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Comparison of effects of forage legumes containing condensed tannins on milk and cheese quality

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LegumePlus 
Marie Curie Initial Training Network



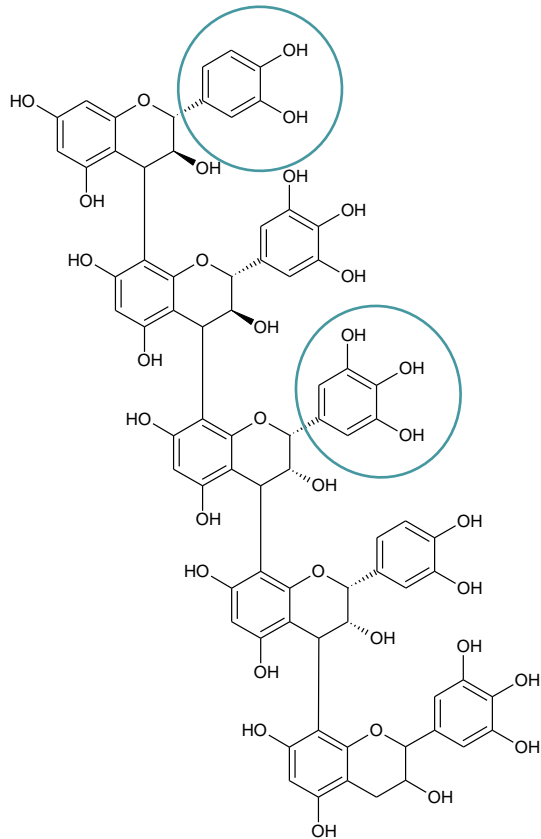
Observations from the literature

- ✦ Western diet are deficient in $n-3$ fatty acids.
- ✦ LA ($18:2n-6$) and ALA ($18:3n-3$) are only provided by food.
- ✦ EPA ($20:5n-3$), DPA ($22:5n-3$) and DHA ($22:6n-3$) are important because of their positive effects on human health (cardiovascular diseases, brain development, inflammatory and oxidative stress)



Dietary PUFA are **biohydrogenated** to saturated fatty acids in the rumen

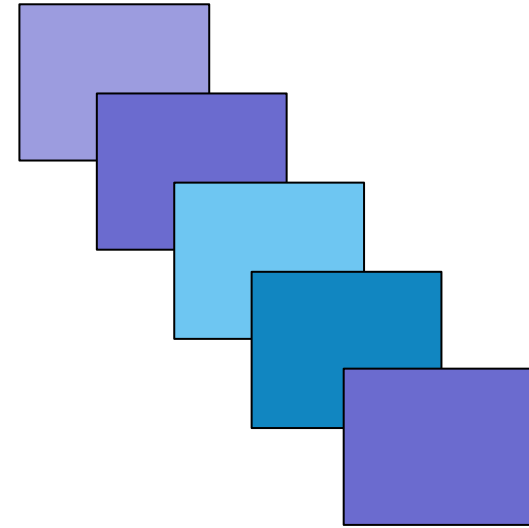
Condensed tannins (CT)...



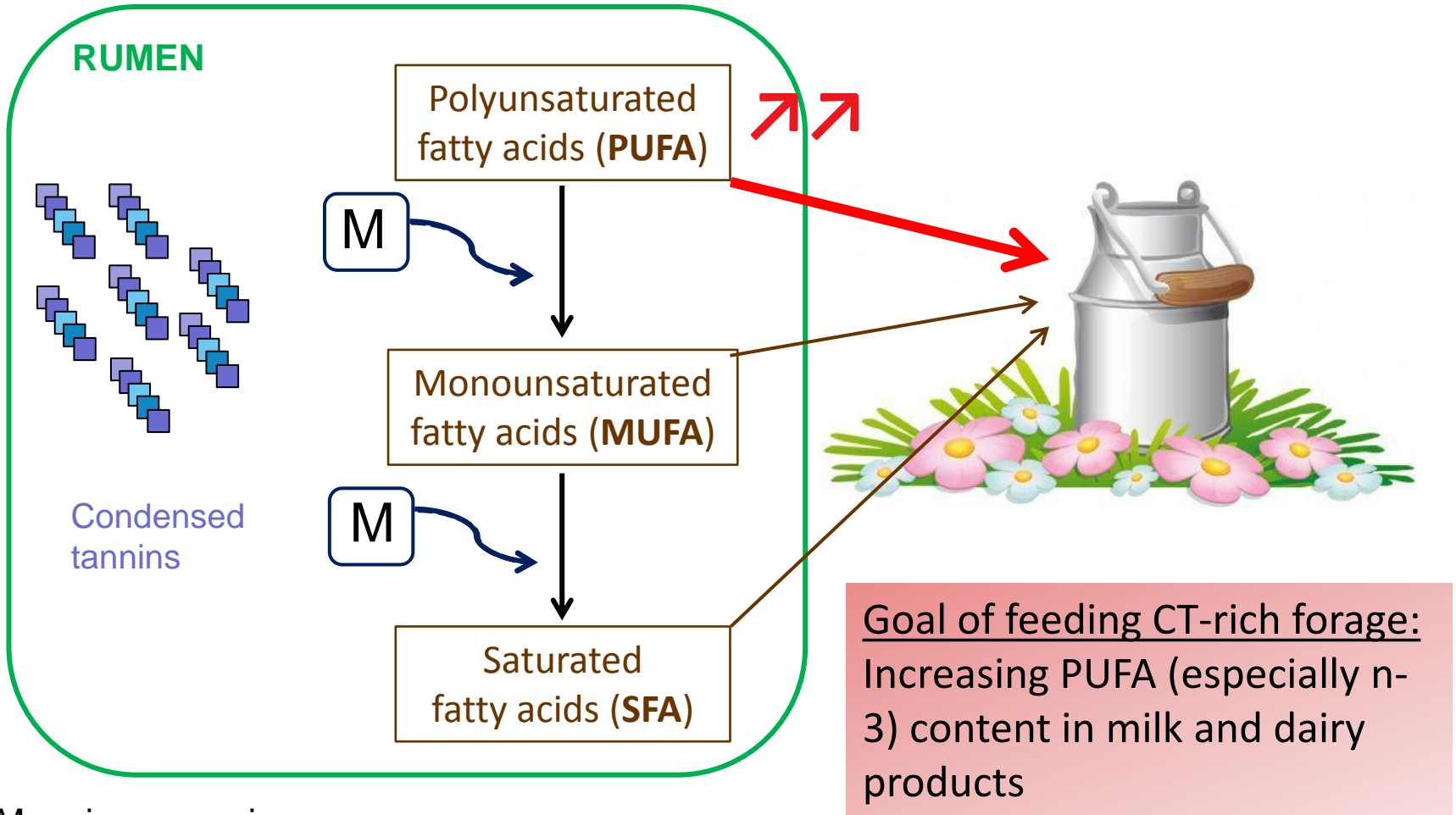
- Family of polyphenols :
polymers of flavan-3-ols
- In forage legumes such as
birdsfoot trefoil and sainfoin
- Very reactive molecules



... can complex with protein, lipids
and carbohydrates.



