







## Miranda's Donkey: a review of published studies with insights on clinical pathology research for breed's preservation

Grasiene Silva<sup>1,2,3</sup>  Ana Cristina Silvestre-Ferreira<sup>1,2,3</sup>   
Miguel Nóvoa<sup>4</sup>  Felisbina Luísa Queiroga<sup>1,2,3\*</sup> 

<sup>1</sup>Departamento de Ciências Veterinárias, Universidade de Trás-os-Montes e Alto Douro (UTAD), 5000-801, Vila Real, Portugal. E-mail: fqueirog@utad.pt. \*Corresponding author.

<sup>2</sup>Centro de Ciência Animal e Veterinária (CECAV), Universidade de Trás-os-Montes e Alto Douro (UTAD), Vila Real, Portugal.

<sup>3</sup>Laboratório Associado para a Ciência Animal e Veterinária—AL4AnimalS, Vila Real, Portugal.

<sup>4</sup>Associação para o Estudo e Proteção Gado Asinino (AEPGA), Atenor, Portugal.

**ABSTRACT:** Miranda's donkey is an autochthonous asinine breed from northern Portugal classified as endangered by the Food and Agriculture Organization. Research on Miranda's donkey in several scientific areas is important to standardize the breed and to assist the breed's preservation. Reference values in clinical pathology are used to determine the health status of individuals and herds, and to control the evolution of diseases and their response to therapy. This review summarized the studies performed on Miranda's donkey and evaluated the state of the art concerning studies performed in the clinical pathology field. Thirty-seven publications on Miranda's donkey published between 2000-2023 in various scientific areas were found. In conclusion, research on Miranda's donkey is scarce, and further studies are needed, especially in hematology and clinical biochemistry areas with respect to physiological and pathological changes that affect the breed, thus helping its conservation. **Key words:** autochthonous breed, clinical pathology, conservation, Miranda's donkey.

### O burro de Miranda: uma revisão dos estudos publicados com perspectivas de investigação em patologia clínica para a preservação da raça

**RESUMO:** O burro de Miranda é uma raça autóctone asinina do norte de Portugal classificada como em perigo pela Organização das Nações Unidas para a Alimentação e a Agricultura. Pesquisas sobre o burro de Miranda em diversas áreas científicas são importantes para padronizar a raça e auxiliar na sua preservação. Os valores de referência em patologia clínica são utilizados para determinar o estado de saúde dos indivíduos e rebanhos, controlar a evolução das doenças e a sua resposta à terapêutica. O objetivo desta revisão é apresentar os estudos realizados no burro de Miranda e avaliar o estado da arte dos estudos realizados no campo da patologia clínica. Foram encontradas trinta e sete publicações sobre o burro de Miranda publicadas entre 2000-2023 em várias áreas científicas. Em conclusão, as pesquisas sobre o burro de Miranda são escassas, sendo necessários mais estudos, especialmente nas áreas de hematologia e bioquímica clínica, com respeito a alterações fisiológicas e patológicas que afetam a raça, ajudando assim à sua conservação.

**Palavras-chave:** burro de Miranda, conservação, Patologia clínica, raça autóctone.

## INTRODUCTION

A breed is designated as native or autochthonous when adapted locally and present in a country long enough to have developed genetic characteristics for one or more traditional production systems (INGRASSIA et al., 2006). In Portugal, there are currently two autochthonous donkey breeds with different phenotypic characteristics: the Graciosa' donkey, from the island of Graciosa in Açores, and Miranda's donkey, which inhabits the Planalto Mirandês (DGAV, 2021).

The Miranda's donkey was first described in 1999, during a project on the donkeys of the Douro Internacional Natural Park and the importance of these

animals for the local community. During the project development, the researchers noticed that some of the donkeys were physically different from the breeds previously described in the region and could constitute a new Portuguese breed (SAMÕES, 2000). The breed was recognized in 2002 by the Ministry of Agriculture, Rural Development, and Fisheries of Portugal, reinforcing the importance of these animals for the genetic diversity of Portuguese breeds, as for the cultural and economic local development (MARQUES, 2006).

Autochthonous breeds constitute a unique and irreplaceable genetic heritage that must be preserved and valued, having been created Portuguese and European strategies and programs

for the conservation and improvement of Miranda's donkey and other autochthonous breeds (MARTA-COSTA et al., 2016). Initially, these efforts consisted of determining its genotypic and morphological characteristics and geographical distribution (ALFRANCA, 2001).

The determination of hematological and biochemical reference values is also important for the preservation of endangered breeds, physiologically characterizing the breed, providing relevant clinical information about the health status of an animal or herd, severity and systemic effects of a disease, and to assess the response to treatment prescribed by veterinarians (SEDLINSKA et al., 2017). When possible, these studies should be performed in all breeds, because factors such as geographic location, climate, feed and types of handling, even in animals of the same species, can interfere with biochemical and hematological parameters (JORDANA et al., 1998; SELINSKA et al., 2017).

This review analyzed the published literature on Miranda's donkey and evaluate which areas have been studied and those needing further research and development, namely on the clinical pathology field. The investigation was divided into two parts: 1) Published research on Miranda's donkey, 2) Clinical pathology research conducted on Miranda's donkey and on other European breeds.

#### *Web based search*

Search was conducted on websites such as Google Web, Google Scholar, ResearchGate and PubMed from January-October 2023. Scientific articles published in journals, PhD Thesis, master dissertations and conference proceedings, in English or Portuguese language were considered. In case the research results were published in article and thesis/dissertation format, only the results published in article format were considered for this study, to avoid duplication of information. The search terms were Miranda's donkey and Planalto Mirandês. Thirty-seven publications on Miranda's donkey from 2000-2023 on various topics were identified and analyzed. The articles published on Miranda's donkey are summarized in figure 1.

