# *Beginning of standardization phenotype and genotype autochthonous breed Busha in Croatia*

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# Introduction

- Busha is one of the autochthonous cattle breeds in Croatia and it belongs to a group of primitive short horned cattle (Bos brachyceros europeus)

- 1903. Busha was dominant breed in Croatia, >92%

- 1908. start point of merging Busha with Oberintal cattle in “Krash” area and use Pinzgauer breed, Mollthalska breed and latter was included Simmental breed in west continental area of Croatia

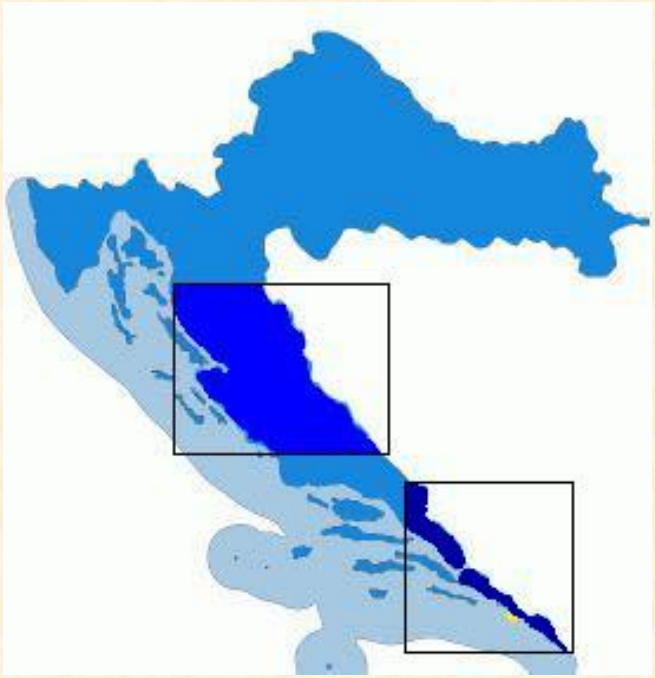
- Till the Second World Busha made more then half of total cattle population in Croatia

- Till the middle XX century
  - There was idea to preserve busha as purebred cattle breed
- Second half of XX. century
  - Idea was not realized
  - purebred of Busha almost disappeared
  - more productive cattle breeds take place instead

# Actual state of Busha population in Croatia

- **2003.** → status of **critically endangered** → list of protective breeds

- **Plan of preservation of Busha**
  - inventarisation inside the breed
  - registration individuals who were "in type"
  - making breed standards
  - establishing Herd-book (CLC)



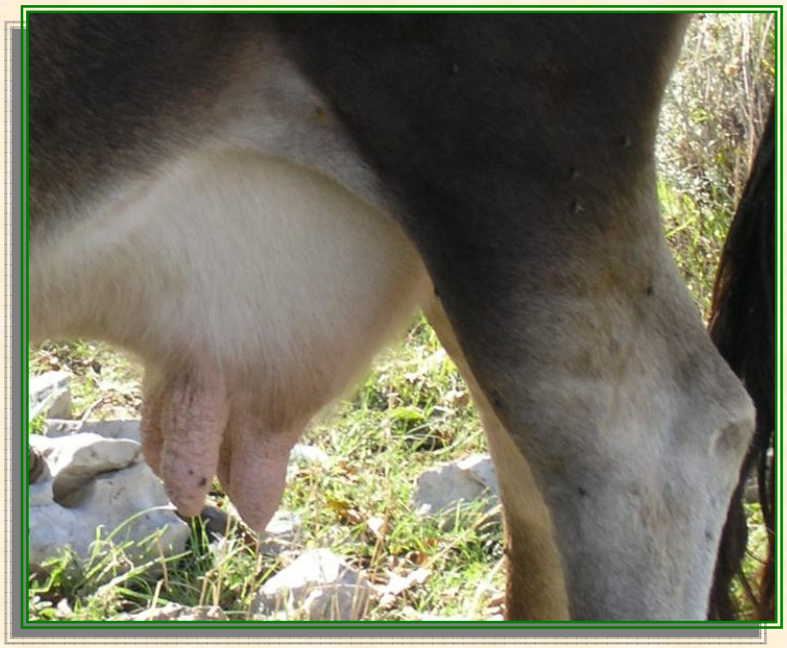
- On the area of Lika and Dalmatia we expected to fined few nucleus herds
- Sperm from 'busak' bulls collected and saved