Hypothesis: Condensed tannins reduce biohydrogenation of PUFA and MUFA











M : micro-organismes

🇨🇭 Experimental design

- 24 dairy cows (4 groups of 6) with an average milk yield of 30.9 ± 5.7 kg/d
- Standard diet: 45% hay, 25% maize silage, 5 % linseed, 7% concentrate



Period	Weeks	Treatments			
		Lucerne	Sainfoin	Birdsfoot Bull	Birdsfoot Polom
Control	1	Standard diet +			
	2				
	3				
	4				
Experimental	5	Standard diet +			
	6	Lucerne pellets	Sainfoin pellets	Birdsfoot Bull pellets	Birdsfoot Polom pellets
	7				
	8				



Cheese making: Gruyère-type

Evaluated traits

✔ Feed intake



✔ Milk : Production

Gross composition (fat, protein, casein, urea)

Fatty acids profile

Sensory assessment



✔ Cheese : Fatty acids profile

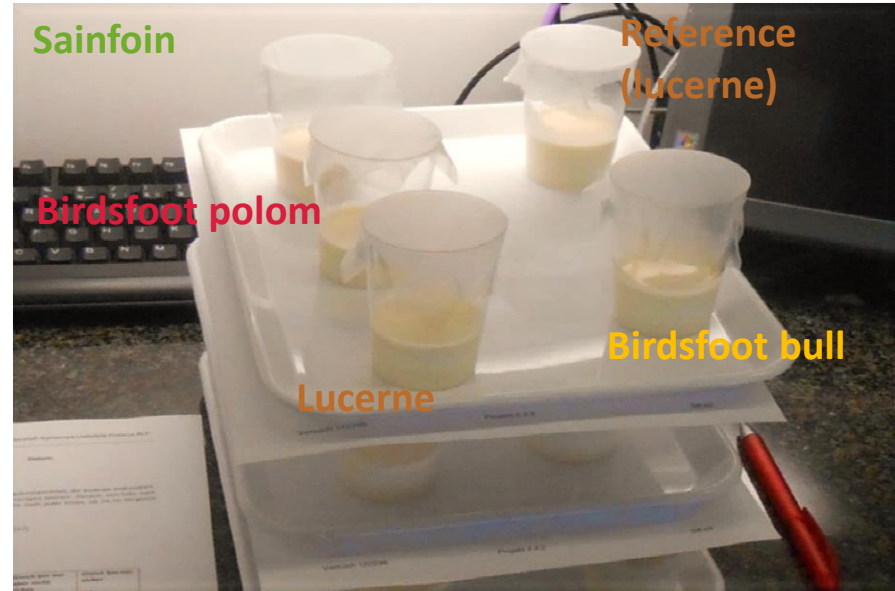
Sensory assessment



Sensory assessment of the milk

Objective:

- Compare the odor of raw milk from each experimental group to a reference sample



Question asked to the panelists:

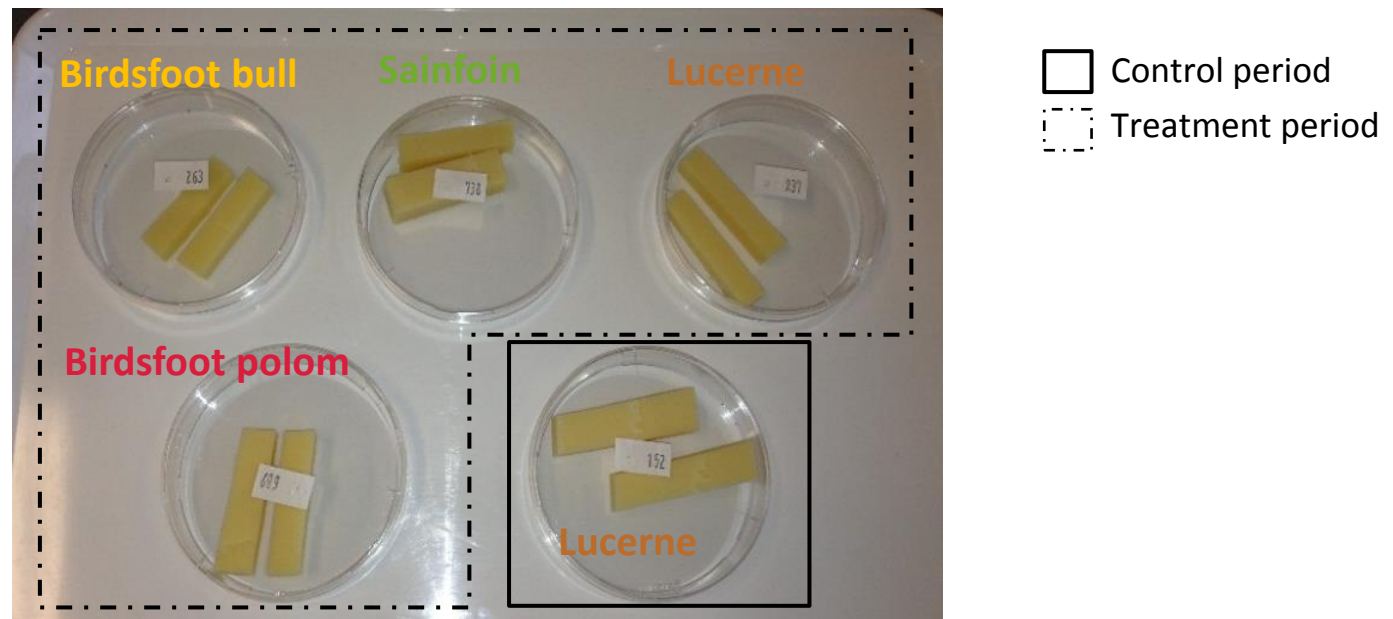
“Does this milk smell different to that of the reference?”

