



## Importance of the study of reproduction in small animals and preservation of species

*Importancia del estudio de la reproducción en animales pequeños y la preservación de especies*

*Importância do estudo da reprodução em pequenos animais e preservação das espécies*

**Martinez-Barbitta Marcelo<sup>123</sup>**

<sup>1</sup>Postgraduate Program, Faculty of Veterinary Medicine, University of the Republic, Montevideo, Uruguay

<sup>2</sup>Doctorate Program, Faculty of Veterinary Medicine, University of Perugia, Perugia, Italy

<sup>3</sup>Reproductive Veterinary Service\_Uruguay®, Nueva Helvecia, Uruguay

### Abstract

*Although reproduction in small animals is relatively current, its objectives are very varied. Initially they were used as a study model in physiology and various pathologies. In the same way, many small animals are produced as food and finally, in the last 20 years, the importance of pets and the intensification in the selection of canine and feline breeds, has caused an important impulse in the development of new scientific information. Although currently technical resources have advanced substantially, there are still many basic and important issues that have not yet been elucidated by formal science. Global threats facing wild species include the consequences of climate change, population growth, urbanization, air and water pollution, and the release of chemicals into the environment, causing, on average, 25% of animals are in danger of extinction. Wild animal populations are small and dispersed in their habitat with little or no opportunity for genetic exchange, which has generated in recent years many programs to preserve wild species and design appropriate strategies that lead to sustainable populations. Fortunately, recent and inspiring advances in the science of wildlife reproduction have been reported that will set directions for future research and will surely lead to further successes in conservation biology.*

**Keywords:** *Small Animal Reproduction, Reproductive Sciences in Animal Conservation, Assisted reproductive technologies.*

### Introduction

Small Animal Reproduction (SAR) is a rather recent topic in the field of Animal Reproduction. Although some continuing educational societies aim to improve the level of general knowledge, published scientific facts are still limited, and there are many hypotheses or affirmations that remain unverified or even sometimes dogmatic or empirical (Fontbonne, 2020). The first International Symposium on Canine and Feline Reproduction was organized in Dublin in 1988, bringing together for the first time around 100 specialized academics or practitioners. It was the beginning of an exponential interest for dog and cat reproduction, including both domestic and wild canids and felids. In 2004, a study based on an open questionnaire was published. The survey was performed in institutions of veterinary education in European countries. It was aimed to provide a view of the general status of education and research in SAR in Europe (Rodríguez-Martínez, 2004). The author concluded that, in the absence of comparative data and research used in education and in the development of new technologies in SAR, there was a risk that sound science-based academic education might be constrained. To counteract these negative trends, SAR should be recognized as a very important branch of the department of animal reproduction. This should lead to the development of a high quality research rejecting empiricism and dogmas. Reproductive Sciences in Animal Conservation (RSAC) is a very recent area, which increasingly assumes a fundamental role, whether in "*in situ*" or "*ex situ*" conservation, whether for the preservation of endangered species and animal study models. The global threats facing wild species include the consequences of climate change, population growth, urbanization, atmospheric and water pollution, and the release of chemicals into the environment, to cite but a few. Reproductive sciences provide important and often unexpected windows into many of these consequences.

Initially we must bear in mind that the SAR is oriented to a significant number of species, with

different objectives. That is why animals such as rodents, fish and amphibians, among others, are used as research models to understand research on basic physiological, pharmacological, molecular or pathological processes. Mammals such as rabbits are used as study models but are also of interest in aspects related to food production. Domestic canines and felines have several peculiarities. Firstly, they are study models in various fields of physiology and pathology, and used in conservation programs for wild animals in danger of extinction in many areas of the world. At the same time, they are also pets, and their implication in clinical reproduction in theriogenological practice acquires another particular significance.

The domestic dog is a genetic enterprise unique in human history. No other mammal has enjoyed such a close association with humans over so many centuries, nor been so substantially shaped as a result. A variety of dog morphologies have existed for millennia, and reproductive isolation between them was formalized with the advent of breed clubs and breed standards in the mid-19th century (Parker et al., 2017). This current situation has generated that, although we speak of the same species, not only due to the intense selection of breeds, but also due to geographical genetic derivations, different groups of dogs and cats behave very differently, even in their morphological characteristics, physiological and behavioral (Ruiz-Garcia, 1999).