#### *Publications on Miranda's donkey*

##### *History and demographics*

Six articles were found on the physical characteristics of the breed, number of animals, challenges and future perspectives (QUARESMA et al., 2005; MARQUES et al., 2006; QUARESMA et al., 2013; QUARESMA et al., 2014; QUARESMA

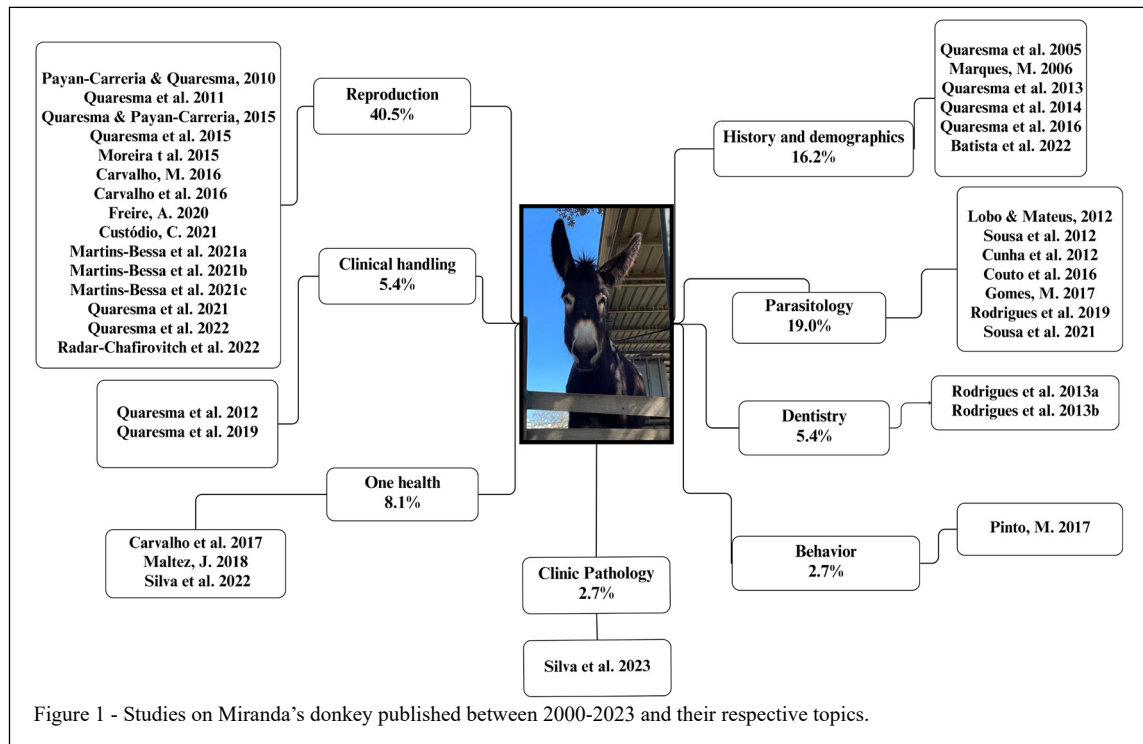
et al., 2016; BATISTA et al., 2022). The first record in the literature on Miranda's donkey was published in 2000 (SAMÕES, 2000), but only in 2002 this breed was officially recognized as an autochthonous breed and presented to the scientific community (QUARESMA et al., 2005). Some of the morphological characteristics of Miranda's donkey described in the Regulation of Zootechnical Registration of the Breed are shown in figure 2.

Most of the animals live in Planalto Mirandês, in the municipalities of Miranda do Douro, Bragança, Vimioso, and Mogadouro (Figure 3). These animals may have developed well in the region due to their good adaptability to poor quality forage and water scarcity. In addition, donkeys were initially used for traction, saddle, loading and production of mules for being rustic, resistant and docile (QUARESMA et al., 2005).

Subsequently, an extensive study was performed on the importance of Miranda's donkey as a cultural and economic heritage by analyzing its involvement in the daily life of Miranda. A chronology was created considering the first reports of the breed, the creation of the Association for the Study and Protection of the Donkey Cattle (AEPGA - Associação para o Estudo e Proteção do Gado Asinino), official recognition, and the change in the perception of the donkey by breeders and incentive programs (MARQUES, 2006).

Researchers and protection organizations have monitored and updated the number of animals. In 2003, 255 animals (213 females, 40 castrated males, and 2 males) were reported (QUARESMA et al., 2005). In 2013, this number increased to 760 animals (681 females, and 79 males), but only 589 were able to reproduce (545 females and 44 males) (QUARESMA et al., 2013). In 2019, 840 animals (780 females and 60 males) were registered. In 2020, there was a decline in the population, with a total of 816 specimens (756 females and 60 males) (FAOSTAT, 2023). According to the latest survey, released in 2023, the current number of sexually mature Miranda's donkey consists of 761 animals, 652 females and 109 males (SPREGA, 2023). Despite efforts, according to the Food and Agriculture Organization classification for breeds, Miranda's donkey remains endangered as it has less than 1000 females and 20 males of breeding age (FAOSTAT, 2023).

Several authors have studied the viability of the breed, the factors that limit the increase in the herd, and alternatives to prevent extinction. The limiting factors are related to characteristics of the breed, herd handling (mainly reproductive), and socioeconomic factors (mechanization, aging



of owners, and rural exodus), (QUARESMA et al., 2005; QUARESMA et al., 2013; QUARESMA et al., 2014; QUARESMA et al., 2016). The alternatives suggested and implemented over the years are government incentives, rural tourism, use as pet animals, participation in cultural events, assisted activities with donkeys, milk production for human consumption, cosmetics production, vegetation control and landscape maintenance (QUARESMA et al., 2005; QUARESMA et al., 2013; BATISTA et al., 2022; AEPGA, 2023).

### Reproduction

Reproduction was the topic with the highest number of studies (n = 15), possibly due to their impact on reproduction rates. The topics involve physiology, pathologies of the female and male reproductive systems, development and use of new technologies to improve reproductive indices (PAYAN-CARREIRA & QUARESMA, 2010; QUARESMA et al., 2011; QUARESMA & PAYAN-CARREIRA, 2015; QUARESMA et al., 2015; MOREIRA et al., 2015; CARVALHO, 2016a; CARVALHO et al., 2016b; FREIRE, 2020; MARTINS-BESSA et al., 2021a; CUSTÓDIO, 2021; MARTINS-BESSA et al., 2021b; MARTINS-BESSA et al., 2021c; QUARESMA et al., 2021; QUARESMA et al., 2022; RADAR-CHAFIROVITCH et al., 2022).