1. “yes, I am sure”
2. “yes, but I am not sure”
3. “no, I am not sure”
4. “no, I am sure”

Sensory assessment of the cheese

Objectives:

- ✦ Compare the flavor and textural structure of the cheeses manufactured in the experimental period and one cheese (lucerne) manufactured in control period.
- ✦ Continuous sensory scale: 0 (low intensity)
10 (high intensity)



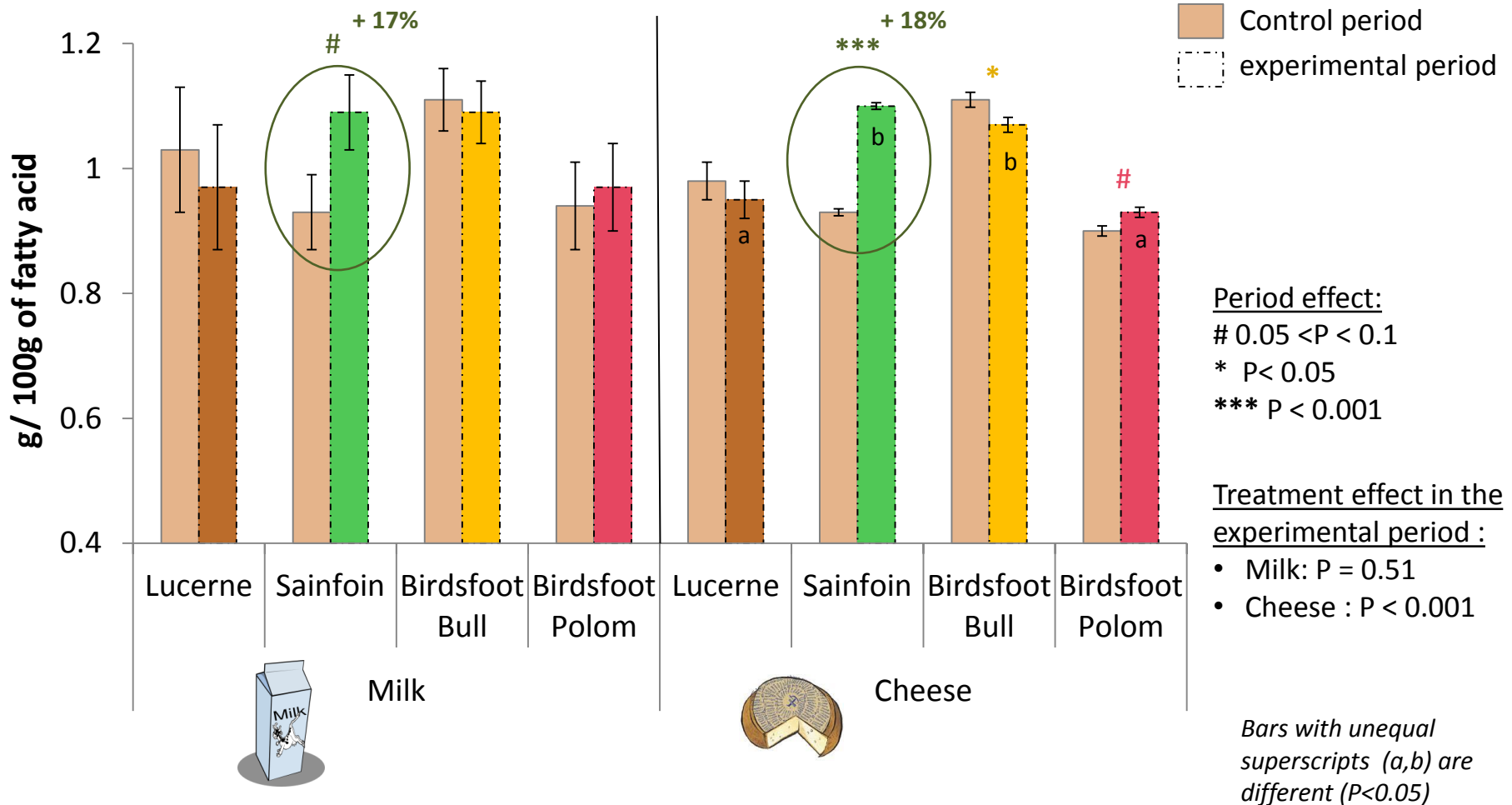
Intake and milk performance

Treatment Period	Lucerne		Sainfoin		Birdsfoot Bull		Birdsfoot Polom		Treatment effect	
	CP	EP	CP	EP	CP	EP	CP	EP	SEM	P-value
Intake										
Total dry matter (kg)	22.0	21.9	22.4	22.7	22.3	23.1	22.9	23.0	1.2	0.86
CT (g/d)	-	0 ^c	-	691 ^a	-	206 ^b	-	120 ^b	47.7	<0.01
Proteins (kg/d)	2.9	3.0	3.0	3.0	2.9	3.3	3.1	3.3	0.2	0.36
α-linolenic acid (g/d)	325	347	325	361	334	392	327	372	15.4	0.17
Milk										
Production (kg/d)	32.8	32.8	33.4	31.9	31.2	31.6	35.2	34	1.4	0.49
Fat (%)	3.99	3.98	4.07	4.10	4.3	4.42	4.07	4.20	0.13	0.06
Protein (%)	3.15	3.24 ^a	3.17	3.20 ^a	3.42	3.45 ^b	3.08	3.20 ^a	0.05	<0.01
Casein (%)	2.45	2.62 ^a	2.46	2.59 ^a	2.72	2.83 ^b	2.36	2.50 ^a	0.05	<0.01
Urea (mg/kg)	159.7	172.1 ^a	169.2	129.9 ^b	152.9	164.3 ^a	165.7	167.7 ^a	4.70	<0.01

