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Présentée par
Sofiane AISSANOU

Thème

Caractérisation de la saison de reproduction chez l'âne commun (*Equus asinus*) en Algérie

Soutenue le : 31 Juin 2022 **Devant le Jury composé de :**

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Dédicaces

Ce travail s'achève avec l'aide de Dieu, le tout puissant !

A mes très chers parents, source de vie, d'amour et d'affection

A ma très cher unique sœur Kenza source de joie et de bonheur

A tous ceux qui porte le nom AISSANOU

A tous mes amis, tout particulièrement, Ilyas et Anis

A vous, chers lecteurs

A la mémoire de mon grand-père, paix à son âme

"What doesn't kill you makes you stronger"

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Abréviations

FAO : Organisation des Nations unies pour l'alimentation et l'agriculture

Ch : Cholestérol total

T : Testostérone

Tg : Triglycéride

AST : Aspartate aminotransférase

ALT : Alanine aminotransférase

ALP : Phosphatase alcaline

TL : Longueur testiculaire

TH : Hauteur testiculaire

TW : Largeur testiculaire

TWe : Poids testiculaire

TV : Volume testiculaire

BW : Poids corporel

BCS : Score du poids corporel

EWe : Poids épididymaire

CED : Diamètre de la queue épididymaire

EV : Volume épididymaire

SC : Circonférence scrotale

GSI : Rapport Gonado-somatique

DL : Diamètre de la lumière

DST : Diamètre des tubes séminifères

GCEH : Hauteur des cellules germinales

ITSA : Surface inter-tubulaire

TTA : Épaisseur de l'albuginée

U/L : Unité internationale

Mmol/l: Millimoles par litre

Ng/ml : Nanogramme par millilitre

µm : Micromètre

$µm^2$: Micromètre carré

Cm : Centimètre

Cm^3 : Centimètre cube

Sommaire

SOMMAIRE

Introduction générale

Depuis sa domestication, l’âne commun (*Equus asinus*) a toujours accompagné l'être humain tout au long de son histoire (**Mitchell, 2018**). L'âne est un ongulé à doigts impairs et la plus petite espèce de la famille des équidés (**Grinder et al., 2006**). L'âne est de nature très amicale, calme, tranquille, patiente, intelligente, prudente, joueuse et désireuse d'apprendre et d'apprécier la compagnie des humains. Il est caractérisé par des pattes courtes et des oreilles exceptionnellement longues.

L'âne commun représente une composante importante du cheptel algérien car il joue un rôle important dans l'économie agricole, notamment comme moyen de transport dans les régions semi-arides et aussi l'utilisation croissante de ces animaux pour la production laitière dans certains pays. Selon les statistiques récentes; la population asine est estimée à **180 160** têtes en Algérie (**FAOSTAT, 2018; Noriss et al., 2018**) que l'on trouve essentiellement dans les régions du nord, où ils sont particulièrement aptes à tolérer les dures conditions de travail. Les populations asines dans le monde ont considérablement diminué avec l'avènement de la motorisation dans les transports et le travail, ce qui constitue un risque d'extinction de l'espèce dans le monde. Cependant, l'âne algérien n'a pas reçu suffisamment de considération scientifique comme les autres espèces animales avec peu de littérature disponible notamment sur le plan reproduction.

Il est très nécessaire d'avoir une parfaite connaissance des changements physiologiques saisonniers afin d'optimiser les caractéristiques de reproduction chez l'âne. Cela peut contribuer à l'augmentation des populations d'ânes, favoriser la biodiversité de l'espèce et éviter le risque de son extinction. En Algérie, la caractérisation de la reproduction des ânes n'a jamais été réalisée pour déterminer la saison de reproduction.

L'analyse morphométrique des testicules a été déjà entreprise chez beaucoup d’espèce animales pour prédire la production de sperme, les potentiels de stockage et la capacité fécondante du mâle reproducteur en fonction de plusieurs facteurs tels que l'âge, la photopériode et la qualité de l’aliment (**Ginther et al.,1987; Henry et al., 1987 ; Alemayehu & Benti, 2009 ; Carluccio et al., 2013 ; Rua et al., 2017**). De même, les données sur la taille des testicules peuvent être utilisées pour comprendre la fonction testiculaire et fournir une meilleure connaissance des différentes phases de la spermatogenèse, et aident à caractériser la puberté et la maturité sexuelle (**Nipken & Wrobel, 1997 ; Assis Neto et al., 2003 ; Moustafa et al., 2005**).

De nombreuses enquêtes quantitatives ont été menées pour évaluer la structure et la fonction testiculaires afin de décrire la fonction de reproduction chez les mammifères (**Macedo et al., 2011 ; Peixoto et al., 2012 ; Silva et al., 2011 ; Ajani et al., 2015 ; Oliveira et al., 2016 ; Rakesh et al., 2014 ; Gemada & Workalemahu, 2017**). L'analyse biométrique de certains paramètres, tels que la circonférence scrotale (SC), le poids testiculaire (TW), la longueur testiculaire (TL) et le volume testiculaire (TV) a également été réalisée chez des boucs (**Ajao et al., 2014**), des béliers (**Ibrahim et al., 2012**), des taureaux (**Jain et al., 2008**) et des chameaux (**Pasha et al., 2011**). Il a été établi que l'étude morphométrique des testicules, la mesure des testicules et les changements saisonniers qui ont lieu tout au long de l'année ont été bien documentés pour les chèvres (**Raji et al., 2008**), les béliers (**Divya et al., 2013**) et les taureaux **(Da silva santos et al., 2013**). Les données générées à partir d'études morphométriques ont été positivement corrélées avec la circonférence, le poids et le volume des testicules chez les taureaux de boucherie (**Cartee et al., 1989**). Cependant, il existe peu d'études dans la littérature concernant l'analyse biométrique testiculaire et épididymaire chez l'âne (**Morais et al., 1993 ; Carluccio et al., 2004 ; Alemayehu & Benti, 2009**).

L'espèce équine présente une reproduction saisonnière atypique, dont la spermatogenèse est continue tout au long de l'année, mais avec une diminution de la spermatogenèse par rapport à celle de la saison de reproduction (**Hafez & Hafez, 2000**). Dans la littérature, la saisonnalité de la reproduction n'est pas aussi claire chez les ânes, bien que les modifications de l'activité sexuelle soient corrélées à la période de l'année, à la zone géographique, aux conditions environnementales et à la disponibilité de la nourriture, mais aussi affectées par des facteurs tels que l'âge et les variations de climat (**Tibary et al., 2006; Miragaya et al, 2018**). L'étude biométrique quantitative des testicules reste insuffisante pour déterminer la dynamique de reproduction des ânes (**Aissanou & Ayad, 2020**). La quantification histologique du parenchyme testiculaire tel que le diamètre des tubules, la hauteur de l'épithélium des cellules germinales sont importantes pour étudier l'appareil reproducteur masculin (**Helbig et al., 2006 ; Ibrahim et al., 2013 ; Shukla et al., 2013**). Les proportions volumétriques des tubules séminifères et des tissus intertubulaires tels que le diamètre des tubules et l'épaisseur des séminifères sont directement liés à l'activité sexuelle du mâle (**Paula et al., 1999**).

Le testicule de mammifère est l'organe reproducteur qui contient de nombreux compartiments différents, à savoir les tubules séminifères, l'interstitium et les types de cellules. Les tubules séminifères sont le siège de la spermatogenèse, tandis que l'interstitium est responsable de la biosynthèse des androgènes et de la sécrétion paracrine (**Petersen et al., 2015**) qui contiennent les cellules de Leydig, les vaisseaux sanguins et lymphatiques ainsi que les nerfs du parenchyme testiculaire (**Diagone et al., 2012**). Les différentes étapes du processus de spermatogenèse ont été bien documentées dans la littérature (**Chiarini-Garcia et al., 2009; Moustafa et al., 2015; Neves et al., 2018**), mais l'impact des variations saisonnières sur les compartiments testiculaires à l'aide des mesures histo-photométriques est limité.

La quantification histologique du parenchyme testiculaire constitue un indicateur important de l'activité spermatogène, fournissant des informations sur le niveau de la spermatogenèse (**Altinsaat et al., 2009; Rua et al., 2017**). La composition et la morphométrie des tissus testiculaires peuvent présenter des inférences sur une compréhension précieuse de la physiologie de la reproduction des espèces d'équidés (**Chiarini-Garcia et al., 2009 ; Han et al., 2016**). Ainsi, de nombreuses études ont été réalisées sur la base de l'examen qualitatif et quantitatif du processus spermatogène afin d'évaluer les paramètres physiologiques de la biologie de la reproduction (**Azvedo et al., 2010 ; Costa et al., 2011**).

Les mécanismes régulant la fonction reproductive reposent sur la relation permanente entre le système nerveux central et les gonades, qui est assurée par les hormones stéroïdiennes et gonadotrophiques. Cependant, cette régulation hormonale est influencée par différents facteurs environnementaux tels que l'âge et la saison (**Vasantha et al., 2016**). Les variations saisonnières des équidés sont dues au changement de la durée du jour tout au long de l'année. Les espèces animales à reproduction saisonnière, notamment les ânes, expriment des variations saisonnières de leur activité sexuelle. Il est connu dans la littérature que la testostérone est une hormone stéroïde qui est la clé principale de la spermatogenèse et du contrôle du comportement sexuel des mâles ; et son dosage est un meilleur outil pour déterminer la saison de reproduction. Malgré la controverse scientifique sur la saisonnalité chez les ânes, elle pourrait probablement être influencée par la photopériode et d'autres facteurs tels que la nutrition et la température.

D'autre part, les paramètres biochimiques sont couramment utilisés pour évaluer l'état physiologique ou pathologique de l'animal, cependant, les valeurs de ces paramètres varient selon les populations, le sexe, l'âge, la nutrition et la saison (**Longodor et al., 2020 ; Tesfaye et al., 2014 ; Girardi et al., 2014**). Il a été rapporté que l'activité de l'enzyme transaminase (AST et ALT) est un bon indicateur de la qualité du sperme (**Corteel et al., 1980**). De plus, une relation a été trouvée entre l'activité enzymatique de l'ALT et de l'ALP et la fonction de reproduction (**Longodor et al., 2020 ; Hussein et al., 2017**). De nombreuses études ont montré que la testostérone a une forte influence sur les hépatocytes et induit plus de variation dans les fonctions enzymatiques (**Azani et al., 2018 ; Charni-Natan et al., 2019**). De plus, il a été rapporté que les androgènes mâles augmentent le cholestérol sanguin **(Guyton, 1981**).

Malgré la valeur économique des ânes en étant que la principale source de transport et de traction dans les zones aux reliefs difficiles, il existe peu d'études publiées sur la physiologie de reproduction. Aussi, à notre connaissance, l'influence de la saison et de l'âge sur la biométrie testiculaire et épididymaire et la morphométrie des tissus testiculaires n'a jamais été rapportée chez l'âne algérien. Ces connaissances peuvent améliorer les informations disponibles sur la biométrie testiculaire des ânes, en particulier la race locale d'Algérie. Il est donc important d'étudier les modifications histologiques des tubules séminifères et des tissus interstitiels, ce qui peut servir de base à une meilleure compréhension de leur reproduction. De même, aucune étude n'a été réalisée pour décrire les concentrations saisonnières de la testostérone de l'âne domestique (*Equus asinus*) dans le nord de l'Algérie. Nous avons émis l'hypothèse que le cholestérol total, les triglycérides, l'aspartate aminotransférase, l'alanine aminotransférase et la phosphatase alcaline pourraient influencer les concentrations de testostérone chez les ânes domestiques.

C'est dans ce contexte que ce travail a été entamé pour contribuer à la caractérisation de la saison de reproduction chez l'âne domestique en Algérie. Les objectifs tracés dans cette thèse sont :

1. Etudier les caractéristiques biométriques testiculaires et épididymaire des ânes algériens tout au long de l'année selon l'âge, le poids corporel et les changements saisonniers. La corrélation entre les mesures morphométriques testiculaires et corporelles a également été estimée.
2. Mettre en évidence les changements histomorphométriques des tissus testiculaires selon la saison et l'âge chez les ânes (*Equus asinus*) dans les conditions du nord de l'Algérie.

3. Déterminer les changements de la testostérone plasmatique chez les ânes du nord de l'Algérie, les coefficients de corrélation entre les concentrations de T et les paramètres biochimiques ont été calculés.

Ce présent travail est subdivisé en deux grandes parties. La première partie est consacrée à une mini-synthèse bibliographique sur quelques aspects de la fonction reproduction chez le male de l'âne (*Equus asinus*) faisant des rappels anatomiques de l'appareil génital et décrivant la saisonnalité de reproduction. La deuxième partie présente les expériences réalisées et les résultats obtenus. Ces travaux de recherches nous ont permis de publier quatre articles, à savoir :

1. **Sofiane Aissanou**, Omar Besseboua, Abdelhanine Ayad (2022). Some reproductive characteristics in common donkey male (*Equus asinus*)-A mini review. *Turkish Journal of Veterinary Research,* (Accepté).
2. **Sofiane Aissanou**, Abdelhanine Ayad. Influence of age, body weight and season on testicular and epidydimal biometrics in donkeys (*Equus asinus*). *International Journal of Morphology*, 2020, 38(5), 1434-1443.
3. **Sofiane Aissanou**, Abdelhanine Ayad (2022). Histomorphometric changes of testicular tissues by season and age of Algerian local donkeys (*Equus asinus*). *Acta Scientiarum-Animal Sciences*, (Accepté)
4. **Sofiane Aissanou**, Omar Besseboua, Mokhtar Benhanifia, Abdelhanine Ayad (2022). Seasonal changes in plasma testosterone and biochemical parameters of male donkey (*Equus asinus*) in northern Algeria. *International Journal of Veterinary and Animal Research (IJVAR)*, (Accepté).

Synthèse bibliographique

Résumé

Contrairement à la plupart des espèces d'élevage domestique, l'âne commun (*Equus asinus*) est largement connu comme un animal avec une saisonnalité marquée dans l'activité de reproduction. Le cycle annuel de la photopériode a été identifié comme le facteur déterminant de l'activité sexuelle.

Une mini-synthèse des particularités de la reproduction des ânes est importante et constitue une base de réflexion scientifique pour gérer les élevages d'ânes et établir un plan de conservation des différentes races à travers le monde. Il est nécessaire d'avoir une parfaite connaissance des changements physiologiques saisonniers afin d'optimiser les caractéristiques de reproduction chez les ânes.

L'objectif de cet article est de faire le point sur l'état actuel des connaissances sur la saisonnalité de la reproduction chez l'âne commun. Nous commençons par un rappel de quelques aspects anatomiques de l'appareil génital et du comportement sexuel. Par ailleurs, les investigations menées par de nombreux auteurs révèlent l'influence de la saison sur la biométrie testiculaire, l'histologie, les paramètres séminaux et hormonaux dans les populations d'ânes mâles.

En conclusion, malgré la controverse scientifique sur le caractère saisonnier de la reproduction chez l'âne, celui-ci pourrait vraisemblablement être influencé par plusieurs facteurs principalement la photopériode.

Some reproductive characteristics in common donkey male (*Equus asinus*)-A mini review

Sofiane Aissanou[1], Omar Besseboua[2], Abdelhanine Ayad[1,*]

[1]Department of Physical Biology and Chemistry, Faculty of Nature and Life Sciences, University of Bejaia, 06000, Bejaia, Algeria.
[2]Department of Agronomic and Biotechnological Sciences, Faculty of Nature and Life Sciences, University H. Benbouali, 02000, Chlef, Algeria.

