The Influence of Mineral Fertilizer on the North-Eastern Romania Permanent Grassland as Investigated by Epithermal Neutron Activation Analysis

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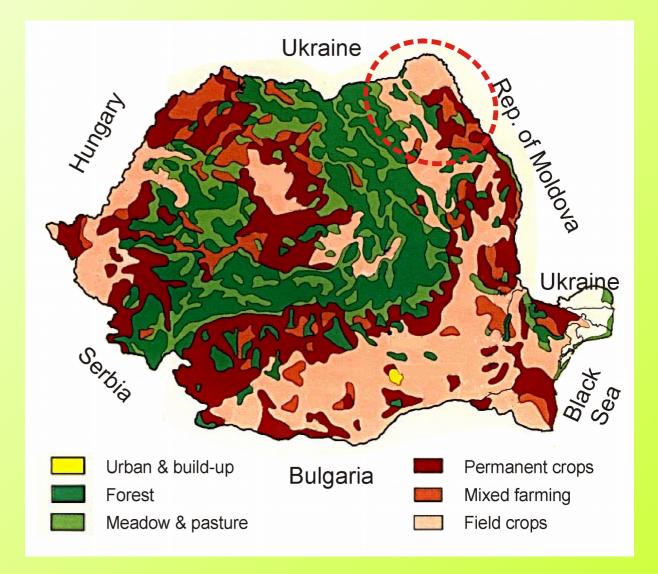
Romania is located in Southeastern Europe, bordering on the Black Sea, the country is halfway between the equator and the North Pole and equidistant from the westernmost part of Europe—the Atlantic Coast and the most easterly—the Ural **Mountains**

Various climatic zone from Humid subtropical to Subarctic and Tundra with an average temperature varying between + 11 °C South and + 8 °C North

Average precipitations are of 589 mm while average relative humidity is of 70 %



As climate and altitude play a paramount role in land using, Romania, from a total of 14.8 Mha agriculture classes, the exploitable grassland (pasture & hayfield) covers about 4.9 Mha



Pasture (kha)	Meadow (kha)	Total (kha)	%
44	2	46	2
154	21	175	8
227	131	358	17
311	291	602	30
488	375	863	43
1244	820	2044	100
	44 154 227 311 488	44 2 154 21 227 131 311 291 488 375 1244 820	44 2 46 154 21 175 227 131 358 311 291 602 488 375 863

* Mountain areas (land over 500 m) occupy about 1/3 of the country

Pastures, which cover about 60 % of the mountain area (18 % of entire country) are in many places overgrazed.

As the number of cattle, sheep and goat is continuously growing, fertilizers appear as the best solution

Organic Fertilizers healthy growing environment

- Add natural nutrients to soil
- Increase soil organic matter
- Improve soil structure
- Improve water holding capacity
- Reduce soil crusting and erosion from wind and water

• Consistent, but slow release of nutrients.

• Non uniform distribution of nutrients

• Cheaper per pound but less richer in essential nutrients

Chemical Fertilizer rapid nutrition

• Rich in essential nutrients needed for plants

• Always ready for immediate supply of nutrients to plants if situation demands.

- Some have acid content
- Long term administration changes soil fertility
- Apparently cheaper as pack more nutrients per pound of weight

• May contain high level of harmful components

Before making a choice between organic and mineral fertilizers, a short differential analysis could be of interest

Major elements in forages for ruminant nutrition

Sodium and chlorine - to maintain proper acidity levels in body fluid and pressure in body cells

Magnesium is necessary for the utilization of energy in the body and for bone growth

Phosphorus is the essential component of ATP, the universals energy vehicle in mammals, and not only

Calcium - blood clothing, of muscles contraction, proper functions of numerous biochemical reactions in the body

Potassium - maintains proper acidity levels in body fluids, optimal pressure in body cells as well as participate to the enzymatic reactions in carbohydrate metabolism and protein synthesis

