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Perspectives on reproductive biotechnologies for FAnGR breeding and conservation

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Abstract. Farm animal breeding and reproduction has to face livestock sector trends, characterization, and monitoring, sustainable use, development and conservation challenges. The increment of assisted reproductive technology (ART) use, next to the advances in the farm animal reproduction field are valuable tools for animal breeding, improvement and conservation programmes, which should be largely implemented. The paper reveals some perspectives on the reproductive biotechnologies available for FAnGR breeding and conservation, and its implementation status in Romania, emphasizing the need of using genomic data also.
Key Words: assisted reproductive technology, artificial insemination, genetic diversity, genomic data, FAnGR.

Résumé. L`élevage e la reproduction des animaux de ferme doit face aux tendances du secteur du bétail, caractérisation, suivi, utilisation durable, développement et conservation défis. L`augmentation des technologies de la reproduction assistées (ART) ensemble aux progrès dans le domaine de la reproduction des animaux sont un moyenne vaillant pour les programmes d`élevage, d`amélioration et de conservation des animaux, que devez largement être mise en œuvre. L`étude relève quelque perspectives sur les biotechnologies de la reproduction disponible pour l`élevage et la conservation des FAnGR, et la situation actuelle de la mise en œuvre en Roumanie, indiquant le besoin d`utiliser l'information génétique aussi.
Mots-clés: technologies de reproduction assistées, insémination artificielle, diversité génétique, information génomique, FAnGR.

Rezumat. Creșterea și reproducția animalelor de fermă trebuie să facă față tendințelor din sectorul zootehnic, caracterizării și monitorizării, utilizării sustenabile, dezvoltării și conservării resurselor animale. Creșterea gradului de utilizare a tehnologiilor de reproducție asistată (ART), alături de progresul realizat în domeniul reproducției animalelor de fermă, reprezintă mijloace importante pentru programele de creștere, ameliorare și conservare, și ar trebui să fie larg implementate. Lucrarea prezintă unele perspective asupra biotehnologiilor de reproducție disponibile pentru creșterea și conservarea FAnGR, precum și statusul implementării acestora în România, evidențiind în același timp necesitatea utilizării datelor genomice.
Cuvinte Cheie: biotehologie, reproducție animală, însămânțare artificială, diversitate genetică.

Introduction. The continuously increment of the world's human population and its shift from country sides to cities runs in the same groove of an augmented demand for food, especially meat and dairy products. Therefore, up to the present, the animal livestock sector still has to face dramatic changes, addressed to increase animal production. Advances in reproductive technologies and in modern genetics implementation in breeding programmes led to a rapid genetic progress towards the development and use of highly specialized breeds related to its production traits (Woelders et al 2006). The highly specialized and productive breeds are often replacing the original indigenous breeds to overstate the human population demand for food, concurring to the loss of genetic diversity in most species of farm animals. In the last decades such destiny was faced by a heritage swine breed, namely by the Mangalitsa all over the world, a breed which is currently stabile in a genetic point of wives (Botha et al 2014; Oroian & Petrescu-Mag 2014). The environmental factors, such us climate change, next to the physiological effects of higher temperatures on individual animals, as well as on production present an increased risk for these highly specialized breeds as well as for

other geographically restricted rare breed populations. Consequently, the breeding programmes may have to be referred to all the above mentioned (Woelders et al 2006; Socol et al 2015a). Considering the potential of significant future changes in animal production background and in the targets of livestock production (Hoffmann 2010), it is essential to consider a better characterization of the population genetics and of different breeds, the reproduction potential and also the implementation of suitable breeding and conservation programmes. Moreover, FAnGR conservation and breeding have to point out also the cultural and historical considerations, rescuing rare or endangered breeds, maintaining indigenous livestock gene pool diversity, disease outbreaks reasons, socio-economic demand and research purposes. In pig breeding to meet this need, very strict objectives are established (Table 1).