Running head:

*Corresponding author: Prof. Dr. Abdelhanine Ayad, Email: abdelhanine.ayad@univ-bejaia.dz
S. Aissanou, Orcid ID: https://orcid.org/0000-0002-2414-4485
O. Besseboua, Orcid ID : https://orcid.org/0000-0003-1365-4479
A. Ayad, Orcid ID: https://orcid.org/0000-0002-9325-7889

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Abstract

In contrary to most domestic livestock species, the common donkey (*Equus asinus*) is widely known as an animal with a marked seasonality in reproductive activity. The annual cycle of daily photoperiod has been identified as the determining factor in the sexual activity. A synthesis of the particularities of donkey reproduction is important and constitutes a basis of scientific reflection for managing asine livestock farms and to establish a well conservation plan for the different breeds around the world. It is necessary to have a perfect knowledge of the seasonal physiological changes in order to optimize the reproductive characteristics in donkeys. The objective of this paper is to review the current state of knowledge on the reproduction seasonality in common donkey. We start with a remainder of some anatomical of the genital apparatus and sexual behavior aspects. Moreover, the investigations undertaken by many authors reveal the influence of the season on testicular biometry, histology, seminal and hormonal parameters in male donkeys' populations. In conclusion, despite scientific controversy on reproductive seasonal character in donkeys, it could likely be influenced by several factors mainly the photoperiod.

Keywords: sexual behavior, sperm, hormone, seasonality, donkey males

Introduction

For as long as donkeys were known, the female was always more scientifically interested than the male. Many of the earliest accounts of donkeys were only interested in the female. However, the male donkey (ass, jackass) has many interesting characteristics and there is an abundant literature dating back to the past century. For some years now, donkeys have been enjoying increasing popularity in Central European countries in equine breeders especially in, Italy, Greece, Spain and France, and this for various therapies involving interactions with horses and other equines which are used for children or people with disabilities, such as horseback riding, equine assisted learning and ecotourism. Field reports revealed continuing decline in world populations of the asinine species (Kugler et al., 2008) with the advent of motorization in transport and work (Figure 1), this can be a risk of extinction of the species and cause negative impact on animal biodiversity. A synthesis of the particularities of donkey reproduction is important and constitutes a basis of scientific reflection for managing asine livestock farms and to establish a well conservation plan for the different breeds around the world (Rodrigues et al., 2021). The aim of the present article is to give a mini-review describing the anatomical and physiological aspects of donkey male's reproduction.

Histoanatomy of the genital apparatus

Genital tract

The donkey penis and prepuce are anatomically similar to the stallions (Hagstrom, 2004). In donkeys, the penis in resting measures 50 cm long for a diameter of 2.5 at 5cm, and presence of two nipples on each side of the sheath (Canisso et al., 2019). The penis body is clearly protruding along the entire length of the copulator organ and is enveloped almost up to its tip by the bulbous spongy muscle (Chabchoub et al., 2007). The dilatation of the glans is more important in donkeys (Tibary et al., 2006) and its length increase clearly in erection, it can measure from 35.5 to 45.5 cm diameter for a small donkey weighing 100 kg (Purdy, 2010).

The prostate forms a large glandular structure which consisted of right and left lobes, extended over the dorsal face and lateral faces of the urethra (Morel, 2003). It shows an isthmus almost a centimeter thick and 2 to 3 cm long in the cranio-caudal direction, of a slate color and becoming yellowish after castration in donkeys. The prostate of the donkey is enveloped by thick capsule which have outer fibrous and inner fibro-muscular layers. It seems that the prostate gland of donkey is more active during spring season compared to the other seasons of the year; this activity could be strongly related to breeding season (Abou-Elhamd et al., 2013).

Testis and epididymis

Donkeys have larger, more globular testicles compared to stallions with smaller, laterally constricted, ovoid-shaped testes (Figure 2). It measure in average 6 to 7 cm in wide, 9 to 10.5 cm in length and 3.7 to 5.2 cm in height (Table 1). Testicular size increases with age, nevertheless degeneration can be noted from the age of 15 years (Chabchoub et al., 2007). Donkeys' testicle is almost horizontal, with its long axis lightly oblique in the ventro-caudal direction without difference between the right and left gonad (Barone, 1990). The testis of the donkey was completely descended into the scrotum around birth. It covered by a thick tunica albuginea consisting of outer and inner fibrous layers in addition to middle vascular one (Banks,1993). Discrete bundles of smooth muscle cells were demonstrated in the outer fibrous layer of the tunica albuginea. These cells give the tunica albuginea a contractile function, which may aid in sperm transport (Abd-Elhafeez et al., 2017).

The epididymis is 12 to 13 cm long in the donkey and the duct can reaches 70 to 85 cm. It weighs an average of 17 to 25 g and its head receives from 12 to 23 efferent cones (Barone, 1990; Aissanou and Ayad, 2020). The corpus is relatively thick. The cauda epididymis is bulging, medially deviated and more prominent in donkeys with an average diameter 2.45±0.08 cm (Aissanou and Ayad, 2020). The testis-specific ligament and the epididymal tail ligament are thick and distinct (Chabchoub et al., 2007). The histomorphological study of the testicular compartments shown that the seminiferous tubule diameter were 222±6µm (Neves et al., 2002); 221.51±2.32 µm (Aissanou and Ayad, 2022) and 205.6±6.65 (Moustafa et al., 2015). Similarly, germ cell epithelial height values in mature individuals were recorded; 68.71±1.1 µm in the Algerian donkey (Aissanou and Ayad, 2022); 72.7±1.98 µm in Egypt (Moustafa et al., 2015) and 70.0±5.3 µm in the wild donkey (*Equus asinus africanus*) (Nipken and Wrobel, 1997)

Sexual behavior

The pre-copulatory behavior manifested by some signs such as naso-nasal contact; flehmen; nibbling and/or sniffing of the head, neck, back of the knee, body, flank, perineum and tail; and olfactory examination of urine or excreta without an erection or immediate copulation (Henry et al., 1998; McDonnell, 1998). It is reported that the donkey sexual behavior is influenced by seasonality and photoperiod.

The role of vocalization appears to be an important factor in successful communication between the two partners in mating, where 78 % of female vocalizations under pasture breeding management were in estrus (Henry et al., 1991). The male donkey is territorial compared to horses which keeps a group of mares and it mate with jennies which approach him or cross his territory (Figure 3). Henry et al. (1991) reported that the first mating of males was recorded during the day on pasture after 39.9±30.4 and 25.9±17.8 min of intermittent teasing periods. While the inter-copulatory interval was 88.4±71.5 and 93.3±54.5 min, when erection is achieved, the jacks return to the jennies to be mate and proceeds straight to the mounts once the males smelled the perinea area. For the jacks, the time from approach in erection to ejaculation was ranged 32±20 and 51±50 s, on the other hand, the time of mount to ejaculation was included 19±5.5 and 19±5.0 s. The ejaculation usually takes place after 4–8 pelvic thrusts lasting 19–30s (Henry et al., 1991; Gastal et al., 1996; Quar-tuccio et al., 2011). It is also noted that from the first contact with the female (Appetitive sexual behavior) to the ejaculation (consummatory sexual behaviour) range from 6 to 32min (Clayton et al., 1981; Gastal et al., 1996; Henry et al., 1991). Other observations were recorded that jacks would relax on the jenny's back for five to ten seconds following ejaculation before assuming the quadrupedal station, with pelvic flares ranging from 5 to 6 before ejaculate (Henry et al., 1998). In the post-copulatory phase which takes about 15–30min, the jack shows a total disinterest for the female, stays in the resting area, and eventually refuses the female approach (Henry et al., 1991; Quartuccio et al., 2011). The variation of erection latency were observed by Costa (1991) and Veronesi et al. (2008), (1 to 45 min and 14 to 39.3 min, respectively) that may well in part be due to the particularly sensitive nature of jacks to environmental disturbances such as noise, weather, and general management (Henry et al., 1991).

Seasonality

There is general factors can influenced the seasonal reproduction of animals, such as related to genotype and individual; and other are related to the animal's environment (Bronson, 1989). It is known that mammals generally use the duration of daylight to regulate not only their reproduction but also many other seasonal processes such as hibernation, growth, moulting, etc. In the literature, the scientific opinion are controversy on the seasonality character in the asine species, it likely could be influenced by photoperiod and other factors such as breed and environmental conditions (Tibary et al., 2006). The investigations undertaken to study the influence of the season on testicular biometry, histology, seminal and hormonal parameters are very limited in male donkey's populations (Aissanou and Ayad 2020; 2022).

Photoperiod is the main environmental factor that influences the seasonal patterns. Long or short days inhibit or stimulate sexual activity in animals. The light detected by the retina is translated into a neuroendocrine message by the epiphysis through the secretion of melatonin which influences seasonal reproductive activity (Reiteret al., 2018). Low or high concentrations of melatonin during the daylight and nighttime respectively, are a molecular indicator of photoperiod (Carcangiu, 2013).

Also, the seasonal changes in reproduction including especially hormonal activity, gametogenesis, and testicular size may show the fluctuations depending on the period of year. It is well known FSH acts specifically on Sertoli cells, playing an important role in the maintenance of qualitatively and quantitatively normal spermatogenesis. LH acts on Leydig cells to stimulate the testosterone secretion (Dutta et al., 2019). Moreover, the testosterone secretion, which is the major testicular androgen, is a necessary prerequisite for the maintenance of established spermatogenesis in the adult testes (Roberts and Chauvin, 2019).

The gel free volume in autumn was more ($P<0.05$) than that of summer and winter. The mass motility score in the autumn was more ($P<0.01$) than other seasons. The pH of semen in winter and summer differed ($P<0.01$) from that of autumn. The sperm concentration during summer was more ($P<0.05$) than that of autumn and winter. The live sperm percentage did not differ significantly due to season. (Roy et al., 2003).

Also, other observations have been reported on the biochemical characteristics of semen, which the means concentrations of glucose, cholesterol, acid phosphatase and aspartate aminotransferase activities in the seminal plasma were significantly higher in autumn and winter compared to summer (Roy et al., 2004).

In Brazilian donkey, the seasonal effect was observed only on seminal pH among all the sperm parameters (Gastal et al., 1997). Whereas Contri et al. (2010) noted a significant difference in the volume and viability of fresh Martina Franca donkey sperm characteristics, which values were higher during the short day length season, *i.e.* in November and December. On the other hand, no significant differences in testicular morphometric traits neither in seminal parameters were observed in Italian Martina Franca donkey during the year (Carllucio et al., 2013). It was also demonstrated that the seasonal change of plasma testosterone level in breeding season (Mar-Sep, 2.18±0.27ng/ml) were significantly higher than in non-breeding season (Oct-Feb, 1.50±0.18ng/ml) in Chinese donkeys and correlated with photoperiod and temperature (Jiaha, 1983). In addition, the seasonal variation of 17β-Estradiol differed by season since the mean level in (Apr-Sept) was higher than in (Oct-March) (Jiaha, 1983).

Schuler et al. (2019) evaluated the testicular endocrine function of male donkeys (*Equus asinus*) in under field conditions in Germany. Highly significant influence of season on the evolution of steroids concentrations namely estrone, estrone sulfate and testosterone through the year was recorded (Figure 4). The concentration values were low from November to January and high in April, May and June. Thus, the results showed that breed also had an effect on the expression of seasonality between dwarf and standard donkeys. In addition, seasonal interactions and geographical location seem to have a considerable influence on reproductive seasonality in donkey's populations around the world (Tibary et al., 2006).

Recently, it has been reported in testis histomorphometrical investigation that sexual activity usually occurs in winter and autumn in local donkeys from northern Algeria (Aissanou and ayad, 2022). Likewise, the highest gonado-somatic index and scrotal circumference values were recorded in the autumn and winter season. As well as the values of the testicular and epididymal biometrics were significantly higher in the short day period than long day period (Aissanou and ayad, 2020).

Sperm quality and cryopreservation

Puberty is the first release of fertile sperm. This is well correlated with the development of the gonads, which the donkeys are under breeding conditions and environmental factors. It has been demonstrates that puberty, by presence of spermatozoa in epididymis, is observed more early in the low density in donkeys having a high density of individuals than the other having a low density of individuals (Choquenot, 1991). The success of the cryopreservation process is conditioned by the survival of plasma membrane of the sperm, the acrosome, and the mitochondria (Sieme et al., 2015). Indeed, the optimal conditions of the cryoconservation is aimed to provide the physico-chemical properties during freezing, as well as the optimal cooling rates, maintaining temperatures, and warming rates for the sperm (Parks and Graham, 1992).

Many studies have aimed to describe the reproductive capacities of the different breeds in different localities (Table1). Carluccio et al. (2013a) were reported that the percentage of viable spermatozoa in the donkey's ejaculates is much higher compared with values reported for horse, (80% vs. 55 to 65 %). Other published results suggest that vitamin C and E could increase the quality of frozen sperm acting on sperm viability, acrosome cell membrane integrity, and mitochondrial activity. Also, the fusion of the both vitamins provides a better effect than the single addition (Yu et al., 2019). According to Álvarez et al. (2019), the use of the extender containing lactose-jenny colostrum can be successfully used for donkey semen cryopreservation (Table3) and could effectively improve donkey sperm qualities after freezing-thawing.

Rota et al. (2012) reported that the use of either glycerol or ethylene glycol as a cryoprotector after thawing and dilution with seminal plasma appears to improve fertility and improve pregnancy rates of inseminated donkey jennies. While, Oliviera et al. (2014) demonstrated that the use of cholesterol-loaded methyl-b cyclodextrin on donkey semen before cryopreservation increased the viability of thawed spermatozoa (Table3).

The vitrification technical by direct exposure of sperm to liquid nitrogen is a method of the biotechnology related to reproduction and seems increasing in popularity as an alternative to conventional freezing. Although, Hidalgo et al. (2020) showed that donkey sperm could not be vitrified using only glycerol as permeable cryoprotectant agents. Vitrification using non-permeable cryoprotectant agents (sucrose 0.1 M and BSA 5%) enhanced sperm motility and viability after warming.

In other study, donkey semen diluted in skimmed milk glucose showed superior sperm motility than lactose - egg yolk. The pregnancy rate for mares inseminated with semen diluted in skimmed milk glucose was superior than that obtained using lactose - egg yolk (56.52% vs. 4.76%, respectively) (Carvalho, et al., 2017**)**. Moreover, total cholesterol (mg/dl), total protein (g/dl) and triglycerides (mg/dl) were significantly higher in donkey's seminal plasma (74.65±9.70, 3.63±0.50 and 61.72±7.3, respectively) than in horse (64.72±10.23, 1.25±0.19 and 39.57±8.35, respectively) (Pal et al., 2009). These results could be explained by the fact that donkeys tend to be more fertile than horses, presenting a conception rate of 78% compared to mares average 65% (Hagstrom, 2004).

Conclusion

Equine professionals recognize that donkeys are an abandoned animal source, and their value is often under-estimated in the equine world. Although the present min-review has focused on the function of reproduction in male donkeys, it would be also need to mention other important physiologic aspects in order to ensure the genetic diversity. Reproductive physiology is a key essential to undertake the research in safeguarding the genetic inheritance of endangered animal species. Therefore, it very important to review the some aspects of the reproductive into understood the asine breeding management. In order to present the aspects related to the reproduction in donkeys males, more contemporary search results have been considered. The donkey belongs to family of equidaes, and exhibits some similar features of reproduction with horse.It appeared that the seasonality is well marked in donkeys, with other factors besides photoperiod and ambient temperature influencing the asine reproduction. Previous studies revealed that there is variability in sperm quality and production in donkeys around the world.