Trace elements in forages for ruminant nutrition

Iron is an essential part of hemoglobin

Cobalt is necessary for the microorganisms in the rumen to synthesize vitamin B12

Copper deficiency can result in anemia, depigmentation in hair, infertility, scouring, and cardiac failure

Zinc affects growth rate, reproduction, skeletal development, the utilization of protein, carbohydrates and fats

Manganese is essential for the utilization of carbohydrates

Molybdenum forms an essential part of some enzymes and also have a stimulating effect on fibre-digesting microorganisms in the rumen

Selenium deficiency may result in nutritional muscular dystrophy

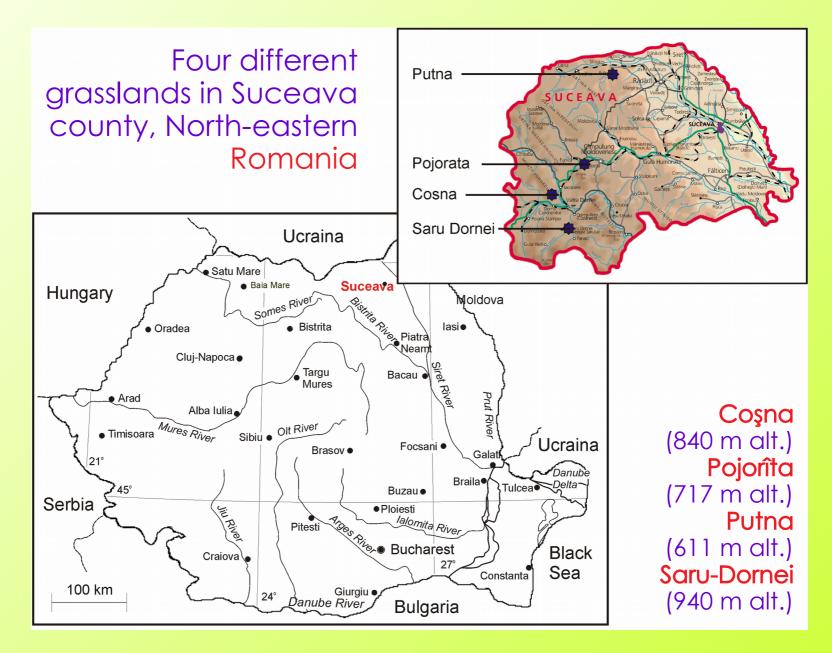
The Aims

Main Goal:

To characterize the quality of permanent grasslands in a potential area of ecological agricultural production in North-Eastern Romania by evaluating the content of essential and potential pollutant elements in order to optimize ruminants nutrition

Specific question:

- Do the organic and mineral fertilization influence the intake of essential minerals into forages ?
- Do the investigated forages fulfill the dietary requirements ?



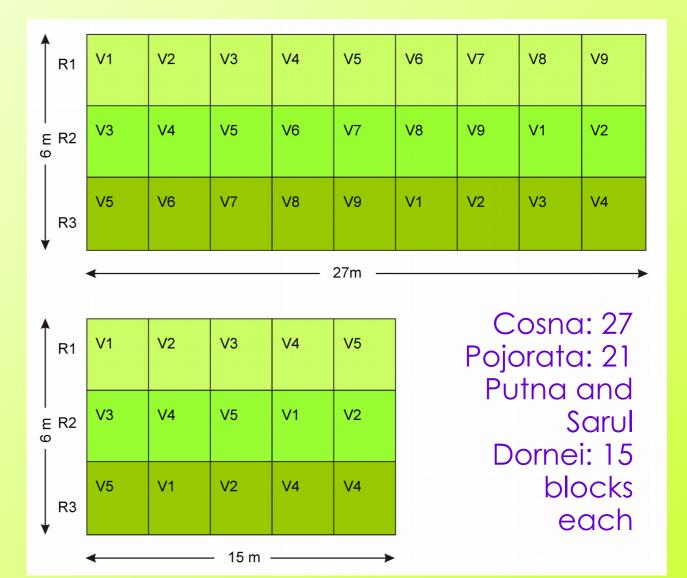


Three different species of grasses, well appreciated by ruminants (cattle, goat, sheep) and commonly found on mountai grasslands