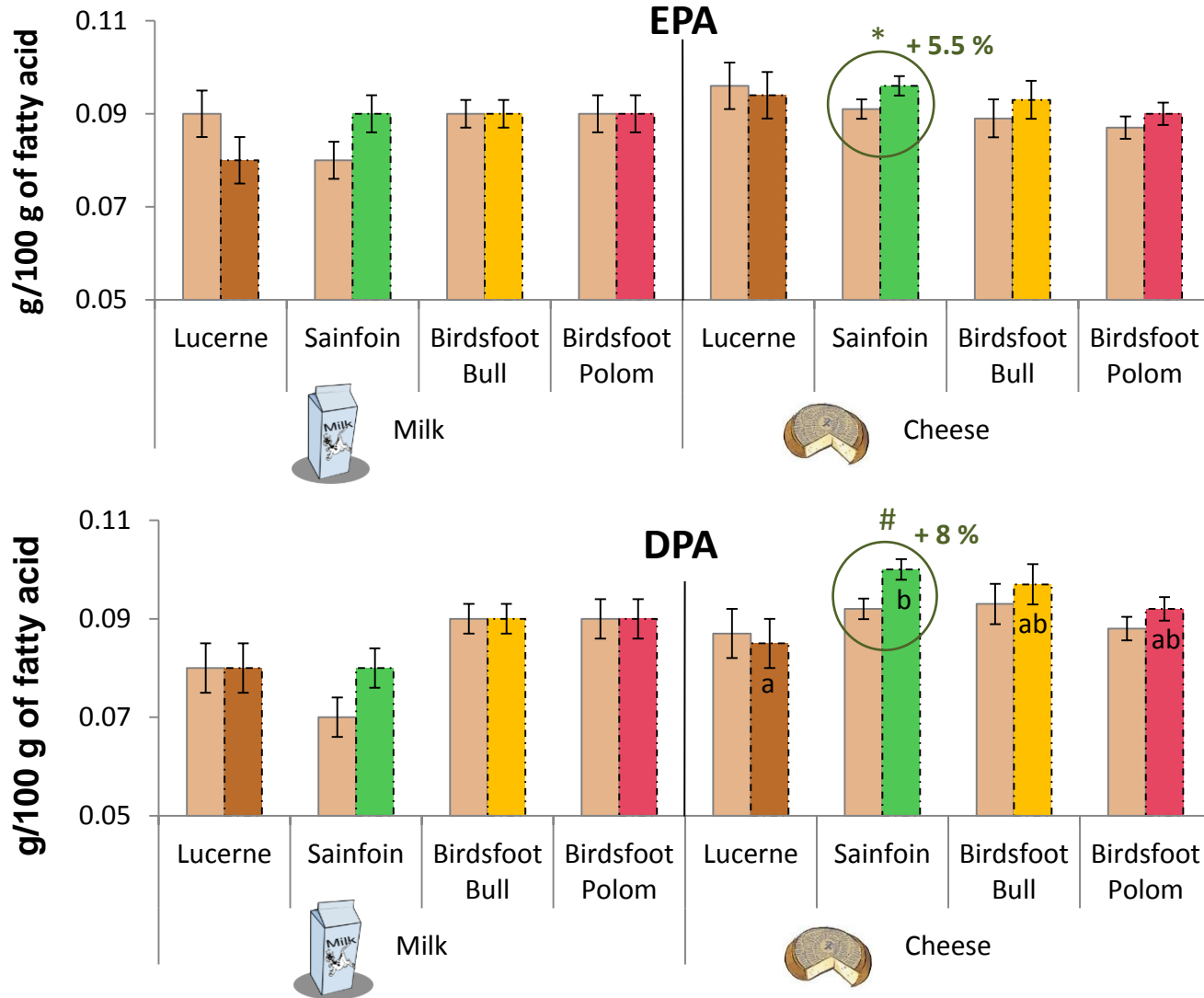
^{abc} Least square means within the same row without a common superscript differ (P < 0.05)

CP: control period ; EP: experimental period

Concentration of α -linolenic acid ($18:3n-3$)



Concentration of EPA (20:5n-3) and DPA (22:5n-3)



Control period
Experimental period

Period effect:
0.05 < P < 0.1

Treatment effect in the experimental period:

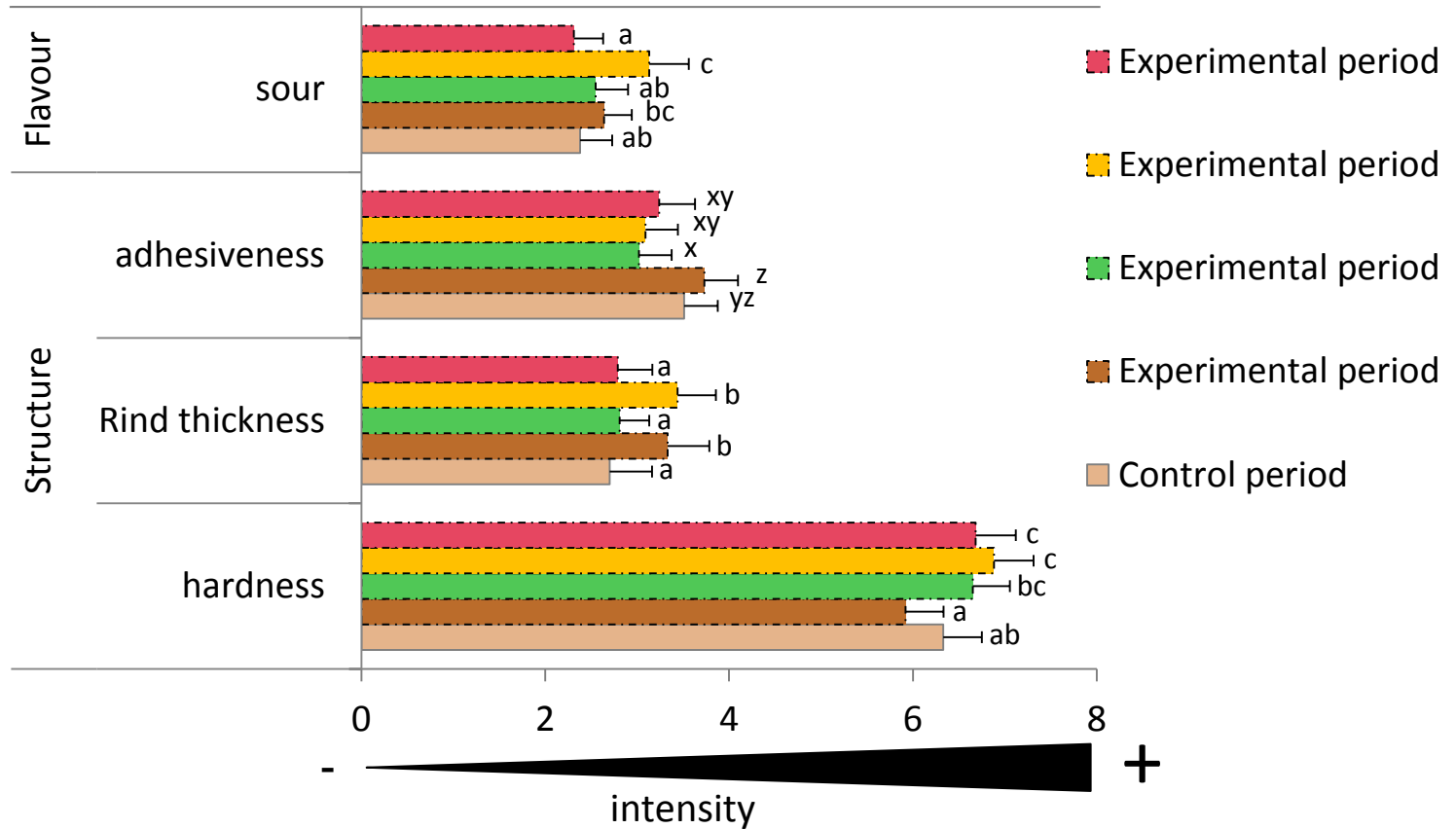
- EPA
- Milk: P = 0.54
 - Cheese: P = 0.23
- DPA
- Milk: P = 0.50
 - Cheese: P = 0.05