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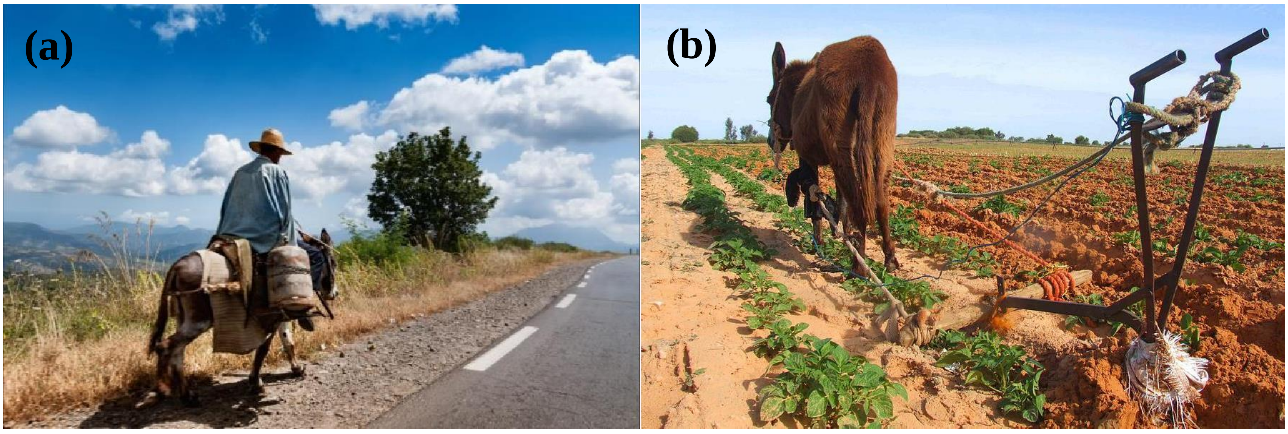

Figure 1. Use of the common donkey (*Equus asinus*) in transport (a) and agriculture (b) before the advent of motorization

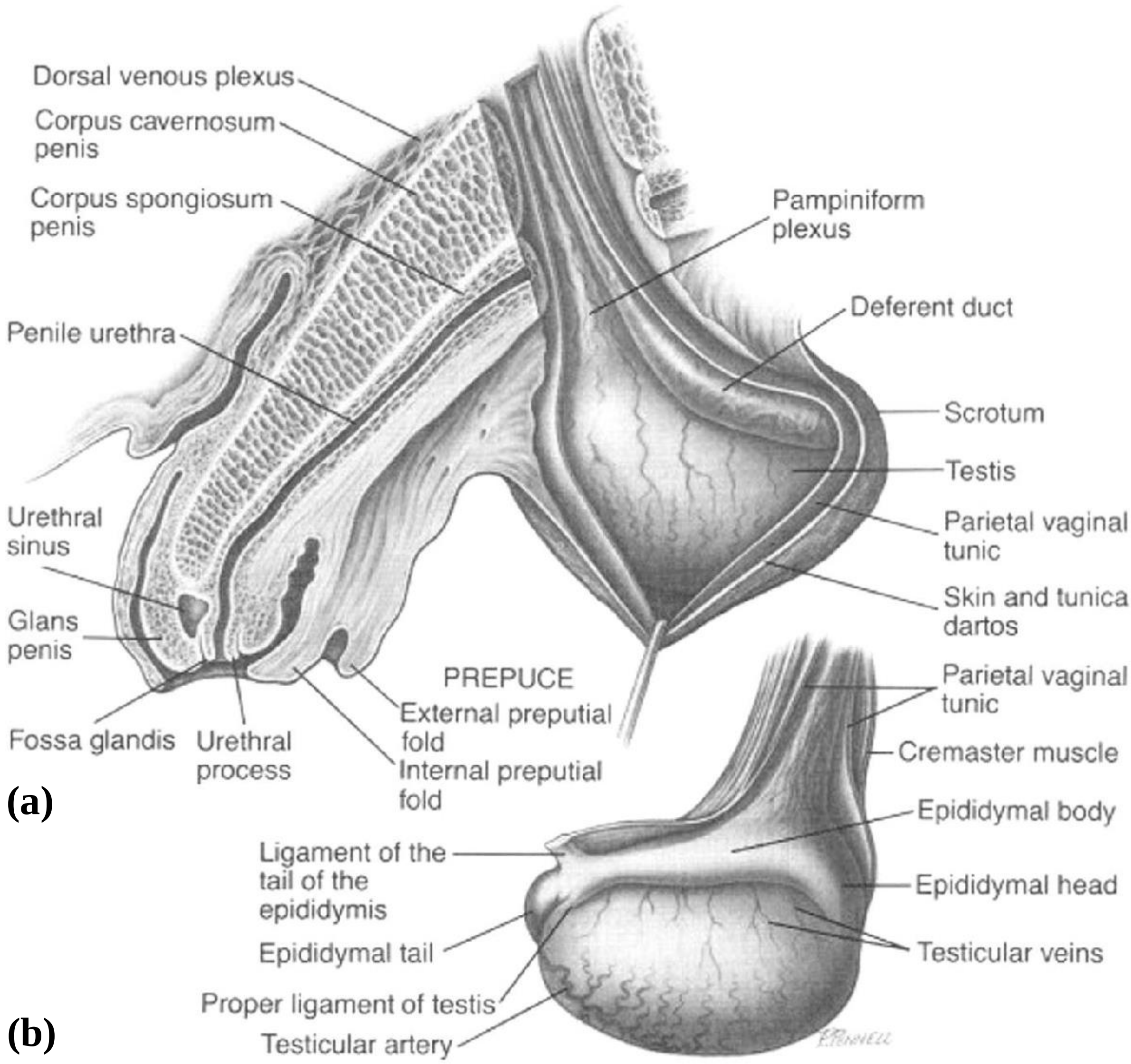

Figure 2. Chart illustrating a longitudinal section of the distal penis (a) and lateral aspect of the testis (b) in the common donkey (*Equus asinus*) (Nickel et al., 1979).

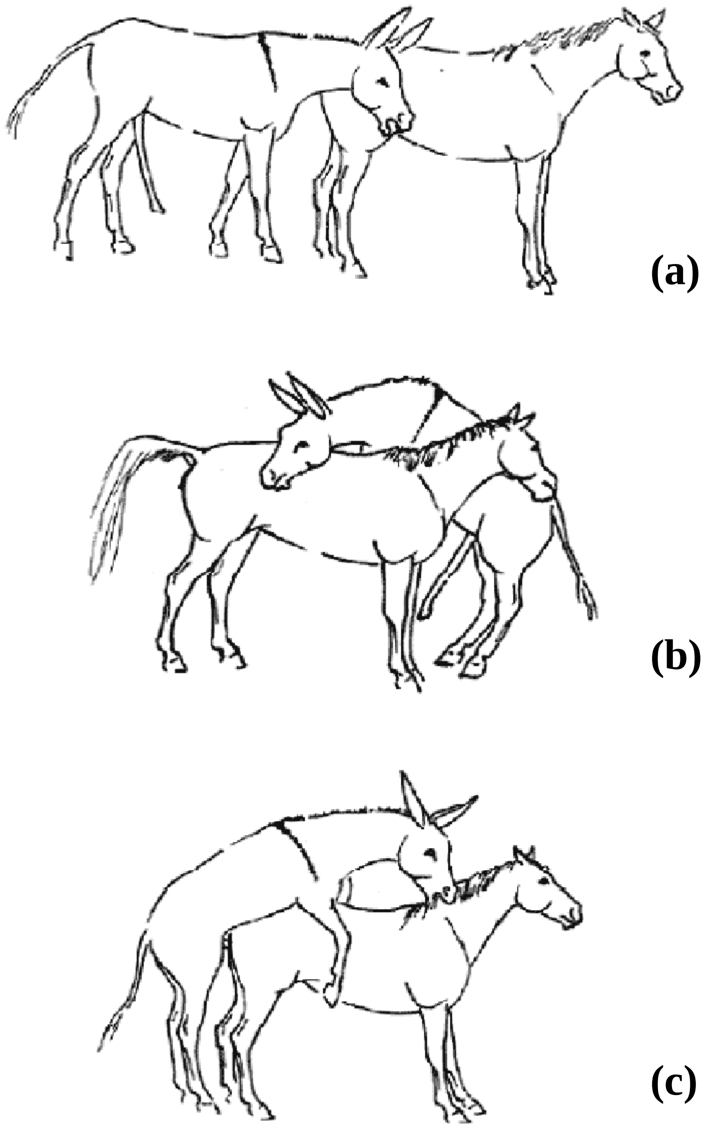

Figure 3. Donkey jack sexual behavior in presence of the female in deep estrus. (a) The donkey, with full erection, approaches of the female that is presenting typical passive behavior (top); (b) the donkey male may mount laterally the female; (c) successful copulation.

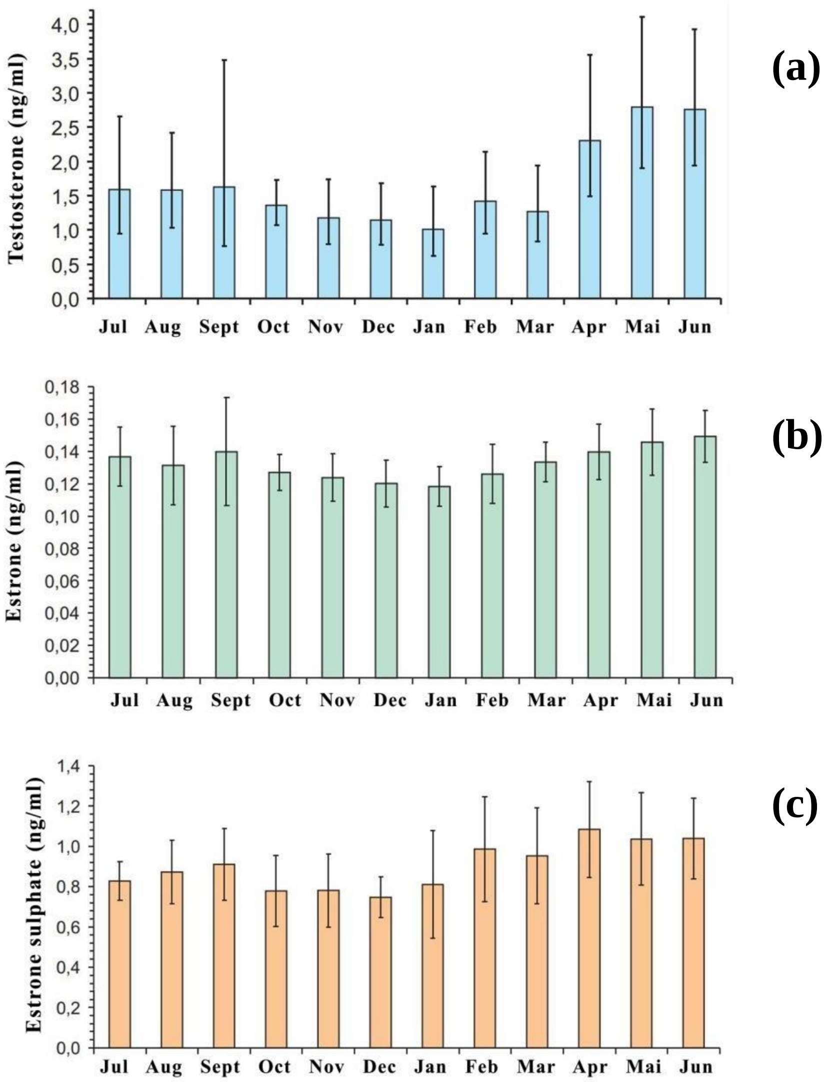

Figure 4. Testosterone (a), estrone (b) and estrone sulfate (c) secretory profile (mean ± SE) in donkeys (*Equus asinus*) in blood serum during different months of the year (Schuler et al., 2019).

Table1. Mean values (± SD) of the donkey (*Equus asinus*) breeds testis biometry.

Measurment	Algerian donkey[1]	Ethiopian donkey[2]	Martina Franca donkeys[3]	Miranda donkey[4]
Testicular length (cm)	6.8±0.2	13.4±3.78	9.6	8.04±1.2
Testicular Width (cm)	4.92±0.13	6.96 ± 1.65	6.8	6.06±1.19
Testicular Height (cm)	3.79±0.12	ND	5.2	4.63±0.86
Testicular Weight (g)	80.91±5.8	277±32.7	ND	ND
Testicular Volume (cm^3)	73.42±5.53	ND	ND	126.28±56.52

[1]Aissanou and Ayad, 2020;[2]Lemma and Deressa, 2009; [3]Carluccio et al., 2004; [4]Martins-Bessa et al., 2021.

ND: not determined

Table 2.Mean (± SD) of donkey (*Equus asinus*) breeds seminal parameters.

Parameters	**Pêga donkey[1]**	**Catalonian donkey[2]**	**Andalusian donkey[3]**	**Martina Franca donkey[4]**
Gel free semen volume (mL)	47.2±28.6	56.61±23.18	80.6±11.1	90±43.4
Gel volume (mL)	71.7±54.8	ND	ND	107.2±41.6
Total motility (%)	84.2±6	68.40±16.59	90.2±2.7	81.9±3.7
Progressive motility (%)	74.4±7	ND	70.1±4.1	76.6±4.8
Sperm concentration (10^6 per mL)	253±91.2	280.88±228.94	259.4±37.6	350.4±139.7
Total abnormalities	7.9±3.0	18.99±8.62	12.2±2.1	ND
pH	ND	7.77±0.35	7.2±00	7.6±0.2
VCL (m/s)	ND	80.20±51.72	106.6±0.2	115.9±14.2
VSL (m/s	ND	49.80±43.18	78.5±0.3	195.9±27.7
VAP (m/s)	ND	59.39±43.04	94.4±0.2	136.8±12.5
LIN (%)	ND	60.50±28.79	73.3±0.2	60.8±5.4
STR (%)	ND	79.21±24.50	82.2±0.2	89±40

[1]Canisso et al., 2010; [2]Miro et al., 2005; [3]Dorado et al., 2013; [4]: Contri et al., 2010.

ND: not determined

Table 3. Effect of different extenders in post thawing donkey semen cryopreservation.

Authors	Extenders	Breed	Concentrations	Hours Post-thaw	Progressive motility (%)	Total motility (%)	VAP (µm/s)
Álvarez et al. (2019)	Lactose–jenny colostrum extender	Zaragoza, Spain	50% of lactose, 20% of jenny colostrum, 25% of Glucose–EDTA and 5% DMF.	0	ND	58.3	50
Oliviera et al. (2014)	CLC	Pega	3mg	0	49.2 ± 2.7	81.1 ± 2.9	89.6 ± 2.7
Rota et al. (2012)	Eg-INRA96	Amiata	2.8 ml	0	33	53	107
				1	31	43	100
				2	21	31	85
Rota et al. (2012)	Gly-INRA96	Amiata	4.4 ml	0	32	49.5	110
				1	28	42	95
				2	21	32	83

ND: not determined

CLC: Cholesterol-loaded cyclodextrin

Eg: Ethylene glycol

Gly: Either glycerol

Partie expérimentale

Etude #01 : Influence of age, body weight and season on testicular and epididymis biometrics in donkeys (*Equus asinus*)

Résumé

L'objectif de cette présente investigation a été entrepris pour étudier les caractéristiques biométriques testiculaires et épididymaires des ânes algériens tout au long de l'année selon l'âge, le poids corporel et les changements saisonniers. L'étude a été menée de février 2019 à janvier 2020. Au total, 24 ânes sexuellement matures (*Equus asinus*) ont été sélectionnés au hasard. Les testicules et l'épididyme ont été prélevés après l'abattage des ânes et séparés des tissus conjonctifs et adhérents. L'épididyme a été soigneusement retiré à la jonction testiculaire. Au total, 10 mesures biométriques ont été sélectionnées et réalisées.