Locality Grass species	Natural fertilizer (tons/ha)	Chemical Fertilizer (kg/ha)
Cosna Nardus stricta L	20 to 50 t of manure applied annually or bi-annually (1 – 4 y)	$\begin{array}{c} N_{100}P_{100};N_{140}P_{140};N_{200}P_{200}\\ N_{100}P_{100}+N_{40}P_{40}\\ N_{100}P_{100}+N_{100}P_{100}\\ N_{80}P_{80}+N_{60}P_{60} \end{array}$
Pojorata Agrostis capillaris L Festuca rubra L. Nardus stricta L.	10 to 50 t of manure applied annually, bi- annually (1 – 4 y)	30 kg -50 kg mineral nitrogen + 10 to 30 t of manure applied annually, bi- annually or every three years
Putna Agrotis capillata L, Festuca rubra L	20 to 50 t of partially or totally fermented manure	
Sarul Dornei Festuca rubra L, Nardus stricta L	20 t to 30 t of manure applied annually and bi- annually 50+0+40+0 t during four years	

The experiments were set up in 2006 as a random block system of 5 to9 plots of land in 3 replications. Plants were collected and analyzed in 2010 and 2011

The experiments were set up in 2006 as a random block system of 5, 7 or 9 plots of land in 3 replications Plants were collected and analyzed in 2010 and 2011



Contents of major and trace elements

Na, Mg, Al, Cl, K, Ca, Sc, V, Cr, Mn, Ni, Fe, Co, Zn, Se, As, Br, Sr, Rb, Mo, Sb, Ba, Cs, La, Sm, Hf, Ta, Th, and U were determined by Epithermal Neutron Activation performed at IBR-2 reactor of JINR P content was determined by ICP-OAS (λ = 420 nm)

Forage quality chemical parameters

Ash (A), % – gravimetrically by muffle furnace at 550 °C ignition Crude protein (CB), % – Kjeldahl method Ether extract (EE), % - Soxhlet method Acid Detergent Fiber (ADF), % - Van Soest method Neutral Detergent Fiber (NDF), % – Van Soest method Sulfuric lignin content (ADL), % – Van Soest method

Major and trace elements – unfertilized parcel

Chronic deficit of Na, partially of Cl and Mo (only Cosna). No elements above the maximum tolerable range

Element	BR	RR	AR	ATL	BR	RR	AR	ATL				
Liement		Co	osna		Pojorîta							
	Major elements											
Na	100	-	-		100	-	-	_				
Mg	-	38	62	12	-	-	100	7				
Cl	10	56	34	-	47	53		-				
K	3	28	69	-	-	-	100					
Ca	9	47	44			20	80	-				
			Т	'race eler	nents							
Mn		-	100	9		-	100	-				
Fe	-	-	100	9	-	-	100	3				
Co		28	72		7	50	43	-				
Zn	-	56	44		3	97						
Se	-	97	3		-	73	27					
Mo	41		59				100					

BR below requirements; RR in requirements range; AR above requirements range; ATL above maximum tolerable range

Major and trace elements – fertilized parcel

	Element -	BR	RR	AR	ATL	BR	RR	AR	ATL		
Chronic	Element		Sarul	Dornei			Putna				
deficit of	Major elements										
Na, and	Na	100			-	100					
Mo.	Mg	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	30	70			80	20			
	Cl		30	70			40	60			
No	K	-	-	100		19-40		100			
elements	Ca	9	_	100		10	90	30	-		
above	- La derina de sua de la desta de selas Las desentes en la della de selas de se			Т	race elen	nents	in i dei i - An i - A Sul i - Sul i - An i - A	ent for first for first for first and for short for share for share	 Jud D. Stal D. Stal D. Stal D. Stal Stal D. Stal D. Stal D. Stal D. Stal 		
the	Mn		-	100	-	-	-	100			
maximu	Fe	-	-	100	_			100	-		
m	Co	20	40	40	-		30	70	-		
tolerable	Zn		-	100	-		90	10	-		
	Se	_	50	50	-	-	90	10	_		
range	Mo	40		60		50		50			