Over 350 inherited disorders have been described in the purebred dog population (Parker et al., 2017). Many are restricted to particular breeds or animal groups as a result of aggressive in-breeding programs used to generate specific morphologies. Similar situations have occurred in domestic felines and sometimes even more intense than canines, due to the small number of animals that have been used as the nucleus for the modern development breeds (Ruiz-Garcia, 1999).

In the field of veterinary sciences, small animal breeding is becoming more important, given the growing interest in companion animal husbandry (Fontbonne, 2020) and in the study of domestic carnivores as a model to develop biotechnologies for conservation of endangered species. However, there are still many unresolved issues, unverified beliefs, and limitations in this field, and despite the large number of published articles and reviews, some questions in the field of small animal reproduction remain unanswered.

At present, in the era of "omics" (genomics, transcriptomics, proteomics, metabolomics, nutrigenomics, epigenomics, among others) where a biological phenomenon can be explained from various perspectives, where possible biomarkers that are associated or predict a biological process, be it normal or that leads to a disease, we still have big problems without consensual and scientifically based solutions. Just to exemplify, we will mention some: a) Unclear data on the physiology of reproduction, such as ovulation in the cat where there are still some unclear physiological pathways, feline placental steroidogenesis or the non-existence of canine seasonality; b) Genetic diversity: on many occasions, studies seek to be conclusive for the entire species, but the enormous physiological and pathological variability between the different canine and feline breeds has been well studied, for the simple reason that their selection makes them genetically very different. therefore, this condition must be taken into account due to the bias that it can generate in the studies and declared conclusions; c) common beliefs based on old studies: on the effect of neutering on the risk of developing mammary tumors in dogs and also on the risk of developing urinary incontinence in bitches are perfect examples; d) unclear nomenclature: in uterine pathology, for example, a more complete and agreed classification would be very useful to ensure that everyone uses the same terminology; e) clinical conditions described mainly through case reports: many diseases or clinical findings give rise to the publication of case reports, which may be merely anecdotal and not representative of the most frequent clinical forms of a disease. For example, Arlt and Haimerl (2016) state that there are no systematic research results available on the clinical and reproductive characteristics of ovarian diseases in canines; f) lack of knowledge and lack of studies: the role of infectious diseases in reproduction is a good example. Another annoying lack of studies concerns the potential promoting effect of recurrent episodes of overt pseudopregnancy with lactation in the development of mammary tumors in bitches. Sometimes the use of a drug or a procedure is too recent and the results are based on an insufficient number of animals to be able to claim that it brings strong scientific data. g) contradictory data showing the need for further investigations: in some topics, there may be contradictory data between different authors, which may complicate some decisions in daily veterinary practice. Contradictory results are common in fields of research in which just a few number of patients or cases can be enrolled or in research protocols that do not take into account bias in the proper way. Only for example: spaying bitches before the first oestrus?, is it necessary to spay a bitch that has been surgically operated for a mammary carcinoma, and if it will increase her chance of survival?. h) fashionable and potentially oriented studies: some topics are discussed more than others in congresses and appear fashionable at some time. But it is dangerous to claim scientific facts before enough data has been collected and enough studies published. One recent example is the potential increased risk of developing cancer after spaying in the dog. i) Peri- and postnatal losses are

relatively high in dogs, with mortality rates ranging from 17% to 30% during the first 8 weeks of life. Stillbirths and deaths within the first week were responsible for the majority of these losses (reviewed by Indrebø et al., 2007). Given these high rates of losses and the obvious impact of the whelping process on the number of stillbirths, one would expect much more information in the literature on the normal course, the physiology and pathophysiology of parturition in the dog. Data on spontaneous and assisted vaginal deliveries do only scarcely show up in the literature and therefore research on canine parturition is not very much encouraged.

There is very little reliable information to interpret the evolution of SAR and RSAC research. Without intending to carry out a bibliometric analysis, it is interesting to verify the number of publications, using some of the most used academic search engines, including some that are geographically significant, in relation to the temporal evolution of the published works. We use keywords and temporary spaces of decades, obtaining the following results:

Time period (decade)	Pubmed		CrossRef		Redalyc*	
	Keyword "Animal Preservation"					
1983-1992	9,981	IV (%)	18,433	IV (%)	174	IV (%)
1993-2002	17,175	72.1	24,294	31.8	6,584	3,683.9
2003-2012	26,437	53.9	80,565	231.6	55,608	744.6
2013-2022	42,916	62.3	107,354	33.3	59,801	7.5
Keyword "Reproduction Small Animal"						
1983-1992	2,871		24,602		196	
1993-2002	3,889	35.5	32,275	31.2	7,331	3,640.3
2003-2012	6,669	71.5	100,267	210.7	88,180	1,102.8
2013-2022	11,596	73.9	131,740	31.4	106,024	20.2

IV – Intemporal variation. Difference between published studies compared to the previous decade.

