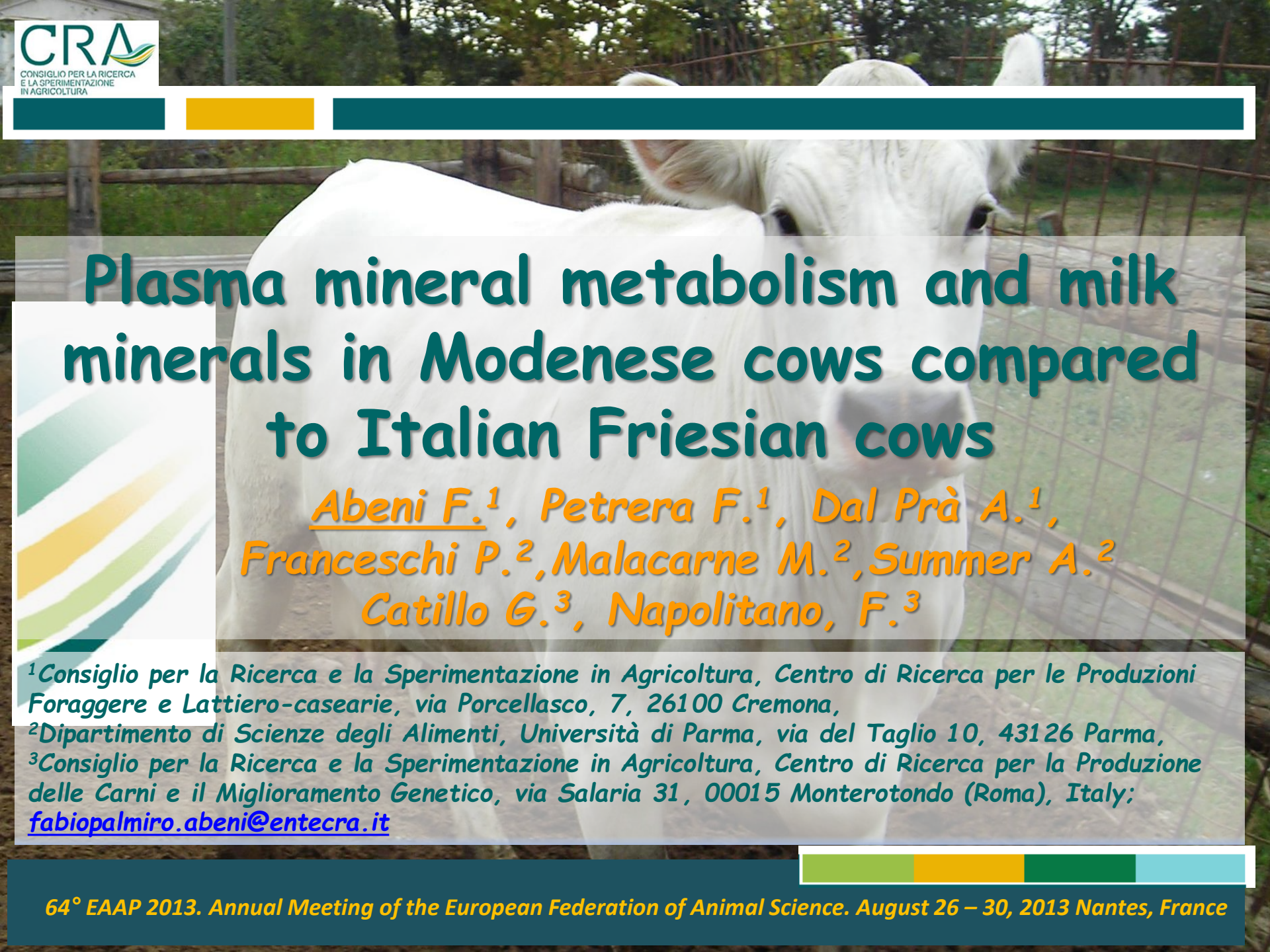

Plasma mineral metabolism and milk minerals in Modenese cows compared to Italian Friesian cows

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Introduction

Agro-biodiversity

Role of autochthonous breeds:

- essential to culture, interaction with the environment, and local economy in Europe
- possess **valuable traits** (resistance to diseases; adaptation to severe conditions and poor quality feeds; specific qualitative features of their product)

Safeguard strategies: are currently under way to repopulate the rural areas with the local breeds to preserve biodiversity in livestock (Gandini et al., 2010)

Introduction

Modenese or Bianca Val Padana breed:

- native from Emilia Romagna region (Italy), product of several crossbreeding between indigenous breeds; **dual purpose** (milk and meat)
- **medium frame size** (breed standards adult female cows: 125-140 cm height; 650 kg average BW), white coated
- **medium-low milk yield** (4700 kg in 305 d, with 3.4% of protein and 3.3% of fat)
- population size ~ **650 cows**, included in the **endangered-maintained breeds** list (**FAO, 2000**), 60% in herds together with Italian Friesian cows
- good **functional traits** (longevity, fertility, robustness, adaptability to climate conditions)
- **advantages in milk components** **(Duclos and Hiemstra, 2010)**

Introduction

Minerals in milk

- Milk phosphate
  - (a) free inorganic P_i(HPO_4^{2-} and $H_2PO_4^-$)
  - (b) esterified phosphate (including casein phosphate)
  - (c) phosphate associated with Ca in the casein micelles *
- P transported from circulation to mammary cells as free inorganic P_i
- Milk Ca
  - (a) free ionized Ca
  - (b) Ca associated with P_i in casein micelles *
  - (c) Ca associated with citrate and phosphate
- free ionized concentration of Ca in milk (~ 3 mM) is greater than that of plasma suggesting that active transport must be taking place (Shennan, 1998)
- h^2 was 0.57 and 0.62 for Ca and P respectively (van Hulzen et al., 2009)

Aim of the study

- to compare Modenese and Italian Friesian cows for their mineral metabolism and the possible relationships with macro-minerals concentrations in milk

- to characterize the milk mineral fractions for a possible valorisation of the Modenese production in Italy

