The impact of organic vs. inorganic selenium on dairy goat productivity and expression of selected genes in milk somatic cells

Daria Reczyńska, Bożena Witek, Justyna Jarczak, Michał Czopowicz, Marcin Mickiewicz, Jarosław Kaba, Lech Zwierzchowski and Emilia Bagnicka

SUPPLEMENTARY FILE

Supplementary Table S1. Ingredients and nutritive composition of the basal diet

Ingredients	**Dry matter (%)**	**% in DM**
Corn silage	31.5	29.7
Meadow hay	85.0	29.2
Oats (grain)	84.8	18.7
Triticale (grain	81.3	11.2
Extracted rapeseed meal	88.0	11.2
Chemical composition and nutritive value (per kg DM)		
NEL (MJ)*	6.3	
Crude protein (g)	142.4	
PDI (g)*	102.6	
NDF (g)	387.4	
ADF (g)	213.7	
ADL (g)	3.2	
Ash (g)	69.0	
Ether extract (g)	29.6	

DM – dry matter, NEL − net energy for lactation, PDI − protein digested in the small intestine, NDF – neutral detergent fibre, ADF – acid detergent fibre, ADL - acid-detergent lignin.
Composition of vitamin-mineral mixture in 1 kg:
Vitamins: Vitamin A – 500,000IU, D3 – 100,000IU, E – 1500mg, B1 – 50mg, B2 – 40mg, B6 – 20mg, B3 – 2,000mg, B5 – 100mg, Folic acid – 20mg, B12 – 0.2mg;
Trace elements:
Copper – 1100mg, Manganese – 4,000mg, Zinc – 9,500mg, Iron – 2,000mg, Iron – (iron chelate hydrate of glycine) 500mg, Iodine – 90mg, Selenium – 45mg
*(IZ PIB-INRA 2009)

Supplementary Table S2. Sequence of primers used in Real-Time PCR analysis

Gene and gene symbol	Primer sequence Forward (F)	Reverse (R)	Product size	Acession of GeneBank	Reference
α S1- casein (CSN1S1)	AACCCAGCTTGCTGCTT	CAAAATCTCAGTTACTGCACA	190	XM_018049133.1	Ramunno *et al*. 2005
α S2-casein (CSN1S2)	CTGGTTATGGTTGGACTGGAAAA	AACATGCTGGTTGTATGAAGTAAAGTG	76	XM_013964678.2	Brenaut *et al*. 2014
κ -casein (CSN3)	AGGTGCAATGATGAAGAGTTTTTTC	CCCAAAAATGGCAGGGTTAA	66	NM_001285587.1	Brenaut *et al*. 2014
Interleukin 8 (IL-8)	TGAGAGTGGGCCACACTGC	CACAACCTTCTGCACCCACTT	103	JN559767.1	Brenaut *et al*. 2014
Serum amyloid A3 (SAA3)	CTGGGCTGCTAAAGTGATCAGTAAC	CCCTTGAGCAGAGGGTCTGTGATT	69	XM_018043001.1	Brenaut *et al*. 2014
Interleukin1 β (IL-1 β)	GACAACAAGATTCCTGTGGCC	TCTACTTCCTCCAGATGAAGTGT	101	XM_013967700.2	Brenaut et al. 2014
Bactenecin7.5 (BAC7.5)	GATCCATCCAATGACCAGTTT	TTGGCCTTGGCAAACGT	91	NM_001285545.1	Jarczak et al. 2014
Bactenecin 5 (BAC5)	CAGTCACCTTGGATCCATCCA	CAGGAAATGGTCCTATGGGT	192	NM_001285577.1	Jarczak et al. 2014
Bactenecin 5 (BAC5)	GAATGGGCTGGTGAAACAGT	AGAGGTCTTCCCTGGGCTTA	274	XM_018066702.1	Simgene.com
Bactenecin 5 (BAC5)	GAGCGGTCCTCAGAAGCTAA	ACTGTTTCACCAGCCCATTC	181	NM_001285577.1	Simgene.com
Cyclophilin A (PPAI)	TGACTTCACACGCCATAATGGT	CATCATCAAATTTCTCGCCATAGA	62	NM_178320.2	Brenaut et al. 2014
β -defensin 2 (GBD2)	CTCAAGGAATAATAAATCA	CATTTTACTGGGGGCCCGTG	110	AJ009877.1	Zhao et al. 1999
Hepcidin (HAMP)	ACCTGCCTTCTGCTCCTTGT	CTCCAGCTGTGTGCTGAGTTT	103	XM_013971234.2	Jarczak et al. 2014
Hepcidin (HAMP)	CCACTTTCCCATCTGCATCT	GGAGGTACTGCGGGTAGACA	119	XM_013971234.2	Simgene.com

