

Perspectives of the selection scheme of the Sarda dairy sheep breed in the era of genomics

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Breeding scheme of Sarda breed

Official recording:

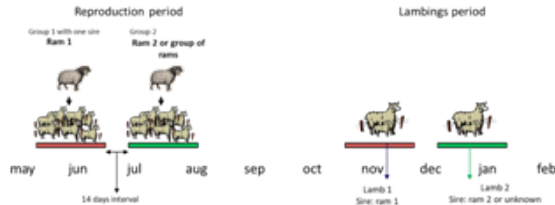
- Milk Yield
- Udder Morphology
- Scrapie resistance

Selection objectives

- Fat and Protein Content

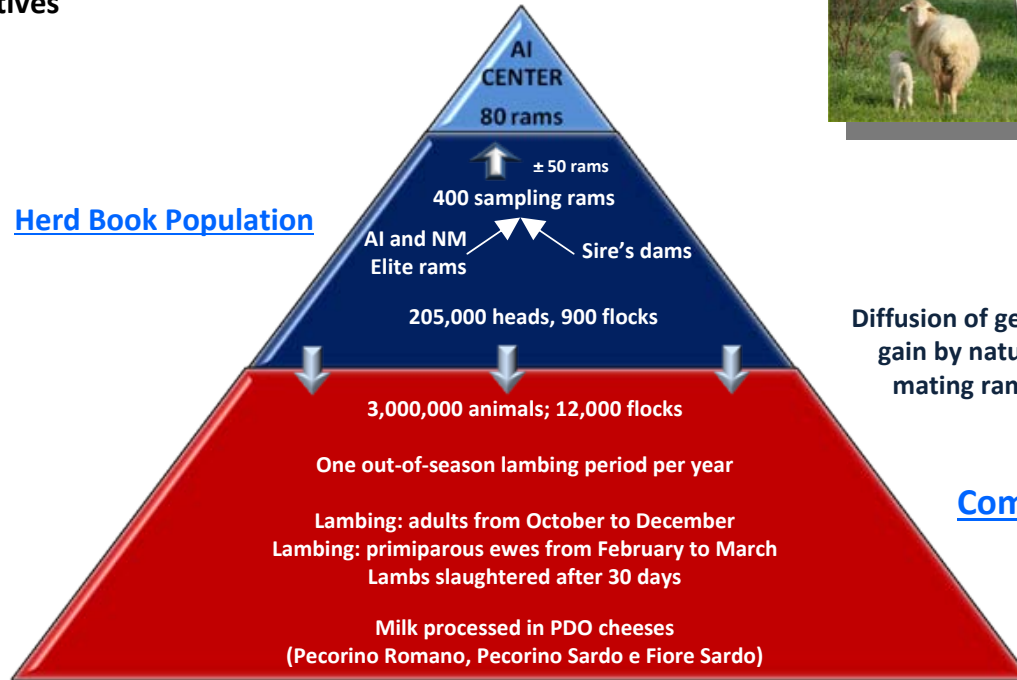
Single Sire Mating:

Management of groups of ewes with just one ram during the reproduction period



rates of AI adequate to achieve accurate genetic evaluations not only of AI rams but also of natural mating ones

around 90% of the national stock reared in Sardinia (3 million heads)