Material & Method: Animals and feeding

11 Modenese (M) and 14 Italian Friesian (IF) pluriparous cows (see Table 1) raised and managed together in the same herd (housed in a free-stall barn), fed the same total mixed ration, once daily

fresh drinking water available

milking: twice a day (at 0500 and 1700 h) in a milking parlour

Characteristics of the cows in the present study

	MO	IF
Lactation number	3.00 ± 1.41	3.07 ± 1.73
Age at calving (mo)	49.1 ± 14.9	57.4 ± 24.5
Previous effective milk yield (kg)	5265 ± 1148 (309.1 ± 43 days)	9556 ± 1930 (373 ± 62 days)
Previous conventional milk yield (kg)	5119 ± 1088	8376 ± 1388

Material & Method

BCS

On a 5-point scale (1=emaciated, 5= obese, scored in 0.25 point intervals) (Ferguson et al., 1994)

BLOOD

From jugular vein, using evacuated tubes (10 ml, Li-heparin)

At 8 and 21 wk of lactation, in the morning, before feed distribution

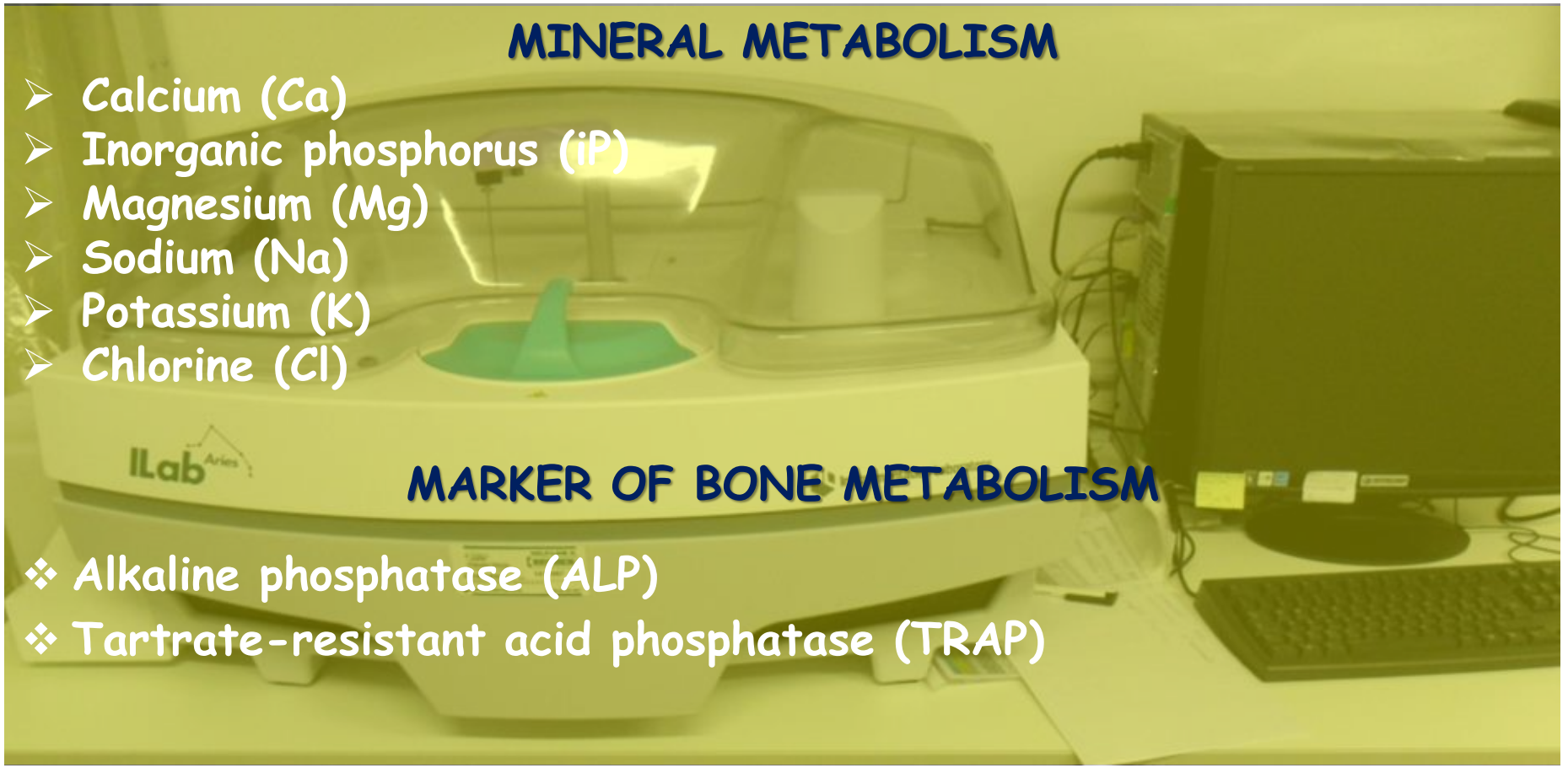
Centrifuged at 3000 g x for 20' at 4°C ⇒ plasma stored at -20°C

MILK

48 samples (22 from MO and 26 from IF cows) collected at 8 and 21 wk of lactation during the morning milking

Milk yield was individually recorded

Plasma metabolites analysis



MINERAL METABOLISM

- Calcium (Ca)
- Inorganic phosphorus (iP)
- Magnesium (Mg)
- Sodium (Na)
- Potassium (K)
- Chlorine (Cl)

MARKER OF BONE METABOLISM

- Alkaline phosphatase (ALP)
- Tartrate-resistant acid phosphatase (TRAP)

Analyzed at 37°C by an automated clinical analyzer (ILAB Aries, Instrumentation Laboratory, Lexington, MA) using commercial kits (Instrumentation Laboratory, Lexington, MA).

Milk analysis

Fat, lactose, protein and casein content in milk:

infrared analysis (Milko-Scan FT 6000, Foss Electric, DK-3400 Hillerød, Denmark).

Milk minerals:

- ✓ **Total Ca in milk and soluble Ca in milk ultrafiltrate (cut off 30,000 D):**

  by atomic absorption spectroscopy (AAS).

- ✓ **Total P in milk, soluble P in milk ultrafiltrate (cut off 30,000 D) and total acid-soluble P in milk (after treatment with TCA 120 g/l) :**

  by colorimetric method (Allen, 1940).

- ✓ **Distribution of Ca and P fractions:**

  calculated according to White and Davies (1958).