Nos résultats ont révélé qu'il existe des différences significatives (P<0,05) entre les groupes dans la plupart des valeurs biométriques. Tous les paramètres biométriques variaient tout au long de l'année et étaient affectés par la saison, différents groupes d'âge et saisons. En revanche, aucune différence significative n'a été observée entre les catégories de poids corporel des ânes. L'analyse des coefficients de corrélation entre les valeurs biométriques montre des corrélations positives élevées, comprises entre 0,98 et 0,72 (P<0,001). Il y avait une forte corrélation positive entre l'âge et tous les paramètres, allant de 0,85 à 0,61 (P<0,001). Cependant, il y avait de faibles corrélations négatives entre la saison et la biométrie testiculaire et épididymaire.

Nos résultats ont montré que des différences essentielles ont été notées entre certains paramètres biométriques et l'âge, la saison et le poids corporel des ânes. En outre, les coefficients de corrélation ont été pris en charge entre les mesures biométriques et ces facteurs. Cependant, d'autres approches sont nécessaires à entreprendre telles que l'histologie des organes reproducteurs et le dosage hormonale, pour une compréhension plus approfondie de la physiologie de la reproduction chez l'âne.

Etude #2 : Histomorphometric changes of testicular tissues by season and age of Algerian local donkeys (*Equus asinus*)

Résumé

La présente étude a été entreprise pour mettre en évidence les changements histomorphologiques des tissus testiculaires selon la saison et l'âge chez les ânes (*Equus asinus*) dans les conditions d'élevage du nord de l'Algérie. L'expérience a été menée de février 2019 à janvier 2020. Un total de 21 ânes sexuellement matures ont été sélectionnés au hasard. Les testicules ont été immédiatement prélevés après l'abattage pour l'observation histologique.

L'analyse des coefficients de corrélation montre des corrélations négatives entre DL et d'autres paramètres tels que DST, GCEH, TTA et ITSA, comprises entre -0,24 et -0,79. De plus, il y avait une forte corrélation négative entre la saison et les paramètres histomorphométriques, sauf que la DL s'est révélée positive (r = 0,65). En revanche, il existait des corrélations positives entre l'âge et les paramètres histomorphométriques testiculaires. Les résultats ont indiqué que les valeurs DST, GCEH, TTA et ITSA étaient significativement plus élevées en hiver et en automne qu'au printemps et en été. Il convient également de noter que les valeurs de DST, GCEH, TTA et ITSA étaient significativement plus élevées chez les ânes adultes et âgés que chez les jeunes ânes.

Notre analyse comparative des paramètres histologiques, suggère que l'activité sexuelle se produit généralement pendant l'hiver et l'automne chez les ânes de race local d'Algérie. De plus, nos résultats d'histomorphométrie des tissus testiculaires sont corrélés avec l'âge des ânes.

Histomorphometric changes of testicular tissues by season and age of Algerian local donkeys (*Equus asinus*)

Sofiane Aissanou, Abdelhanine Ayad*

Department of Physical Biology and Chemistry, Faculty of Nature and Life Sciences, Université de Bejaia, 06000, Bejaia, Algeria.

Running head: Histomorphometric of testicular in donkeys

*Corresponding author: Prof. Dr. Abdelhanine Ayad, Email: abdelhanine.ayad@univ-bejaia.dz

S. Aissanou, Orcid ID: https://orcid.org/0000-0002-2414-4485

A. Ayad, Orcid ID: https://orcid.org/0000-0002-9325-7889

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Etude #3 : Seasonal changes in plasma testosterone and biochemical parameters of male donkey (*Equus asinus*) in northern Algeria

Résumé

Cette présente étude visait à déterminer les variations de concentrations de la testostérone plasmatique chez les ânes du nord algérien, et les coefficients de corrélation entre les concentrations de T et les paramètres biochimiques ont été estimés. Des échantillons de sang ont été prélevés le matin à partir la veine jugulaire dans un tube contenant de l'EDTA. Un total de 24 ânes matures en bonne santé a été sélectionné au hasard sous condition de période de stabulation libre. Les concentrations de T chez les ânes ont été mesurées par un kit de dosage immuno-enzymatique.

Les concentrations d'AST, d'ALT et d'ALP ont été dosées conformément aux recommandations du kit. En ce qui concerne la concentration en Ch et Tg, le dosage a été effectué à l'aide d'un analyseur biochimique automatique. Les résultats ont indiqué que les valeurs de concentration de T étaient significativement ($P<0,05$) plus élevées en hiver et en automne (2,468±0,66 ng/ml et 2,785±0,49 ng/ml, respectivement) qu'au printemps et en été (0,95±0,343 ng/ml et 0,745 ± 0,236 ng/ml, respectivement).

De même, les valeurs de Ch et Tg étaient significativement ($P<0,05$) plus élevées en hiver et en automne qu'au printemps et en été. En outre, il est également à noter que les valeurs d'ALT étaient significativement ($P <0,05$) plus élevées en hiver et en automne qu'au printemps et en été. Alors que les valeurs d'AST étaient significativement ($P <0,05$) plus faibles en été qu'en hiver, en automne et au printemps. Quant à l'ALP, les valeurs moyennes étaient pratiquement similaires en toutes saisons.

Notre découverte suggère que l'activité sexuelle se produit probablement pendant l'hiver et l'automne chez les ânes de race locale du nord de l'Algérie. De plus, nos résultats des niveaux de Ch et de Tg sont corrélés avec la saison, ce qui peut être une bonne indication des performances de reproduction chez les ânes.

Seasonal changes in plasma testosterone and biochemical parameters of male donkey (*Equus asinus*) in northern Algeria

Sofiane Aissanou[1], Mokhtar Benhanifia[2], Omar Besseboua[3], Abdelhanine Ayad[1,*]

[1]Department of Physical Biology and Chemistry, Faculty of Nature and Life Sciences, University of Bejaia, 06000, Bejaia, Algeria.

[2]Department of Agricultural Sciences, Faculty of Life and Nature Sciences, University Mustapha Stambouli, 29000, Mascara, Algeria.

[3]Department of Agronomic and Biotechnological Sciences, Faculty of Nature and Life Sciences, University H. Benbouali, 02000, Chlef, Algeria.

Running head: Seasonal changes in T concentrations in Algerian local donkeys

*Corresponding author: Prof. Dr. Abdelhanine Ayad, Email: abdelhanine.ayad@univ-bejaia.dz

S. Aissanou, Orcid ID: https://orcid.org/0000-0002-2414-4485

A. Ayad, Orcid ID: https://orcid.org/0000-0002-9325-7889

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Seasonal changes in plasma testosterone and biochemical parameters of male donkey (*Equus asinus*) in northern Algeria

Abstract

This present study was aimed to determine the changes of plasma testosterone in North Algerian donkeys, the correlation coefficients between T concentrations and biochemical parameters were estimated. Blood samples were withdrawn in the morning from the jugular vein into a tube containing EDTA. A total of 24 mature healthy donkeys were selected randomly under condition of free stabling period. T concentrations in male donkeys were measured by enzyme-linked immunosorbent assay kit. AST, ALT and ALP concentration were measured according to the manufacturer recommendation. As regards the concentration of Ch and Tg, the assay was carried out using the automatic biochemical analyzer. The results indicated that T concentration values were significantly ($P<0.05$) higher in winter and autumn (2.468±0.66 ng/ml and 2.785±0.49 ng/ml, respectively) seasons than in spring and summer (0.95±0.343 ng/ml and 0.745±0.236 ng/ml, respectively) seasons. Likewise, Ch and Tg values were significantly ($P<0.05$) higher in winter and autumn seasons than in spring and summer seasons. Also, it is also noteworthy that ALT values were significantly ($P<0.05$) higher in winter and autumn seasons than in spring and summer seasons. Whereas, AST values were significantly ($P<0.05$) lower in summer than in winter, autumn and spring seasons. As regards ALP, values mean were practically similar in all seasons. Our finding suggests that the sexual activity probably occurs during winter and autumn in local donkeys of Northern Algeria. In addition, our results of Ch and Tg levels are correlated with season, which may be a good indication of the reproductive performance in donkeys.

Keywords: Testosterone; biochemical parameters; season; donkeys, Algeria.

Running head: Seasonal changes in T concentrations in Algerian local donkeys.

Introduction

The domestic donkey (*Equus asinus*) represents a unique equine species that descended from wild donkeys (*Equus africanus*), and that evolved in inhospitable, mountainous and arid desert environments (Epstein 1984). The common donkey is mainly kept as a domestic companion animal in Europe and North America, but is commonly used as a working animal in developing countries. The number of donkeys worldwide considerably decreased with the advent of motorization in transport and work, which constitutes a risk of extinction of the species in the worldwide (Vlaeva et al.2017; Kugler et al.2008). It appears a good knowledge of the reproductive function seems to be a very essential tool to preserve and reintroduce the donkey populations.

The mechanisms regulating reproductive function are based on the permanent relationship between the central nervous system and the gonads, which is provided by the steroidal and gonadotrophic hormones. However, this hormonal regulation is influenced by different environmental factors such as age; season (Vasantha et al.2016). Seasonal variations of equides are due to the change of day duration throughout the year. The seasonally reproductive animal species, notably donkeys, express seasonal variations in their sexual activity. It is known in the literature that the testosterone is a steroid hormone that is the main key to the spermatogenesis and control males' sexual behavior; and its dosage is a best tool to determination of the reproductive season. Despite the scientific controversy on seasonality in donkeys, it likely could be influenced by photoperiod and other factors such as nutrition and temperature.

In other hand, biochemical parameters are commonly used for the assessing the physiological or pathological condition of animals, however, the values of these parameters vary between populations, sex, age, nutrition and season (Longodor et al.2020; Tesfaye et al.2014; Girardi et al.2014). It has been reported that transaminase enzyme activity (AST and ALT) is a good indicator of semen quality (Corteel et al.1980). Moreover, a relationship was found between ALT and ALP enzymatic activity and reproductive function (Longodor et al.2020; Hussein et al.2017). Numerous studies that have shown that testosterone has a strong influence on hepatocytes and induces more variation in enzyme functions (Azani et al.2018; Charni-Natan et al.2019). In addition, it was reported that male androgens increase blood cholesterol (Guyton 1981).

To our knowledge, no study has been done to describe the seasonal testosterone (T) concentrations of the domestic donkey *(Equus asinus*) in north Algeria. We hypothesized that total cholesterol (Ch), triglyceride (Tg), aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP) could influence T concentrations in domestic donkeys. Therefore, this present study was aimed to determine the changes of plasma testosterone inNorth Algerian donkeys,the correlation coefficients between T concentrations and biochemical parameters were estimated.

Materials and methods

This research was approved by the Scientific Council of the Faculty of Nature and Life Sciences (Report of Faculty Scientific Council #05 dated October 30, 2018), University of Bejaia, Algeria). Concerning the ethical aspects, the experimental procedure was performed according to good veterinary practice under farm conditions.

Study area and animals

The study was carried out from January 2020 to February 2021 in Northern Algeria, Bejaia (36° 43' N, 5° 04' E) and Jijel (36° 42' N, 4° 2' E) province were chosen randomly. The study region has four distinct seasons: winter (January to March), spring (April to June), summer (July to September) and autumn (October to December) with a seasonal mean of day length of 610.6; 785.88; 851.03 and 677.16 minutes/day, respectively. The mean maximum summer temperature reaches 36.9 °C (September) and the mean minimum winter temperature falls to 9.8 °C (February).

Animals and samples

Blood samples were withdrawn in the morning (8:00 am to 10:00 am) from the jugular vein into a tube containing EDTA (Sarstedt®, Numbrecht, Germany). The samples were immediately centrifuged (15 min at 1500 ×g), and stored at -20 °C until assayed. A total of 24 mature healthy donkeys were selected randomly under condition of a free stabling period. The age of the animals ranged between 3 and 20 years. Male donkeys had a body corporal score between 2.5 and 3.5. Animals were checked by a veterinarian and presented no signs of disease clinical.

Testosterone and biochemical parameters assay

T concentrations in male donkeys were measured by enzyme-linked immunosorbent assay kit (Human Diagnostics Worldwide: ELISA Testosterone, direct REF: 55010, Germany) according to the kit instruction. Each sample was analyzed in duplicate. The detecting antibody was rabbit anti-T IgG as biotin-conjugate. The enzyme substrate was avidin-horseradish peroxidase (HRP). Intra- and inter-assay coefficients of variation were 7.5% and 11%, respectively.

AST, ALT and ALP concentration (nmol/ml) were measured according to the manufacturer recommendation (Spinreact, S.A./S.A.U. Sant Esteve de Bas, Spain). As regards the concentration of Ch and Tg, the assay was carried out using automatic biochemical analyzer (COBAS INTEGRA 400 plus Analyzer REF: 5179282).

Statistical analyses

Statistical analyses were carried out in Statview, version 4.55 (Abacus concepts Inc., Berkeley, CA, USA). The concentration values were expressed as mean ± SE, and $P < 0.05$ was considered significant. The one-way variance analysis (ANOVA) was used to evaluate the obtained data. Statistical analysis was performed using *t*-test to compare different seasons.

Results

The length day and mean temperature during the period of study are presented in Figure 1. The overall mean (±SE) T concentration and biochemical parameters of donkeys were shown in Table 1. Mean value of T concentration is 1.47 ± 0.28 ng/ml, ranged between 0.21 and 4.77 ng/ml. Mean values of Ch and Tg are 0.87 ± 0.11 and 0.81 ± 0.1 mmol/l, respectively. Mean values of ALT, AST and ALP are 21.4 ± 1.66, 199.63 ± 10.08 and 123.6 ± 4.8 U/l, respectively.

Figure 1. Mean (± SE) values of daylight and temperature of the study area, Northern Algeria

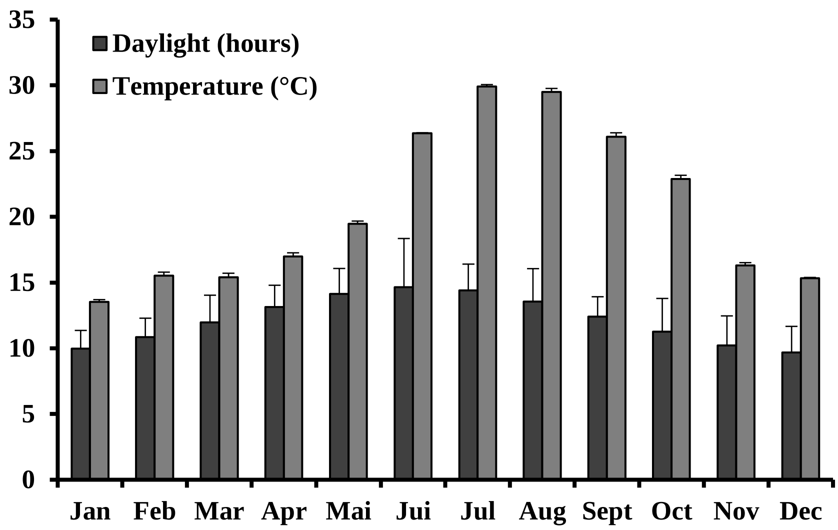


Table 1. The overall descriptive data (Mean ± SE) of plasma testosterone, total cholesterol, triglyceride, alanine aminotransferase, aspartate aminotransferase and alkaline phosphatase in Algerian local donkeys (*Equus asinus*).