Diffusion of genetic gain by natural mating rams

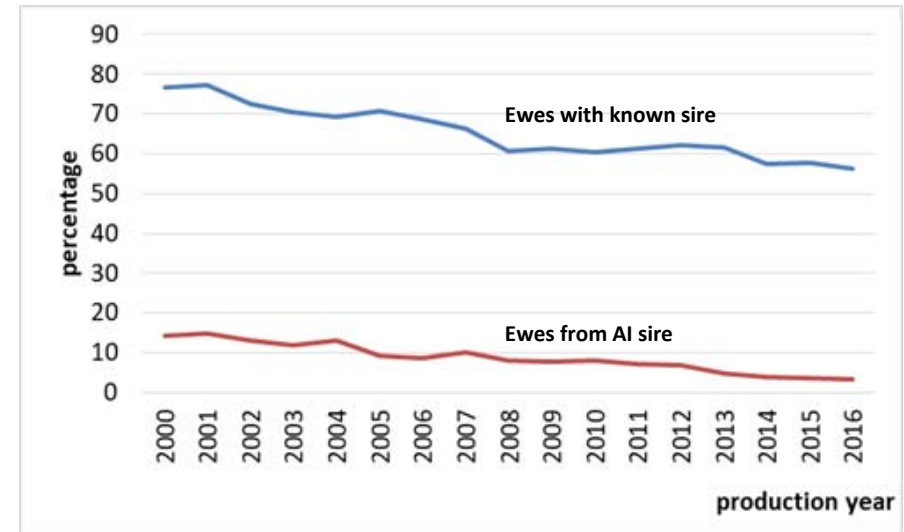
Commercial Flocks

Current state of the selection scheme: milk yield

Evolution of the effective size of the Sarda herd book

Production Year	flocks	Recorded Ewes (<= 4 yr)	Recorded ewes (1 yr)	Breeding Sires	Progeny Testing sires
2000	1'168	123'737	31'758	1'594	575
2001	1'176	126'111	33'255	1'625	482
2002	1'167	129'290	37'343	1'603	497
2003	1'157	135'858	35'781	1'617	552
2004	1'141	147'883	41'807	1'688	519
2005	1'091	142'939	35'326	1'711	501
2006	1'062	143'581	34'609	1'613	560
2007	1'075	141'320	31'197	1'449	461
2008	1'079	135'060	35'216	1'407	405
2009	1'042	127'231	31'083	1'371	390
2010	1'035	130'776	31'744	1'260	416
2011	1'025	126'920	30'491	1'204	365
2012	972	119'746	28'555	1'136	350
2013	953	117'404	26'372	1'093	368
2014	969	112'024	25'172	1'046	328
2015	943	114'877	27'482	1'048	307
2016	936	120'604	32'627	1'142	344

Percentage respect to ewes recorded for milk yield (1 yr)



Over the last decade a progressive decline of all parameters was observed

Current state of the selection scheme: milk composition and udder morphology

Recording

- on more than 50% and 40% between 2000 and 2006
- over the last years decreased to around 40% and less than 20%

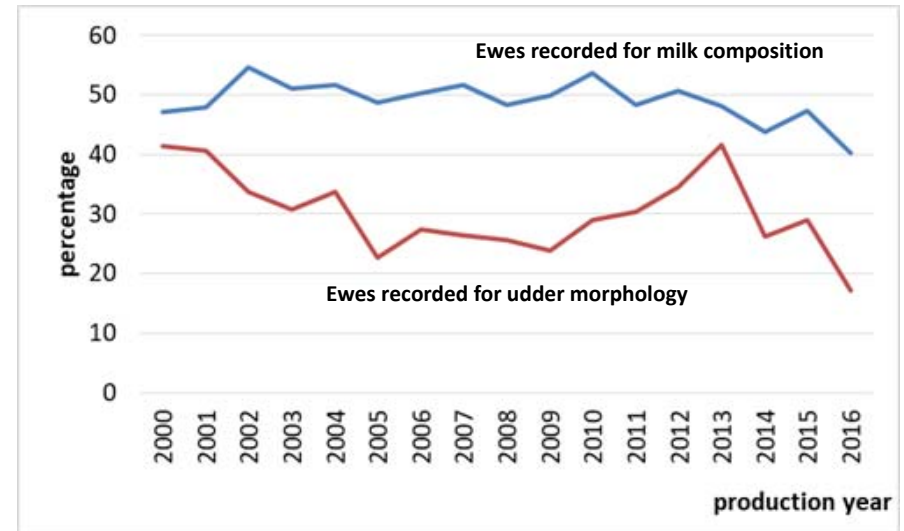
Milk composition

- low number of records per lactating yearling (on average 2.3)
- small size of the recorded population
- lower heritability (fat content) compared to literature
- no payment system based on the chemical quality of milk

Udder morphology

- limited percentage of indexed rams
- low number of scored ewes
- limitations due to organizational constraints