Rennet-coagulation parameters:

measured at 35 °C (McMahon and Brown, 1982) using a Formagraph (Foss Electric, DK-3400 Hillerød, Denmark)

pH: with a potentiometer

Titratable acidity: by titration with 0.25 M-NaOH (Soxhlet-Henkel method)

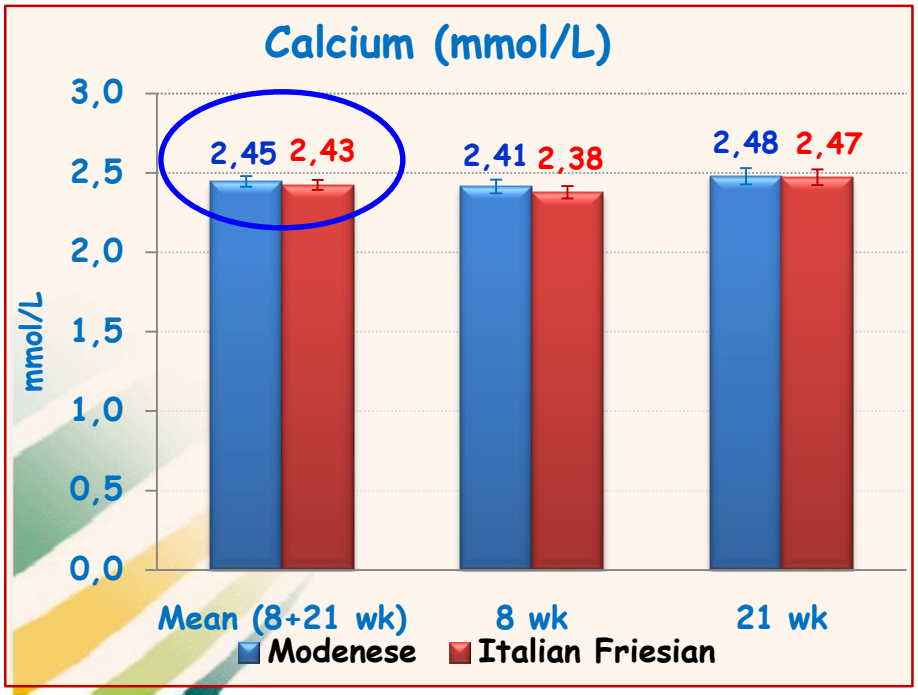
Statistical analysis

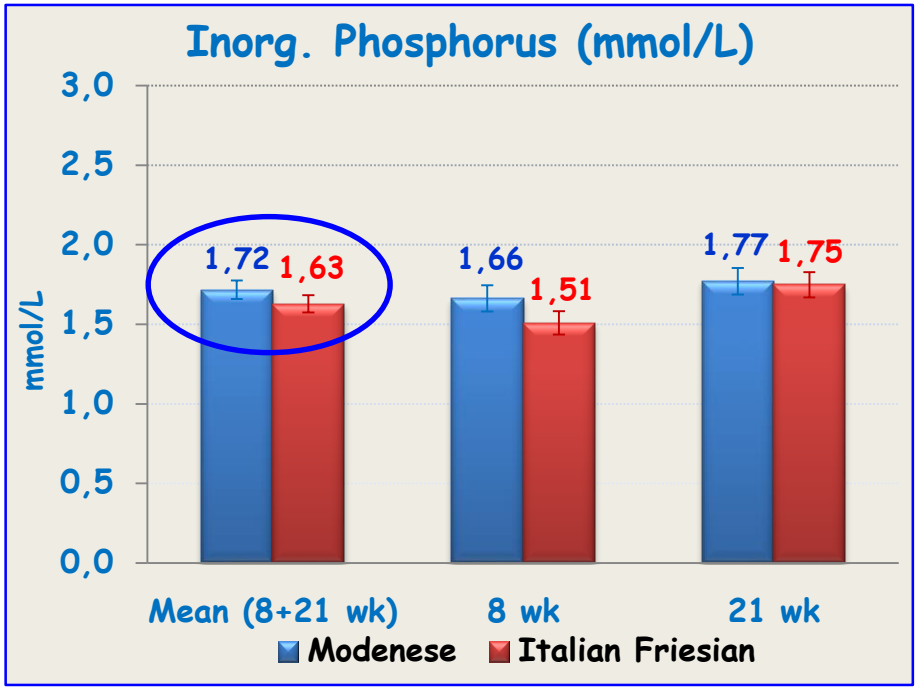
- **NORMAL DISTRIBUTION TEST:** PROC UNIVARIATE (**SAS, 2009**) with the Shapiro-Wilk test
  - The variables that did not fit the normal distribution were re-tested after log-transformation to match the assumption for a parametric analysis; their results are presented in the original scale after re-transformation

- BCS, blood, and milk data were analysed as repeated measures by a mixed model, with breed (**B**), week from calving (**T**), and their interaction (**B** × **T**) as main factors, with cow within breed considered as random
  - COVARIANCE STRUCTURE (according to the AIC) the one which best fitted the data among **SIM, CS, ANTE(1), AR(1), UN**

- Means ± s.e. (c.i. for re-transformed data); **significant** = $P < 0.05$; **trend**:= $0.05 > P < 0.10$