Bars with unequal superscripts are different (P<0.05)

Swiss Milk and cheese assessment



	Lucerne	Sainfoin	Birdsfoot Bull	Birdsfoot Polom
Control period	n.s	n.s	n.s	n.s
Experimental period	n.s	n.s	<0.05	n.s



Comparison of effects of forage legumes containing condensed tannins on milk and cheese quality | Results

Marion Girard

Summary of the effects of condensed tannins

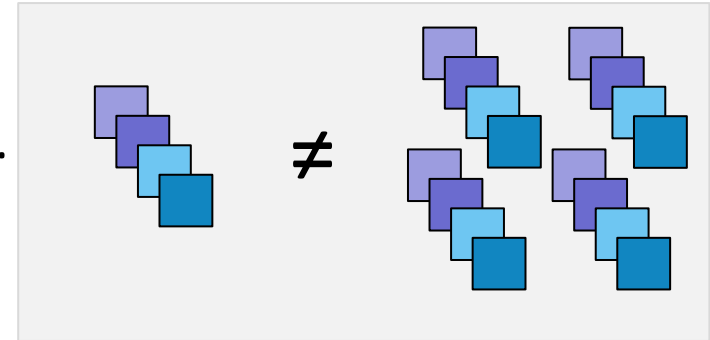
Control period (lucerne) vs experimental period (condensed tannins):

		Birdsfoot Polom	Birdsfoot Bull	Sainfoin	
Intake	Condensed tannins (g/j)	120	206	691	
	Total dry matter	=	=	=	
	18:3n-3	+ 14%	+ 17%	+ 11%	Blue : tendency
Milk	Production, fat, protein	=	=	=	
	Urea	=	+ 7%	- 23%	
	18:3n-3	=	=	+ 17%	
Cheese	18:3n-3	+ 3%	- 4%	+ 18%	
	20:5n-3	=	=	+ 5.5%	
	22:5n-3	=	=	+ 8%	

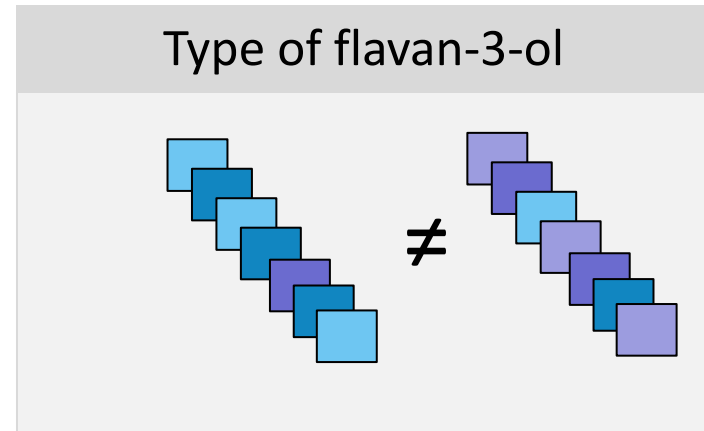
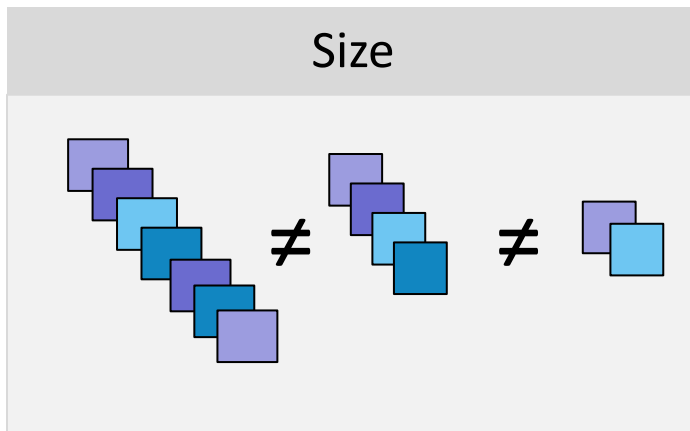
Sainfoin was more promising to improve milk and cheese quality than the 2 birdsfoot trefoil

What can explain these differences?

➤ The concentration of condensed tannins ...



➤ The structure of tannins (which can be different even in the same species)...



➤ Other molecules (polyphenols...)



Thank you for your attention !!!