- Busha phenotype standardisation began at 2003. At the same time started project: “Phenotype and genotype characteristics Busha in Croatia” main reasearcher prof. dr. sc. Pavo Caput

| Body measurements | $\bar{x}$ | s | Min. | Max. | μ (95%) |
|---|---|---|---|---|---|
| Withers height | 114.12 | 4.58 | 104 | 123 | 112.83-115.61 |
| Hip Height | 116.72 | 4.80 | 109 | 126.5 | 115.26-118.18 |
| Body length | 135.90 | 9.86 | 112 | 153 | 132.90-138.90 |
| Chest with | 36.13 | 3.28 | 28 | 45 | 35.13-37.12 |
| Chest depth | 60.89 | 3.74 | 50 | 66 | 59.75-62.02 |
| Rump width | 44.10 | 3.66 | 33 | 51.5 | 42.99-45.21 |
| Rump length | 45.63 | 3.30 | 38 | 51 | 44.62-46.63 |
| Chest circumference | 162.13 | 9.53 | 140 | 180 | 159.23-165.02 |
| Leg circumference | 16.15 | 1.06 | 14 | 18 | 15.83-16.47 |
| Head length | 47.60 | 2.95 | 42 | 55 | 46.70-48.50 |
| Front head length | 20.24 | 1.89 | 17 | 26 | 19.67-20.81 |
| Horn length | 22.41 | 6.74 | 6 | 38 | 20.33-24.48 |

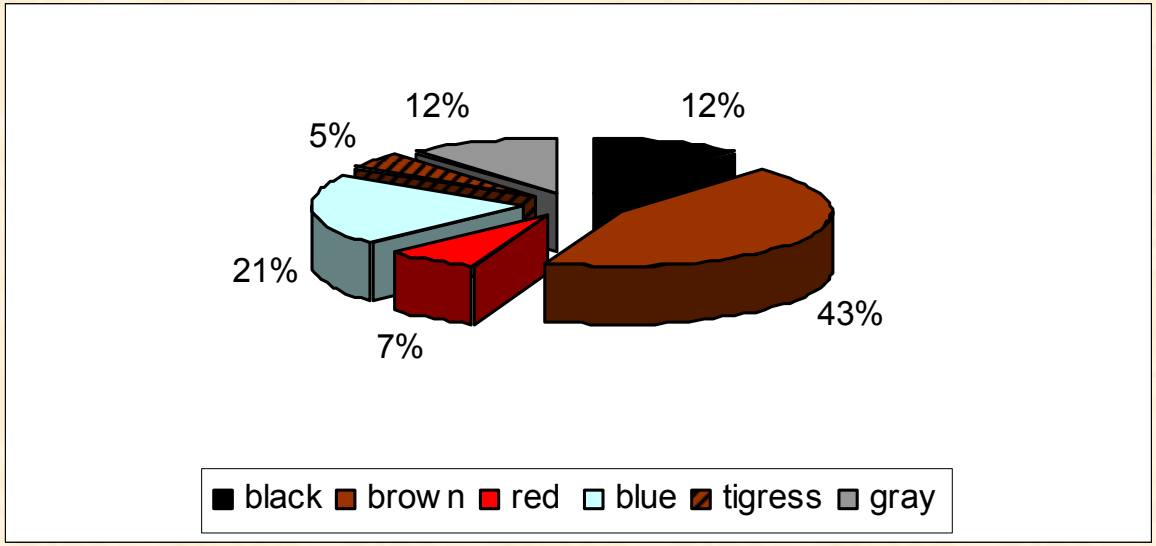
# Breed standard of Busha

- small, modest, resistant cattle
- firm legs and hoofs
- head is small and oblong, forehead small and short
- neck is long, sheet slightly developed
- wither mostly short
- chest is small and quite deep (52-57% of the withers height),
- pretty short trunk (115-120%  withers height)
- pelvis is short, peaky to the tail root, thighs are downcast