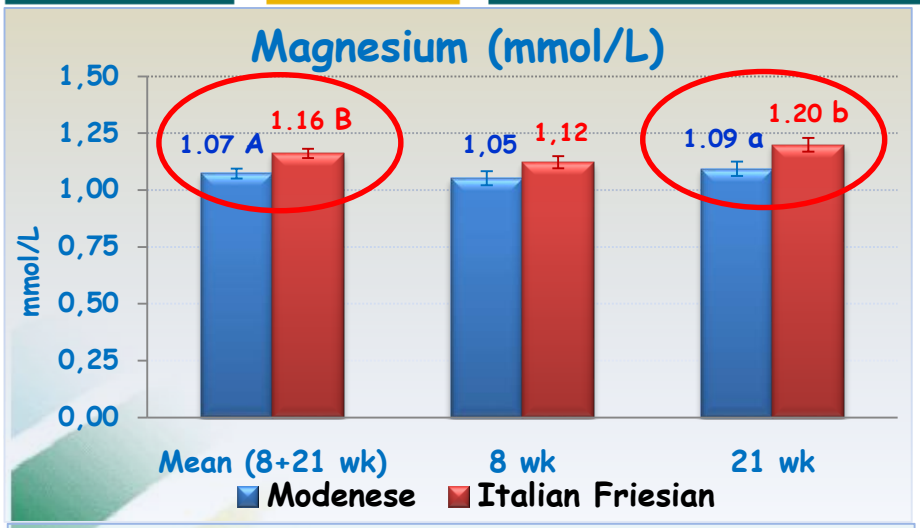
Results: BLOOD

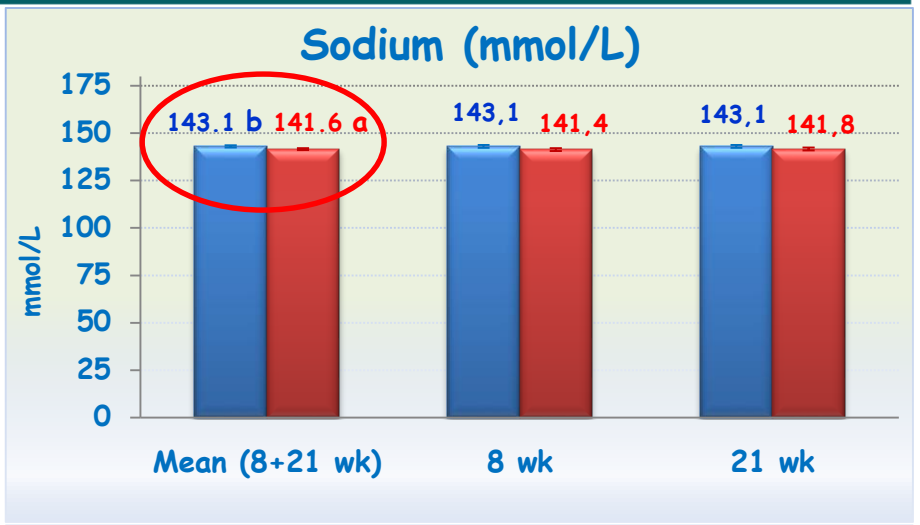


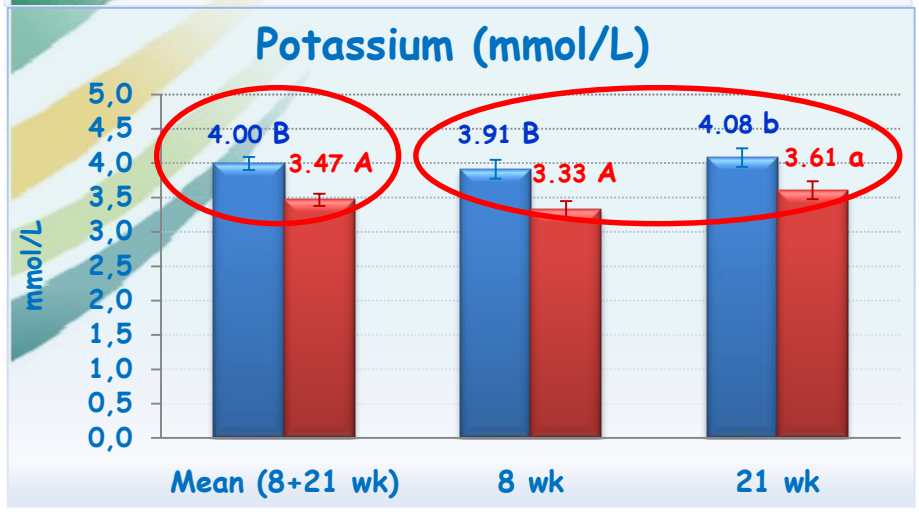


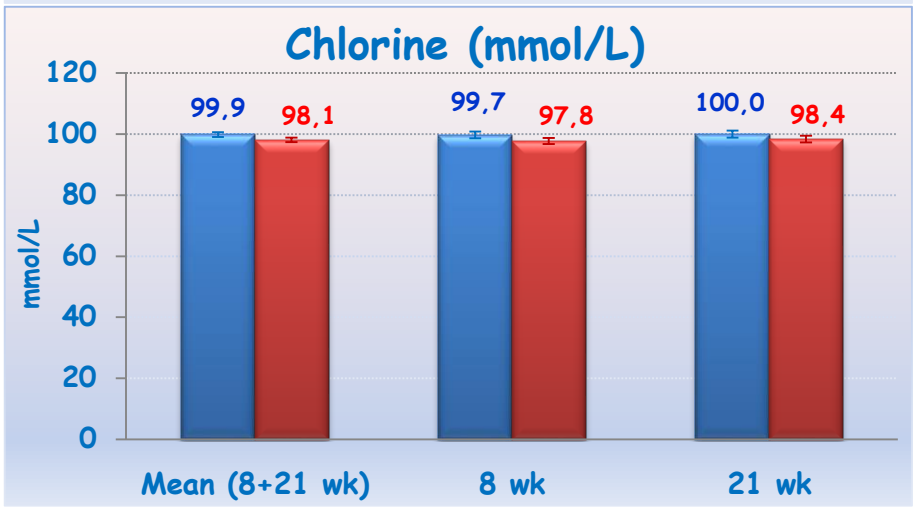
Correlations			
Modenese		Italian Friesian	
Plasma	iP	Plasma	iP
Ca	0.48 0.02	Ca	- -