Percentage respect to ewes recorded for milk yield (1 yr)



effectiveness of the selection scheme

official genetic
evaluation
1983 -2016



N: 3'267'456

CG with at least five lactation records
of ewes with known sire, including
daughters of sires having offspring in at
least one other CG including daughters
of other sires

genetically
connected
population



N: 1'192'864

milk yield
heritability
estimate



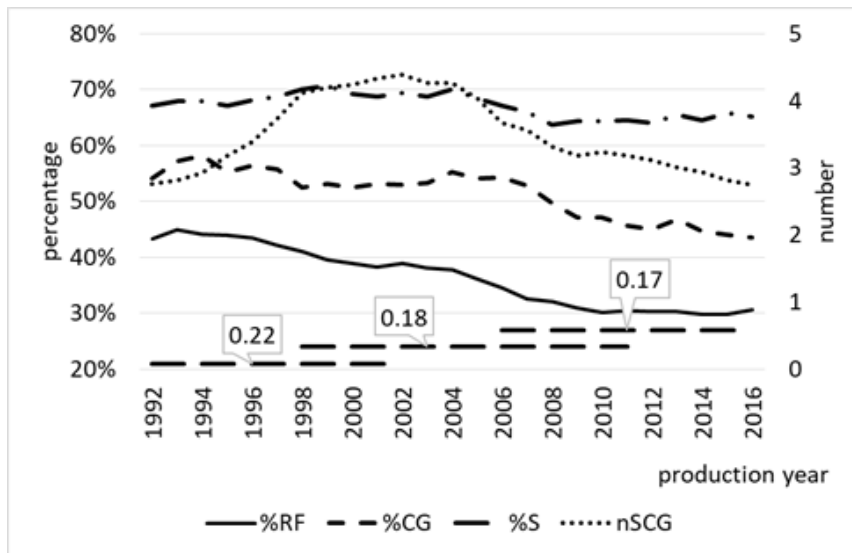
from 1983 to 2002



from 1998 to 2011



from 2006 to 2016



size of the recorded population which is actually exploitable for selective breeding

- rate respect to the official dataset of the number:
 - number of recorded ewes (%RF)
 - number of contemporary group (%CG)
 - number of sires of lactating ewes (%S)
- number of sire per CG per production year were calculated (nSCG)

Potential impact of the implementation of genomic tools

Context

- current decline of the selection scheme
- difficulty to measure on large scale innovative traits (production quality, health and sustainability)

Genomic application:

selection assisted by causal or linkage disequilibrium (LD) mutations (MAS)

- needs long trials based on large population measured for a lot of traits

genomic selection (dairy cattle approach)

- applied in some dairy breeds such as the French Lacaune
- large use of AI
- accurate recording schemes for functional and health traits



Sarda approach

a female reference population (FRP)

- ✓ for detecting QTLs
- ✓ predicting genomic effects to be used for GS

Female reference population

experimental flock



3,949 ewes have been generated by 161 rams.

Description:

- a typical Sardinian dairy sheep farming system
- 1,000 milking ewes with a replacement of about 25% (from Sarda HB rams)

Original aim:

detecting QTL segregating in the Sarda breed

Management

- 40 daughters per ram until 2009
 - 9 daughters per ram since 2010
- Increase the number of represented *bloodlines* from HB

Measured traits:

- **production** (milk yield, fat, protein and lactose content, body weight and body condition score)
- **milkability and udder morphology**
- **health** (somatic cell count, clinical mastitis, faecal egg count, ELISA test for paratuberculosis, ELISA test for visna-maedi, histo-pathological examination for paratuberculosis),
- **milk fatty acids content**
- **reproduction** (fertility and prolificacy).

Genotyping and whole genome resequencing

FRP

3,949 ewes have been generated by 161 rams.