\* Data from the Network of Scientific Journals of Latin America and the Caribbean, Spain and Portugal is added as it is a regional initiative that generates qualitative and quantitative indicators of Ibero-American Science.

This exercise only seeks to understand the importance that the scientific community has given to these areas of research and dissemination, but does not intend, in any way, to characterize the quality of the published information. Similarly, other components are not evaluated here, such as the evolution of scientific communication media, nor the number of existing journals, nor the criteria that search engines have for inclusion of scientific media.

Banchi et al. (2022) when analyzing the bibliometric results of the last decade detected that, within Latin American country, the distribution of the affiliations that publishes the most in SAR is Brazil, which leads the origin of studies worldwide, followed by USA and Italy. The same authors emphasize that neonatology is an expanding theme in published research. The trend lines show that canine reproductive research is also increasing more than feline reproductive research in terms of the number of articles published. However, the importance of the feline species as a research topic in animal reproduction is increasing, especially in relation to the interest in studying cats to improve assisted reproductive technologies (ART) in wild felids in danger of extinction. Most wild felines are considered at risk and cats represent a good model for research in this area (Farstad, 2000). Interest in dogs as a research model for wild canid conservation is limited for two reasons: there are not many endangered species (Farstad, 2000) and ART such as *in vitro* fertilization (IVF) are not now routine practice.

Literature on neonatology (ie, studies of puppies and kittens from birth to weaning) appears to be lacking, and topics such as neonatal mortality are considered niche topics. However, neonatology and pediatrics represent everyday interests for veterinarians, breeders, and pet owners, and neonatal mortality is still considered a major problem in domestic carnivores (Sparkes et al., 2006; Indrebø et al., 2007; Mila et al., 2014).

Extinction of animal and plant species is currently occurring at a much higher rate than speciation due to detrimental human activities such as habitat destruction, overhunting/fishing, and poaching. In addition, climate change modifies natural habitats and promotes the appearance of new challenges that affect wildlife and the environment. From the modification of migration patterns to the spread of the disease (Sutherland et al., 2018). Conservation biology aims to understand and maintain a thriving biodiversity because the disappearance of even a single species can compromise the functioning of an entire ecosystem (Holt et al., 2014). The core of animal conservation is the maintenance of genetic diversity within

populations large enough to be sustainable in the long term. In addition to protecting viable populations in their natural habitat (*in situ* conservation), it is essential to maintain populations in captivity (*ex situ* conservation) for subsequent reinforcement or reintroduction. Unfortunately, the International Union for Conservation of Nature (IUCN) estimates that 25% of mammals, 12% of birds, 20% of reptiles, 30% of amphibians, 20% of fishes, 30% of invertebrate, and 55% of plant species are threatened with extinction (IUCN 2020, <http://www.iucnredlist.org/>). Furthermore, many wild animal populations are small and scattered in their habitat with little or no opportunity for genetic exchange, which increases homozygosity and inbreeding that, in turn, leads to a poor adaptive capacity to environmental changes as well as fertility problems (Wildt et al., 2010).

Due to higher rates of extinction due to human and natural factors, research in basic and applied reproductive biology is required to preserve wild species and design appropriate strategies, leading to sustainable populations (Comizzoli & Holt, 2019). Recent and inspiring advances in RSAC have been reported that will set directions for future research and will surely lead to further successes in conservation biology.

Comparative studies in multiple animal models have been essential to improve our understanding of aging mechanisms and to develop mitigation strategies. Different models, have provided organisms for the study of reproductive aging (Jones et al., 2014). Interestingly, some processes of reproductive aging are common among species (Ottinger, 2010).

Studies in wild species (in their natural habitats or in conservation breeding centers) are invaluable because we can learn about unique adaptations especially in long-lived species, which will not only improve conservation efforts, but also provide new comparative models for other domestic species, and importantly for humans, oriented to preventive medicine (Comizzoli & Ottinger, 2021).

It is imperative that the reproductive sciences are recognised by the community and policy makers as important contributors to future health and welfare of animals, humans and the planet if these potential benefits are to be captured and utilised (Findlay et al., 2019).