Results: BLOOD



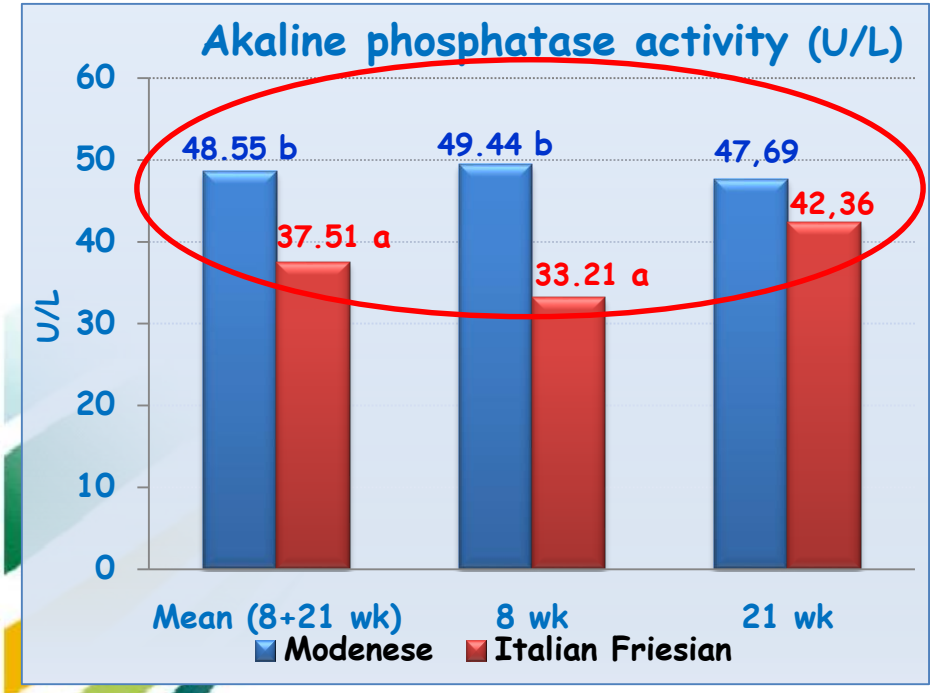




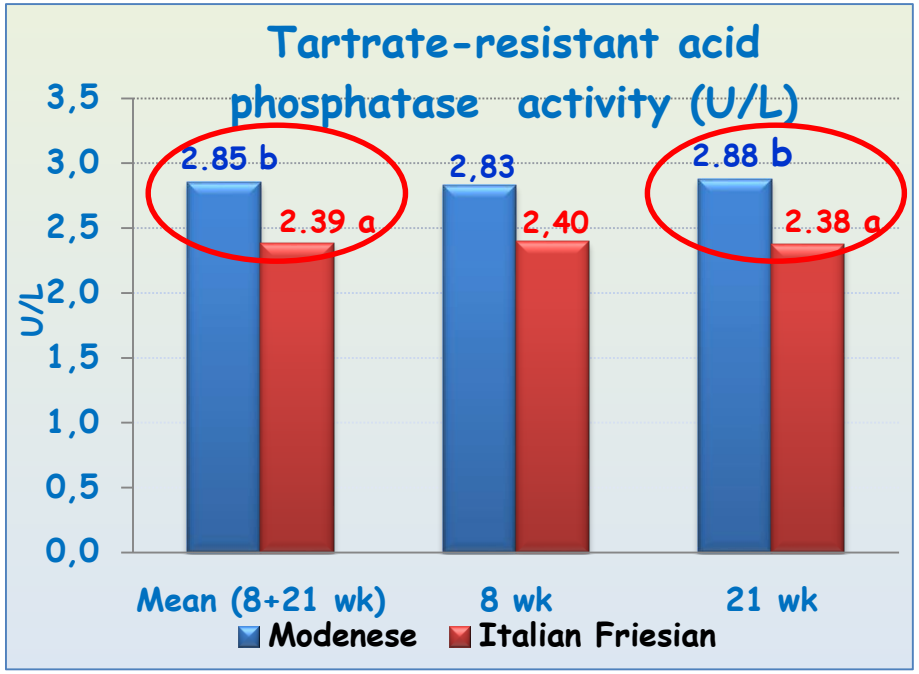


Letters indicate significant differences between MO and IF cows: (a, b: P <0.05; A, B: P<0.01).

Results: BLOOD



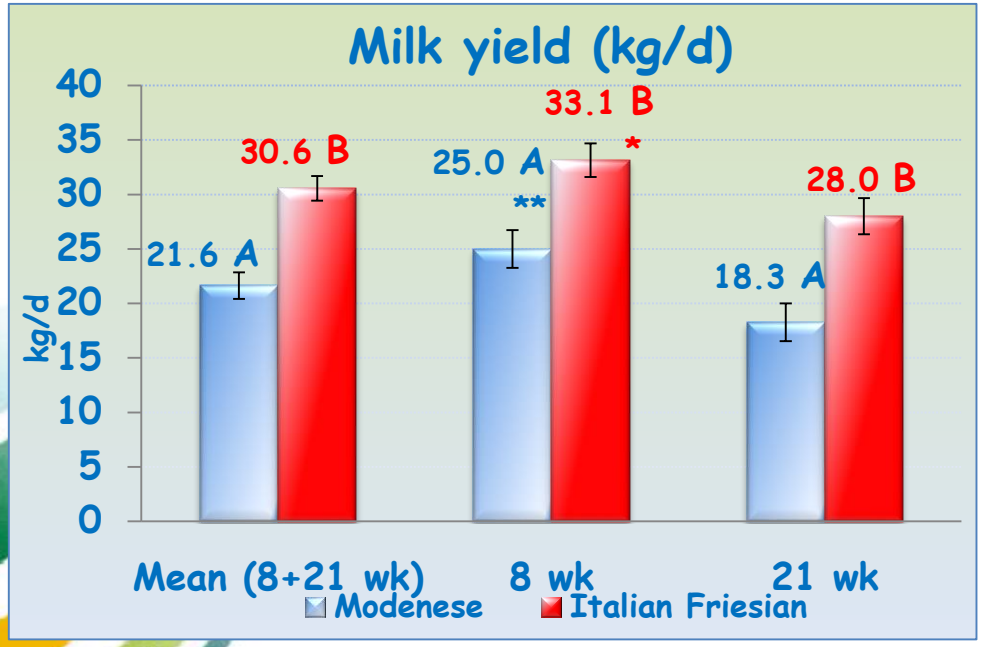
95% Confidence interval (C.I.)				
ALP activity				
Time	Modenese		Italian Friesian	
Mean (8+21 wk)	41.11	57.36	32.19	43.70
8 wk	38.38	63.68	26.53	41.57
21 wk	38.40	59.24	34.42	52.14
TRAP activity				
Mean (8+21 wk)	2.56	3.19	2.16	2.64
8 wk	2.38	3.37	2.05	2.80
21 wk	2.52	3.29	2.09	2.70