### Parasitology

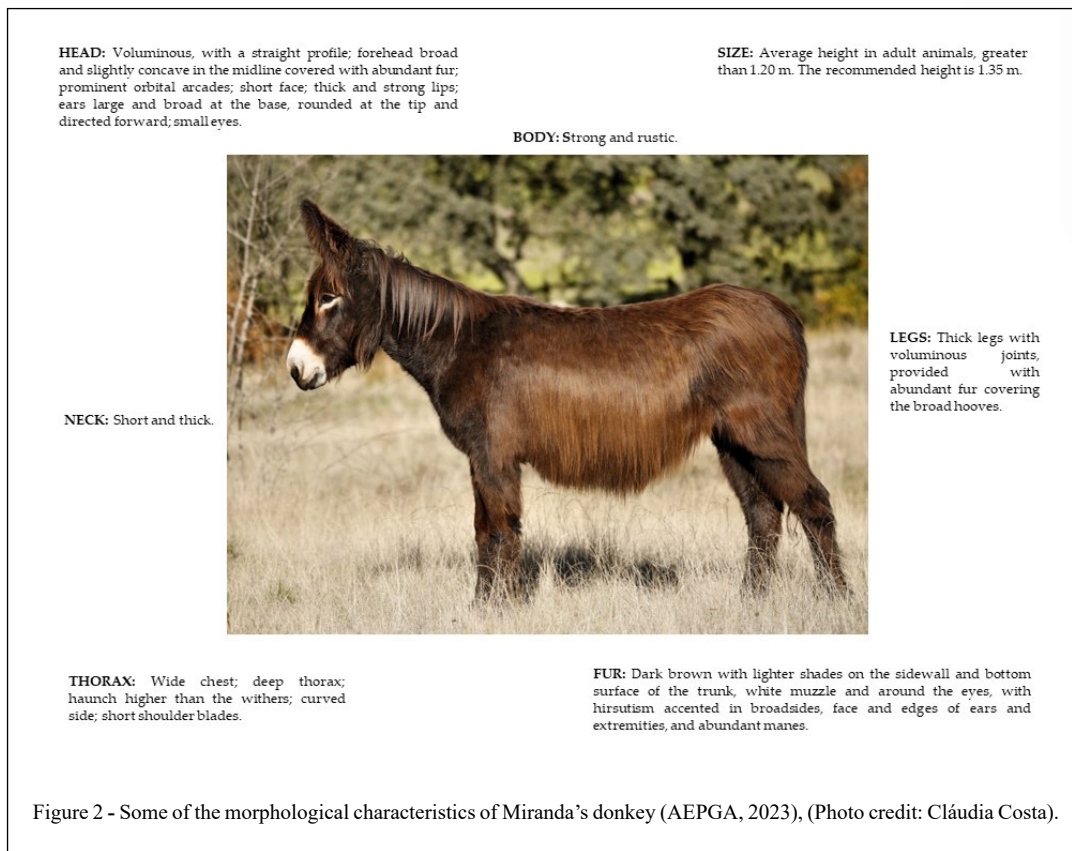
Parasites, especially those of the gastrointestinal tract, are clinically relevant in donkeys, and are present in animals of the Miranda's donkey breed in rehabilitation centers, small herds and on dairy farms. Seven studies on parasitology were reported, most were about epidemiology and control of gastrointestinal parasites, such as those of the genus *Strongyloides*, *Triodontophorus*, *Trichostrongylus*, *Cyathostomum* and *Oxyuris equi*, and another on the prevalence of protozoa of the genus *Toxoplasma gondii* and *Leishmania spp.* (LOBO & MATHEUS, 2012; CUNHA et al., 2012; SOUSA et al., 2012; COUTO et al., 2016; GOMES, 2017; RODRIGUES et al., 2019; SOUSA et al., 2021).

A seroprevalence analysis was performed to assess susceptibility to infection by *Toxoplasma gondii* and *Leishmania spp.* Although, a low number of reactive animals was identified it was confirmed that donkeys in northern Portugal are exposed to these parasites (RODRIGUES et al., 2019).

### Dentistry

Two studies were carried out on the prevalence of incisor and cheek teeth disorders in the Miranda's donkey. Both studies reported several changes that compromised the welfare of animals and reinforced the importance of regular dental care in





endangered breeds, improving their well-being and contributing to their preservation (RODRIGUES et al., 2013a; RODRIGUES et al., 2013b).

#### Clinical handling

Two studies that used technology and anatomy to facilitate veterinarians' handling of these animals were identified. The first study determined the best body points to evaluate the body condition score through ultrasonography, more accurate method than the traditional subjective visual methods (QUARESMA et al., 2012). The second consisted of the development of a weight tape for determining the animals weight without the need for a scale (QUARESMA et al., 2019).

#### Behavior

Only one study was reported in the area of behavior (PINTO, 2017). The researchers studied the individual and social behavior of Miranda's donkey and showed that these animals spend approximately 70% of their time feeding, 20% at rest and 10% in other activities, varying the frequency of these according to age and reproductive status. This

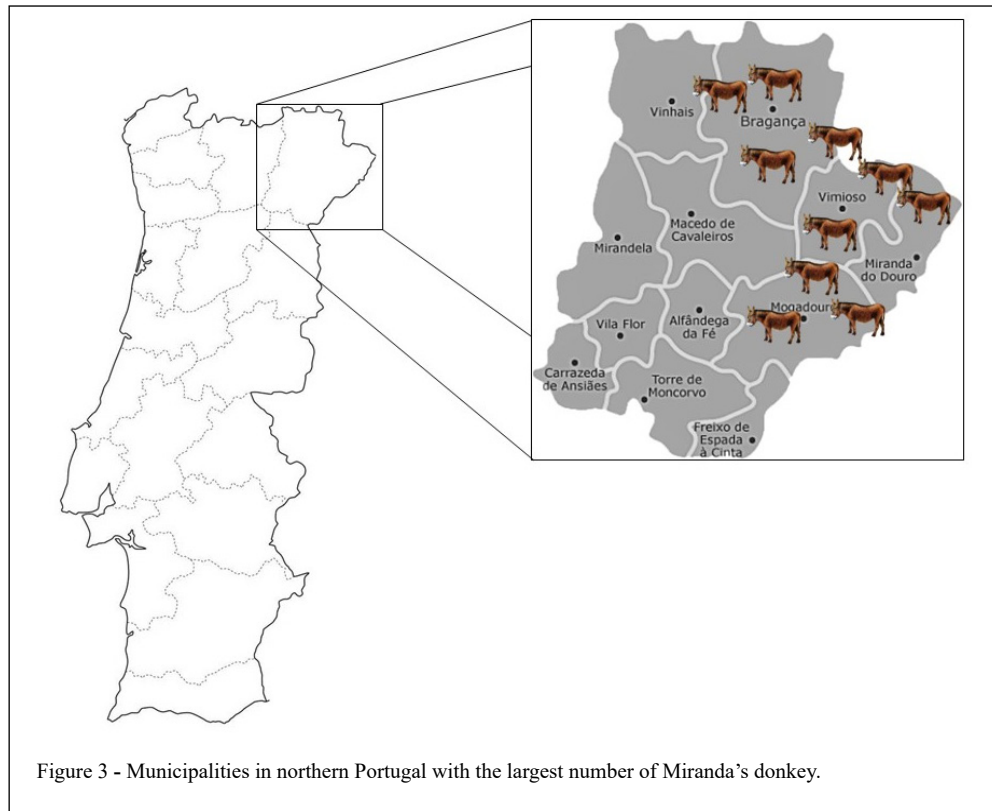
information helps in the management and interaction of these animals, improving research, the management of food resources and time, and the physical and psychological well-being of animals (PINTO, 2017).

