University of Montenegro
Biotechnical Faculty – Podgorica
Department of Livestock Science

FAO and ERFP Workshop on *In situ* and *Ex situ* gene conservation

In-situ and *Ex-situ* Gene conservation of AnGR in Montenegro

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Presentation outline

- Introduction - background
- Current state of farm animal genetic resources
- In-situ conservation of AnGR
- The main research results
- Ex situ conservation of AnGR
- Challenges and recommendations

Background – general trends

- General trends of development of livestock production imply uniformity of animal genetic resources, caused by loosing endangered breeds and increased inbreeding within commercial breeding populations.

- Very intensive development of livestock sector globally have caused big changes:
  - Highly productive breeds have been created but less resistant, with shorter productive life and lower fertility;
  - Low productive breeds were become non competitive, their population size have rapidly been reduced, while many of them disappeared completely.

Trends of AnGR in Montenegro

- High economic importance of livestock sector for Montenegro (the biggest sector in the agriculture ≈ 50% of total value in agricultural production),
- During the last 50 years: global trends were present in the Montenegrin livestock sector causing many changes.
- Local breeds have been significantly decreased, especially local cattle breeds, pig and poultry populations
- In sheep and goat sectors there are still local breeds, however general trend is that the size of their populations are decreasing
- Challenge: How to preserve farm animal genetic diversity, specialy autochthonous – low productive breeds, knowing all new tendencies ?

Actaul status of animal genetic resources in Montenegro

Species	Population size (2014)	Exotic breeds	Autochthonous (native) & local breeds
Cattle	93550	- Holstein, - Brown Swiss, - Simmental	- Busha - Grey cattle
Sheep	204400	- Württemberg, - Tsigaja, - Romanovska	- Pivska pramenka
			- Bardoka
			- Ljaba
			- Žuja
			- Sora
			- Sjenička sheep
Goat	33000	- Alpine - Saanen	Balkan red goat

Actaul status of animal genetic resources in Montenegro

Species	Population size (2014)	Exotic breeds	Autochthonous or local & native breeds
Horse	**4980**	**- Arabian horse, - Thoroughbred , - Cold blood breeds**	**Montenegrin hilly horse**
Donkey	**< 1000**	-	**Domestic grey donkey**
Pig	**22050**	**- Landrace, - Yorkshire, - Duroc and - Pietren**	-
Poultry	**623000**	Lines of: **Rhode Island and White Plymouth Rock**	Local varieties of hens: **grašasta, jarebičasta** etc.
Bee (Beehives)	**42300**		Kranjska bee - **Apis melifera carnica**

Population size and trends of autochthonous breeds in MNE

Species	Autochtonous and local breeds	Number of breeding animals	Trends
Cattle	Busha	up to 250	↓
Sheep	- Pivska pramenka	≈ 5000	↓
	- Bardoka	≈ 2500	↔
	- Ljaba	≈ 1500	↓
	- Žuja	up to 150	↓
	- Sora	≈ 1500	↔
	- Sjenička sheep	8000	↑
Goat	Balkan red goat	> 5000	↔
Horse	Montenegrin hilly horse	≈ 2000	↓
Donkey	Domestic grey donkey	< 500	↓
Pig	Šiška pig	** extinct	
Poultry	Grašasta & jarebičasta	** no data	

Busha cattle in Montenegro

Autochthonous sheep breeds

Pivska pramenka

Žuja

Sora

Bardoka

Ljaba

Autochthonous breeds: goats, horses and donkeys

Balkan red goat breed

Mne . hilly horse

Domestic donkey

THE *IN SITU* GENE CONSERVATION of AnGR – CONCEPT –

- Officially established in 2008.
- Strategic background:
  - Strategy of Agriculture and Rural development adopted in 2006 and updated 2014
  - National program of conservation and sustainable use od AnGR with the Action Plan, adopted in 2008
- Legal frame – Law on Livestock Sector, the last one from 2010
- National focal point institution was appointed - Biotechnical Faculty (University of Montenegro) is the National focal point for international activities referred to AnGR
- The Ministry of agriculture and Rural development is in charge for AnGR at governmental level

Implementation of the program of *In-situ* conservation of AnGR

The main bodies involved – responsible for the implementation:

- The Ministry of agriculture and rural development provides legal framework and financial support through the yearly budget for agriculture (for limited number of animals),
- Biotechnical Faculty was appointed to be responsible body for monitoring, inventory, characterization and conservation of AnGR at the national level;
- Farmers – breeders of autochthonous breeds are included in the program on the contractual bases with the Ministry
- Others - temporary

Number herds and animals per breed included in the program of In situ conservation in 2015.

