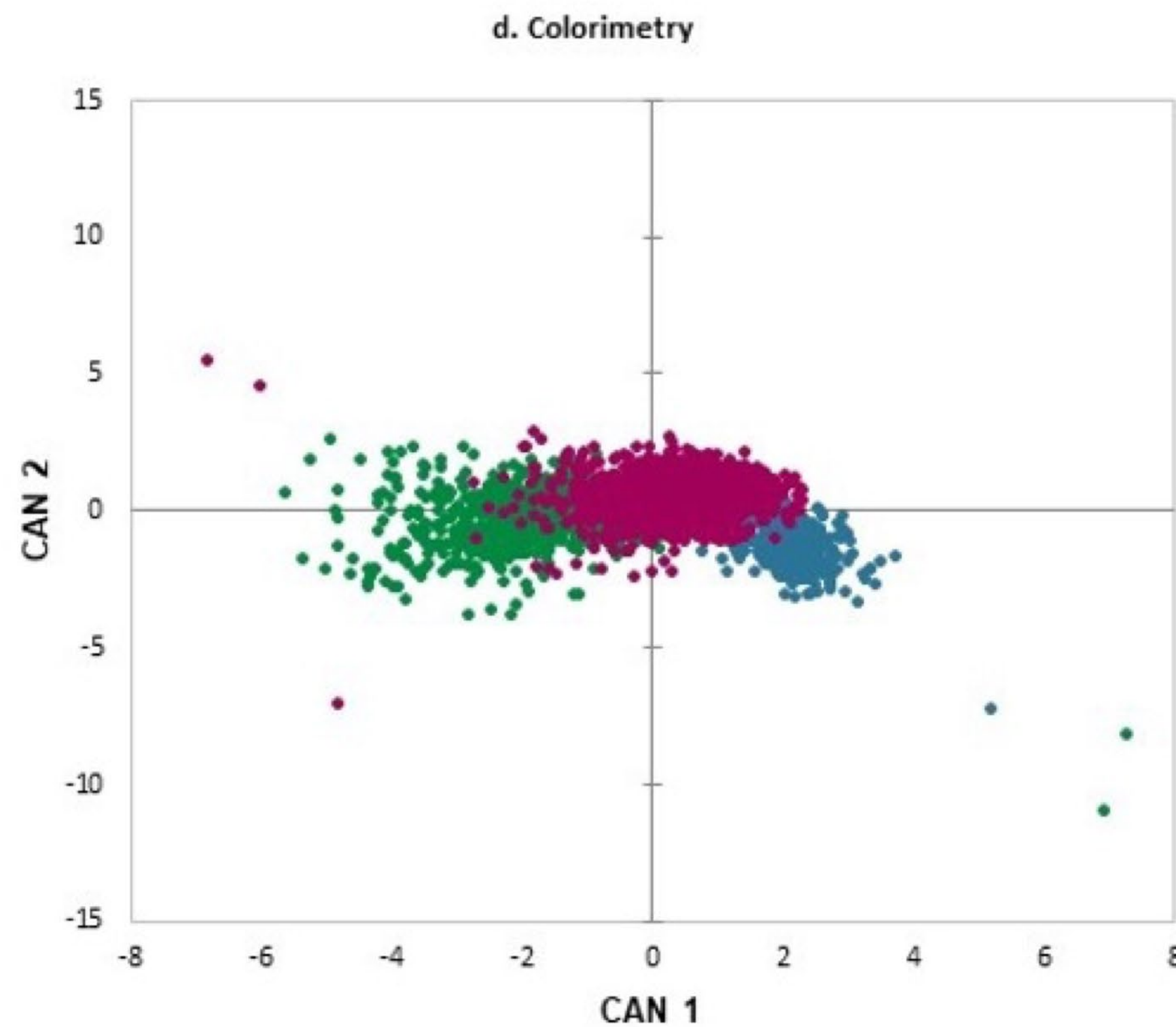
● COW ● GOAT ● SHEEP





Canonical discriminant analysis (colour)

Graphical representation of the results obtained by canonical discriminant analysis for the colour variables, defined by the axes of the first 2 canonical variables.



● COW ● GOAT ● SHEEP





Discriminant analysis

Assignment percentages in the predefined groups and classification errors.

Model	Goat	Cow	Sheep	
<i>All variables</i>				
Goat	94.72	1.51	3.77	97.8%
Cow	0.15	98.66	1.19	
Sheep	1.49	0.59	97.92	
Error level	0.08	0.02	0.01	
<i>Composition</i>				
Goat	87.92	3.02	9.06	91.9%
Cow	3.86	94.95	1.19	
Sheep	8.25	0.59	91.16	
Error level	0.37	0.02	0.01	
<i>Coagulation</i>				
Goat	76.98	6.04	16.98	82.2%
Cow	10.57	86.90	2.53	
Sheep	17.25	1.78	80.97	
Error level	0.60	0.06	0.05	
<i>Colour</i>				
Goat	96.23	0.38	3.40	88.8%
Cow	0.00	85.88	14.12	
Sheep	3.64	7.50	88.86	
Error level	0.16	0.15	0.08	