Acknowledgements

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Treatment	Alfalfa				Sainfoin				Birdfoot trefoil Bull				Birdfoot trefoil Polom				Treatment effect	
	CoP	ExP	SEM	<i>P</i> value	CoP	ExP	SEM	<i>P</i> value	CoP	ExP	SEM	<i>P</i> value	CoP	ExP	SEM	<i>P</i> value	SEM	<i>P</i> value
Period	n = 6	n = 6			n = 6	n = 6			n = 6	n = 6			n = 6	n = 6				
Item	n = 6				n = 6				n = 6				n = 6					
Feed intake (kg DM/cow per d)																		
Mixture	16.4	16.6	0.64	0.81	16.4	17.2	0.63	0.39	17.0	17.8	0.92	0.51	16.4	16.9	1.02	0.75	0.87	0.73
Concentrate	1.4	1.5	0.45	0.85	1.7	1.8	0.57	0.89	1.1	1.0	0.45	0.95	2.1	2.2	0.70	0.87	0.58	0.50
Pellets	4.3	3.8	0.29	0.25	4.3	3.6	0.36	0.23	4.3	4.2	0.24	0.94	4.4	3.9	0.25	0.18	0.37	0.61
Total	22.0	21.9	0.89	0.91	22.4	22.7	0.79	0.81	22.3	23.1	1.43	0.69	22.9	23.0	1.47	0.96	1.20	0.86
Nutrient intake (per cow per d)																		
CT ¹ (g)	0	0 ^c	-	-	0	691 ^a	59.70	<0.01	0	206 ^b	5.03	<0.01	0	120 ^b	7.31	<0.01	47.68	<0.01
CP (kg)	3.0	3.0	0.13	0.78	3.0	3.0	0.11	0.84	3.0	3.3	0.20	0.23	3.1	3.3	0.21	0.45	0.17	0.36
ADF (kg)	5.9	5.7	0.17	0.33	5.9	5.6	0.18	0.29	6.0	5.9	0.31	0.82	6.0	5.6	0.25	0.31	0.23	0.73
NDF (kg)	10.4	10.1	0.33	0.48	10.5	9.9	0.32	0.27	10.6	10.5	0.57	0.85	10.6	10.0	0.54	0.45	0.46	0.77
NE _L (MJ)	138	139	6.37	0.92	141	146	5.90	0.56	139	146	9.68	0.62	144	148	10.79	0.82	7.87	0.87
Fatty acid ² intake (g/cow per d)																		
14:0	2.77	2.88	0.08	0.35	2.78	2.89	0.08	0.39	2.82	3.17	0.15	0.14	2.82	3.05	0.12	0.21	0.12	0.22
16:0	104.7	106.4	4.62	0.80	106.5	110.8	4.19	0.49	105.7	115.8	7.18	0.35	109.0	115.4	7.76	0.57	6.34	0.64
16:1 <i>c</i> -9	1.04	5.61 ^b	0.09	<0.01	1.05	5.29 ^b	0.12	<0.01	1.05	6.71 ^a	0.22	<0.01	1.07	6.50 ^a	0.15	<0.01	0.22	<0.01
18:0	19.6	20.8	0.75	0.28	19.8	21.1	0.71	0.21	19.8	21.4	1.21	0.38	20.1	20.8	1.34	0.70	1.07	0.96
18:1 <i>c</i> -9	110.4	112.2	5.67	0.84	112.8	118.9	5.79	0.47	111.4	116.7	8.11	0.65	115.0	118.6	10.15	0.81	7.94	0.91
18:1 <i>c</i> -11	5.02	5.63	0.26	0.13	5.12	6.08	0.25	0.02	5.05	6.00	0.38	0.11	5.22	6.18	0.46	0.18	0.38	0.72
18:2n-6	240.4	236.5	13.59	0.84	246.7	249.1	13.97	0.91	241.1	247.2	19.04	0.82	253.5	259.5	23.59	0.86	19.04	0.85
18:3n-3	325.2	346.8	10.51	0.18	325.1	360.7	11.19	0.05	333.9	391.9	18.4	0.05	327.4	372.5	16.58	0.09	15.42	0.17
20:0	3.20	3.20 ^b	0.11	0.99	3.22	4.14 ^a	0.13	<0.01	3.24	3.53 ^b	0.19	0.31	3.28	3.38 ^b	0.19	0.70	0.17	<0.01
20:1	1.03	1.06	0.07	0.78	1.07	1.14	0.08	0.59	1.03	1.16	0.10	0.37	1.11	1.16	0.13	0.77	0.11	0.88
22:0	3.24	3.04 ^b	0.10	0.16	3.26	3.63 ^a	0.12	0.06	3.30	3.23 ^{ab}	0.17	0.80	3.30	3.06 ^b	0.15	0.30	0.14	0.01
SFA	138.2	142.3	5.77	0.63	140.3	148.5	5.19	0.29	139.8	153.7	9.17	0.31	143.3	151.8	9.73	0.55	7.97	0.71
MUFA	122.8	127.7	6.17	0.59	125.3	134.5	6.24	0.32	124.0	134.2	8.97	0.44	127.8	136.0	11.03	0.61	8.69	0.89
PUFA	566.2	583.7	23.18	0.61	572.5	610.4	21.93	0.25	575.7	639.9	36.95	0.25	581.5	632.9	39.58	0.39	32.93	0.57
Total	852.8	879.7	35.56	0.60	863.6	918.3	33.50	0.28	865.4	956.8	56.27	0.28	878.4	949.3	61.06	0.44	50.21	0.64