Chemokine 4 (CCL4)	CAGCCGTGGTATTCCAGACC	CTGGGAGCAGCTCAGTTCAGT	109	XM_005693171.3	Brenaut et al. 2014
Tumor necrosis factor α (TNF α)	CAGAGGGAAGAGCAGTCCCC	TGGGCTACCGGCTTGTTATTT	101	NM_001286442.1	Brenaut et al. 2014
Toll-like receptor 2 (TLR2)	TAAACTTGAGAGTGGAGGTCAAATCA	TCAGAGGCTCCTTCCGTGG	101	JQ911706.1	Brenaut et al. 2014
Cathelicidin-7 (MAP34)	TGGAGCAGTGTGACTTCAGG	GTCCAGAAGTCTGAGCCAGG	258	NM_001285620.1	Simgene.com
Cathelicidin-7 (MAP34)	AGAGGTGGGCAGAAAATCCT	GTCCAGAAGTCTGAGCCAGG	112	NM_001285620.1	Simgene.com
Cathelicidin-6 (MAP28)	TGAACATCACCTGCGAAGAG	CAGAATCCAGAAGCCTGAGC	179	NM_001285546.1	Simgene.com

Supplementary Table S3. Real-Time PCR program for primer annealing

Gene symbol	Primer annealing		
	Temperature (°C)	Time (s)	No. of cycles
α S1- casein (CSN1S1)	62	15	45
α S2- casein (CSN1S2)	62	15	50
κ - casein (CSN3)	62	15	50
Interleukin 8 (IL-8)	60	15	40
Serum amyloid A3 (SAA3)	62	15	50
Interleukin 1 β (IL-1 β)	62	15	50
Bactenecin 7.5 (BAC7.5)	60	15	45
Bactenecin 5 (BAC5)	60	15	45
Bactenecin 5 (BAC5)	60	15	50
Bactenecin5 (BAC5)	56	15	45
β - defensin 2 (GBD2)	60	15	45
#Hepcidin (HAMP)	60	15	45
^Hepcidin (HAMP)	60	15	45
Chemokine 4 (CCL4)	62	15	50
Tumor necrosis factor α (TNF α)	62	15	50
Toll-like receptor 2 (TLR2)	62	15	50
Cathelicidin-7 (Map34)	60	15	50
Cathelicidin-7 (MAP34)	62	15	50
Cathelicidin-6 (MAP28)	62	15	50

Supplementary Table S4. Least square means and their standard errors (SE) of biochemical parameters in blood serum from inorganic and organic Se-treated of dairy goats subjected to inorganic and organic treatments, at the beginning (top within cell) and at the end (bottom within cell) of experiment

Trait	Day of lactation	Treatment		SE	Reference values# [Unit of measure]
		Inorganic	Organic		
Glucose	21	59.8	64.6	3.9	54-93
	180	63.9	55.6		[mg/dl]
Aspartate aminotransferase (AST)	21	173	150	9.4	122-321
	180	158	144		[U/l]
Alanine aminotransferase (ALT)	21	35.5	30.3	2.5	23-44 [U/l]
	180	33.5	27.7		
Alkaline phosphatise (ALP)	21	104	114	12.3	75-228[U/l]
	180	141	103		
Gamma-glutamyltransferase (GGT)	21	65.6	69.4	4.1	60-101 [U/l]
	180	67.1	63.9		
Lactate dehydrogenase (LDH)	21	939	889	39.0	811-1250
	180	879	855		[U/l]
Albumins	21	32.2	33.0	1.3	25-44 [g/l]
	180	31.5	28.1		
Total cholesterol (TCHOL)	21	81	108	11.6	61-108
	180	107	101		[mg/dl]
Low density cholesterol (LDL)	21	23.7	27.6	3.2	[mg/dl]
	180	33.4	25.9		No information
High density cholesterol (HDL)	21	58.6	61.6	5.5	[mg/dl]
	180	30.2	50.5		No information
Bilirubin	21	0.33	0.34	0.06	0.1-1.9
	180	0.55	0.52		[mg/dl]
Triglyceride (TRIG)	21	11.7 A	16.5	2.6	9-27
	180	25.6 B	17.9		[mg/dl]
Creatinine (CRE)	21	1.40	1.38	0.04	1.0-2.2
	180	1.39	1.77		[mg/dl]

Creatinine	21	126 a	119	24.1	28-130 [U/l]
kinase (CK)	180	44 b	89		
Urea	21	31.5	33.7	2.6	28-94
	180	28.5	32.1		[mg/dl]
Total	21	63.7	62.7	2.92	59-78 [g/l]
protein (TP)	180	64.6	56.6		
Globulin	21	35.6	39.6	2.8	31-55 [g/l]
(GLOB)	180	38.3	36.6		
Calcium	21	9.5	10.1	0.6	8.8-12.2
(Ca)	180	8.8	9.3		[mg/dl]
Phosphorus	21	7.0	7.9	0.5	5.0-13.7
(P)	180	4.5	6.6		[mg/dl]
Magnesium	21	2.7	3.1	0.2	1.8-4.0
(Mg)	180	2.4	2.1		[mg/dl]
Sodium	21	159	154	7.5	141-157
(Na)	180	149	141		[mmol/l]
Potassium	21	3.7	3.4	0.2	2.5-4.1
(K)	180	3.8	3.3		[mmol/l]
Chloride	21	112	108	5.2	98-111
(Cl)	180	100	93		[mmol/l]

Inorganic – sodium selenite
Organic – selenized yeast
SE – standard error
A, B – different letter within the columns indicate significant differences at ($p<0.01$)
a, b – different letter within the columns indicate significant differences at ($p<0.05$)
#according to Winnicka (2004) – reference values of biochemical parameters in goat blood serum according to the International System of Units.

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