	No. of herds	No. of female animals	Number of males	Total number of heads	Subsidies €/head/year
Buša – cattle	9	81	13	94	150€
Pivska pramenka	4	335	11	345	16€
Žuja	3	147	5	152	23€
Sora	3	245	9	254	16€
Ljaba	2	155	6	160	16€

Biotechnical Faculty as focal point institution – its unit Department of Livestock Sciences takes care of the program implementation – based on its report subsidies are paid to the farmers by Ministry of agriculture and rural development

The results of phenotypic and genetic characterization

Exterior measures of Busha breed

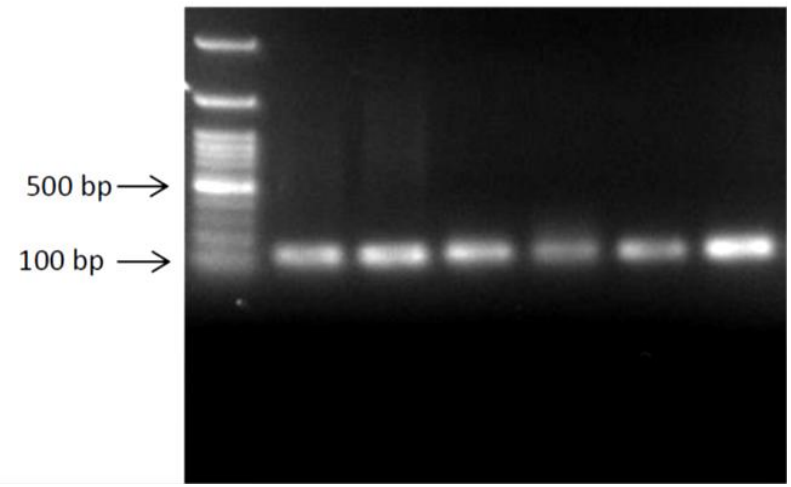
Parameter	Height to withers	Height at rump (to hips)	Chest circumf erences	Body length	Body depth	Cannon bone circumf erences	Body weight
Number of animals	40	15	39	38	11	17	38
Mean	112,9	118,1	157,7	126,6	57,1	15,8	290,8
Standard Error	0,70	0,99	1,25	1,19	0,43	0,32	5,98
Standard Deviation	4,44	3,86	7,80	7,33	1,42	1,30	36,87
Minimum	100	111,5	135	107	54	11,5	193,8
Maximum	122	123	175	142	59	17,5	353,1

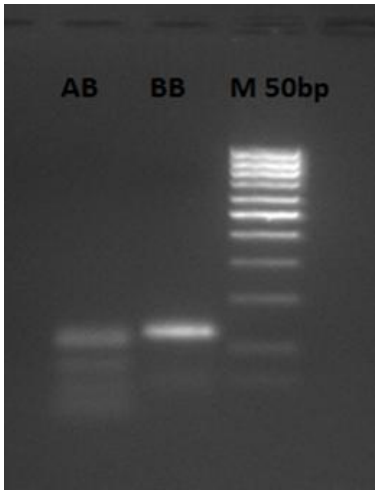
Eksterior measures of the some sheep breeds (2012-2015)