#### One health

Miranda's donkeys are present in three surveys involving One health approach (CARVALHO et al., 2017; MALTEZ, 2018; SILVA et al., 2022). The first mentioned Miranda's donkey as a reservoir of antibiotic-resistant bacteria (*Escherichia coli* and *Enterococcus spp.*), which were found in the digestive tract of these animals and presented higher resistance to some antibiotics than those found in other domestic and wild animal species from Portugal and Spain (CARVALHO et al., 2017).

The second study used ultraviolet radiation to reduce the amount the total aerobic microorganisms in donkey milk used for human food. Donkey milk is highly suitable for consumption by individuals with dietary restrictions such as lactose intolerance (MALTEZ, 2018).

More recently, *Staphylococcus aureus* was isolated and identified in oral and nasal swab samples



of Miranda's donkey and its handlers. The donkey isolates were different from the human isolates, suggesting no zoonotic transmission between species. However, interestingly, *Staphylococcus aureus* isolates resistant to some antibiotics were reported in both species (SILVA et al., 2022).

#### *Clinical pathology*

Only one study was found in the area of clinical pathology that included 75 healthy animals (SILVA et al., 2023). This recent study determined the reference range for the hematological parameters of Miranda Donkey and evaluated the influence of gender and age on these parameters. No significant differences were found between gender (42 females and, 33 males), but for the age factor several hematological parameters showed statistical differences between young ( $n = 20$ ) and adult ( $n = 55$ ) animals (SILVA et al., 2023).

#### *Clinical pathology research conducted on Miranda's donkey and other european breeds*

In Europe, there are currently 28 autochthonous breeds with relatively complete phenotypic descriptions, and most of them are in danger of extinction (FAOSTAT, 2023), (Table 1).

Clinical pathology research has been performed only in 9 of those 28 European breeds, with a total of 16 published studies: Catalana ( $n = 2$ ), Ragusana ( $n = 1$ ), Âne du Cotentin and Âne Normand ( $n = 2$ ), Andaluza ( $n = 3$ ), Martina Franca ( $n = 4$ ), Balkan ( $n = 1$ ), Herzegovinian ( $n = 2$ ) and Miranda's donkey ( $n = 1$ ). The summarized results of those studies are described in table 2.

In all the studies, the authors emphasized the importance of determining clinical pathology parameters for the conservation and preservation of breeds and encourage further research with a larger number of animals and considering factors such as gender and age with in each population. Moreover, they also report that divergences between reference values of different breeds could also be explained by geographical, physiological, season, and climate differences, or even by body conditions of the animals, management, and sample size.

#### *Future perspectives on clinical pathology setting for Miranda's donkey*

Miranda's donkey constitutes a valuable Portuguese genetic heritage that must be protected, and efforts are still needed to increase the number of individuals, especially those of reproductive and fertile

Table 1 - List of European donkey breeds, countries of origin, and risk situation in 2022 (FAO).

Country	Breed	Trend
Croatia	Istrian donkey	Endangered maintained
	Littoral Dinaric donkey	Endangered
	North Adriatic donkey	Critical maintained
France	Âne Bourbonnais	Critical maintained
	Âne de Provence	Endangered maintained
	Âne des Pyrénées	Endangered maintained
	Âne du Cotentin	Endangered maintained
	Âne Grand Noir du Berry	Endangered maintained
	Âne Normand	Endangered maintained
	Baudet du Poitou	Endangered maintained
Hungary	Hungarian Plain donkey	Endangered
Italy	Asino dell'Amiata	Endangered maintained
	Asino dell'Asinara	Critical
	Asino di Pantelleria	Critical
	Asino Sardo	Endangered
	Grigio Viterbese	Critical
	Martina Franca	Endangered
	Ragusano	Endangered maintained
	Romagnolo	Endangered
Portugal	Burro de Graciosa	Critical
	Miranda's donkey	Endangered
Spain	Andaluza	Endangered
	Asno de las Encartaciones	Critical
	Balear	Critical maintained
	Catalana	Endangered
	Majorena	Extinct
	Zamorano-Leonés	Endangered
Serbia	Balkan donkey	Endangered maintained

age, to prevent the extinction of the breed (MARTA-COSTA et al., 2016). Knowledge on hematology and serum biochemistry and establishment of the respective reference values is essential as it allows to recognize and monitor sick individuals affected by acute or chronic diseases, as well as to promote conservation strategies and preservation measures (SILVA et al., 2023).

Miranda's donkey is a relatively recent identified autochthonous breed considered by FAO as an endangered breed. Moreover, amongst sexually mature animals there is a high predominance of females and elderly animals (QUARESMA et al., 2016). Although, there is a recent published research on reference values for this breed (SILVA et al., 2023), the authors defend continued studies on this topic as there are several aspects that need to be clarified. Since the pregnancy in this breed is long (12 months) and decisive for the success of conservation programs, it is necessary to explore changes on hematological and biochemical parameters in the different phases of the gestation period. Additionally,

it would also be very interesting to understand the changes associated with the geriatric age phase. Many animals are now at or over 15 years of age (data not shown), which is why it is important to know the physiological variables and how they may fluctuate in older animals. From the above, much remains to be studied in this breed. The authors; therefore, strongly encourage further research in this area.

## CONCLUSION

In the last decade, researchers' interest in the Miranda's donkey has grown and the number of publications has increased. We concluded that there are areas of greater interest to researchers, such as reproduction and parasitology. Clinical pathology, conversely, is still little explored, despite its importance, with only one recently published study. We hope that this review can be a valuable contribution so that researchers realise the need to continue studying this breed in greater depth, in all

Table 2 - Studies on the clinical pathology of autochthonous European donkey breeds.