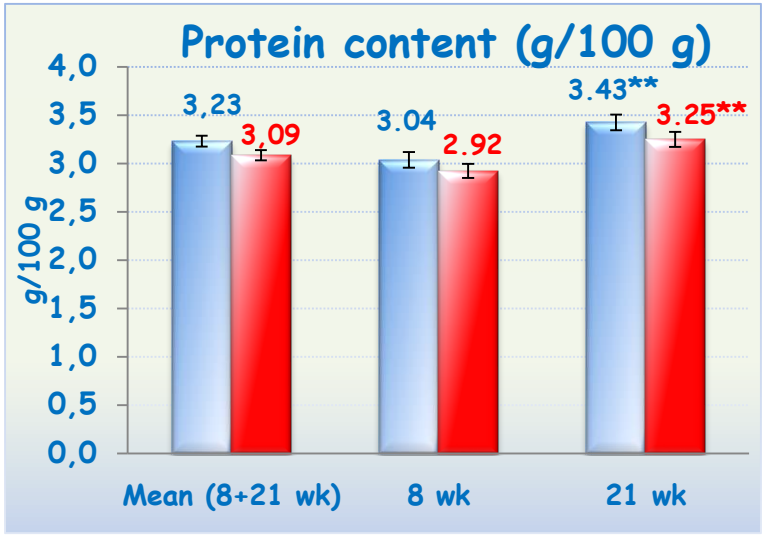
Correlations					
Modenese			Italian Friesian		
Plasma	ALP	TRAP	Plasma	ALP	TRAP
Ca	- -	0.34 0.12	Ca	0.56 0.003	- -
iP	- -	-0.31 0.16	iP	- -	0.31 0.12

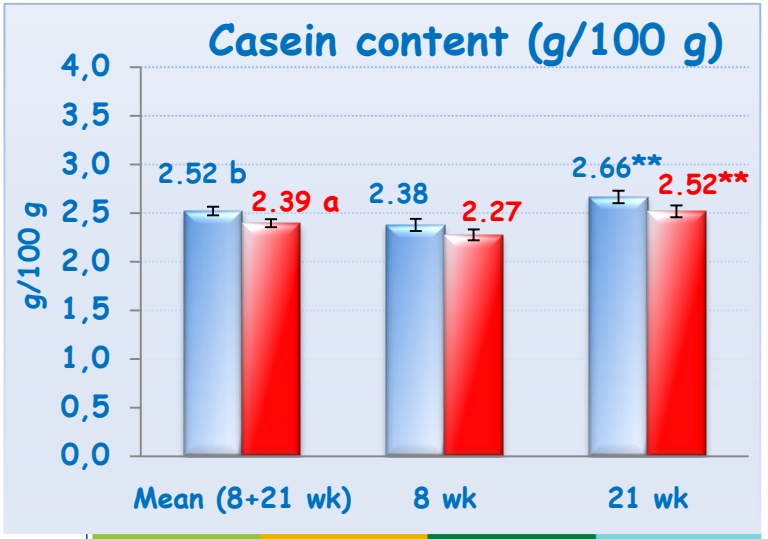
Letters indicate significant differences between MO and IF cows: (a, b: P <0.05).

Results: MILK



Item		Modenese Mean	Modenese S.E.	Italian Friesian Mean	Italian Friesian S.E.
Fat (g/100 g)	Mean (8+21 wk)	3.97	0.158	3.87	0.146
Lactose (g/100 g)		4.77	0.038	4.74	0.035
Casein number		78.00	0.249	77.56	0.230





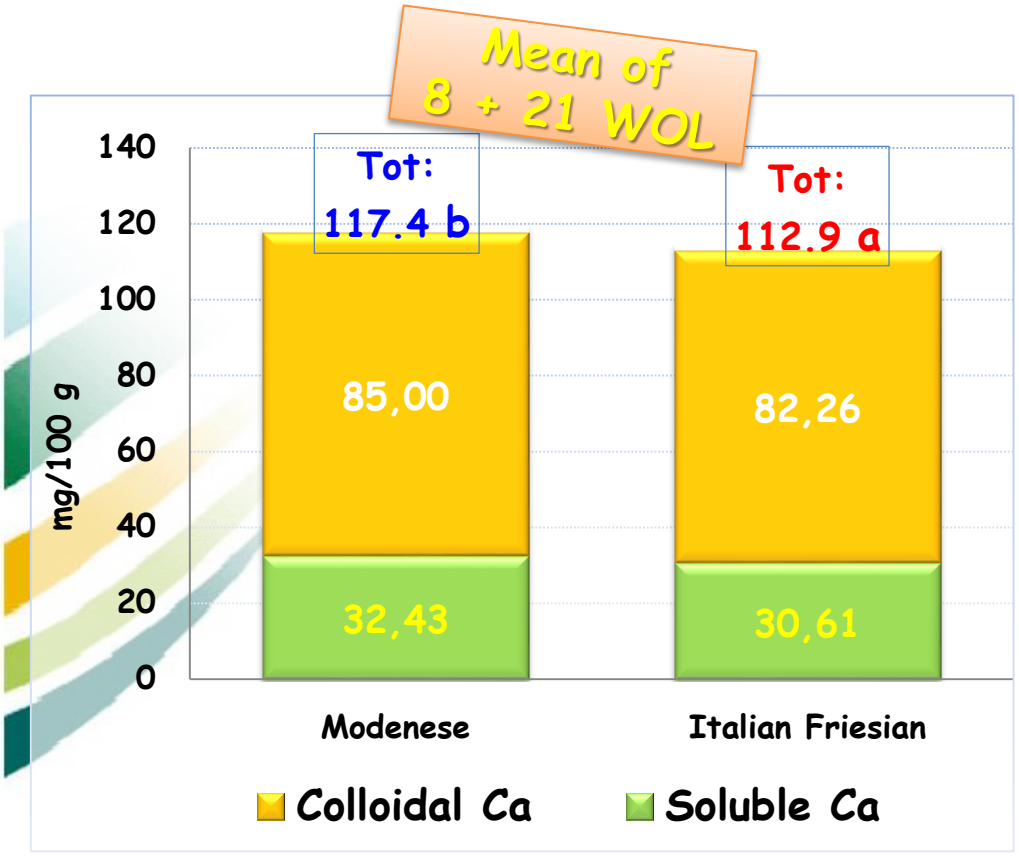
Letters indicate significant differences between MO and IF cows: (a, b: P <0.05; A, B: P<0.01).
Asterisks indicate significant differences (within breed) between 8 WOL and 21 WOL: (* P<0.05; ** P <0.01).