BR below requirements; RR in requirements range; AR above requirements range; ATL above maximum tolerable range

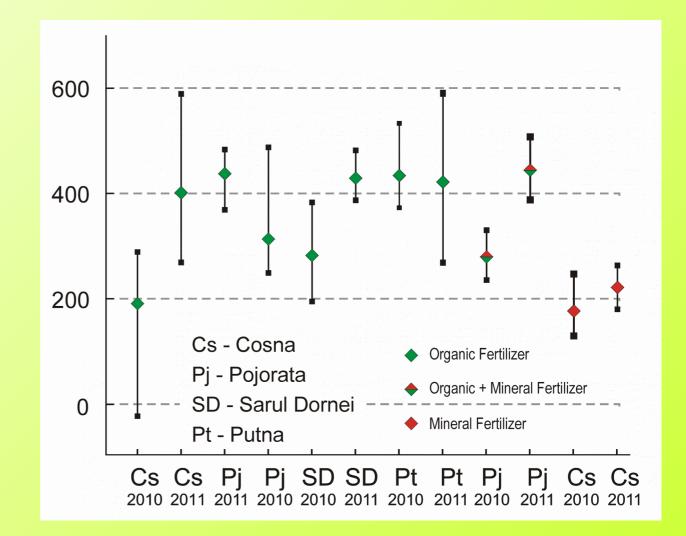
Dietary Cation-Anion Difference (DCAD): Na + K – Cl represents the main descriptor of the influence of the grass major elements on cattle lactation and meat productivity

Location	Year	Mean ± St.Dev	Min	Max
Cosna	2010	194 ± 103	-21	293
Organic Fertilizers	2011	271 ± 88	401	593
Cosna	2010	181 ± 40	129	247
Mineral Fertilizers	2011	255 ± 30	225	265
Pojorata	2010	441 ± 44	366	493
Organic Fertilizers	2011	315 ± 75	244	488
Pojorata	2010	284 ± 16	269	306
Organic + Mineral Fertilizers	2011	449 ± 38	380	509
Sarul Dornei	2010	287 ± 71	196	359
Organic Fertilizers	2011	432 ± 42	386	483
Putna	2010	436 ± 67	369	535
Organic Fertilizers	2011	423 ± 21	401	451

In dry cows, a negative DCAD can help prevent metabolic problems; in lactating cows, a positive DCAD help increase milk production and milk components

Final results concerning the influence of fertilizer type on the DCAD evolution during experiment

Excepting one single case (Cosna, 2010), all other experiments with exclusive or blend organic and mineral fertilization indicate a higher DCAD than mineral fertilization



Final results concerning the influence of fertilizer on the content of major and some essential trace elements

Locality	Fertilizer	Na	Mg	Cl	K	Ca	Mn	Fe	Со	Zn	Se	Mo
Cs	0/2010										nsd	
Cs	0/2011		nsd	nsd				nsd				
Pj	0/2010			nsd			nsd	nsd	nsd		nsd	nsd
Pj	0/2011			nsd	nsd			nsd	- 		-	nsd
SD	0/2010	nsd	nsd	nsd	nsd				nsd	nsd		nsd
SD	0/2011		nsd		nsd	nsd	nsd			nsd		nsd
Pt	0/2010		nsd	nsd				nsd	nsd			nsd
Pt	0/2011	nsd	nsd			an an taon an taon An an taon an t	nsd					nsd
Pj	(O+M)/2010		nsd	nsd	nsd	nsd		-	nsd	nsd	<u> </u>	nsd
Pj	(O+M)/2011	nsd	nsd	nsd	nsd	nsd				nsd		nsd
Cs	(O+M)/2010			nsd	nsd	nsd			nsd	nsd	nsd	
Cs	(O+M)/2011							nsd	nsd		· · ·	