Genotyping of all ewes and their sires (Illumina Inc. OvineSNP50 Beadchip)

Whole genome resequencing of target animals is in progress

- genetic impact on FRP
- high probability to be homozygous at causal mutations

24 animals genomic re-sequencing (12X coverage)

further individuals will soon be resequenced with higher coverage

whole genome sequences jointly to genotypes from DNA arrays allow the imputation of large genome blocks to many individuals of FRP

Detection of causal or LD mutations

QTL detection analysis based on 50K data and a combined Linkage and LD mapping method

QTL regions significant at 5% genome-wide threshold for measured traits

Measured Trait	Chromosome (number of regions in the chromosome)
<i>Production traits¹</i>	
Milk Yield	1(1), 2(2), 3(1), 5(2), 10(2), 11(2), 13(1), 16(1), 18(1), 19(1), 20(1), 24(1), 25(1)
Fat Yield	1(1), 2(2), 3(1), 4(1), 5(1), 10(1), 12(1), 14(1), 16(1), 20(1), 25(1)
Protein Yield	2(2), 3(1), 5(1), 12(1), 14(1), 16(1), 20(1), 25(1)
Fat Content	1(3), 2(1), 3(3), 4(2), 6(3), 7(1), 10(2), 11(2), 12(2), 14(1), 15(1), 16(1), 17(1), 19(2), 20(1)
Protein Content	1(2), 2(3), 3(3), 4(1), 7(2), 8(2), 9(3), 10(1), 11(2), 12(2), 15(1), 17(2), 19(1), 24(1), 26(1)
<i>Udder morphology²</i>	
Udder depth	9(1), 24(1)
Teat position	3(3), 6(1), 7(1), 8(1), 9(1), 10(1), 16(2), 17(1), 20(1)
Degree of separation of the two halves	2(1), 5(1)
Degree of suspension of the udder	1(1), 2(1), 4(1), 5(2), 7(1), 8(1), 9(3), 10(1), 14(1), 16(1), 20(1), 25(1)
<i>Milk quality traits³</i>	
C4:0	9(1), 11(1), 12(1), 14(1), 15(1), 17(1)
C14:0	7(1), 11(1)
CLA/vaccenic acid ratio	22 (1)
Total Saturated Fatty Acids	11(1), 16(1)
Total Fatty Acids	1(1), 18(1), 22(1)
<i>Health Traits⁴</i>	
Somatic Cell Count	2(1), 3(2), 4(1)
Faecal Egg Count	1(1), 3(1), 4(1), 8(1), 12(1), 15(2), 17(1), 19(1), 20(1), 21(1)
ELISA test for paratuberculosis	20(1)
Histo-pathological examination for paratuberculosis	20(1)

Detection of causal or LD mutations

the **most significant ones**:

- are **screened** to verify whether they harbour evident **candidate genes** for the traits of interest
- candidate polymorphisms can be **annotated** to the reference genome to search for **variants potentially affecting gene expression** or SNPs in coding regions
- As an alternative to causative mutation, **informative LD SNPs will be identified** using the sequences of haplotypes of interest.

Causal or LD mutations will be used to design customer low-density SNP panel

Genomic selection

the feasibility of **genomic predictions based on FRP records using milk yield as reference trait**

- identifying useful criteria **to optimize the size and the structure** of the FRP
- **modulate the flow of breeding animals** from and toward the HB

A sample of 537 HB rams representing categories of breeding males (official EBV or PA for milk yield were available):

- 144 AI rams with daughters in FRP
- 105 AI rams without daughters in FRP
- 288 NM rams with no daughters in FRP

genomic breeding values (GBV) from FRP lactation records

theoretical accuracy of GBV (r_{GBV_i}) =
$$\sqrt{1 - \left(\frac{SE_i^2}{G_{ii} \times \sigma_g^2} \right)}$$

The traditional correlation between GBV and EBV with high accuracy is not suitable in the Sarda population.

- low rate of AI
- simplified recording schemes

less than 50% of genotyped rams had a number of daughters higher than 50 in HB.