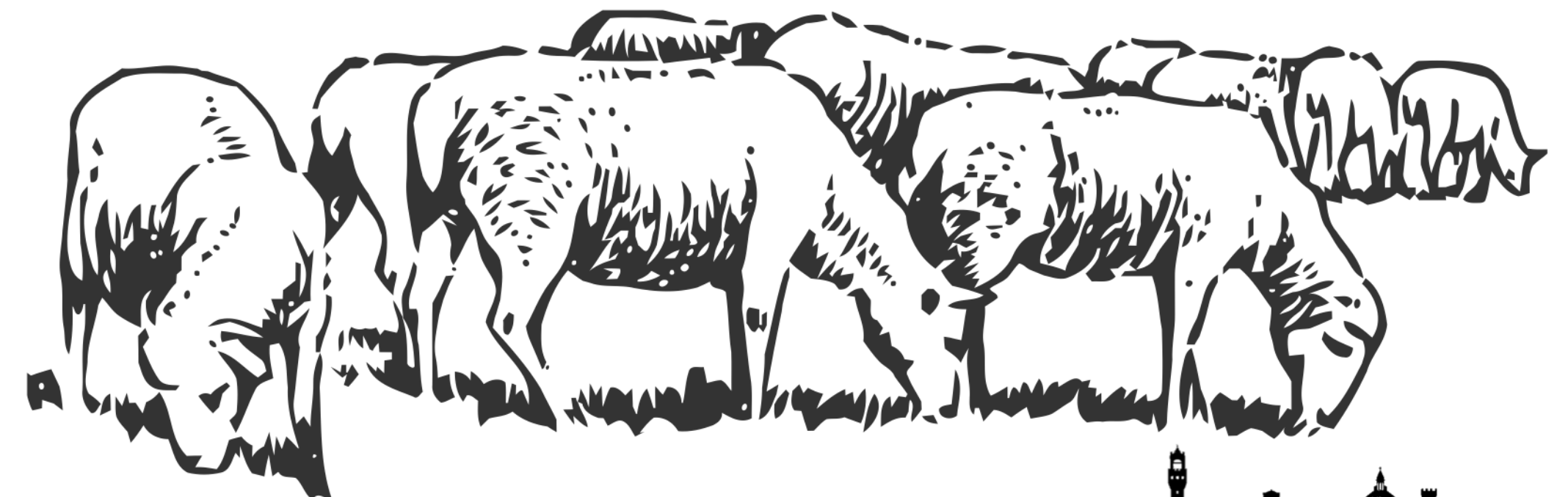
Canonical correlation analysis

Correlation coefficients between variables and canonical variables included in the canonical correlation analysis.

Variable	CANONICAL COMPONENT					
	COW		GOAT		SHEEP	
	F ₁	F ₂	F ₁	F ₂	F ₁	F ₂
<i>Composition—coagulation models</i>						
pH	-0.324	0.823	-0.150	-0.104	0.217	0.890
FAT	-0.829	-0.359	0.809	0.569	-0.954	-0.127
CP	-0.800	-0.112	0.825	-0.519	-0.864	0.184
LAC	-0.119	-0.455	-0.076	-0.126	-0.087	-0.301
RCT	-0.449	0.661	-0.078	-0.322	-0.117	0.898
k ₂₀	0.151	0.702	-0.397	0.409	0.499	0.478
A ₆₀	-0.633	0.251	0.234	-0.563	-0.627	0.161
CY	-0.952	-0.157	0.968	0.010	-0.948	0.103
DCY	-0.561	-0.231	0.130	-0.346	-0.555	-0.446

<i>Composition—colorimetric models</i>						
pH	-0.091	0.677	-0.199	0.272	-0.099	-0.774
FAT	-0.865	0.260	0.943	0.257	0.899	0.165
CP	-0.796	-0.154	0.618	-0.755	0.823	-0.228
LAC	-0.337	-0.671	-0.360	-0.239	-0.793	0.350
L*	-0.920	-0.166	0.653	-0.136	0.472	0.840
a*	-0.055	0.869	0.746	0.516	0.748	0.371
b*	-0.791	0.490	0.985	-0.040	0.951	-0.186

Variable	CANONICAL COMPONENT					
	COW		GOAT		SHEEP	
	F ₁	F ₂	F ₁	F ₂	F ₁	F ₂
<i>Coagulation—colorimetric models</i>						
RCT	-0.255	0.630	0.028	-0.820	0.191	-0.630
k ₂₀	0.227	0.735	0.041	-0.693	-0.119	-0.712
A ₆₀	-0.493	-0.426	0.138	0.793	0.324	-0.401
CY	-0.966	0.096	-0.852	0.153	0.840	-0.080
DCY	0.314	-0.501	-0.361	0.026	0.265	0.271
L*	-0.889	-0.104	-0.673	0.590	0.526	0.836
a*	-0.187	0.982	-0.821	-0.120	0.735	0.272
b*	-0.851	0.150	-0.983	0.185	0.953	-0.212





Improvements & **further actions**

This field of research should be continued considering several aspects.

LIMITATIONS OF THE STUDY

SCC

SIMPLE MODELS

OTHER MILK COMPOUNDS



Article

Exploring Interrelationships between Colour, Composition, and Coagulation Traits of Milk from Cows, Goats, and Sheep

Ana Garzón ¹, José M. Perea ¹, Elena Angón ¹, Eoin G. Ryan ², Orla M. Keane ³
and Javier Caballero-Villalobos ^{1,*}

FURTHER ACTIONS

- 1 Application of the developed methodology on milk from individual animals from different breeds.
- 2 Development and assessment of more complex models that include other variables in order to fine-tune the predictive ability of colour values in milk processing into cheese.
- 3 Exploration of colour measurements as a potential tool to assess the presence of bacteria in milk.



CONCLUSIONS





Conclusions and proposed actions

- 1 Colour can be used to effectively discriminate milk at a species level. It can also help predict if milk will coagulate under laboratory conditions.
- 2 While milk quality traits show some common patterns of variations, strong species-specific relationships emerge, highlighting the connection between milk colour and curd yield.
- 3 These findings may allow to model these relationships to estimate milk technological performance, potentially leading to an optimization of the coagulation process beneficial for the cheesemaking industry.



THANK YOU

@jcaballerovet