Milk FA

Treatment	Alfalfa				Sainfoin				Birdsfoot trefoil Bull				Birdsfoot trefoil Polom				Treatment effect	
	CoP	ExP	SEM	P value	CoP	ExP	SEM	P value	CoP	ExP	SEM	P value	CoP	ExP	SEM	P value	SEM	P value
Period	n = 6		n = 6		n = 6		n = 6		n = 6		n = 6		n = 6					
Item ¹	n = 6	n = 6			n = 6	n = 6			n = 6	n = 6			n = 6	n = 6				
4:0	3.47	3.77	0.08	0.03	3.71	4.02	0.10	0.06	3.56	3.72	0.09	0.25	3.74	3.85	0.24	0.73	0.14	0.39
6:0	2.41	2.56	0.08	0.22	2.52	2.61	0.06	0.26	2.45	2.53	0.07	0.40	2.56	2.58	0.07	0.85	0.07	0.83
8:0	1.75	1.54	0.07	0.06	1.77	1.55	0.03	<0.01	1.77	1.55	0.07	0.06	1.81	1.64	0.04	0.01	0.06	0.61
10:0	3.41	3.35	0.20	0.82	3.34	3.28	0.11	0.73	3.49	3.40	0.23	0.79	3.50	3.51	0.12	0.96	0.19	0.84
12:0	3.56	3.54	0.21	0.95	3.46	3.47	0.14	0.96	3.72	3.64	0.28	0.85	3.71	3.76	0.16	0.81	0.21	0.76
14:0	11.59	11.80	0.33	0.65	11.48	11.61	0.33	0.79	11.29	11.54	0.42	0.68	11.72	11.82	0.43	0.87	0.37	0.92
15:0	1.11	1.11	0.05	0.99	1.08	0.98	0.04	0.10	1.10	1.06	0.03	0.39	1.12	1.08	0.05	0.52	0.04	0.13
16:0	26.28	26.84	0.57	0.50	26.22	26.69	1.31	0.81	26.56	27.31	1.25	0.68	27.10	26.28	0.62	0.38	1.02	0.90
16:1 c	1.18	1.07	0.08	0.38	1.14	0.98	0.05	0.07	1.19	1.08	0.05	0.15	1.08	0.98	0.05	0.21	0.07	0.54
18:0	13.04	12.24	0.59	0.36	13.70	13.25	0.58	0.59	11.98	11.88	0.47	0.89	12.39	12.73	0.32	0.49	0.49	0.20
18:1 t-9	0.29	0.36 ^y	0.04	0.20	0.29	0.29 ^{xy}	0.01	0.77	0.29	0.28 ^x	0.02	0.71	0.31	0.29 ^{xy}	0.01	0.30	0.03	0.10
Σ 18:1 t-10/t-11	1.81	1.74	0.15	0.76	1.75	1.54	0.07	0.08	1.96	1.86	0.17	0.67	1.89	1.85	0.11	0.79	0.14	0.34
18:1 t-13-14,c-6-8	0.93	1.05	0.06	0.24	1.01	0.96	0.04	0.41	1.05	1.06	0.06	0.93	1.06	1.03	0.08	0.79	0.05	0.51
18:1 c-9	17.94	17.83	0.62	0.91	17.69	17.43	0.68	0.80	17.84	17.59	1.22	0.89	16.78	17.27	0.72	0.65	0.75	0.95
18:2n-6	1.41	1.35	0.10	0.68	1.33	1.34	0.05	0.80	1.38	1.32	0.03	0.19	1.34	1.32	0.05	0.79	0.07	0.99
18:3n-6	0.03	0.03 ^x	0.01	0.67	0.02	0.02 ^{xy}	0.01	0.51	0.02	0.02 ^y	0.01	0.23	0.02	0.02 ^{xy}	0.01	0.56	0.01	0.06
18:3n-3	1.03	0.97	0.10	0.66	0.93	1.09	0.06	0.07	1.11	1.09	0.05	0.76	0.94	0.97	0.07	0.79	0.09	0.51
20:0	0.17	0.15	0.01	0.06	0.17	0.16	0.01	0.42	0.16	0.15	0.01	0.23	0.16	0.16	0.01	0.65	0.01	0.45
20:3n-6	0.04	0.04	0.01	0.27	0.05	0.04	0.01	0.28	0.05	0.05	0.01	0.66	0.05	0.05	0.01	0.96	0.01	0.15
20:4n-6	0.06	0.06 ^y	0.01	0.41	0.07	0.07 ^{xy}	0.01	0.80	0.07	0.07 ^{xy}	0.01	0.39	0.08	0.07 ^x	0.01	0.50	0.01	0.06
20:5n-3	0.09	0.08	0.01	0.20	0.08	0.09	0.01	0.26	0.09	0.09	0.01	0.50	0.09	0.09	0.01	0.23	0.01	0.54
22:5n-3	0.08	0.08	0.01	0.93	0.07	0.08	0.01	0.30	0.09	0.09	0.01	0.51	0.09	0.09	0.01	0.91	0.01	0.50
SCFA ²	11.48	11.67	0.40	0.75	11.78	11.96	0.23	0.58	11.72	11.67	0.39	0.92	12.11	12.08	0.22	0.94	0.33	0.71
MCFA ³	46.63	47.40	0.72	0.47	46.37	46.68	1.37	0.88	46.94	47.71	1.77	0.76	47.81	47.04	1.10	0.63	1.16	0.91
LCFA ⁴	42.06	41.13	1.07	0.55	42.04	41.53	1.48	0.81	41.53	40.79	2.00	0.80	40.26	41.05	1.20	0.65	1.33	0.98
SFA ⁵	69.30	69.50	1.25	0.91	70.02	70.23	0.95	0.88	68.70	69.47	1.52	0.73	70.33	70.08	1.04	0.87	1.16	0.94
MUFA ⁶	25.89	25.88	0.91	0.99	25.63	25.25	0.86	0.77	26.29	25.66	1.40	0.76	24.98	25.29	0.86	0.81	0.96	0.95
PUFA ⁷	4.80	4.63	0.39	0.76	4.37	4.51	0.14	0.48	5.01	4.86	0.18	0.55	4.66	4.60	0.23	0.85	0.27	0.82
Σ 18:1	23.06	23.13	0.84	0.95	22.81	22.62	0.90	0.88	23.36	22.89	1.46	0.83	22.11	22.59	0.89	0.72	0.97	0.97
Σ 18:2	3.40	3.33	0.28	0.85	3.10	3.07	0.10	0.84	3.52	3.39	0.13	0.49	3.34	3.26	0.14	0.69	0.20	0.62
Σ CLA ⁸	0.67	0.69	0.09	0.89	0.59	0.56	0.03	0.46	0.77	0.77	0.07	0.97	0.72	0.71	0.07	0.88	0.08	0.22
Σ n-3 ⁹	1.69	1.59	0.16	0.65	1.50	1.67	0.08	0.18	1.83	1.78	0.08	0.67	1.59	1.59	0.13	0.99	0.13	0.67
Σ n-6 ¹⁰	2.67	2.67	0.17	0.97	2.56	2.72	0.10	0.25	2.73	2.64	0.07	0.39	2.62	2.62	0.08	0.94	0.13	0.93
% of total CLA																		
18:2 c-9,t-11	76.2	78.1	1.10	0.25	75.6	77.4	0.83	0.15	77.0	78.4	1.02	0.35	76.4	78.2	0.55	0.05	0.91	0.87
18:2 t-10,c-12	0.45	0.32	0.06	0.14	0.44	0.34	0.03	0.02	0.33	0.32	0.04	0.30	0.33	0.28	0.03	0.65	0.04	0.57