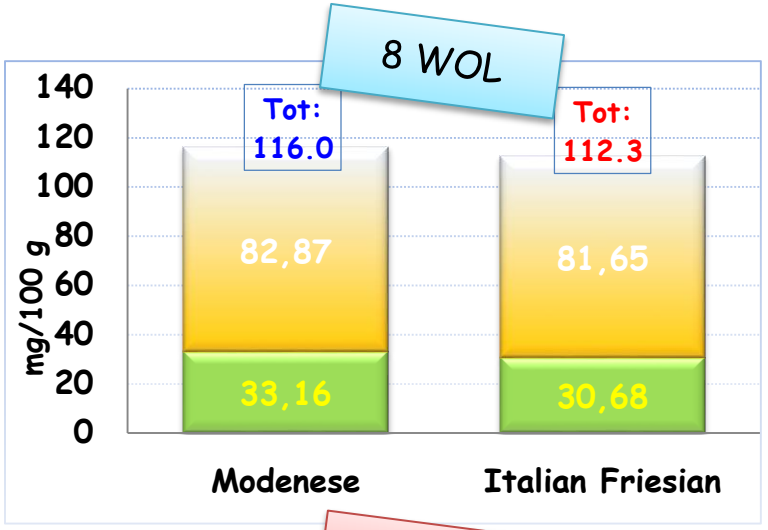
Results: MILK

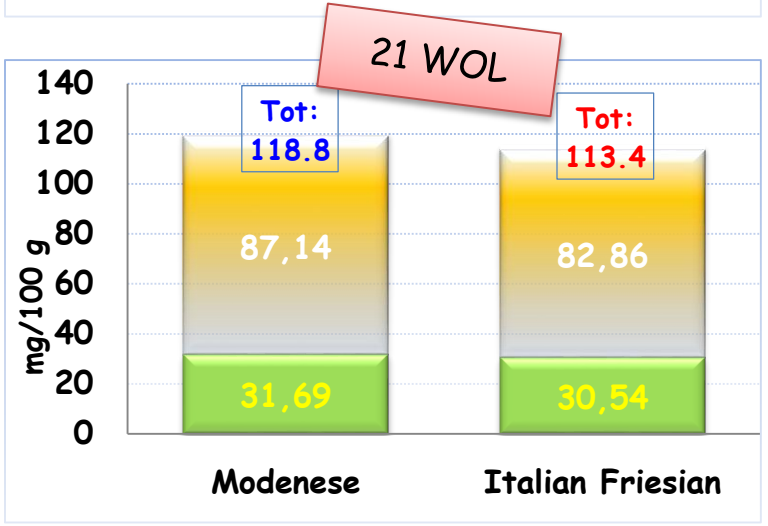
Rennet coagulation properties							
Item	WK	Modenese			Italian Friesian		
		Mean		S.E.	Mean		S.E.
pH	8	6.70		0.019	6.72		0.017
	21	6.73		0.019	6.73		0.019
Titrat. acid. (°SH/50 mL)	8	3.45	b	0.101	3.14	a	0.090
	21	3.38	(b)	0.101	3.13	(a)	0.097
Clotting time, r (min)	8	16.2	(b)	1.677	20.3	(a)	1.395
	21	21.7	(*)	2.250	19.1		1.901
Curd firming time, K_{20} (min)	8	3.47		0.399	3.82		0.427
	21	3.42		0.652	3.29		0.461
Curd firmness a_{30} (mm)	8	31.70		4.645	24.85		4.074
	21	21.81		5.997	32.33		5.552

Letters indicate significant differences between MO and IF cows: (a, b: $P < 0.05$; (a), (b): $0.05 < P < 0.10$). Asterisks indicate significant differences between 8 WOL and 21 WOL: (*) $0.05 < P < 0.10$.