Country	Breed	Area of clinical pathology	n	Gender	Age	*Gender and Age effect (P < 0,05)		Authors/Year
						Gender	Age	
Spain	Catalana	Biochemistry Hematology	98	45♀; 26♂	71A; 27Y		x	FOLCH et al. 1997
Spain	Catalana	Biochemistry	97	45♀; 26♂	71A; 26Y	x	x	JORDANA et al. 1998
Italy	Ragusano	Biochemistry Hematology	54		30A; 10Y; 14F		x	CALDIN et al. 2005
France	Normand and Cotentin	Biochemistry Hematology	83	57♀; 26♂	32A; 29Y; 22F	x	x	PITEL et al. 2006
France	Normand and Cotentin	Biochemistry Hematology	140	105♀; 35♂	68A; 22Y; 50F		x	COUROUCE-MALBLANC et al. 2008
Spain	Andaluza	Coagulation	38	20♀; 18♂				MENDOZA et al. 2011
Italy	Martina Franca	Biochemistry	10	5♀; 4♂	10F		x	D'ALESSANDRO et al. 2012
Italy	Martina Franca	Biochemistry Hematology	15		15F		x	VERONESI et al. 2014
Serbia	Balkan	Biochemistry Hematology	74		52A; 22Y		x	STANISIC et al. 2015
Bosnia	Herzegovinian	Hematology	30	18♀; 12♂				RUKAVINA et al. 2016
Bosnia	Herzegovinian	Hematology	30	18♀; 12♂		x	x	RUKAVINA et al. 2018
Spain	Andaluza	Hematology	80			x	x	GONZÁLEZ, 2017
Italy	Martina Franca	Biochemistry	10	10♀				GLORIA et al. 2018
Spain	Andaluza	Biochemistry	73	54♀; 13♂				PEREZ-ECIJA et al. 2020
Italy	Martina Franca	Biochemistry Hematology	81	64♀; 17♂	29A; 36Y; 16F		x	TRIMBOLI et al. 2020
Portugal	Miranda's donkey	Hematology	75	42♀; 33♂	55A; 20Y		x	SILVA et al. 2023

♀ - Females; ♂ - Males; A - Adults; Y - Young; F - Foals; \*Gender and Age effect on Hematology and/or Biochemistry values;

areas of knowledge and especially in the context of clinical pathology.

### ACKNOWLEDGEMENTS

The authors would like to thank the Centro de Ciência Animal e Veterinária (CECAV) and Associação para o Estudo e Proteção do Gado Asinino (AEPGA). This work was supported by National Funds by the Fundação para a Ciência e a Tecnologia (FCT) and Ministério da Ciência e Tecnologia (MCT) under the project UIDP/00772/2020. The authors also want to acknowledge the support received from projects LA/P/0059/2020, UIDB/04033/2020 and a Ph.D. Grant number UI/BD/150840, also financed by FCT.

### DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflict of interest. The funders had no role in the design of the study, in the collection, analyses, or interpretation of data, in the writing of the manuscript, or in the decision to publish the results.

### AUTHORS' CONTRIBUTIONS

Conceptualization, F. L. Q; writing-original draft preparation, G. S; writing-review and editing, G. S.; A. C. S-F; M. N.; F. L. Q; data analysis, G. S; visualization, M. N.; and supervision and re-sources, F. L. Q and A. C. S-F. All authors have read and agreed to the published version of the manuscript.