Parameters	Mean	SE	Minimum	Maximum	CV
Testosterone (ng/ml)	1.74	0.28	0.21	4.77	0.8
Total cholesterol (mmol/l)	0.87	0.11	0.25	1.77	0.62
Triglyceride (mmol/l)	0.81	0.1	0.22	2.2	0.59
Alanineaminotransferase (U/l)	21.4	1.66	5.83	37.91	0.38
Aspartateaminotransferase(U/l)	188.63	10.08	109.66	268.91	0.26
Alkaline phosphatase (U/l)	123.6	4.8	91.3	184.8	0.2

The changes of T concentrations and biochemical parameters expressed by seasons in donkeys (*Equus asinus*) are shown in Figure 1 and 2, respectively. The results indicated that T concentration values were significantly ($P<0.05$) higher in winter and autumn (2.468±0.66 ng/ml and 2.785±.0.49 ng/ml, respectively) seasons than in spring and summer (0.95±0.343 ng/ml and 0.745±0.236 ng/ml, respectively) seasons. Likewise, Ch and Tg values were significantly ($P<0.05$) higher in winter and autumn seasons than in spring and summer seasons. Also, it is also noteworthy that ALT values were significantly ($P<0.05$) higher in winter and autumn seasons than in spring and summer seasons. Whereas, AST values were significantly ($P<0.05$) lower in summer than in winter, autumn and spring seasons. As regards ALP, values mean were practically similar in all seasons.

Figure 2. Seasons changes in testosterone concentrations (A), total cholesterol (B), Triglyceride (C) in Algerian local donkeys (*Equus asinus*). [a,b] Means (±SE) with the same superscripts in each groups of different seasons are significantly different ($p<0.05$).

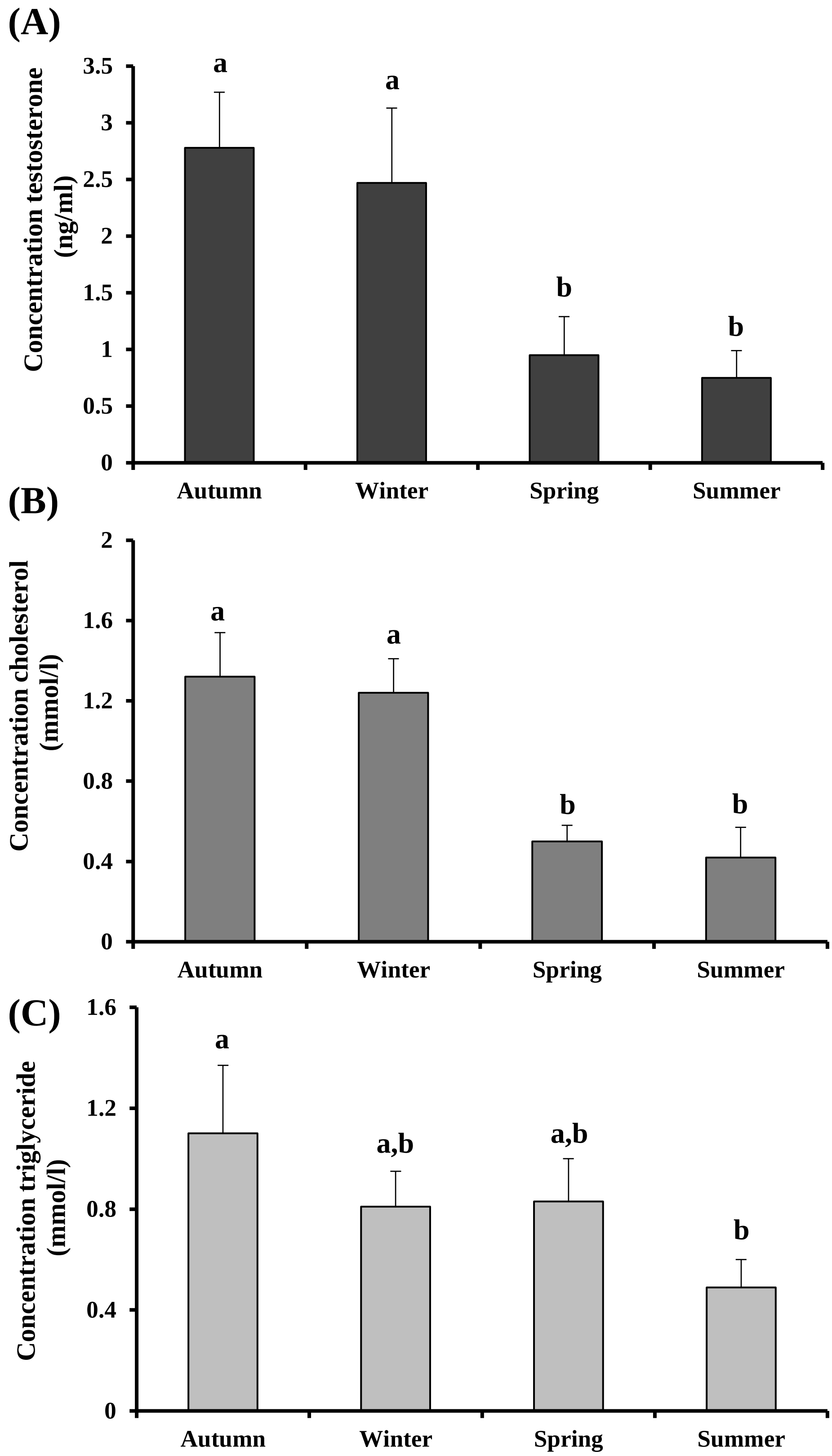
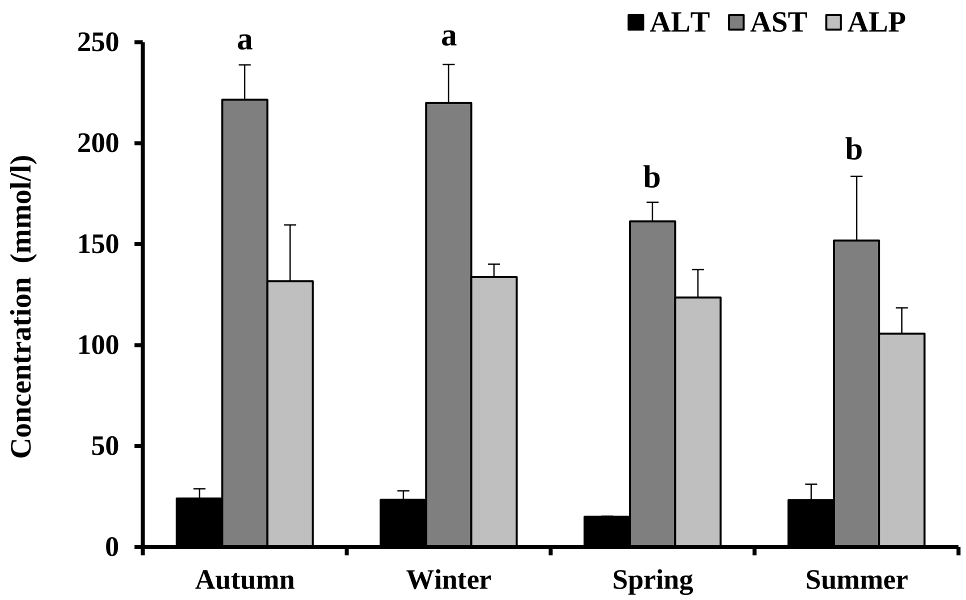


Figure 3. Season changes of transaminase activities (ALT, AST and ALP) in Algerian local donkeys (*Equus asinus*). [a,b]Means (±SE) with the same superscripts in each groups of different seasons are significantly different (p<0.05).



The correlation coefficients between T concentrations and different biochemical parameters are summarized in Table 2. The analysis of the correlation coefficients between T concentration and cholesterol (r=0.894), AST (r=0.537) and ALP (r=0.51) shows high positive correlations (P<0.01). Also, the highest correlation coefficient found between cholesterol and triglyceride (r=0.642), and AST (r=5.28) (P<0.01). In the other hand, there was a high negative correlation (P<0.001) between photoperiod and T concentrations, cholesterol and AST (-0.61, -0.74 and -0.64, respectively) (Table 3).

Table 2. Correlation coefficients (r) between, testosterone, total cholesterol, triglyceride, alanine aminotransferase, aspartate aminotransferase and alkaline phosphatase in donkeys (*Equus asinus*).

	T	**Ch**	**Tg**	**ALT**	**AST**	**ALP**
T	1					
Ch	0.894***	1				
Tg	0.446*	0.642***	1			
ALT	0.073	0.105s	-0.175	1		
AST	0.537**	0.528**	0.145	0.252s	1	
ALP	0.51**	0.19	0.3	-0.152	0.183	1

*P <0.05),**P <0.01,***P < 0.001

Table 3. Correlation coefficients (r) between photoperiod and testosterone, total cholesterol, triglyceride, alanine aminotransferase, aspartate aminotransferase and alkaline phosphatase in donkeys (*Equus asinus*).

Parameters	**Correlation coefficient**
Photoperiod and testosterone	-0.61*
Photoperiod and cholesterol	-0.74*
Photoperiod and triglyceride	-0.24
Photoperiod and ALT	-0.31
Photoperiod and AST	-0.64*
Photoperiod and ALP	-0.35

*P <0.001

Discussion

It is known that equines have been considered for a long time as seasonal breeders with sexual activity being related to long days. However some studies have demonstrated that donkey females (*Equus asinus*) can cycle throughout the year (Contri et al.2014). Also, the ovarian activity, pregnancy, and parturition appear to be much less seasonal in domestic donkeys than in wild donkeys (Ginther et al.1987). In another investigation, it has been reported that female donkeys can presented an oestral cycle year-round in the southern United States, while 64% of the females have a cycle in December in Wisconsin (Tibary 2004). In Morocco, field observations show that the birthing peak season is in March and a few sporadic births in February and from June to September (Tibary et al.2006). Further, the results obtained by Carluccio et al. (2013) showed that the season has no effect on testicular morphometry and semen characteristics. One study conducted by Canisso et al. (2019) reported an increase of percentage of sperm morphology and decreased of motility in the spring and summer in comparison with the autumn. Another study concluded that there were no apparent seasonal variations in semen quality (Gastal et al.1997; Canisso et al.2019). To our knowledge, the present study is the first to assess the influence of season on T concentration and biochemical parameters in Algeria local breed donkeys, and this could help to determine of the reproduction season in the northern of Algeria.

Testosterone is known as the male sexual hormone that controls the changes of epididymis and testis, as well as accessory glands. It is essential and responsible for increasing sexual activity and the testicular size expressed by the proliferation of Leydig cells and germ cells. The results obtained revealed that the T concentrations in the short days were significantly higher than in the long day in Algeria local donkey breeds. According to Aissanouand Ayad (2020; 2022), the donkey breeds living in northern Algeria express a seasonal influence on biometry and histophotometry of testis, *i.e.* during short days length; this is in full agreement with data registered under same latitudes. This could be explained by coincident with spermatogenesis and peak testosterone levels in winter and autumn seasons. In addition, the increased testosterone may be caused by increased androgen levels paralleling increased sexual activity in winter and autumn. Also, Schuler et al. (2019) recorded plasma T concentration values in donkeys were similar to the results of the present study. However, Jiaha(1983) revealed that the mean T concentrations in the breeding season (Mar-Sep) were higher than in the non-breeding season (Oct-Feb) in Guan-Zhong donkey. These differences among studies may be due to differences betweenbreeds, feeding level, season, photoperiod, and different latitude, as described in other specie animals (Aldori et al. 2017; Ismaeel 2018; Ait-Amran et al. 2013), it is likely to be due to heterogeneous management in various agro-climatic regions and feed practices (Gupta et al. 2016; Kabukçu et al. 2020).

Lipids have an important position in the organism function and serve as hormones or hormonal precursors such as steroidogenesis. (Welteand Gould, 2017;Maxfield andTabas, 2005). The results of this study revealed that the serum biochemical parameters of Algerian local donkeys recorded in this study are very similar to those published previously in wild donkeys (*Equus africanus asinus*) (da Silva et al. 2018) and German donkeys (*Equus asinus*) (Schuler et al. 2019). In our study, Ch and Tg levels were significantly higher ($P<0.001$) in winter and autumn, which corresponds to a period of short days. This result is in agreement with previous reports (Roy et al. 2004; Longodor et al. 2020), in which Ch and Tg levels were significantly higher in winter than in summer in Indian and Romanian donkeys. Also, Seri et al. (2010) has reported that the values of (ALT, AST, Ch and Tg) showed significant seasonal changes in Sudanese donkeys and the highest concentrations were detected during the rainy season. Moreover, HaskovićandSuljevi(2011) recorded similar results to the present study in Bosnian mountain horse, whose enzymatic activity of AST and ALT showed significant variation in seasonal changes between summer and autumn season, while ALP shows no significant seasonal changes. Conversely, our findings noticed that Tg, Chol and AST values were lower in Algerian local donkeys compared to England donkeys, mentioned in previously by Burden et al. (2016). Our results could be explains by the existence of a close relationship between AST, located in the spermatozoa mid-piece part, and sperm motility; therefore, membrane defect and loss of AST lead to blockade of ATP production (Pesch et al. 2006). In the literature, the enzyme system (AST, ALT, and ALP) plays an essential role in the sperm metabolic processes, providing energy for sperm survival, motility, and fertility in stallions (Kosiniak et al. 2000; Turner and McDonnell, 2003). Likewise, it has been reported that transaminase activities (AST and ALT) in semen are a good indicator of sperm quality as they measure the stability of the sperm membrane. (Sirat et al. 1996; Corteel et al. 1980).

A significant positive correlation was recorded between the T concentrations and biochemical parameters; this probably due to steroidogenesis increased and the sperm production. Cholesterol is one of the precursors for steroid biosynthesis and is used for the synthesis of testosterone (Amerkhanov et al. 2013). In addition, several enzymes have been identified in the accessory glands in seminal plasma of stallions. These correlations study of enzyme activities with testosterone might be provides information on semen quality and fertility of stallions (Kareskoski, 2011). It was reported that photoperiod is the most important environmental factor that affect reproduction function in donkeys (Cappai et al.2017; Deikman et al.2002), this because of its influence on circulating levels of gonadotropins and gonadal hormones. The data of this study showed a correlation of high negative correlation ($P<0.001$) photoperiod with T, Ch, and AST in Algerian donkeys. The findings of this study are also consistent with those published previously, which showed significant correlations between T values with photoperiodic data in the Guan Zhong donkey (Jiaha, 1983). Note that the photoperiod influences seasonal breeders through changes in melatonin secretion that affects the function of the hypothalamic– pituitary–testicular axis (Vividand Bentley, 2018).