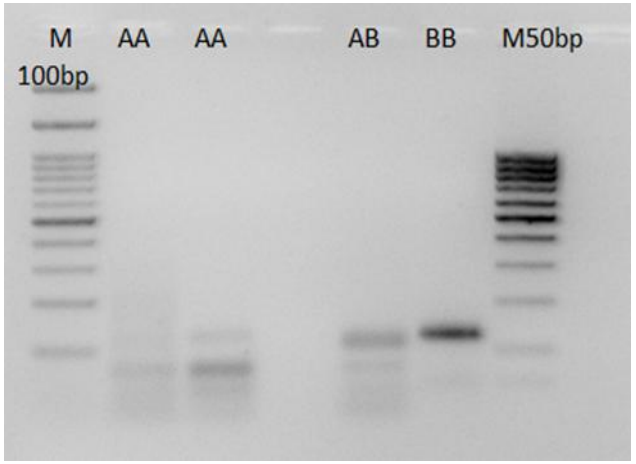
Measures	Zetska žuja	SORA	PIVSKA
Height to wither, cm	63,10	69,95	71,7
Body lenght, cm	59.41	69.92	70,7
Chest circumference, cm	81,85	99,22	100,2
Chest width, cm	15.13	20.29	21,3
Chest depth, cm	27.26	30.64	32,2
Fore leg circumference, cm	7.60	9.11	9,0
Body weight, kg	37,10	60,83	71,6