Table 1

Breeding objectives in pigs (FAO 2007)

Objectives	*Criteria*	*Further specification*
Production traits	Growth rate	At different ages
	Carcass weight	-
	Carcass quality	Uniformity, leanness of carcass
	Meat quality	Water holding capacity, colour, flavour
Functional traits (Health and welfare)	General resistance	Robustness
	Vital piglets	Maternal ability, teat number
	Survival of pigs	
	Stress	Elimination of stress (halothane) gene in dam lines, and where possible, in male lines
	Congenital effects	Examples: atresia ani, cryptorchism, splay leg, hermaphrodism and hernia
	Leg problems	Leg weakness and lameness
Efficiency	Litter size	Number of slaughter pigs per sow per year
	Feed conversion efficiency	-
Longevity	Functional herd life	Lifetime production with minimal health problems

The basic of maintaining a wide portfolio of FAnGR is the modulation and development of agricultural production systems, considering some important factors such as the climate change and the emergence of new and virulent animal diseases. All these are underlying even more the need of implementing appropriate measures to overcome the loss of genetic diversity and to retain the adaptive capacity of old autochthonous breeds (Rischkowsky & Pilling 2007).

The loss of genetic diversity within and between animal breeds is still challenging the implementation of a wise management, able to maintain the animal biodiversity and at the same time to satisfy the economical needs and the population demand of meat and other animal products. However, the insufficient or the lack of data related the genetic and production traits of each breed available for a breeding programmes and the development of local breeds are often ignored in favor of using germplasm from exotic and high productive breeds, generally showing more such information available. However, breeds characterization, both at animal phenotypes and genetic level and its interaction with production systems is essential. Thus a critical item for national and global farm animal breeding and conservation efforts is the characterization of the population and individual genetics, next to the further development of FAnGR gene

banking (FAO 2011). The additional knowledge obtained on the genetic variability can provide data for conservation decisions making, and has already been used to set conservation priorities in various species (FAO 2012; Socol et al 2015a). In the last decades, molecular data have become more and more relevant for the characterization of genetic diversity (Groeneveld et al 2010). On the same lines subscribe also the ART, which are largely directed for farm animal genetic improvement and breeding goals, being specifically employed to overcome reproductive issues (Wajid et al 2013). Animal livestock trends is predicting an increased future impact regarding the quality and quantity demand for livestock products, scoring out the level of changes in international trade in animal products, retailing, marketing infrastructure and access; all these are standing next to the changes in technology and other factors affecting animal resources (FAO 2014). The design of breeding plans refers to the breeding objectives, the available infrastructure for the selection process (animal resources, reproductive technologies, software for data analyses) (König et al 2009; Henryon et al 2014), the genotyping and phenotyping strategies, the prediction of genetic merit, the effective selection and mating, considering the interacting components (Henryon et al 2014), next to progress monitoring and dissemination of genetic improvement (Simm 1998; FAO 2007).

The present paper provides an overview of the reproductive biotechnologies available for FAnGR breeding and conservation, and some aspects regarding the implementation of the reproductive methods and genomic data also for FAnGR breeding and conservation programmes in Romania.

Material and Method. The paper is an assesment of the actual situation of the reproductive biotechnologies implementation and the FAnGR available, throughout the data recorded by the Romanian National Agency for Animal Husbandry "Prof. dr. G. K. Constantinescu". The present approach is carried out to point some perspectives that should be considered for future FAnGR breeding and conservation programmes emphasizing the status of the reproduction methods applied and other relevant aspects.

Results and Discussion

The status of the reproductive biotechnologies implementation and the impact on FAnGR. Since 2005 a significant outcome has been registered based on the data provided by 103 countries form various continent regions related to an increased use of artificial insemination and embryo transfer reproductive technologies (FAO 2014). Artificial insemination, embryo transfer, in vitro fertilization, cloning, transgenics and genomics are all components of the reproductive tools that can contribute to a significant improvement in animal production (Betteridge 2003; Sejian et al 2010; Zăhan et al 2014). Sex determination of sperm and embryos are having a great impact on the animal breeding sector, being a valuable tool for managing female animal resources and its associated production traits. Manipulating the sex of offspring is a goal of livestock industry for decades (Chen et al 1999; Sejian et al 2010). Regarding the Romanian status of the reproductive biotechnologies implementation for farm animal breeding, the artificial insemination, embryo transfer and germplasm sexing techniques need to be enhanced.