Cs – Cosna, PJ – Pojorata, SD – Sarul Dornei, Pt – Putna; nsd – no significant difference, horizontal line - below detection limit; O – organic fertilizer, M – mineral fertilizer; all results were significant at p < 0.01 according to Student's t test

CI, K and Ca have quite similar behavior The most significant differences are in Cosna region, most probable due to differences in geographic position and soil properties

Final results concerning the content of some essential trace elements in all investigated grass species

Zlomont -	Cos	na	Pojo	rita	Saru-I	Dornei	Put	na	NATT	
Element -	min	max	min	max	max min max		min max		- MTL	
Al	90	1750	84	1370	157	1030	143	324	1000	
V	0.4	2.2	<0.2	1.7	<0.4	1.7	0.5	1.0	50	
Cr	<1.7	11	1.6	7.3	<2.3	6.1	2.9	4.9	100	
Ni	<1.2	4.8	1.4	5.2	<1.8	6.0	3.4	7.6	100	
As	0.6	3.4	0.4	2.8	0.8	2.6	0.9	2.0	30	
Sr	13	48	9	209	28	51	21	41	2000	

MTL Maximum Tolerable Level

Only the Aluminum content has overpasses the Maximum Tolerable Level

Final results concerning the influence of organic fertilization on chemical composition of forages obtained from *Nardus stricta* L. grasslands

Fertilization variant	СР	Α	EE	NDF	ADF	ADL
V1 Unfertilized control	6.4	5.4	1.6	75	47	11
V2 20 t ha-1 every year	9.3	9.8	2.0	60	38	10
V3 30 t ha-1 every year	12	9.0	1.6	55	37	10
V4 40 t ha-1 every year	9.7	11	1.9	50	38	10
V5 50 t ha-1 every year	11	10	2.2	54	41	10
V6 20 t ha-1 every 2 years	12	8.8	1.7	57	38	10
V7 30 t ha-1 every 2 years	12	9.0	1.3	58	39	9.5
V8 40 t ha-1 every 2 years	13	9.4	1.3	56	39	9.6
V9 50 t ha-1 every 2 years	13	10	1.5	55	43	9.2
LSD at p <0.05	0.9	1.9	0.2	1	1	0.4

CP - crude protein; A – ash; EE - ether extract; NDF- neutral detergent fibre; ADF - acid detergent fibre; ADL -sulphuric lignin content; LSD – Lowest Significant Difference

Final results concerning the influence of mineral fertilization on chemical composition of forages obtained from *Nardus stricta* L. grasslands

Fertilization variant	СР	Α	EE	NDF	ADF	ADL
V1 Unfertilized control	6.5	7.6	2.7	72	46	10
$V2 N_{100}P_{100}$	6.6	6.3	3.0	59	34	9.6
$V3 N_{140}P_{140}$	8.1	6.2	3.1	59	33	9.8
$V4 N_{200}P_{200}$	10	7.1	3.2	52	33	9.5
$V5 N_{100}P_{100} + N_{40}P_{40}$	11	6.0	3.0	55	34	9.7
$V6 N_{100}P_{100} + N_{100}P_{100}$	14	7.3	3.3	54	32	10.5
$V7 N_{80}P_{80} + N_{60}P_{60}$	11	6.2	2.6	59	40	9.6
LSD at p <0.05	0.2	0.3	0.2	1.2	0.7	0.4

CP - crude protein; A – ash; EE - ether extract; NDF- neutral detergent fibre; ADF - acid detergent fibre; ADL -sulphuric lignin content; LSD – Lowest Significant Difference

Summary statistics regarding the content of major and trace elements at the end of 2010 – 2011 experiment (83 samples, ENAA results)