## REFERENCES

- ALFRANCA, I. S. El concepto de raza: Evolución y realidad. **Archivos de Zootecnia**, v.50, p.547-564, 2001. Available from: <<https://www.researchgate.net/publication/28057711>>. Accessed: May, 04, 2023. ISSN 0004-0592.
- ASSOCIAÇÃO PARA O ESTUDO E PROTEÇÃO DO GADO ASININO (AEPGA). 2023. Online. <<https://www.aepga.pt/>>. Accessed: Jan. 29, 2023.
- BATISTA, H. P. S. et al. Association for the study and protection of donkeys (APEGA): Burro de Miranda a species to be preserved. **Revista Internacional de Ciências, Tecnologia e Sociedade**, v.5, n.2, p.72-87, 2022. Available from: <[https://oasisbr.ibict.br/vufind/Record/RCAP\\_82737a6a53d1ce84316c142bb2f3107b](https://oasisbr.ibict.br/vufind/Record/RCAP_82737a6a53d1ce84316c142bb2f3107b)>. Accessed: Oct. 25, 2023. doi: 10.37334/riets.v5i2.74.
- CALDIN, M. et al. Reference ranges for haematology, biochemical profile and electrophoresis in a single herd of Ragusana donkeys from Sicily (Italy). **Comparative Clinical Pathology**, v.14, p.5-12, 2005. Available from: <<https://www.researchgate.net/publication/225929795>>. Accessed: Apr. 17, 2023. doi: 10.1007/s00580-005-0544-8.
- CARVALHO, I. et al. Antimicrobial-resistant *Escherichia coli* and *Enterococcus* spp. Isolated from Miranda donkey (*Equus asinus*): an old problem from a new source with a different approach. **Journal of medical microbiology**, v.66, n.2, p.191-202, 2017. Available from: <<https://pubmed.ncbi.nlm.nih.gov/28260587/>>. Accessed: Apr. 17, 2023. doi: 10.1099/jmm.0.000423.
- CARVALHO, M. C. B. **Biópsias uterinas na avaliação da integridade funcional do endométrio em burras do Planalto Mirandês**. 2016a. 80f. Dissertation (Integrated Master's in Veterinary Medicine), Universidade de Trás-os-Montes e Alto Douro, Vila real, Portugal. Available from: <<https://catalogo.biblioteca.utad.pt/cgi-bin/koha/opac-detail.pl?biblionumber=69044>>. Accessed: Apr. 17, 2023.
- CARVALHO, S. et al. Vaginal bacterial microbiota of an endangered donkey breed: a comparison between Miranda donkey breed (*Equus asinus*) jennies with and without reproductive problems. **Journal of Integrated OMICS**, v.6, n.1, p.18-22, 2016b. Available from: <<https://www.jiomics.com/index.php/jiomics/article/view/1116>>. Accessed: Feb. 17, 2023. doi: 10.5584/jiomics.v6i1.193.
- COUROUCE-MALBLANC, A. F. G. et al. Reference values on hematologic and biochemical parameters in French donkeys. In: INTERNATIONAL CONGRESS OF WORLD EQUINE VETERINARY ASSOCIATION, 10., 2008, Moscow, Russia. **Proceeding of the 10<sup>th</sup> International Congress of World Equine Veterinary Association**, Moscow: WEVA, 2008. p.458-460. Online. Available from: <<https://www.ivis.org/library/weva/weva-internal-congress-russia-2008/reference-values-on-hematologic-and-biochemical>>. Accessed: Jun. 28, 2023.
- COUTO, M. et al. Grazing behaviour of Miranda donkeys in a natural mountain pasture and parasitic level changes. **Livestock Science**, v.186, p.16-21, 2016. Available from: <<https://www.sciencedirect.com/science/article/abs/pii/S1871141316300051?via%3Dihub>>. Accessed: Apr. 17, 2023. doi: 10.1016/j.livsci.2016.01.005.
- CUNHA, B. et al. Gastrointestinal parasitism of the Miranda breed in Couço, Portugal. **Acta Parasitológica Portuguesa**, v.19, p.47-51, 2012. Available from: <<https://www.rcaap.pt/detail.jsp?id=oai:repositorio.ipsantarem.pt:10400.15/2691>>. Accessed: Apr. 17, 2023. doi: 10400.15/2691.
- CUSTÓDIO, C. A. P. **Analysis of behavior and seminal characteristics in the collection of semen with artificial vagina in Burro de Miranda: effect of pre-collection administration of PgF2a**. 2021. 115f. Dissertation (Integrated Master's in Veterinary Medicine), Universidade de Trás-os-Montes e Alto Douro, Vila Real, Portugal. Available from: <<https://www.aepga.pt/area/repositorio/teses/>>. Accessed: Apr. 17, 2023.
- D'ALESSANDRO, A. G. et al. Values of energetic, proteic and hepatic serum profiles in neonatal foals of the Martina franca donkey breed. Macedonian. **Journal Animal Science**, v.2, p.213-217, 2012. Available from: <<https://www.researchgate.net/publication/249992054>>. Accessed: Feb. 28, 2023. doi: 10.54865/mjas1222213da.
- DIREÇÃO GERAL DE ALIMENTAÇÃO E VETERINÁRIA (DGAV). 2021. **Catálogo Oficial de Raças Autoctóneas Portuguesas**. Online. Available from: <<https://www.dgav.pt/wp-content/uploads/2021/04/Catalogo-Oficial-Racas-Autoctones-Portuguesas.pdf>> Accessed: Apr. 29, 2023.
- FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS (FAOSTAT). 2023. **World Livestock: Domestic Animal Diversity Information System (DAD-IS)**. Online. Available from: <<https://www.fao.org/dad-is/browse-by-country-and-species/en/>>. Accessed: Apr. 14, 2023.
- FOLCH, P. et al. Reference ranges and the influence of age and sex on haematological values of the endangered Catalanian donkey. **Veterinary Journal**, v.154, p.163-8, 1997. Available from: <<https://www.sciencedirect.com/science/article/abs/pii/S1090023397800548>>. Accessed: Apr. 17, 2023. doi: 10.1016/S1090-0233(97)80054-8.
- FREIRE, A. F. C. **Determination of fetal age in Mirandese and Zamorano-Leonesa asinines by transrectal ultrasound**. 2020. 110f. Dissertation (Integrated Master's in Veterinary Medicine), Universidade de Trás-os-Montes e Alto Douro, Vila Real, Portugal. Available from: <<https://www.aepga.pt/area/repositorio/teses/>>. Accessed: Apr. 14, 2023.
- GLORIA, A. et al. Biochemical blood analysis along pregnancy in Martina Franca jennies. **Theriogenology**, v.115, p.84-9, 2018. Available from: <<https://pubmed.ncbi.nlm.nih.gov/29734125/>>. Accessed: Apr. 17, 2023. doi: 10.1016/j.theriogenology.2018.04.026.
- GOMES, M. C. S. **Perfil Parasitário de uma Exploração de Burras de Leite**. 2017. 61f. Dissertation (Integrated Master's in Veterinary Medicine), Universidade Lusófona de Humanidades e Tecnologia, Lisboa, Portugal. Available from: <<https://recil.ensinolusofona.pt/handle/10437/8486>>. Accessed: Apr. 14, 2023.
- GONZÁLEZ, C. C. A. **Validación del analizador hematológico lasercyte en Burros sanos**. 2017. 285f. Thesis (Doctorate in Veterinary Medicine), Universidad de Córdoba, Córdoba, Spain. Available from: <<https://helvia.uco.es/xmlui/handle/10396/14823>>. Accessed: Apr. 18, 2023.
- INGRASSIA, A. et al. The legal framework for the management of animal genetic resources. In: **FAO Legislative Study**. Rome, 2006. 150p. Online. Available from: <<https://www.fao.org/3/a0276e/a0276e.pdf>>. Accessed: Feb. 15, 2023.