The constant challenge to increase animal production, for carrying out economic demands, population's feeding, next to the insurance of the natural resource base focus both on the need to gain genetic advantage the minimization of the genetic disadvantages. Therefore, it stands for a constant challenge in developing and applying different improvement methods and technologies for farm animals (Nicholas 1996). Thus older methods up to advanced one are applied to this end, such us animal genetic improvement and reproduction methods referring to natural mating, artificial insemination, multiple ovulation, embryo transfer, pure/straight breeding, cross-breeding, selection methods, quantitative genetics and molecular genetics methods. The maintenance of genetic variation is a condition for continuous genetic improvement, a high rate of inbreeding determining the loss of genetic variation within a breed (Rischkowsky & Pilling 2007). Nevertheless, in terms of a high efficiency animal selection,

the pedigree and the individual performance data shall be accurately registered. Reproductive capacity and techniques are definitely playing an important role regarding the number of parents required for the production of the next generation, and thereby on the rate of genetic improvement (Rischkowsky & Pilling 2007). The limits of implementing selection and reproduction in pedigree breeds, mainly referring to increased consanguinity, lack of effectiveness of selecting characteristics with poor inheritance ability, and other aspects concur to find out solutions for overdrawing these boundaries. Cross-breeding allows the combining the advantages of several breeds, pointing out the heterosis and complementarity between breeds' traits. Based on the reproductive technology benefits on animal genetic improvement, the high use of artificial reproduction techniques *e.g.* artificial insemination and, embryo transfer is recommended to push up genetic progress; a higher reproduction rate implies a lower number of breeding animals alongside a higher intensity of selection. FAnGR breeding and conservation along with gene banking is standing for artificial insemination and embryo transfer backing up, concurring for higher selection intensity, due to a preciseness selection of the males based on their genetic value, semen quality and thus reproductive capacity. One of the major advantages of the artificial reproduction techniques consist in allowing quick distribution of high value genetic in different animal populations. The implementation of reproductive biotechnologies as artificial insemination and multiple ovulation and embryo transfer, allow to high up the rate of genetic improvement, showing acceptable rates of inbreeding, meanwhile other reproductive techniques as semen sexing, embryo sexing and embryo cloning lead to limited increases in the rates (Nicholas 1996). In the last decades significant progress in genetic improvement methods was achieved applying targeted methods related to gene-based selection, marker assisted selection and introgression (Zăhan et al 2010, 2009).

According to the female farm animal livestock data on catagraphy registered in 2014 by the Reproduction Department of The Romanian National Agency for Animal Husbandry "Prof. dr. G. K. Constantinescu" 1.606.005 heads account for cattle, 16.870 heads for buffalo, 9.862.367 heads for sheep and goat, 339.636 heads for pig (Table 2). Referring to the similar data registered in 2013, an increment is indicated in cattle, buffalo, sheep and goat (1.5%, 2.8%, 3.72% respectively) and a decline in swine (0.36%).

Table 2

Farm animal female livestock catagraphy data registered in 2014*

Species	*Female livestock's catagraphy (no. of individuals)*
Cattle	1.606.005
Buffalo	16.870
Sheep and goat	9.862.367
Swine	339.636

*Source: ANZ 2015.

Focusing on the artificial insemination data registered in 2014, a number of 621.563 were in bovine, 1.331 in buffalo, 1.169 in sheep and goat, and 298.418 in pig.