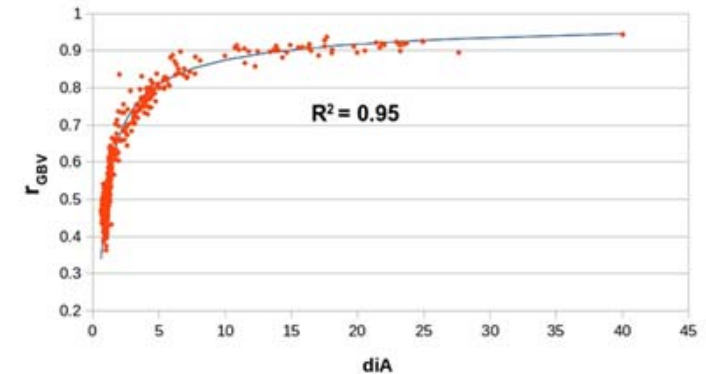
Genomic selection

the impact of the information on relatives in the FRP on rGBV of rams of the sample

indicator of the relationship of HB rams with FRP:

diA: diagonal element corresponding to the ram
of the inverted section of the relationship matrix (**A**)
that included all FRP ewes and the ram itself

- good predictor of rGBV
- to maximize number of rams that will be accurately evaluated by GBLUP
- outliers may be used for detecting pedigree errors



Group	Rams n	FRP sires ¹ n	FRP daug. ² n	rGBV	cor(GBV,EBV) ³
High related rams (diA ≥ 2.5)	134	130	19.0	0.82	0.47
Medium related rams (diA ≥ 1.25 and < 2.5)	103	14	0.3	0.61	0.42
Low related rams (diA < 1.25)	300	0	0	0.47	0.18
Whole sample	537	144	4.8	0.58	0.24

¹ number of rams used as sire in the female reference population

² average number of daughters per ram in the female reference population

³ coefficient of correlation

Genomic selection

to provide practical criteria to:

- modulate the flow of animals from and toward the FRP
- allow farmers to reach a sufficient estimated rGBV for their rams

linear model ($R^2= 0.82$)

expected rGBV = n. of relatives in FRP with r of 0.50 + n. of relatives in FRP with r of 0.25

r: relationship coefficient

Rams having five daughters showed a predicted rGBV around 0.58

Sons of rams with five daughters (i.e five half sisters in FRP) showed 0.54

In the practical management

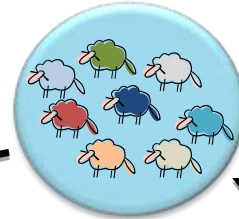
- only young rams entering progeny test will be genotyped and part of those will be used as sires in FRP allowing genomic evaluation of them and their sons.
- considering a replacement rate in FRP of 250 ewes, at least 50 new rams and their sons will be genotyped and GBV calculated with an accuracy higher than that of young rams in progeny test in HB (parent average accuracy of 0.52)

Perspectives

1. Results of QTL detection and accuracies of HB rams genomic predictions realized on the basis of the female reference population showed that it is a realistic option to increase the effectiveness of the current selection program of the Sarda breed.
2. The female reference population is crucial to trigger the application of genomic tools to the breeding scheme.
3. For traits costly to measure on large scale, the female reference population will allow to produce genomic predictions for adult and young rams with sufficient accuracies.
4. For traits routinely measured in HB, the progressive pile up of males genotypes jointly to the application of ssGBLUP methodologies will allow the increase of the accuracies of genomic enhanced breeding values.
5. The use of customer DNA chips including LD or causal SNPs may help to accelerate the genetic progress and to make more feasible the pedigree recording.

Potential Evolution of the selection scheme of Sarda dairy sheep breed

Female Reference Population
 ≈ 1,000 ewes/year
 genotyped 50K beadchip
 whole genome sequences



AI centre rams
 genotyped 50K
 whole genome sequences
 GBV for routinely or cost to measure traits

NM rams
 genotyped 50K
 GBV for routinely or cost to measure traits
Female
 Customer Chip (150-3000)
 Parentage assignment
 GBV for routinely or cost to measure traits

- Measured traits:**
- milk yield, fat, protein and lactose content, body weight and body condition score
 - milkability and udder morphology
 - somatic cell count, clinical mastitis, faecal egg count, ELISA test for paratuberculosis, ELISA test for visna-maedi, histopathological examination for paratuberculosis
 - fatty acids content
 - fertility and prolificacy

ssGBLUP for routinely measured traits
GBLUP for cost to measure traits



- routinely measured traits:**
- Milk Yield
 - Udder Morphology
 - Scrapie resistance
 - Fat and Protein Content

??? Fine Milk composition and diseases resistance from milk sample ???

Estimation of Genomic Segments Effects (b)

Thanks for your attention