	(Cosna		Po	jorita		Saru	-Dorne	ei]	Putna		_	
Ele- ment	non-	fertil	ized	non-	fert	ilized	non-	ferti	ilized	non-	ferti	ilized	RDMR	MTL
mone	fertilized	min	max	fertilized	min	max	fertilized	min	max	ferti- lized	min	max		
DACD elements (mg/kg)														
Na	84 ± 52	51	248	95 ± 25	70	176	<mark>70</mark> ± 17	37	82	75 ± 19	61	107	1000-2200	40000
Cl	1960 ± 1300	634	6290	1350 ± 490	732	2760	$\begin{array}{c} 2450 \pm \\ 2100 \end{array}$	1950	8020	1750 ± 1700	1660	5670	1300-2900	40000
K	9500 ± 4500	1520	26600	16500 ± 3600	10300	22200	16200 ± 3500	13200	23200	16800 ± 3700	16100	26500	4700- 10000	30000
Metabolic and structural elements (mg/kg)														
Mg	2800 ± 2400	1100	1140	5700 ± 1300	2520	7160	$\begin{array}{c} 2500 \pm \\ 800 \end{array}$	1700	4100	1900 ± 550	1140	2990	1100-2100	6000
Р		700	4300		880	1740		1600	2800		1200	3300	2200-3800	7000
Ca	5150 ± 2600	2700	13200	1420 ± 4700	3920	19200	8650 ± 830	7950	11100	4100 ± 1550	3250	7630	3700-6200	18000
					Enz	ymatic	elements (1	ng/kg))					
Mn	550 ± 350	160	1410	87 ± 18	43.4	122	285 ± 40	197	323	540 ± 90	357	618	13-24	1000
Fe	300 ± 180	60	811	310 ± 150	<60	567	250 ± 130	<83	408	$165\pm\!85$	<86	330	12-40	500
Co	0.23 ± 0.10	0.12	0.45	0.21 ± 0.11	<0.06	0.30	0.1 ± 0.05	0.09	0.21	0.20 ± 0.10	0.15	0.24	0.10-0.15	25
Zn	70 ± 15	40	99	45 ± 3	<2.2	52	70 ± 10	57	82	48 ± 6	37	64	21-55	500
Se	0.10 ± 0.05	<0.14	0.34	0.20 ± 0.10	0.13	0.44	bdl	<0.24	0.38	bdl	<0.25	0.34	0.1-0.3	2
Mo	0.48 ± 0.41	<0.01	1.88	0.70 ± 0.10	0.24	0.89	0.10 ± 0.05	5 0.10	0.40	0.10 ± 0.10	<0.16	0.50	0.2	5
					Poter	tial tox	ic elements	(mg/l	(g)					
Al	510 ± 44	to 90	1750	520 ± 390	84	1370	600 ± 330	157	1030	200 ± 50	143	324		1000
As	2.0 ± 1.0	0 0.6	3.4	1.0 ± 0.05	0.4	2.8	2.0 ± 1.0	0.8	2.6	1.5 ± 0.5	0.9	2.0		30
Sr	20 ± 9	13	48	bdl	9	209	bdl	28	51	bdl	21	41		2000

RDMR Recommended Dietary Mineral Requirements

> MTL Maximum Tolerable Level

Concluding remarks

- The content of major elemets, except for Na, CI and Mo (some localities) are in the requirement range or above.
- Only few grass samples contain essential minerals Mg, Mn and Fe in concentrations slightly higher than maximum tolerable limits
- The content of non-essential elements (except Al in very few cases) were well below the maximum tolerable limits
- The use of organic or blended organic and mineral fertilizers showed, excepting Cosna location, higher DCAD than mineral fertilizer ones
 - The fertilization of *Nardus stricta* L. grasslands with organic and mineral fertilizers significantly improves its quality
 - No significant differences between the use of organic and mineral fertilization were observed for the essential grass chemical parameters

Thank you for attention

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