# Breed standard of Busha

- udder is poorly developed, colored, covered with dens, rough hair
- short lactation (50 to 150 days) – 600 to 1 400 liters of milk with 5,2% milk fat
- bulls usually have one third or one half of lower part of testicles in black color
- easy calving-body weight at birth (15 to 18 kg)
- modest fattening potential


12%
12%
5%
21%
43%
7%
black
brown
red
blue
tigress
gray

# Breed standard of Busha

- *deer muzzle*
- *eel-like* stripe along the back
- hoofs are darker color-almost black

# GENETIC CHARACTERISATION OF BUSHA

- Mitochondrial DNA was analysed on ten sequences
- Sequencing of the proximal part (nt 15900-16225) of the mtDNA D-loop region revealed 10 polymorphic sites
- Differences between sequences were found in 1-5 polymorphic sites
- Analysed sequences showed diversity of 0.31-1.54%, level of genetic distances from 0.0062 to 0.0156

GenBank: EU017530.1

# Bos taurus haplotype BU6 control region, partial sequence; mitochondrial

Features Sequence

```
LOCUS       EU017530                326 bp    DNA     linear   MAM 31-JUL-2007
DEFINITION  Bos taurus haplotype BU6 control region, partial sequence;
            mitochondrial.
ACCESSION   EU017530
VERSION     EU017530.1  GI:154082334
KEYWORDS    .
SOURCE      mitochondrion Bos taurus (cattle)
  ORGANISM  Bos taurus
            Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi;
            Mammalia; Eutheria; Laurasiatheria; Cetartiodactyla; Ruminantia;
            Pecora; Bovidae; Bovinae; Bos.
REFERENCE   1  (bases 1 to 326)
  AUTHORS   Konjacic,M., Ivankovic,A. and Dovc,P.
  TITLE     Analysis of the mtDNA control region of Busha cattle
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 326)
  AUTHORS   Konjacic,M., Ivankovic,A. and Dovc,P.
  TITLE     Direct Submission
  JOURNAL   Submitted (05-JUL-2007) Department of Animal Production, Faculty of
            Agriculture, Svetosimunska Cesta 25, Zagreb 10000, Croatia
FEATURES             Location/Qualifiers
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ORIGIN
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       61 ggtaatgtac ataacattaa tgtaataaag acataatatg tatatagtac attaaattat
      121 atgccccatg catataagca agtacatgac ctctatagca gtacataatc catataatta
      181 ttgactgtac atagtacatt atgtcaaatt cattcttgat agtatatcta ttatatattc
      241 cttaccatta gatcacgagc ttaattacca tgccgcgtga aaccagcaac ccgctaggca
      301 gggatccctc ttctcgctcc gggccc
//
```
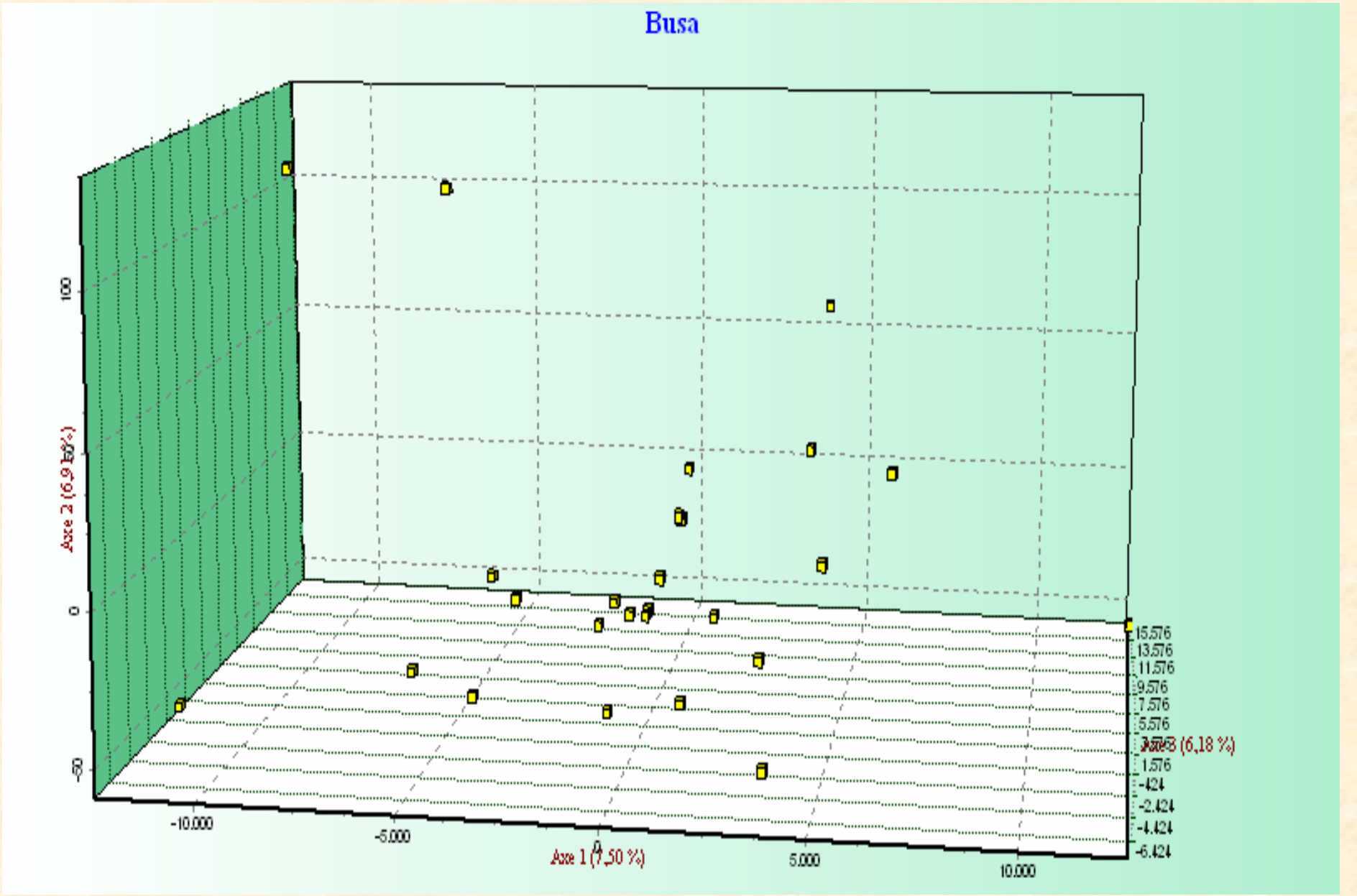