Analyzing the breed structure data for the female livestock on various farm species recorded in 2014 the mainly breeds listed in cattle are: 30.96% Romanian Spotted Cattle – Simmental, 13.58% Brown Cattle, 20.28% Romanian Black Spotted Cattle – Holstein Friesian, 0.6% Pinzgau, 0.16% meat breeds, 32.86% crossbreeds, 0.54% undefined; in sheep and goat: 56.19% Tsurcana, 20.77% Tsigai, 1.06% Spanca variety, 5.6% Merinos, 4.11% Karakul; 0.46% other breeds, 11.81% crossbreeds; in swine mainly, the following breeds are mentioned: Large White, Duroc, Pietrain, Landrace. Mangalitza and other hybrids account for the pig species, precise data being not provided.

The above data is showing that progress still should be made related the implementation of ART for farm animal livestock breeding sector, a special concern being

addressed to the pig sector, due to the decline showed and the in the red data recorded (Figure 1 & 2). Also data for unregistered artificial insemination and unauthorized activity into the reproduction field should be taken into account, state measures being still needed to overdraw this issue. Progress is still required into the reproduction area for increasing the number of artificial inseminations, eradicating clandestine natural mating and implementation of embryo transfer and other reproductive techniques.

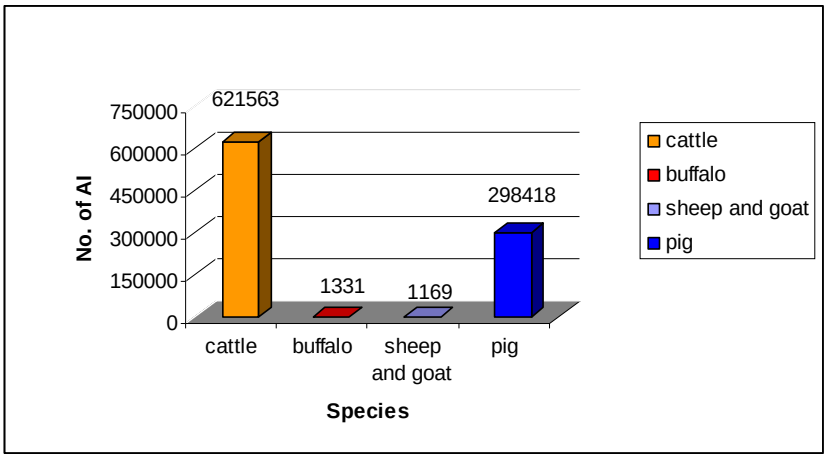

Figure 1. The artificial insemination data registered in 2014 (Source: AZ 2015).

For the same year (2014), the authorized natural mating data recorded were 21.204 in bovine, 6.618 in buffalo, 4.264.952 in sheep and goat, and 9.200 in swine (ANZ 2015).

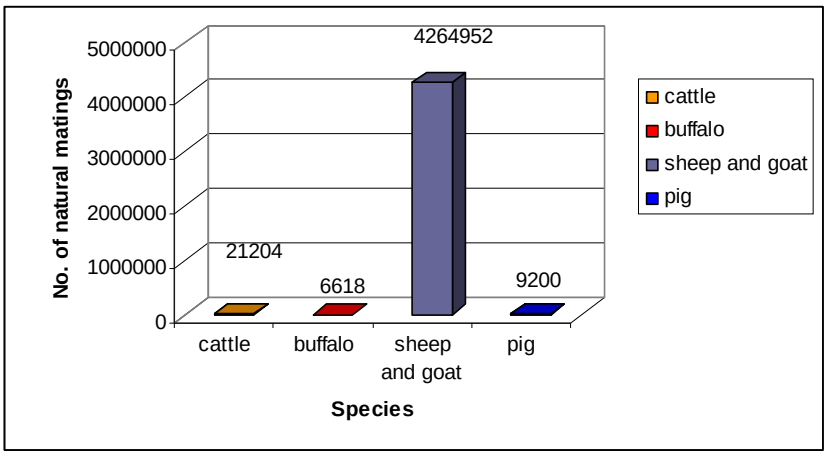

Figure 2. The natural mating data registered in 2014 (Source: ANZ 2015).