- JORDANA, J. et al. Clinical biochemical parameters of the endangered Catalan donkey breed: normal values and the influence of sex, age, and management practices effect. **Research in Veterinary Science**, v.64, p.7-10, 1998. Available from: <<https://www.sciencedirect.com/science/article/abs/pii/S0034528898901072>>. Accessed: Apr. 17, 2023. doi: 10.1016/S0034-5288(98)90107-2.
- LOBO, L.; MATEUS, T. Andam Caudas de Rato nos Asininos da Raça de Miranda! **Acta Parasitológica Portuguesa**, v.19, p.43-45, 2012. Available from: <<https://www.researchgate.net/profile/Teresa-Mateus/publication/316582897>>. Accessed: Apr. 17, 2023.
- MALTEZ, J. P. F. V. **Efeito da radiação ultravioleta na contagem de microrganismos aeróbios totais no leite de burra da raça asinina de Miranda**. 2018. 91f. Dissertation (Integrated Master's in Veterinary Medicine), Universidade Lusófona de Humanidades e Tecnologias, Lisboa, Portugal. Available from: <<https://recil.ensinulusofona.pt/handle/10437/8827>>. Accessed: Apr. 29, 2023.
- MARQUES, M. H. A. G. **“O burro mirandês: A definição de um património. Estudo de caso numa aldeia da Terra de Miranda”**. 2006. 299f. Dissertation (Master's in Anthropology), Instituto Superior de Ciências do Trabalho e da Empresa, Lisboa, Portugal. Available from: <<https://www.aepga.pt/area/repositorio/teses/>>. Accessed: Apr. 16, 2023.
- MARTA-COSTA, A. et al. **Raças Autóctones no Espaço Ibérico: um recurso sustentável**. UTAD, Portugal, 2016. 138p. Online. Available from: <<https://www.researchgate.net/publication/311103718>>. Accessed: Feb. 20, 2023. ISBN: 978-989-704-039-9.
- MARTINS-BESSA, A. et al. Age-related linear and nonlinear modelling of semen quality parameters in Miranda donkeys. **Italian Journal of Animal Science**, v.20, n.1, p.1029-1041, 2021a. Available from: <<https://www.researchgate.net/publication/353334758>>. Accessed: Apr. 18, 2023. doi: 10.1080/1828051X.2021.1933629.
- MARTINS-BESSA, A. et al. Bayesian linear regression modelling for sperm quality parameters using age, body weight, testicular morphology, and combined biometric indices in donkeys. **Animals**, v.11, p.176, 2021. Available from: <<https://www.mdpi.com/2076-2615/11/1/176>>. Accessed: Apr. 20, 2023. doi: 10.3390/ani11010176.
- MARTINS-BESSA, A. et al. Linear and non-linear regression model fitting of testicular three-dimensional growth in Miranda donkeys. **Italian Journal Animal Science**, v.20, n.1, p.1518-1531, 2021. Available from: <<https://www.researchgate.net/publication/355379583>>. Accessed: Apr. 23, 2023. doi: 10.1080/1828051X.2021.1965046.
- MENDOZA, F. J. et al. Coagulation profiles of healthy Andalusian donkeys are different than those of healthy horses. **Journal of Veterinary Internal Medicine**, v.25, p.967-970, 2011. Available from: <<https://pubmed.ncbi.nlm.nih.gov/21736621/>>. Accessed: Apr. 25, 2023. doi: 10.1111/j.1939-1676.2011.0748.x.
- MOREIRA, J. et al. Early embryonic development and indicators of early embryonic death in Asinina de Miranda jennies (*Equus asinus*). **Reproduction in Domestic Animals**, v.50, n.3, p.67, 2015. Available from: <<https://onlinelibrary.wiley.com/journal/14390531>>. Accessed: May, 11, 2023. doi: 10.1111/rda.12581 2015.
- PAYAN-CARREIRA, R.; QUARESMA, M. Cervical hypoplasia in jennets: a description of two cases. **Reproduction in Domestic Animals**, v.45, n.3, p.540-544, 2010. Available from: <<https://onlinelibrary.wiley.com/doi/10.1111/j.1439-0531.2008.01231.x>>. Accessed: Apr. 25, 2023. doi: 10.1111/j.1439-0531.2008.01231.x
- PEREZ-ECIJA, A. et al. Reference intervals of acute phase proteins in healthy Andalusian donkeys and response to experimentally induced endotoxemia. **Journal of Veterinary Internal Medicine**, v.35, p.580-589, 2020. Available from: <<https://pubmed.ncbi.nlm.nih.gov/33336874/>>. Accessed: Apr. 25, 2023. doi: 10.1111/jvim.16015.
- PINTO, M. P. **O burro como animal de companhia – Etograma e Estudo do comportamento**. 2017. Online. Available from: <<https://www.aepga.pt/area/repositorio/estudos/>>. Accessed: May, 10, 2023.
- PITEL, P. et al. Approche des valeurs hématologiques et biochimiques chez deux races asines. **Veterinary Equine Practice**, v.38, p.19-25, 2006. Available from: <<http://asineriedubocage.com/documents/anes%20PVE.PDF>>. Accessed: May, 01, 2023.
- QUARESMA, M. et al. The Asinine race of Miranda. **Portuguese Journal of Veterinary Sciences**, v.553, p.173-177, 2005. Available from: <<https://www.researchgate.net/publication/240615272>>. Accessed: Apr. 15, 2023.
- QUARESMA, M. et al. Bilateral ovulation fossa inclusion cysts in Miranda jennets. **Journal of Comparative Pathology**, v.145, n.4, p.367-372, 2011. Available from: <<https://www.sciencedirect.com/science/article/abs/pii/S0021997511000338>>. Accessed: Apr. 18, 2023. doi: 10.1016/j.jcpa.2011.03.004.
- QUARESMA, M. et al. Pedigree and herd characterization of a donkey breed vulnerable to extinction. **Animals**, v.8, p.354-359, 2013. Available from: <<https://www.sciencedirect.com/science/article/pii/S1751731113002218>>. Accessed: Apr. 10, 2023. doi: 10.1017/s1751731113002218.
- QUARESMA, M. et al. Viability analyses of an endangered donkey breed: The case of the Asinina de Miranda (*Equus asinus*). **Animal Production Science**, v.55, n.9, p.1184-1191, 2014. Available from: <<https://www.researchgate.net/publication/266206694>>. Accessed: Apr. 20, 2023. doi: 10.1071/AN13307.
- QUARESMA, M.; PAYAN-CARREIRA, R. Characterization of the estrous cycle of Asinina de Miranda jennies (*Equus asinus*). **Theriogenology**, v.83, n.4, p.616-624, 2015. Available from: <<https://pubmed.ncbi.nlm.nih.gov/25467767/>>. Accessed: May, 11, 2023. doi: 10.1016/j.theriogenology.2014.10.021.
- QUARESMA, M. et al. Reproductive Patterns in the Non-Breeding Season in Asinina de Miranda Jennies. **Reproduction in Domestic Animals**, v.50, p.784-792, 2015. Available from: <<https://pubmed.ncbi.nlm.nih.gov/26281788/>>. Accessed: May, 10, 2023. doi: 10.1111/rda.12588.
- QUARESMA, M. et al. Demographic studies of the Miranda Donkey breed – The Challenges To Avoid Extinction. In: MARTA-COSTA, A. et al. **Raças Autóctones no Espaço Ibérico: um recurso sustentável**. UTAD: Portugal, 2016. p.56-64. Online. Available from: <<https://www.researchgate.net/publication/311103718>>. Accessed: Apr. 20, 2023.
- QUARESMA, M. et al. “Relationship between ultrasound subcutaneous fat depth, body condition score and body