Genetic characterization of the some sheep breeds

Polimorphysm beta lactoglobulingene of three sheep breeds, PCR-RFLP





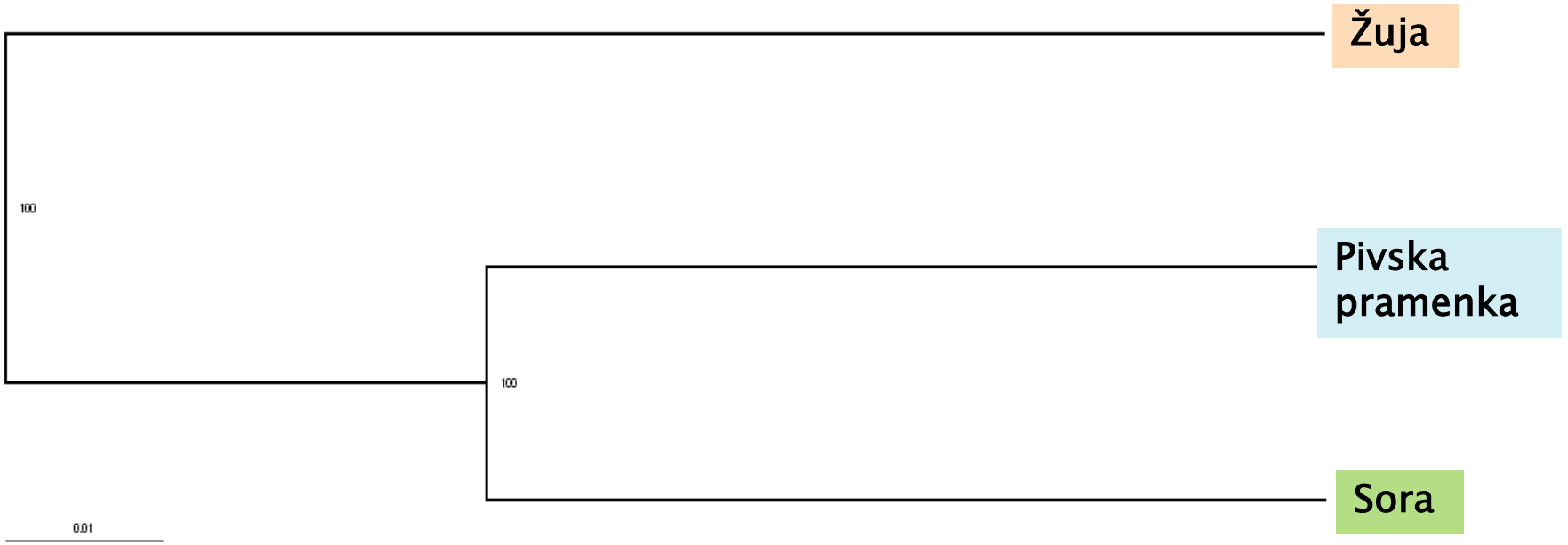


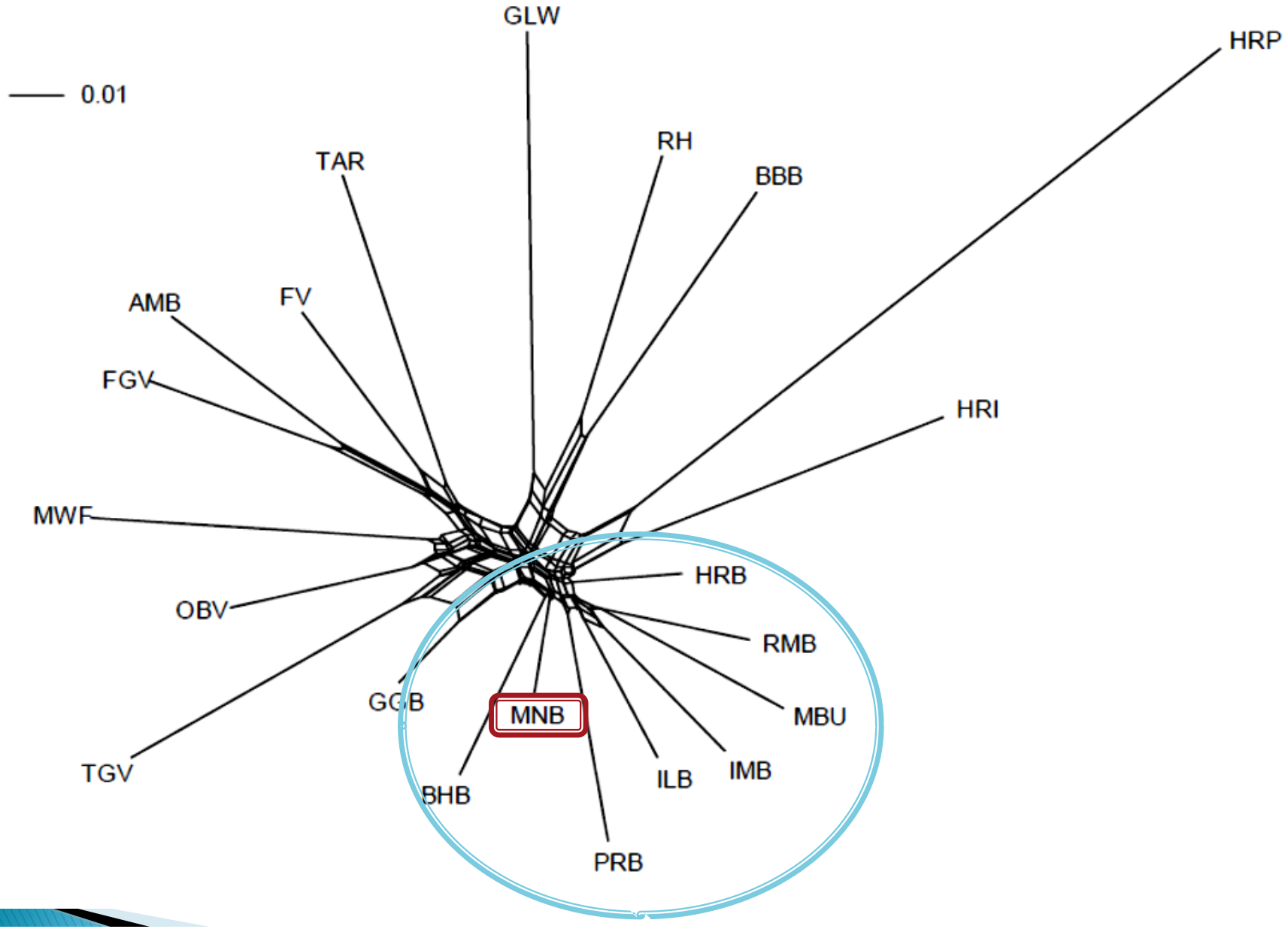
Breed	Beta – LG genotype				Frequency of genes	
	Parameters	AA	AB	BB	A	B
PIVSKA	Ho	0	16	12	0,214	0,786
	He	1,3	9,4	17,3		
	Frequency	0	0,429	0,571		
SORA	Ho	4	14	11	0,379	0,621
	He	3,1	12,8	13,1		
	Frequency	0 ,144	0,471	0,385		
ŽUJA	Ho	2	8	6	0,375	0,625
	He	2,25	7,5	6,25		
	Frequency	0,128	0,459	0,413		