The National Centre for Professional Training in Animal Husbandry department of the National Agency for Animal Husbandry "Prof. dr. G. K. Constantinescu" since 2011 was authorized by the county authorization commissions from the National Agency for Qualifications of the Ministry of Education and Scientific Research for qualification and specialization courses for farm animal artificial insemination; up to this moment a number of 240 graduates are reported for the above qualification and specialization professional training courses (2015), contributing in this way to the development of the reproduction field in Romania.

Considering the monitoring data of the reproduction material from accredited farm units allover the country and the Romanian strategy for a sustainable management of farm animal genetic resources, listing the following species and breeds of native or other origin as being at risk, showing an endangered status (cattle: Romanian Grey Steppe; sheep: Corkscrew Walachian, Palas Meat sheep, Palas Milk sheep, Rusetu Tsigai,

Polwarth, Stavropol Merino, Caucasus Merino, Australian Merino, Romney March, Coopwarth; pig: Romanian saddleback [Bazna], Red Magalitsa, Hampshire;) or an extinct status (cattle: Romanian Grey Steppe varieties Bucsana, Transilvaneana, Ialomiteana, etc., Walachian (Mocanitsa Mountain) and its varieties, Red Danish, Polish, Latvian, Lithuanian, Red Poll, Jersey, Shorthorn, Santa Gertrud; sheep: Karnabat, Ascanian Merino, Ile de France, Ostfriesian, Southdown, Texel, Border Leicester, Romanov, Finish Landrace, Perendal, Drysdal, Awasi; pig: Stocli, Palatin, Strei, Banat White, Black Dobrogea Pig; horse: Romanian local breed Moldavian, Mountain, Ialomitsan etc.) (FAO 2003; Socol et al 2015b) a special concern should be addressed to the reproduction area from this point of view. The genetic conservation of these breeds may be focused for genetic diversity purposes, but also for farm animal breeding ones.

The use of genomic data for farm animal breeding and conservation programmes. Genomic information is an exciting prospect for animal breeding aiming to maximize long-term genetic gains in breeding schemes (Henryon et al 2014). The progress registered in animal genome sequencing guide to applied gene-based selection especially related to Mendelian traits (mainly diseases and genetic defects) and provides new tools for selecting healthy individuals. DNA based methods were also successfully applied to eradiate genetic disorder in many species. Beside all these aspects, it should be considered that significant economic traits in animal production involve quantitative aspects and a large number of loci (Andersson et al 1994; Le Roy et al 1990). The widely used marker assisted selection methods enable the genotyping of a gene locus showing a major effect, but also adjacent chromosomal region of the target gene can be used as marker (Rischkowsky & Pilling 2007; Zăhan et al 2010, 2009). Data available on animal genome structure and function can be used in breeding programmes to improve various target traits (Williams 2005), marker assisted selection being one of the latest selection method applied for improving the genetic structure of a population. Databases for genome projects (harboring molecular markers) and other databanks for animal genetic resources should be a component of FAnGR breeding, improvement and conservation programmes in Romania too, highlighting its relevance for the implementation of the reproduction programmes.

Conclusions. The need to increase the number of artificial inseminations, to eradicate clandestine natural mating and to implement embryo transfer techniques, next to the use of genomic data and other reproductive techniques should concur to the development of the reproduction field in Romania, this need is most evident especially in swine, were the registered female livestock follow an decreasing trend despite of the growing demand for pork, and implicit for breeding stock. The farm animal artificial insemination professional training programs should be maintained or extended in the future, to ensure highly specialized human resources for the animal reproduction field. The reproductive biotechnologies, next to the genomic profile of data stand for offering a modern perspective for considering FAnGR breeding and conservation programmes in Romania. Such consideration should be largely applied in farm animals, and especially in swine sector, allover the country to ensure rapid progress in animal husbandry, addressing pure breeding, high specialized breeding and biodiversity preservation, in the specific socio-economic, environmental, cultural and farm animal production context of Romania.

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