# GENETIC CHARACTERISATION OF BUSHA

- **S**imple **S**equence **T**andem **R**epeats Mikrosatellites (**SSTRs**)

- We used 30 microsatellites from *Utrecht* list from LMU München /also those are markers recommended from FAO for easier comparison genetic diversity of cattle breeds among countries)

# Busa


100
0
-50
Axe 2 (6,9
-10.000
-5.000
0
5.000
10.000
Axe 1 (7,50 %)
15.576
13.576
11.576
9.576
7.576
5.576
1.576
-424
-2.424
-4.424
-6.424
(6,18 %)

- Average number alleles per locus was 8.7 (ranged from 3 to 10)
- Average Ho (Heterozygosity observed): 0.66
  He (Heterozygosity expected): 0.74
- from Hardy-Weinberg equlibrium 11 loci deviated
- average (Polymorphism Information Content) PIC was 0.68
- average $F_{IS}$ was 0.112
- AMOVA
  - 11% variability between individuals within busa populations
  - 89% variability within each individuals of busha

# Conclusion

- For phenotype characteristics has been observed high variability. This has been largely determined by non-consolidation of the breed and non-existence of breeding guidelines in the last decades.
- The reaserch of D-loop region of mtDNA shows moderate variability of population
- Busha kept certain amount of genetic variability
- Systeatic monitoring and evaluation of breeding programs are needful


Thank you for attention