MILK CALCIUM CONTENT



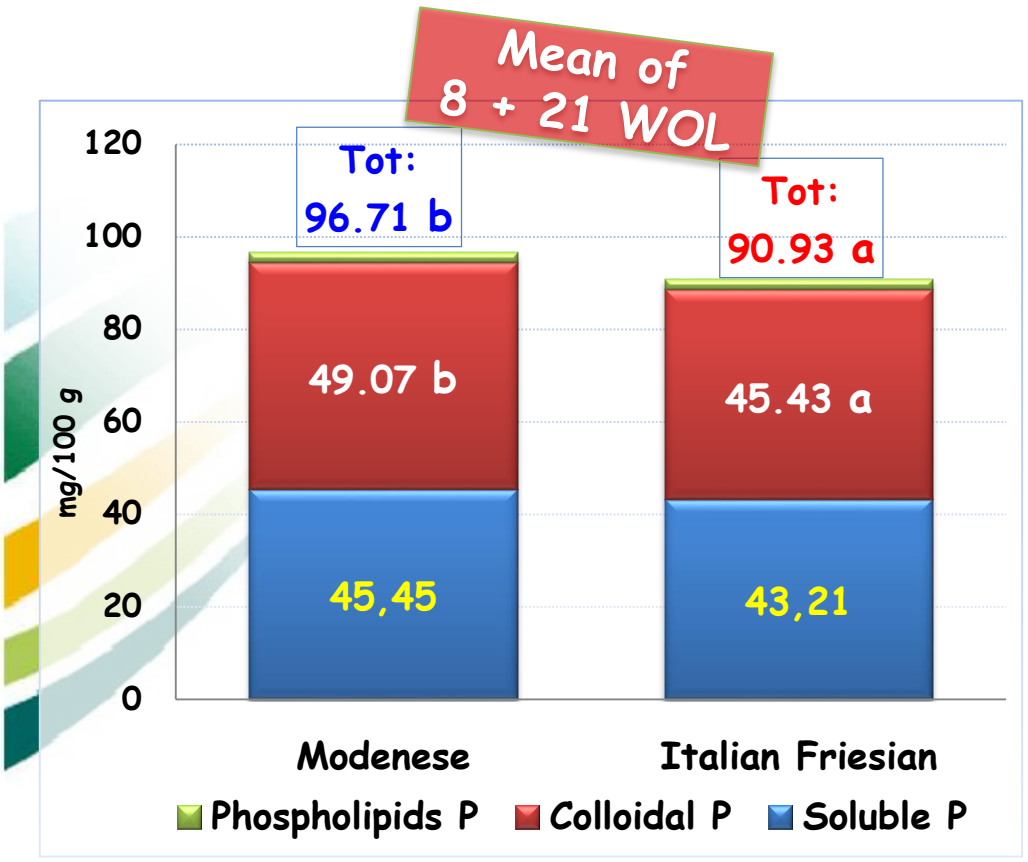


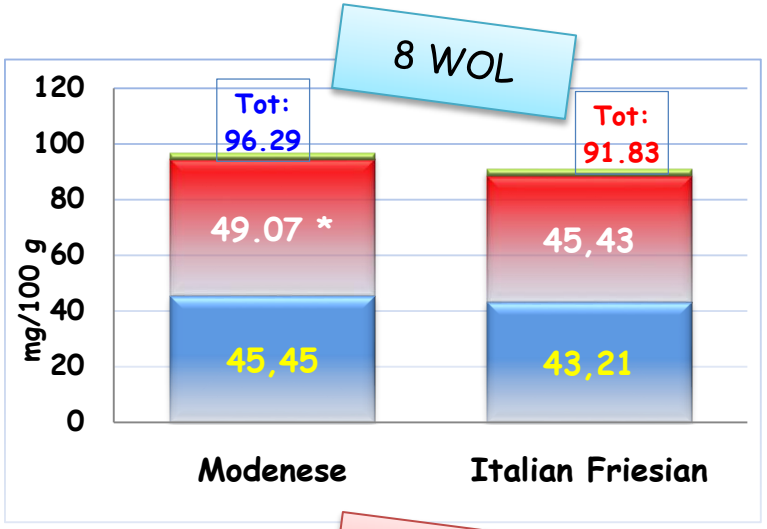


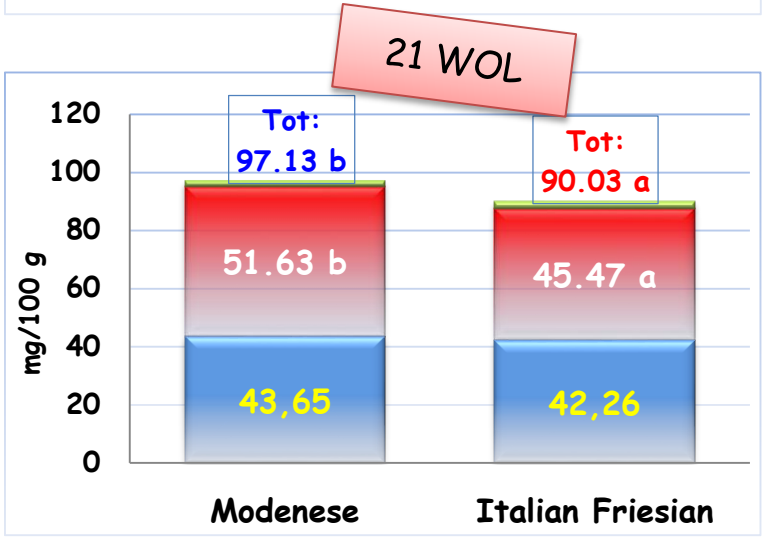
Letters indicate significant differences between MO and IF cows: a, b: P <0.05.

MILK PHOSPHORUS CONTENT

Letters indicate significant differences between MO and IF cows: a, b: P <0.05.







Correlations: Milk-Plasma

Modenese		Plasma			
		iP	Ca	ALP	TRAP
Milk Ca and P concentrations	Phosphorus	0.31 0.158	0.70 0.0003	- -	- -
	Total P excreted daily	-0.60 0.003	- -	0.34 0.126	0.34 0.124
	Soluble P	0.46 0.032	0.69 0.0004	0.41 0.061	- -
	Colloidal P	- -	- -	- -	- -
	Calcium	- -	0.43 0.048	- -	- -
	Total Ca excreted daily	-0.65 0.001	-0.35 0.107	- -	- -
	Soluble Ca	- -	- -	- -	- -
	Colloidal Ca	- -	0.39 0.073	- -	- -

Italian Friesian		Plasma			
		iP	Ca	ALP	TRAP
Milk Ca and P concentrations	Phosphorus	- -	- -	0.32 0.110	- -
	Total P excreted daily	-0.40 0.044	- -	- -	- -
	Soluble P	- -	- -	- -	- -
	Colloidal P	- -	- -	- -	- -
	Calcium	- -	- -	0.34 0.091	- -
	Total Ca excreted daily	-0.45 0.020	- -	- -	- -
	Soluble Ca	-0.49 0.012	- -	- -	- -
	Colloidal Ca	0.36 0.072	- -	- -	- -

Correlations: Milk-Milk

Modenese		Phosphorus	Soluble P	Colloidal P
Milk Ca and P concentrations	Phosphorus	1	**0.68** 0.0006	**0.63** 0.002
	Total P excreted daily	- -	- -	- -
	Soluble P	- -	1	- -
	Colloidal P	- -	- -	1
	Calcium	**0.59** 0.004	- -	**0.55** 0.0079
	Total Ca excreted daily	- -	- -	- -
	Soluble Ca	- -	- -	**-0.38** 0.082
	Colloidal Ca	**0.59** 0.004	- -	**0.68** 0.0005

Italian Friesian		Phosphorus	Soluble P	Colloidal P
Milk Ca and P concentrations	Phosphorus	1	**0.74** <.0001	**0.60** 0.001
	Total P excreted daily	**0.57** 0.002	**0.45** 0.020	- -
	Soluble P	- -	1	- -
	Colloidal P	**0.60** 0.001	- -	1
	Calcium	**0.56** 0.003	**0.62** 0.0006	- -
	Total Ca excreted daily	**0.44** 0.026	- -	- -
	Soluble Ca	- -	**0.37** 0.060	- -
	Colloidal Ca	**0.40** 0.045	**0.36** 0.074	- -

Discussion

- Plasma Ca and iP concentrations (MO = IF) were not mirrored in milk Ca and P contents (MO > IF)
- Marked differences in ALP and TRAP only partly mirrored in milk minerals content

- Differences in plasma Mg and K not related to diet concentration: different metabolism/requirement or genetic regulation
- Milk titratable acidity difference more pronounced than expected from milk proteins

Conclusions

- The differences in plasma ALP and TRAP activities need further study to explain their meaning

- The differences in milk mineral contents need further study to understand the reasons for a preferential secretion of Ca and P in MO: polymorphisms in membrane proteins involved in their transport? (a study on SNPs in these 2 breeds is in progress)

CRA
CONSIGLIO PER LA RICERCA E LA SPERIMENTAZIONE IN AGRICOLTURA

Thank… … you … … for … … your … … attention!

Acknowledgment

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