## References

- Arlt SP, Haimerl P. Cystic ovaries and ovarian neoplasia in the female dog - a systematic review. *Reprod Domest Anim* 51(Suppl 1):3–11, 2016.
- Banchi P, Rota A, Bertero A, Domain G, Ali Hassan H, Lannoo J, Van Soom A. Trends in Small Animal Reproduction: A Bibliometric Analysis of the Literature. *Animals* 12: 336, 2022.
- Comizzoli P, Holt WV. Breakthroughs and new horizons in reproductive biology of rare and endangered animal species. *Biol of Reprod*, 101(3), 514–525, 2019.
- Comizzoli P, Ottinger MA. Understanding Reproductive Aging in Wildlife to Improve Animal Conservation and Human Reproductive Health. *Front Cell Dev Biol*. 19;9: 680471, 2021.
- Farstad W. Current state in biotechnology in canine and feline reproduction. *Anim. Reprod. Sci.* 60–61, 375–387, 2000.
- Findlay JK, Holland MK, Wong BBM. Reproductive science and the future of the planet. *Reproduction*. 158(3):R91-R96. 2019.
- Fontbonne A. Small animal reproduction: Scientific facts versus dogmas or unverified beliefs. *Theriogenology*. 150: 464-470, 2020
- Holt WV, Brown JL, Comizzoli P. Reproductive science as an essential component of conservation biology. *Adv Exp Med Biol* 753:3–14, 2014.
- Indrebø A, Trangerud C, Moe L. Canine neonatal mortality in four large breeds. In: O'lafsson Th (ed.), *Perinatal Death in Domestic Animals. Proceedings 20<sup>th</sup> NK-Vet Symposium, Reykjavik, Iceland*, pp. 62–68. 2007.
- Jones OR, Scheuerlein A, Salguero-Gómez R, Camarda CG, Schaible R, Casper BB, Dahlgren JP, Ehrlén J, García MB, Menges ES, Quintana-Ascencio PF, Caswell H, Baudisch A, Vaupel JW. Diversity of ageing across the tree of life. *Nature* 505 169–173, 2014.
- Mila H, Feugier A, Grellet A, Anne J, Gonnier M, Martin M, Rossig L, Chastant-Maillard S. Inadequate passive immune transfer in puppies: Definition, risk factors and prevention in a large multi-breed kennel. *Prev. Vet. Med.* 116, 209–213, 2014.
- Ottinger MA. Mechanisms of reproductive aging: conserved mechanisms and environmental factors. *Ann. N. Y. Acad. Sci.* 1204 73–81, 2010.
- Parker HG, Dreger DL, Rimbault M, Davis BW, Mullen AB, Carpintero-Ramirez G, Ostrander EA. Genomic analyses reveal the influence of geographic origin, migration and hybridization on modern dog breed development. *Cell Rep.* 25; 19(4): 697–708. 2017.



**Rodríguez-Martínez H.** Reflections upon the trends of education and research in small animal reproduction in Europe. *J Vet Med Educ* 31(1):38-44, 2004.

**Ruiz-Garcia M.** Genetic structure of different cat populations in europe and south america at a microgeographic level: importance of the choice of an adequate sampling level in the accuracy of population genetics interpretations *Genetics and Molecular Biology*, 22, 4, 493-505. 1999.

**Sparkes AH, Rogers K, Henley WE, Gunn-Moore DA, May JM, Gruffydd-Jones TJ, Bessant CA.** A questionnaire-based study of gestation, parturition and neonatal mortality in pedigree breeding cats in the UK. *J. Feline Med. Surg.* 8, 145–157, 2006.

**Sutherland WJ, Butchart SHM, Connor B, Culshaw C, Dicks LV, Dinsdale J, Doran H, Entwistle AC, Fleishman E, Gibbons DW, Jiang Z, Keim B, Roux XL, Lickorish FA, Markillie P, Monk KA, Mortimer D, Pearce-Higgins JW, Peck LS, Pretty J, Seymour CL, Spalding MD, Tonneijck FH, Gleave RA.** A 2018 horizon scan of emerging issues for global conservation and biological diversity. *Trends Ecol Evol* 33:135–149, 2018.

**Wildt DE, Comizzoli P, Pukazhenti B, Songsasen N.** Lessons from biodiversity-the value of nontraditional species to advance reproductive science, conservation, and human health. *Mol Reprod Dev* 77:397–409, 2010.

---