- subcutaneous fat distribution in Miranda breed jennets". **EAAP Scientific Series**, v.132, n.1, p.391-398, 2012. Available from: <<https://www.researchgate.net/publication/256654691>>. Accessed: Jun. 07, 2023. doi: 10.3920/978-90-8686-755-4\_56.
- QUARESMA, M. et al. Estimation of live weight by body measurements in the Miranda donkey breed. **Journal Equine Veterinary Science**, v.79, p.30-34, 2019. Available from: <<https://www.sciencedirect.com/science/article/abs/pii/S0737080619301078>>. Accessed: May, 14, 2023. doi: 10.1016/j.jevs.2019.05.014.
- QUARESMA, M. et al. Selenium and vitamin E concentrations in Miranda Jennies and Foals (*Equus asinus*) in Northeast Portugal. **Animals**, v.11, n.1772, p.1-13, 2021. Available from: <<https://pubmed.ncbi.nlm.nih.gov/34198534/>>. Accessed: May, 15, 2023. doi: 10.3390/ani11061772.
- QUARESMA, M. et al. Follicular dynamics during the non-reproductive season. **Reproduction in Domestic Animals**, v.57, p.616-624, 2022. Available from: <<https://pubmed.ncbi.nlm.nih.gov/35191106/>>. Accessed: Jun. 10, 2023. doi: 10.1111/rda.14102.
- RADAR-CHAFIROVITCH, A. et al. Is endometriosis in Miranda Donkeys influenced by eosinophils and age? In: CISSA CONGRESS 2022, Lisboa, Portugal. **Proceedings of CISSA Congress 2022 – Innovation in Animal and Veterinary Biomedical Research**, Lisboa, CISSA's, 2022. p.115. Online. Available from: <<https://congressociisa.fmv.ulisboa.pt/assets/files/AbstractsCISSACongress.pdf>>. Accessed: Nov. 01, 2023.
- RODRIGUES, J. B. et al. A Clinical survey Evaluating the Prevalence of Incisor Disorders in Zamorano-Leonés and Mirandês Donkeys (*Equus asinus*). **Journal Equine Veterinary Science**, v.33, n.9, p.710-718, 2013a. Available from: <<https://www.sciencedirect.com/science/article/abs/pii/S0737080612008763>>. Accessed: Jun. 12, 2023. doi: 10.1016/j.jevs.2012.11.012.
- RODRIGUES, J. B. et al. A clinical survey on the prevalence and types of cheek teeth disorders present in 400 Zamorano-Leonés and 400 Mirandês donkeys (*Equus asinus*). **Veterinary Record**, v.179, p.581-588, 2013b. Available: <<https://pubmed.ncbi.nlm.nih.gov/24296357/>>. Accessed: Jun. 12, 2023. doi: 10.1136/vr.101747.
- RODRIGUES, F. T. et al. Seroprevalence of *Toxoplasma gondii* and *Leishmania* spp. in domestic donkeys from Portugal. **Revista Brasileira de Parasitologia Veterinária**, v.28, n.1, p.172-176, 2019. Available from: <<https://pubmed.ncbi.nlm.nih.gov/30892383/>>. Accessed: Jun. 07, 2023. doi: 10.1590/S1984-296120180091.
- RUKAVINA, D. et al. Preliminary study of hematological parameters in Herzegovinian Donkey. **Veterinarska Stanica**, v.65, n.1, p.25-26, 2016. Available from: <<https://www.researchgate.net/publication/303228743>>. Accessed: Jul. 11, 2023.
- RUKAVINA, D. et al. The influence of gender and age on haematological parameters in Herzegovinian donkey. **Veterinarska Stanica**, v.49, p.22-26, 2018. Available from: <<https://hrcak.srce.hr/file/325701>>. Accessed: Jul. 11, 2023.
- SAMÕES, L. M. R. **Estudo do Gado Asinino no Parque Natural do Douro Internacional**. 2000. Dissertation (Master's in Zootechnics), Instituto Politécnico de Bragança, Bragança, Portugal. Available from: <<https://bibliotecadigital.ipb.pt/handle/10198/5464>>. Accessed: Apr. 15, 2023.
- SEDLINSKA, M. et al. Biochemical and haematological profile of donkeys in the Czech and Slovak Republics: influence of age and sex. **Acta Veterinaria**, v.86, p.151-157, 2017. Available from: <<https://actavet.vfu.cz/86/2/0151/>>. Accessed: Aug. 01, 2023. doi: 10.2754/avb201786020151.
- SILVA, V. et al. One Health Approach Molecular Analysis of *Staphylococcus aureus* Reveals Distinct Lineages in Isolates from Miranda Donkeys (*Equus asinus*) and Their Handlers. **Antibiotics**, v.11, n.374, p.1-11, 2022. Available from: <<https://pubmed.ncbi.nlm.nih.gov/35326837/>>. Accessed: Jun. 14, 2023. doi: 10.3390/antibiotics11030374.
- SILVA, G. et al. Establishment of reference Intervals of Hematological Parameters and Evaluation of Sex and Age Effect in the Miranda Donkey. **Animals**, v.13, n.2331, p.1-10, 2023. Available from: <<https://www.mdpi.com/2076-2615/13/14/2331>>. Accessed: Oct. 18, 2023. doi: 10.3390/ani13142331.
- SOUSA, S. et al. Parasite control in Miranda Donkeys as a way of keeping animal welfare. **EAAP Scientific Series**, v.132, n.1, p.425-431, 2012. Available from: <<https://www.researchgate.net/publication/285066244>>. Accessed: Jun. 22, 2023. doi:10.3920/978-90-8686-755-4\_60.
- SOUSA, S. R. et al. Gastrointestinal parasitism in Miranda donkeys: Epidemiology and selective control of strongyles infection in the northeast of Portugal. **Animals**, v.11, n.1, p.1-18, 2021. Available from: <<https://pubmed.ncbi.nlm.nih.gov/33440886/>>. Accessed: Jun. 25, 2023. doi: 10.3390/ani11010155.
- SPREGA- SOCIEDADE PORTUGUESA DE RECURSOS GENÉTICOS ANIMAIS. **Asininos – Raça Burro de Miranda**. 2023. Online. Available from: <<https://www.sprega.com.pt/conteudo.php?idesp=asininos&idraca=Burro%20de%20Mirand>>. Accessed in: May, 13, 2023.
- STANISIC, L. et al. Morphological, biochemical and hematological characterization of endangered Balkan donkey breed. **Acta Veterinaria**, v.65, p.125-136, 2015. Available from: <<https://www.researchgate.net/publication/272069643>>. Accessed: Aug. 11, 2023. doi:10.1515/acve-2015-0010.
- TRIMBOLI, F. et al. Reference Ranges for Hematological and Biochemical Profile of Martina Franca Donkeys. **Frontiers in Veterinary Science**, v.7, p.1-7, 2020. Available from: <<https://www.frontiersin.org/articles/10.3389/fvets.2020.602984/full>>. Accessed: Jul. 07, 2023. doi: 10.3389/fvets.2020.602984.
- VERONESI, M. C. et al. Blood analysis in newborn donkeys: hematology, biochemistry, and blood gases analysis. **Theriogenology**, v.82, n.2, p.294-303, 2014. Available from: <<https://pubmed.ncbi.nlm.nih.gov/24831574/>>. Accessed: Jul. 27, 2023. doi: 10.1016/j.theriogenology.2014.04.004.