Cheese FA

Treatment Period Item ¹	Alfalfa				Sainfoin				Birdsfoot trefoil Bull				Birdfoot trefoil Polom				Treatment effect	
	CoP	ExP	SEM	P value	CoP	ExP	SEM	P value	CoP	ExP	SEM	P value	CoP	ExP	SEM	P value	SEM	P value
	n = 3	n = 3			n = 3	n = 3			n = 3	n = 3			n = 3	n = 3				
4:0	3.79	3.72 ^y	0.09	0.63	3.94	4.05 ^x	0.05	0.21	3.48	3.76 ^{xy}	0.14	0.25	3.89	3.90 ^{xy}	0.05	0.95	0.08	0.08
6:0	2.53	2.51	0.04	0.71	2.59	2.59	0.01	0.94	2.44	2.49	0.05	0.47	2.52	2.55	0.02	0.30	0.03	0.19
8:0	1.55	1.53	0.02	0.47	1.52	1.51	0.01	0.35	1.69	1.61	0.08	0.53	1.51	1.56	0.01	<0.01	0.05	0.49
10:0	3.45	3.35 ^a	0.05	0.23	3.22	3.15 ^b	0.01	<0.01	3.50	3.40 ^a	0.04	0.10	3.28	3.41 ^a	0.01	<0.01	0.03	<0.01
12:0	3.70	3.62 ^b	0.05	0.26	3.46	3.40 ^c	0.01	0.02	3.79	3.71 ^{ab}	0.04	0.19	3.59	3.76 ^a	0.02	<0.01	0.02	<0.01
14:0	11.9	11.9 ^a	0.11	0.76	11.5	11.4 ^b	0.01	0.05	11.3	11.6 ^b	0.06	0.06	11.7	11.9 ^a	0.01	<0.01	0.06	<0.01
15:0	1.08	1.10 ^a	0.01	0.38	1.04	0.98 ^c	0.01	<0.01	1.07	1.04 ^b	0.01	0.11	1.10	1.06 ^{ab}	0.01	<0.01	0.01	<0.01
16:0	26.4	26.9 ^b	0.11	0.04	26.5	26.8 ^b	0.05	<0.01	26.9	27.5 ^a	0.15	0.04	27.3	26.4 ^b	0.07	<0.01	0.11	<0.01
16:1c	1.00	1.06 ^{xy}	0.02	0.09	1.03	1.05 ^{xy}	0.01	0.13	1.09	1.11 ^x	0.02	0.57	1.06	1.04 ^y	0.01	0.22	0.02	0.07
18:0	12.9	12.5 ^{bc}	0.21	0.35	13.6	13.4 ^a	0.04	0.02	11.9	11.9 ^c	0.21	0.96	12.7	12.9 ^{ab}	0.06	0.07	0.14	<0.01
18:1 c-9	17.2	17.7	0.34	0.38	17.5	17.9	0.09	0.04	17.6	17.4	0.10	0.24	17.0	17.3	0.04	<0.01	0.20	0.33
Σ 18:1 t-10/t-11	1.62	1.67 ^b	0.04	0.43	1.66	1.63 ^b	0.02	0.26	1.88	1.84 ^a	0.01	0.03	1.91	1.89 ^a	0.02	0.53	0.03	<0.01
18:2n-6	1.36	1.31	0.04	0.45	1.35	1.38	0.01	0.02	1.40	1.31	0.01	<0.01	1.29	1.31	0.01	0.32	0.02	0.14
18:3 n-6	0.02	0.03	0.01	0.09	0.03	0.03	0.01	0.51	0.02	0.03	0.01	0.71	0.03	0.02	0.01	0.08	0.01	0.17
18:3n-3	0.98	0.95 ^b	0.02	0.39	0.93	1.10 ^a	0.01	<0.01	1.11	1.07 ^a	0.01	0.07	0.90	0.93 ^b	0.01	0.04	0.01	<0.01
20:0	0.22	0.22 ^{bc}	0.01	0.75	0.23	0.23 ^a	0.01	0.54	0.24	0.22 ^b	0.01	0.01	0.22	0.21 ^b	0.01	0.38	0.01	<0.01
20:3 n-6	0.04	0.04 ^b	0.01	0.74	0.05	0.05 ^{ab}	0.01	0.58	0.05	0.05 ^{ab}	0.01	0.05	0.05	0.05 ^a	0.01	0.41	0.01	0.05
20:3 n-3	0.02	0.02 ^b	0.01	0.34	0.02	0.02 ^{ab}	0.01	0.83	0.02	0.02 ^a	0.01	0.76	0.02	0.02 ^b	0.01	0.17	0.01	0.03
20:4 n-6	0.07	0.07	0.01	0.18	0.08	0.07	0.01	0.29	0.08	0.07	0.01	0.03	0.07	0.07	0.01	0.25	0.01	0.24
20:5n-3	0.10	0.09	0.01	0.43	0.09	0.10	0.01	0.05	0.09	0.09	0.01	0.42	0.09	0.09	0.01	0.15	0.01	0.23
22:5n-3	0.09	0.09 ^b	0.01	0.73	0.09	0.10 ^a	0.01	0.10	0.09	0.10 ^{ab}	0.01	0.59	0.09	0.09 ^{ab}	0.01	0.32	0.01	0.05
SCFA ²	11.8	11.6	0.20	0.40	11.7	11.7	0.05	0.84	11.6	11.7	0.14	0.55	11.7	11.9	0.06	0.10	0.13	0.43
MCFA ³	46.9	47.6 ^a	0.18	0.07	46.5	46.7 ^b	0.05	0.12	47.2	48.0 ^a	0.25	0.11	47.9	47.3 ^{ab}	0.10	0.02	0.15	<0.01
LCFA ⁴	41.4	41.0 ^{ab}	0.35	0.50	42.0	41.8 ^a	0.05	0.07	41.4	40.5 ^b	0.35	0.16	40.6	41.0 ^{ab}	0.04	<0.01	0.24	0.03
SFA ⁵	70.2	70.0	0.45	0.69	70.3	70.1	0.11	0.24	69.1	69.8	0.10	<0.01	70.5	70.2	0.06	0.04	0.31	0.79
MUFA ⁶	25.1	25.5	0.32	0.45	25.2	25.4	0.10	0.39	25.9	25.4	0.14	0.06	25.0	25.3	0.03	0.02	0.38	0.89
PUFA ⁷	4.6	4.5	0.13	0.62	4.5	4.6	0.03	0.22	5.0	4.8	0.04	0.02	4.5	4.5	0.038	0.32	0.09	0.13
Σ CLA ⁸	0.60	0.64 ^{bc}	0.03	0.42	0.58	0.57 ^c	0.01	0.40	0.74	0.74 ^a	0.01	0.98	0.70	0.67 ^b	0.010	0.13	0.02	<0.01
Σ n-3 ⁹	1.64	1.58 ^b	0.06	0.38	1.57	1.71 ^a	0.01	<0.01	1.83	1.76 ^a	0.02	0.06	1.54	1.54 ^b	0.013	0.84	0.03	<0.01
Σ n-6 ¹⁰	2.76	2.57	0.04	0.03	2.63	2.55	0.02	0.04	2.77	2.55	0.03	<0.01	2.52	2.47	0.021	0.16	0.03	0.14



Cheese quality at 8 months

Treatment	Lucerne (n=3)		Birdsfoot Bull (n=3)		Sainfoin (n=3)		Birdsfoot Polom (n=3)		Treatment effect	
	CP	EP	CP	EP	CP	EP	CP	EP	SEM	P value
Chemistry										
Moisture (g/kg)	305	309	310	311	305	309	304	312	2.7	0.85
Fat (g/ kg DM)	542	533 ^a	542	539 ^{ab}	542	546 ^b	541	536 ^{ab}	1.5	< 0.01
Sodium chloride (g/kg)	13.0	12.7	13.0	13.1	12.5	13.1	13.3	14.0	0.56	0.47
Biochemistry (mmol/kg)										
Total volatile fatty acids	12.7	16.4	6.9	8.3	7.0	7.4	8.0	8.3	4.13	0.42
Total lactic acid	129	124	137	124	134	124	132	122	3.9	0.97
Free amino acids	276	262	266	277	275	285	297	290	29.4	0.91

^{ab} Least square means within the same row without a common superscript differ (P < 0.05)

No effects on cheese quality at 8 months, except for the fat ...