Conclusion

The present study is the first to describe the seasonal variations of T concentrations and biochemical parameters in Algerian local donkeys. Our finding suggests that sexual activity probably occurs during winter and autumn in local donkeys of Northern Algeria. In addition, our results of Ch and Tg levels are correlated with the season, which may be a good indication of the reproductive performance in donkeys. However, other approaches such as sperm analysis are necessary to undertake to strengthen these results. Further the measuring of the melatonin level is needed to confirm the photoperiodic aspect in male donkey.

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Discussion générale

Etude #1. Influence de l'âge, du poids corporel et de la saison sur la biométrie des testicules et de l'épididyme chez l'âne (*Equus asinus*) :

Notre travail de recherche a d'abord porté sur la biométrie testiculaire et épididymaire chez l'âne commun en Algérie. La biométrie testiculaire est utilisée comme indicateur du statut reproducteur et sa caractérisation ont été bien documentés chez de nombreux mammifères (**Ibrahim et al., 2012 ; Murta et al., 2013 ; Peixoto et al., 2012**). L'état de la fonction de la reproduction semble être influencé par de nombreux facteurs tout au long de l'année chez les espèces animales à reproduction saisonnière, en particulier la durée du jour (**Machado et al., 2011**) et l'âge (**Da silva santos et al., 2013 ; Jain et al., 2008**). On sait que les méthodes quantitatives sont d'une grande valeur pour évaluer la structure et la fonction testiculaire dans diverses conditions physiologiques et pathologiques (**Ekhoye et al., 2013 ; Omar et al., 2013**). La connaissance de la fonction reproductrice mâle chez les ânes commence par la description biométrique testiculaire et épididymaire, car il s'agit d'un moyen très important pour comprendre la physiologie de reproduction de cette espèce et peut aider à prédire le potentiel de reproduction.

Les résultats de cette première partie ont révélé que le volume testiculaire des ânes était supérieur à celui de la race nord-est des ânes brésiliens âgés de 5 à 7 ans (**Rocha et al., 2018**). À l'inverse, plusieurs études ont rapporté des valeurs élevées de volume de testicules dans différentes races par rapport à nos résultats (**Canisso et al., 2009; Omar, 2013**). La taille globale des testicules des ânes dans l'étude actuelle est inférieure à celle des enquêtes antérieures du Brésil (**Gastal et al., 1997**) et de l'Italie (**Carluccio et al., 2004**). La valeur du GSI enregistrée chez les ânes de race locale algérienne est très supérieure à celle des ânes Pêga (**Neves et al., 2004**). Cependant, les valeurs de GSI étaient beaucoup plus élevées que celles rapportées chez les ruminants domestiques tels que le taureau (**Andreussi et al., 2014**), le bouc (**Leal et al., 2004**) et le bélier (**Al kawmani et al., 2014**). Par conséquent, selon nos résultats du GSI liés à l'âge, il convient de noter que les testicules se développent jusqu'à ce que l'âge des ânes soit atteint. Les différences entre la moyenne de nombreuses mesures biométriques peuvent s'expliquer par la différence de race, les emplacements géographiques, le niveau nutritionnel, les conditions agroclimatiques et la gestion d'élevage.

Les paramètres morphométriques corporels (WH, TC et BL) enregistrés dans cette étude étaient très similaires à ceux publiés récemment chez les ânes de race locale algérienne (**Ayad et al., 2019**). Les résultats de cette étude ont remarqué que les ânes sont de petite taille par rapport aux les autres mentionnés dans les articles précédents. Cela peut s'expliquer par la quantité et la qualité de la nourriture et l'intensité du travail.

Nos résultats ont révélé que les individus jeunes, adultes et âgés présentent une différence significative (P <0,05) des paramètres biométriques testiculaires et épididymaires et cela augmente concomitamment avec l'âge des ânes. De même, le même constat a été obtenu concernant le poids corporel où il existait une différence significative (P<0,05) entre les valeurs de certaines mesures biométriques selon les catégories de poids corporel des animaux. Il y avait une différence significative (P<0,05) entre les valeurs de GSI et SC selon l'âge des individus et leurs catégories de poids corporel. Ces résultats corroborent des résultats obtenus précédemment chez les ânes de race Pêga (**Canisso et al., 2009**) et d'autres espèces animales domestiques (**Ajao et al., 2014; Eljarah et al., 2017; Al Sadoon et al., 2019**). De même, **El wishy** (**1974**) a observé le même poids épididymaire chez les ânes avec un poids corporel de 256,7 kg.

De plus, **Alemayehu & Benti** (**2009**) ont enregistré une SC élevée chez les ânes éthiopiens allant de 15,2 à 26,7 cm. Ces différences peuvent être attribuées aux altérations BCS des ânes considérés dans cette étude. Cependant, il a été déterminé que le BCS et l'âge peuvent influencer la circonférence scrotale (**Alemayehu & Benti, 2009**). De nombreuses recherches ont rapporté que les paramètres de taille des testicules augmentaient avec différents groupes d'âge, cela pourrait indiquer une structure normale des spermatozoïdes et prouver leur grande capacité de fécondation (**Leal et al., 2004; Abba & Igbokwe, 2015**). Ceci serait dû au fait que l'évolution physiologique selon l'âge des animaux conduit à un développement de l'appareil génital et à une croissance morphologique. Nos résultats sur le changement des paramètres biométriques suggèrent que les activités des testicules et de l'épididyme semblent être plus actives chez les ânes âgés (≥6 ans) et pesés (≥220 kg) que les autres ânes. De plus, la croissance testiculaire est soutenue dans une période appropriée à la prolifération de l'épithélium des tubes séminifères associée à une augmentation de la production de sperme (**Nipken & Wrobel, 2009**).

Dans la littérature, les valeurs biométriques testiculaires et épididymaires semblent affectées par la saison. Des études antérieures ont démontré des variations saisonnières de l'activité de reproduction tout au long de l'année dans des conditions de photopériode naturelles chez certaines espèces animales (**Rakesh et al., 2014 ; Blottner & Jewgenow, 2007**). Dans le présent travail, les paramètres biométriques ont changé au cours des différentes saisons, des valeurs plus élevées ont été enregistrées pour la durée des jours courts (hiver et automne) par rapport à la durée des jours longs (printemps et été) ($P < 0{,}05$). En ce qui concerne les valeurs GSI et SC des ânes, les résultats de cette étude ont également remarqué que ces valeurs étaient significativement plus importantes en hiver et en automne qu'au printemps et en été. Cela pourrait s'expliquer par une augmentation de l'activité des tubes séminifères et de la production de sperme (**Carluccio et al., 2013**). En effet, nos observations sont en accord avec de nombreuses investigations démontrant une augmentation de l'activité physiologique des testicules par des mesures biométriques au cours des différentes saisons chez différentes espèces animales (**Martinez et al., 2012 ; Pasha et al., 2010 ; Machado et al., 2011 ; Perumal et al., 2017**). Nos résultats sont controversés par rapport à ceux précédemment publiés, qui démontraient que des ânes mâles de race Martina Franca et Pêga ne présentaient pas de différences significatives dans les caractéristiques morphométriques testiculaires tout au long de l'année (**Kreuchauf. 1984 ; Gastal et al., 1996 ; Carluccio et al., 2013**). Cette différence pourrait être attribuée à l'environnement nutritionnel et photopériodique, elle est aussi probablement due à une différence génétique de capacité sexuelle des races. Les résultats de cette première étude suggèrent que le comportement sexuel avec une libido intense pourrait être présenté pendant l'hiver et l'automne par rapport au printemps et à l'été chez les ânes locaux algériens, ce qui renforce le concept qu'ils sont contrôlés par la photopériode.

Nos résultats ont aussi révélé qu'il y avait des corrélations significativement très élevées ($P < 0{,}001$) entre les valeurs biométriques testiculaires et épididymaires, celles-ci fournissent des informations sur l'état gonadique des ânes. Cela pourrait prédire que la structure des testicules et de l'épididyme des ânes est normale chez les ânes de la présente investigation. Ces résultats corroborent avec ceux rapportés chez certains animaux domestiques (**Ajao et al., 2014; Ibrahim et al., 2012; Jain et al., 2008 ; Pasha et al., 2011 ; Al Bulushi et al., 2019**).

Etude #2. Variations histomorphométriques des tissus testiculaires en fonction de la saison et de l'âge des ânes de race local d'Algérie (*Equus asinus*) :

La deuxième partie est la suite de la précédente étude et qui a pour but de confirmer les changements physiologiques des testicules au cours des différentes saisons chez l'âne commun dans les conditions d'élevages algériennes. Les observations histologiques sont utiles pour fournir des informations sur les cellules cibles et pour comprendre la physiologie de l'organe. Les caractéristiques histomorphologiques des testicules ont été décrites chez plusieurs espèces animales telles que le taureau, le chameau, le bouc, le bélier, le lapin...etc. (**Alkafafy et al., 2012; Andreussi et al., 2014; Okpe & Ezeasor, 2016 ; Vaškas et al., 2018 ; Umar et al., 2017**). La seconde étude est la première enquête d'histomorphométrie du testicule chez les ânes locaux algériens, montrant les changements dans les compartiments testiculaires selon la saison et l'âge de l'animal. Les données obtenues de cette présente investigation sont essentielles pour mieux comprendre la biologie de la reproduction des ânes en Algérie, et aussi pour expliquer la corrélation entre les mensurations histomorphométriques des tissues testiculaires et la biométrie testiculaire, qui est un aspect important des programmes de conservation des espèces animales en danger d'extinction.

Les résultats obtenus ont révélé qu'il y avait des similitudes dans le diamètre des tubes séminifères des testicules de l'âne algérien avec ceux précédemment rapportés chez les ânes sauvages (*Equus asinus africanus*) (**Nipken & wrobel, 1997**), les ânes de race Pega (**Neves et al., 2002**), les ânes égyptiens (**Moustafa et al., 2015**). À l'inverse, une autre étude (**Han et al., 2016**) a rapporté de faibles valeurs de diamètre des tubes séminifères chez les ânes chinois par rapport à nos résultats. Aussi, **Nipken & Wrobel.** (**1997**) et **Moustafa et al.** (**2015**) ont enregistré des valeurs similaires de la hauteur de l'épithélium des cellules germinales chez les ânes aux résultats de la présente étude. Cependant, l'épaisseur de l'albuginée chez les ânes algériens était inférieure à celle du cheval Chamurthi (**Shukla et al., 2013**). De même, la longueur, la largeur et la hauteur moyennes des testicules sont inférieures à celles décrites chez l'âne brésilien (**Gastal et al., 1997**) et chez les ânes Martina Franca (**Carluccio et al., 2004**). Cela est probablement dû aux différences d'âge et au poids des ânes investigués dans la présente étude. D'autre part, la valeur moyenne du volume testiculaire des ânes obtenue dans cette étude est supérieure à celle rapportée chez la race d'ânes du nord-est du Brésil (**Rocha et al., 2018**).

Les valeurs histomorphométrique du parenchyme testiculaire varient considérablement d'une race à une autre, reflétant probablement l'activité spermatogénèse de chaque race. On se basant sur les mesures histologiques des tissus testiculaires, les présents résultats décrivent l'activité sexuelle de l'âne algérien. Nos données ont révélé que DST, GCEH, TTA et ITSA étaient significativement plus élevés en hiver et en automne (octobre à mars) par rapport au printemps et à l'été (avril à septembre). En revanche, les valeurs de DL étaient significativement plus élevées au printemps et en été. Ces résultats corroborent avec ceux obtenus précédemment chez d'autres espèces (**Dorostghoal et al., 2009; Sudhakar et al., 2010; Ibrahim et al., 2013**), dont le diamètre des tubes séminifères diminuent faiblement pendant les jours longs et augmentent pendant les jours courts.

Dans le présent travail, les changements histologiques des tissus testiculaires des ânes dans les jours courts (*e.i.* automne-hiver) étaient considérables par rapport aux longs jours (*e.i.* printemps-été), ce qui corrobore aussi ceux rapportés par **Beenish** (**2019**) chez les taureaux Buffalo. En outre, **Ovcharenko et al,** (**2018**) ont rapporté que l'épaisseur de l’albuginée augmente considérablement chez le cerf élaphe adulte pendant la saison estivale. Cela pourrait signifier que la période spermatogénèse commence à l'automne, où les spermatocytes grossissent, le diamètre des tubes séminifères augmente également; le poids, la longueur et la largeur des testicules augmentent et la lumière séminifère diminue et se charge de spermatozoïdes. De plus, les valeurs histomorphométriques des tissus testiculaires étaient négativement corrélées ($P < 0,001$) à l'exception du diamètre de la lumière. L'augmentation du diamètre des tubes séminifères pendant la saison automnale et hivernale est due à une prolifération importante de cellules germinales en plein développement dans leur épithélium, provoquant l'extension du tube séminifère.

Il est connu que l'organisation du tissu interstitiel associé au système lymphatique change au cours du cycle de reproduction, y compris la morphologie de la population de cellules de Leydig (**Abd-Elaziz et al., 2012**). Nos observations ont révélé que les cellules de Leydig pendant l'hiver et l'automne sont très abondantes et bien serrées occupant la zone intertubulaire avec peu de tissu conjonctif. D'autre part, l'observation des coupes histologiques de testicules d'âne au printemps et en été a montré que les cellules de Leydig sont dispersées dans un tissu conjonctif abondant avec de vastes sinusoïdes lymphatiques péritubulaires et de très petits lymphatiques interstitiels. En outre, plusieurs études ont signalé une diminution de la taille des cellules de Leydig chez les espèces saisonnières, telles que le daman des rochers, le chameau, la viscache et le hamster, pendant la saison de non-reproduction (**Neaves, 1973 ; Sinha et al., 1988; Muñoz et al., 1997**).

Une corrélation significativement négative a été enregistrée entre le diamètre de la lumière des tubes séminifères et l'âge des ânes ; cela est probablement dû à une diminution du nombre de différents types de cellules dans l'épithélium séminifère. Parallèlement, il existait une corrélation négative entre le diamètre de la lumière et la hauteur des cellules germinales et le diamètre des tubules séminifères. Nos résultats sont en accord avec les conclusions de travaux antérieurs. **Hondo et al.,** (**1998**) ont rapporté que la surface de l'épithélium séminifère et le poids de l'albuginée sont plus élevés chez les chevaux plus âgés et que la surface de l'épithélium séminifère augmente en fonction de l'âge. **Saber** (**1994**) a également observé que les cellules occupaient environ 4 % de la surface intertubulaire chez les ânes âgés de 2 ans, et est devenu 8% à l'âge de 6 ans, puis a diminué à environ 6% à l'âge de 9 ans.

Cependant, chez l'âne sauvage d'Afrique (*Equus asinus africanus*), le diamètre de l'épithélium séminifère diminue dès l'âge de 5 ans de 3 µm par an et atteint 70 µm à 10 ans (**Nipken & wrobel, 1997**). Ces différences pourraient s'expliquer par le type d'espèces sauvages ou domestiques, la diversité génétique, la qualité des nutriments et les facteurs environnementaux.