Microsatellite analyses of genetic diversity of some sheep breeds

Rasa	Number of animals	H_o	H_e	n	n_e	F	PIC
PIVSKA pramenka	48	0,779	0,773	9,778	5,051	-0,003	0,745
SORA	44	0,754	0,762	9,500	4,705	0,011	0,732
ŽUJA	33	0,747	0,704	7,722	3,708	-0,064	0,668

Phylogenetic tree based on Nei's standard genetic distances





I. Medugorac et al. 2011. **Phylogenetic network of BUSHA META POPULATIONS (9) and 12 western European cattle breeds – on the base 105 microsatelite**

National data on AnGR were entered into EFABIS and regularly updated

EUROPEAN FARM ANIMAL BIODIVERSITY I

EFABIS

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Breed Data Sheets

Buša/Cattle/Montenegro

Breed names

Most common name	Buša
Language	mne
Transboundary or brand name	Busha

Breed local names

Other name	Language
Busha	English

Images

Bull of busha breed, old three year.

Year: 2009
Gender: male
Photo credit: DR. Bozidraka Markovic, Biotechnical Faculty, Department of Livestock science email: bmarkovic@t-com.me
Locality:
Niksic - Slano

Cow of busha breed

Year: 2009
Gender: fem.
Photo credit: Dr. Bozidraka Markovic, Biotechnical faculty, Department of Livestock Science. Podgorica - Montenegro
Locality:
Ulcinj - Montenegro

Origin and development

Ex-situ conservation of AnGR

- Ex situ program of conservation (national GENE BANK for AnGR) has not been established yet,
- The capacities (human and technical) for establishing of ex situ conservation are limited.
- The genetic materials of some autochthonous breeds have been collected and preserved (-20℃ or -40℃) at Biotechnical faculty, primarily for the research purposes.
- The semen of exotic cattle breeds for the purpose of AI are permanently imports.

Collected and laboratory preserved genetic material of autochthonous breeds

– number of sampled animals

Species	Breeds	Blood	Hair	DNA
Cattle	Busha	54	25	35
Sheep	- Pivska pramenka	75	x	60
	- Bardoka	53	x	45
	- Ljaba	48	x	43
	- Žuja	55	23	40
	- Sora	58	x	42
	– Sjenička sheep	52	x	45

The samples of genetic materials are permanently renewed and expands on the other breeds.

Bull semen used for AI of cows in MNE
(anual level, example of 2014)

Parameter	Breed of bull				Total
	HF	Brown Swiss	Simmental	Tyrolean Grey	
Number of doses	15415	14690	14670	2650	47425
Number of bulls used	11	8	7	3	29
Average No. of doses per bull	1401	1836	2096	883	1635
Breed share, %	32.5	31.0	30.9	5.6	100.0

OPEN ISSUES AND CHALLENGES

- Farmers readiness to implement program strictly is not at an appropriate level (subsidies are not so stimulative),
- Human resources for the Program implementation are not sufficient,
- Financial support for both aspect of AnGR (the subsidies and the institutional support) has to be increased,
- Public awareness on the importance AnGR is not at a satisfying level – it needs to be strengthen,
- Many endangered breeds are transboundary breed – thus regional initiatives should be reinforced.

THE NEXT STEPS ON IMPLEMENTATION OF THE PROGRAM

- Continuation of the current tasks and duties on implementation of the program (inventory, monitoring, characterization, databases, international cooperation etc.)
- Preparation of the new (update the current) National program of AnGR and its harmonization with FAO guidelines.
- Creating breeding program for autochthonous breeds of livestock.
- Enlarging a scope of the ***in situ*** program by increasing number of herds/flocks and animals in the program.
- Promoting native & local breeds and their products in much better way by using positive experience from other countries.

THANK YOU FOR ATENTION!