Etude #3. Variations saisonnières de concentrations de testostérone plasmatique et des paramètres biochimiques des ânes (*Equus asinus*) dans le nord de l'Algérie :

Ensuite nos recherches se sont poursuivies par une troisième investigation afin de confirmer l'aspect de saisonnalité de l'activité sexuelle chez l'âne local d'Algérie. On sait pertinemment que les équidés ont longtemps été considérés comme une espèce à reproduction saisonnière avec une activité sexuelle liée aux périodes de longues journées. Cependant, certaines études ont démontré que les femelles d'ânes (*Equus asinus*) peuvent cycler tout au long de l'année (**Contri et al., 2014**). De plus, l'activité ovarienne, la gestation et la parturition semblent être beaucoup moins saisonnières chez les ânes domestiques que chez les ânes sauvages (**Ginther et al., 1987**). Dans une autre étude, il a été rapporté que les ânesses peuvent présenter un cycle oestral toute l'année dans le sud des États-Unis, tandis que 64 % des femelles ont un cycle en décembre dans le Wisconsin (**Tibary, 2004**). Au Maroc, les constatations de terrain montrent que le pic des naissances est en mars et quelques naissances sporadiques en février et de juin à septembre (**Tibary et al., 2006**). De plus, les résultats obtenus par **Carluccio et al.,** (**2013**) ont montré que la saison n'a aucun effet sur la morphométrie testiculaire et les caractéristiques du sperme. Une étude menée par **Canisso et al.** (**2019**) a rapporté une augmentation du pourcentage de la morphologie des spermatozoïdes et une diminution de la motilité spermatique au printemps et en été par rapport à l'automne. Une autre étude a conclu qu'il n'y avait pas de variations saisonnières apparentes dans la qualité du sperme (**Gastal et al., 1997; Canisso et al., 2019**).

À notre connaissance, la présente étude est la première à évaluer l'influence de la saison sur la concentration en T et les paramètres biochimiques chez les ânes de race locale d'Algérie, et cela pourrait aider à déterminer la saison de reproduction dans le nord de l'Algérie.

La testostérone (T) est connue comme l'hormone sexuelle mâle qui contrôle les changements de l'épididyme et des testicules, ainsi que des glandes accessoires. Il est essentiel et responsable de l'augmentation de l'activité sexuelle et de la taille des testicules exprimée par la prolifération des cellules de Leydig et des cellules germinales. Les résultats obtenus ont révélé que les concentrations de T pendant les jours courts étaient significativement plus élevées que pendant les jours longs.

Selon **Aissanou & Ayad** (**2020, 2022**), la race d'ânes vivant dans le nord de l'Algérie expriment une influence saisonnière sur la biométrie et l'histophotométrie des testicules, c'est-à-dire lors de journées courtes ; ceci est en plein accord avec les données enregistrées sous les mêmes latitudes de la présente investigation. Cela pourrait s'expliquer par la coïncidence avec la spermatogenèse et les pics de testostérone en hiver et en automne. De plus, l'augmentation de T peut être causée par une augmentation des niveaux d'androgènes parallèlement à une activité sexuelle accrue en hiver et en automne. Aussi, **Schuler et al.,** (**2019**) ont enregistrés des valeurs de concentration plasmatique de T similaires aux résultats de la présente étude. Cependant, **Jiaha** (**1983**) a révélé que les concentrations moyennes de T pendant la saison de reproduction (mars-septembre) étaient plus élevées que pendant la saison de non-reproduction (oct-février) chez l'âne de Guan-Zhong. Ces différences entre les études peuvent être dues à des différences entre les races, le niveau d'alimentation, la saison, la photopériode et la latitude, comme décrit chez d'autres espèces animales (**Aldori et al., 2017 ; Ismaeel, 2018 ; Ait-Amran et al., 2013**), il est probablement dû à une gestion hétérogène dans diverses régions agro-climatiques et pratiques alimentaires (**Gupta et al., 2016 ; Cihan Kabukçu et al., 2020**).

Les lipides occupent une place importante dans le fonctionnement de l'organisme et servent d'hormones ou de précurseurs hormonaux tels que la stéroïdogenèse (**Welte & Gould, 2017 ; Maxfield & Tabas, 2005**). Les résultats de cette troisième étude ont révélé que les paramètres biochimiques sériques des ânes locaux algériens sont très similaires à ceux publiés précédemment chez les ânes sauvages (**da Silva et al., 2018**) et les ânes d'Allemagne (**Schuler et al., 2019**). Dans notre étude, les taux de Ch et de Tg étaient significativement plus élevés ($P<0,001$) en hiver et en automne, ce qui correspond à une période de jours courts. Ce résultat est en accord avec les rapports précédents (**Roy et al., 2004 ; Longodor et al., 2020**), dans lesquels les niveaux de Ch et Tg étaient significativement plus élevés en hiver qu'en été chez les ânes indiens et roumains. Aussi, **Seri et al.** (**2010**) a rapporté que les valeurs de (ALT, AST, Ch et Tg) ont montré des changements saisonniers significatifs chez les ânes soudanais et que les concentrations les plus élevées ont été détectées pendant la saison des pluies. De plus, **Hasković & Suljević** (**2011**) ont enregistré des résultats similaires à la présente étude chez le cheval de montagne bosniaque, dont l'activité enzymatique de l'AST et de l'ALT a montré des variations saisonnières significatives entre l'été et l'automne, tandis que l'ALP ne montre aucun changement saisonnier. À l'inverse, nos résultats ont remarqué que les valeurs de Tg, Chol et AST étaient plus faibles chez les ânes locaux algériens par rapport aux ânes anglais, rapportés précédemment par **Burden et al.** (**2016**).

Nos résultats pourraient s'expliquer par l'existence d'une relation étroite entre l'AST, située dans la partie médiane des spermatozoïdes, et la motilité des spermatozoïdes ; par conséquent, le défaut de membrane et la perte d'AST entraînent le blocage de la production d'ATP (**Pesch et al., 2006**). Dans la littérature, le système enzymatique (AST, ALT et ALP) joue un rôle essentiel dans les processus métaboliques des spermatozoïdes, fournissant de l'énergie pour la survie, la motilité et la fertilité des spermatozoïdes chez les étalons (**Kosiniak et al., 2000; Turner & McDonnell, 2003**). De même, il a été rapporté que les activités transaminases (AST et ALT) dans le sperme sont un bon indicateur de la qualité du sperme car elles mesurent la stabilité de la membrane spermatique (**Sirat et al., 1996; Corteel et al., 1980**).

Une corrélation positive a été enregistrée entre les concentrations de T et les paramètres biochimiques ; ceci est probablement dû à l'augmentation de la stéroïdogenèse et à la production de sperme. Le cholestérol est l'un des précurseurs de la biosynthèse des stéroïdes et est utilisé pour la synthèse de la testostérone (**Amerkhanov et al., 2013**). De plus, plusieurs enzymes ont été identifiées dans les glandes accessoires du plasma séminal des étalons. Cette corrélation entre les activités enzymatiques et les concentrations de T pourrait fournir des informations sur la qualité de la semence et la fertilité des étalons (**Kareskoski, 2011**).

Il a été rapporté que la photopériode est le facteur environnemental le plus important qui affecte la fonction de reproduction chez les ânes (**Cappai et al., 2017 ; Deikman et al., 2002**), ceci en raison de son influence sur les niveaux circulants de gonadotrophines et d'hormones gonadiques. Les données de cette étude ont montré de haute corrélation négative ($p<0,001$) de la photopériode avec T, Ch et AST chez les ânes algériens. Les résultats de la présente étude sont également cohérents avec ceux publiés précédemment, qui ont montré des corrélations significatives entre les valeurs de T et les données photopériodiques chez l'âne de Guan Zhong (**Jiaha, 1983**). Il est à noter que la photopériode influence les espèces animales à reproduction saisonnière par des changements dans la sécrétion de mélatonine qui affecte la fonction de l'axe hypothalamo-hypophyso-testiculaire (**Vivid & Bentley, 2018**).

Conclusions générales et perspectives

La présente thèse a un principal objectif qui est de caractériser la saison de reproduction chez l'âne commun (*Equus asinus*) en Algérie. Comme nous l'avons rappelé dans l'introduction, il s'agit de la première investigation qui décrit les organes reproducteurs mâles chez l'âne de la race locale algérienne (*Equus asinus*), sur la base de mesures biométriques testiculaires et épididymaires ; les modifications histomorphométriques des tissus testiculaires et également les variations saisonnières des concentrations de T et des paramètres biochimiques sanguins en fonction des saisons.

Nos résultats ont montré que dans :

- **L'étude #01 :** des différences essentielles ont été enregistrées entre certains paramètres biométriques testiculaires en fonction de l'âge, la saison et le poids corporel des ânes. De plus, les coefficients de corrélation étaient considérables significatives entre les mesures biométriques et ces facteurs.
- **L'étude #02 :** l'analyse comparative des paramètres histologiques, tels que le diamètre des tubes séminifères, la hauteur de l'épithélium des cellules germinales, le diamètre de la lumière, l'épaisseur de l'albuginée et la surface inter-tubaire, suggère que l'activité sexuelle se produit généralement pendant l'hiver et l'automne chez les ânes de race local du nord d'Algérie. De plus, les valeurs histomorphométriques des tissus testiculaires sont corrélées avec l'âge des ânes.
- **L'étude #03 :** l'activité sexuelle se produit probablement pendant l'hiver et l'automne chez les ânes vivants au nord de l'Algérie. Egalement, les niveaux de Ch et de Tg sont corrélés avec la saison.

Les conclusions de la présente thèse nous permettent de dégager un certain nombre de perspectives à savoir :

- Élargir la recherche sur l'effet de la saison et l'âge sur les paramètres biochimiques du sperme chez l'âne.
- Entreprendre des investigations sur l'analyse spermatique chez l'âne commun en Algérie en fonction de la saison et l'âge (Concentration, mobilité et viabilité ...etc.).
- Entreprendre des essais sur la cryoconservation de la semence en améliorant les milieux de conservation avec des composés bioactifs, ainsi que l'application de l'insémination artificielle chez l'âne.

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Annexes

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MORPHOLOGICAL CHARACTERISTICS OF DONKEYS (*EQUUS ASINUS*) IN KABYLIE AREA, ALGERIA

Abdelhanine AYAD*, Sofiane AISSANOU, Karim AMIS, Amel LATRECHE, Mokrane IGUER-OUADA

Department of Biological Sciences of the Environment, Faculty of Nature and Life Sciences, University Abderrahmane Mira, Bejaia, Algeria

ABSTRACT

The survey was to define some morphometric characteristics and body biometric indexes of donkeys sampled in the Kabylie area, Algeria. The study was carried out from February to June 2018 in Bejaia and Tizi-Ouzou province. The study population included 124 males and 2 females. In total, 17 body measures were selected for morphometric characterization including and seven body biometric indexes were calculated. Body weight estimated the two equations was 144.3 ± 23.9 and 171.5 ± 28.8 kg, respectively. Significant higher body weight was recorded in the age group ≤ 5 years and the lower body weight in the age group ≥ 6 - ≤ 10 years and ≥ 11 years. Morphological variables of chest width (CW) and Cannon length (CL) were significant longer ($P < 0.02$) in aged donkeys (25.2 ± 1.3 and 20.5 ± 0.7 cm, respectively) compared to adult donkeys (24.7 ± 2.3 and 20 ± 1.4 cm, respectively). Aged donkeys (114.8 ± 5.8 cm) were also significantly superior ($P < 0.01$) concerning the thoracic circumference (TC) compared to adult donkeys (112.2 ± 9.8 cm). The highest values were found between WH and BH ($r = 0.80$); HR and BH ($r = 0.72$) HR and WH ($r = 0.72$) ($P < 0.05$). Dactyl thoracic Index (DTI), Compact Index (CI), Massive index (MI) and Relative body index (RBI) appeared to be influenced by donkey ages ($P > 0.05$). This is a first report on the phenotypic characterization in donkeys in Kabylie area (Algeria) based on corporal measurements. Our comparative analysis of morphometric parameters; such as back length, body length, neck length; suggests that donkeys of Kabylie area are typically invariant among breeds and it has not been changed through the periods.

Key words: donkeys; morphometric characterization; Kabylie; Algeria

INTRODUCTION

Donkey (*Equus asinus*) is an odd-toed ungulate and the smallest species in the Equidae family (Grinder *et al*., 2006). Donkeys in their nature are very friendly, calm, quite, patient, intelligent, cautious, playful, and eager to learn and enjoy the company of humans. It is characteristically short-legged with exceptionally long ears. Importance of donkeys is also conferred through their use in riding tourism and as eco-friendly means of pack and transportation when compared with horses (GOVS, 2005). Donkeys (*Equus asinus*) represent an important component of Algerian livestock and make a significant contribution to the agricultural economy; serving as draft animals.

According to the year 2001 inventory; the donkeys population is estimated 180160 heads in Algeria (FAO, 2003) found essentially in the northern regions, where they are particularly appropriate to tolerate the hard conditions of works. As draught animals, donkeys play a major role in the economy of developing countries by being the main source in transport and traction, particulary in areas with difficult reliefs. However, despite the donkey's popularity, information regarding various morphological characteristics in this species is limited (Labbaci *et al*., 2018).

***Correspondence:** E-mail: hanine06@gmail.com
Prof. Dr. Abdelhanine Ayad (DVM, MSc, PhD), Department of Biological Sciences of the Environment, Faculty of Nature and Life Sciences, University Abderrahmane Mira, Bejaia 06000, Algeria
Tel.: +213 7727 722 595

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The capacity performance of donkeys could be assessed by the description of the morphological characteristics, such as umbilical girths, body length and height. This has been suggested as donkey draft power is directly proportional to size parameters (Nengomasha, 1999). In donkey, during domestication, some morphological and genetic changes have taken place in order to survive better in given conditions (Rossel *et al*., 2008). In African continent, the typical factors (high daily temperatures, minimal amount of precipitation and lack of nutriment quality) enabled donkeys to develop typical aptitudes, which played a key role to survive in dry areas (Pearson and Ouassat, 2000). The knowledge of morphometric measurements in donkey is of great importance for the genetic diversity preservation and development and taxonomic affiliation. Thus, the general objective of the current study was to contribute to a better knowledge of donkey in Algeria, especially in Kabylie region, known for its typical mountains. The survey was to define some morphometric characteristics and body biometric indexes of donkeys sampled in Bejaia and Tizi-Ouzou province. The correlation coefficients different between body measurements were estimated.

MATERIAL AND METHODS

Area study

The study was carried out from February to June 2018 in the Kabylie area, Algeria. Different localities of Bejaia (36° 43' N, 5° 04' E) and Tizi-Ouzou (36° 42' N, 4° 2' E) province were chosen randomly. The topography of Kabylie area is mostly predominated by mountainous. The vegetation is mainly composed of several species of trees and natural or cultivated herbs. Constitute part of climate is Mediterranean region. The maximum summer temperature are ranged from 30.3 to 36.3 °C (July) and the minimum winter temperature are ranged from 6.6 to 6.7 °C (February).

Animal and measurements

The study population included 124 males and 2 females. The donkeys are divided in 3 age groups namely ≤ 5 (young), ≥ 5 - ≤ 10 (adulte), ≥ 11 (aged). In total, 17 body measures were selected for morphometric characterization including. Linear measures (Figure 1) as head length (HL), ear length (EL), neck length (NL), chest width (CW), back length (BaL), body length (BoL), hips width (HW), umbilical circumference (UC), back height (BH), height at the rump (HR), thoracic circumference (TC), chest depth (CD), withers Height (WH), front leg length (FLL), cannon circumference (CC), cannon length (CL), cannon height (CH) were performed using a specially graduated measuring tape. The ages of donkeys were determined from the donkey owners and controlled by dentition analysis (Daveze and Raveneau, 2002). The identification of robe color was performed by direct observation under natural daylight and the frequency distribution of each phenotype was estimated.

From some measured morphometric donkeys, seven body biometric indexes were calculated according to the following formulas. Body Profile Index (BPI) = WH/BoL (Mariante *et al*., 2002); > 0.90: long and good animal for speed; 0.86 - 0.88: medium conformation animal or < 0.85: small conformation animal, fit for traction. Pectoral height index (PHI) = CD/FLL (Marcenac *et al*., 1980); 0.50 ≤ PHI ≤ 0.55: leggy animal or PHI > 0.56: leg shorted. Dactyl thoracic index (DTI) = CC/TC (Chabchoub *et al*., 2004); this index define three animal types: hypermetric, eumetric and elliptical. Compact index (CI) = BW/WH (Boujenane *et al*., 2008). Front-back height in (FBH) = WH/HR (Marcenac *et al*., 1980); FBH ≤ 1: straight back (no overload) or FBH > 1: the anterior region is higher than the posterior (overload). Massive index (MI) = TC/WH (Mariante *et al*., 2002); MI ≤ 1: support well its weight or MI > 1: massive overload. Relative Body Index (RBI) = BoL/TC (Nicks *et al*., 2006); RBI ≥ 0.90: longilinear, 0.84 ≤ RBI ≤ 0.89: mediolinear or RBI ≤ 0.83: brevilinear.

The body weight (BW) for each animal was calculated according to two validated formulas: BW-1 = $TC^{2.65}$/2188 (Pearson and Ouassat, 1996) or BW-2= ($WH^{0.24}$) x ($TC^{2.576}$) x 0.000252 (Eley and French, 1993).

Statistical analysis

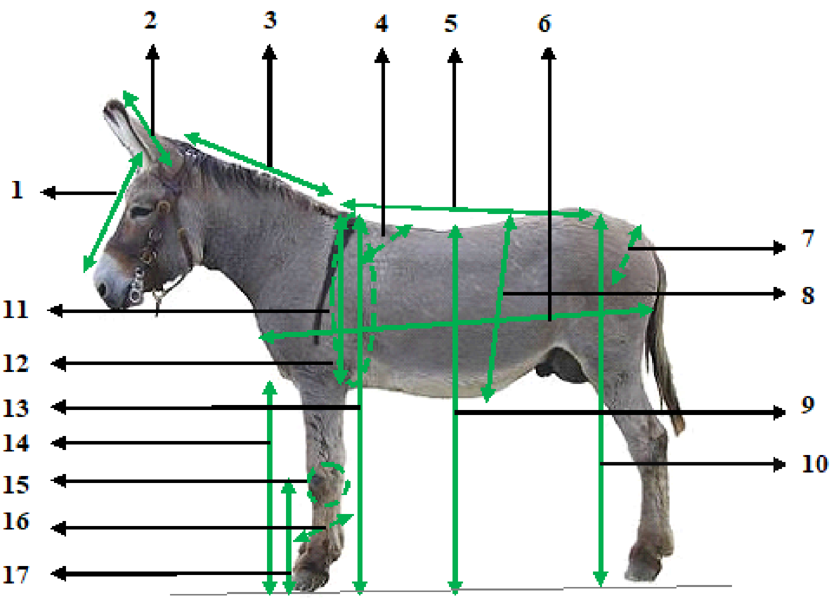
Data were analyzed using a mixed model for repeated measurements (Statview Software, Version 4.55) taking into account an autocorrelation between data obtained successively on the same animal. The data (± SD) were expressed as values of the donkey body measurements (cm). The animal weight and donkey body measurements were analyzed using age (young: ≤ 5 years aged; adult: ≥ 5 - ≤ 10 years aged; aged: ≥ 11 years) as factors of variation. The one way variance analysis (ANOVA) was used to evaluate the obtained data. The values were statistically different when the *P*-value was < 0.05.

RESULTS

The frequency of the coat color (Figure 2) showed that 59.5 % of the donkeys had various shades of brown, 27 % grey and 13.5 % black. The body weight according to the age of donkeys is illustrated in Figure 4. Body weight estimated the two equations was 144.3 ± 23.9 and 171.5 ± 28.8 kg, respectively. The higher body weight was recorded in the age group ≤ 5 years and the lower body weight in the age group ≥ 6 - ≤ 10 years and ≥ 11 years (Figure 3). A significant difference of body weight was observed between the young donkey group and the aged donkey group (P < 0.05).

Descriptive statistics of morphological variables including mean, standard deviation, minimal-maximal and coefficient of variation are depicted in Table 1. Mean values of morphological variables and their standard for each age group are shown in Table 2. Morphological variables of chest width (CW) and Cannon length (CL) were significant longer (P < 0.02) in aged donkeys (25.2 ± 1.3 and 20.5 ± 0.7 cm, respectively) compared to adult donkeys (24.7 ± 2.3 and 20 ± 1.4 cm, respectively). Aged donkeys (114.8 ± 5.8 cm) were also significantly superior (P < 0.01) concerning the thoracic circumference (TC) compared to adult donkeys (112.2 ± 9.8 cm).

Phenotypic correlation coefficients (r) among morphologic variables and body weight are given in Table 3. The highest values were found between WH and BH (r = 0.80); HR and BH (r = 0.72) HR and WH (r = 0.72) (P < 0.05). Other high values were found between CW and BLL (r = 0.60), WH and CH (r = 0.56), WH and HR (r = 0.56) (P < 0.05). The correlation values of TC-CH, WH-CH, WH-HR, HL-TC, BaL-WH, BoL-WH and TC-WH presented values



1 - Head length (HL); 2 - Ear length (EL); 3 - Neck length (NL); 4 - Chest width (CW); 5 - Back length (BaL); 6 - Body length (BoL); 7 - Hips width (HW); 8 - Umbilical circumference (UC); 9 - Back height (BH); 10 - Height at the rump (HR); 11 - Thoracic circumference (TC); 12 - Chest depth (CD); 13 - Withers Height (WH); 14 - Front leg length (FLL) 15 - Cannon circumference (CC); 16 - Cannon length (CL); 17 - Cannon height (CH)

Figure 1. The different body measurements performed in donkey

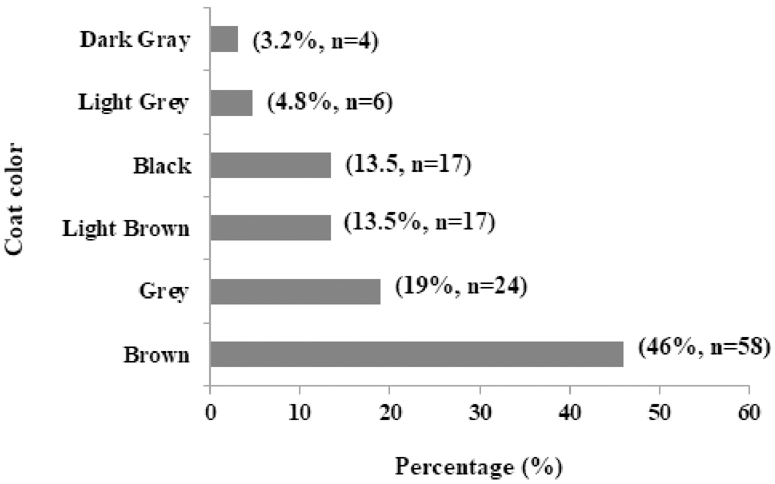
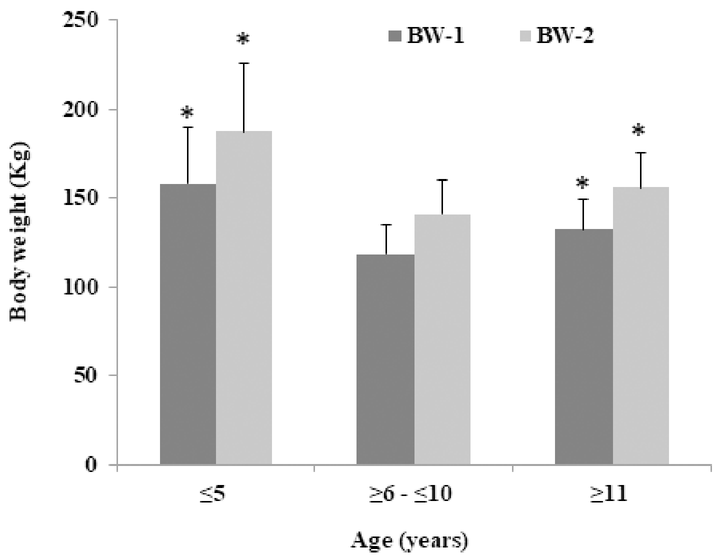


Figure 2. Frequency distribution of coat color of donkey in Kabylie area, Algeria

ranged between 0.51 and 0.58 ($P < 0.05$). Other low or very low correlation values were found between the others morphological parameters. There were also no high negative correlations between all other traits. For the body weight, the correlations were more marked with TC ($r = 0.99$), moderately marked with HL; WH and CH ($0.50 \geq r \leq 0.70$), and weakly marked with the rest of the morphological parameters ($P < 0.05$).

The results of body biometric indexes are summarized in Table 4. Dactyl thoracic Index (DTI), Compact Index (CI), Massive index (MI) and relative body index (RBI) appeared to be influenced by donkey ages ($P > 0.05$). The averages of the DTI, CI, MI and RBI index are 0.18 ± 0.01, 1.34 ± 0.2, 1.11 ± 0.06 and 0.93 ± 0.06, respectively. The BPI, FBH and PHI indexes are 0.97 ± 0.05; 0.66 ± 0.04 and 0.98 ± 0.04 respectively.



Means with the same superscripts in each weight of different ages are significantly different (*P < 0.05).

Figure 3. Weight of donkey by age groups (young: ≤ 5 years aged; adulte: ≥ 5 - ≤ 10 years aged; aged: ≥ 11 years)

Table 1. Descriptive analysis of donkey body measurements in Kabylie area, Algeria

	Mean ± SD	Min-Max	Median	CV (%)	95 % CI
BW-1 (kg)	144.3 ± 23.9	91.2 - 107.5	146.2	0.165	473 - 718.1
BW-2 (kg)	171.5 ± 28.8	107.5 - 250.8	172.8	0.168	679.7 - 1031.9
HL (cm)	48.5 ± 3.3	40 - 56	48	0.069	9.1 - 13.8
EL (cm)	24.4 ± 1.8	20 - 28	25	0.074	2.7 - 4.1
NL (cm)	46 ± 4.7	33 - 56	47	0.102	18.1 - 27.4
CW (cm)	25.6 ± 1.9	20 - 29	26	0.073	7.1 - 10.8
BaL (cm)	63.2 ± 2.5	58 - 72	63	0.039	5 - 7.6
BoL (cm)	110.1 ± 5.9	91 - 130	110	0.054	28.6 - 43.4
HW (cm)	32.4 ± 1.6	29 - 40	32	0.051	2.2 - 3.4
UC (cm)	141.1 ± 10.3	108 - 161	142	0.073	86.3 - 131
BH (cm)	107.2 ± 5.3	92 - 120	107.5	0.039	23.2 - 35.2
HR (cm)	109.6 ± 4.8	97 - 118	110	0.044	18.9 - 28.7
TC (cm)	118.5 ± 7.5	100 - 137	119.5	0.063	45.9 - 69.8
CD (cm)	49.2 ± 1.94	44 - 56	49	0.039	3.1 - 4.7
WH (cm)	106.9 ± 5.4	94 - 118	107	0.051	24.3 - 36.9
FLL (cm)	75 ± 3.9	51 - 82	75	0.052	12.6 - 19.2
CC (cm)	14.7 ± 1.1	12 - 23	15	0.078	1.1 - 1.6
CL (cm)	21.07 ± 1.72	14 - 25	21	0.081	2.4 - 3.7
HC (cm)	31.98 ± 2.92	17.5 - 38	32	0.091	7 - 10.6

Min: minimal value; Max: maximal value; CV: coefficient of variation; CI: confidence interval.

Table 2. Morphometric measurements of the donkeys in Kabylie area, Algeria

Body variables (cm)	Young donkeys (≤ 5 years) (n = 13) (Mean ± SD)	Adult donkeys (≥ 6 - ≤ 10 years) (n = 62) (Mean ± SD)	Aged donkeys (≥ 11 years) (n = 51) (Mean ± SD)
HL	47.8 ± 5.3[a]	46.3 ± 2.3[a]	47.1 ± 2.1
EL	24.8 ± 1.9	23.6 ± 2.4	23.7 ± 1.6
NL	44.4 ± 5.4	48.9 ± 1	48.2 ± 1.6
CW	25.6 ± 2.1[a]	24.7 ± 2.3[a,b]	25.2 ± 1.3[b]
BaL	63.5 ± 3.8[a]	61.7 ± 1.5[b]	62.1 ± 2.2[a,b]
BoL	107.3 ± 8.7[a]	109.6 ± 3.5	108.4 ± 6.4[a]
HW	33.2 ± 2.9[a,b]	32.5 ± 0.9[a]	32.1 ± 2[b]
UC	137.5 ± 14.6	142.8 ± 12.5	143.4 ± 9.3
BH	105.8 ± 6	107.8 ± 5.3[a]	105.8 ± 4.2[a]
HR	110.2 ± 5.8	108.7 ± 5.3[a]	107.2 ± 3.7[a]
TC	112.2 ± 9.8[a,b]	110.2 ± 5.9[a]	114.8 ± 5.8[b]
CD	48.8 ± 3.2[a,b]	49.6 ± 1.2[a]	48.4 ± 2.1[b]
WH	106 ± 6.3	105.3 ± 4.8	103.2 ± 5.2
FLL	73.5 ± 7.5[a]	77.2 ± 1.6[a,b]	73.5 ± 3.2[b]
CC	14.5 ± 1.3[a]	14.8 ± 0.6[a,b]	14.7 ± 0.9[b]
CL	21.5 ± 1.6	20 ± 1.4	20.5 ± 0.7
CH	32.5 ± 2.1[a]	30.3 ± 0.6[a,b]	30.8 ± 1.1[b]

[a,b] Means with the same letters superscripts in each row of different ages are significantly different ($P < 0.05$).

Table 3. Phenotypic correlation coefficients (r) between body measurements in donkeys (*P < 0.05)

	BW-1	BW-2	HL	EL	NL	CW	BaL	BoL	HW	UC	BH	HR	TC	CD	WH	FLL	CC	CL	CH
BW-1																			
BW-2	**0.99***																		
HL	**0.53***	**0.55***																	
EL	033*	0.34*	0.23*																
NL	-0.08	-0.08	-0.35*	0.07															
CW	0.40*	0.40*	-0.01	0.12	0.13														
BaL	0.48*	0.50*	0.43*	0.28*	-0.08	0.34*													
BoL	0.34*	0.37*	0.27*	0.47*	0.14	0.05	0.29*												
HW	0.29*	0.30*	0.15	0.31*	0.02*	0.23	0.32*	0.25*											
UC	0.17	0.19*	0.32*	0.30*	-0.08	-0.13	0.26*	0.33*	0.22*										
BH	0.36*	0.40*	0.36*	0.38*	0.05	0.09	0.41*	0.46*	0.40*	0.46*									
HR	0.49*	**0.52***	0.32*	0.40*	-0.04	0.08	0.41*	0.49*	0.36*	0.44*	**0.72***								
TC	**0.99***	**0.99***	**0.54***	0.33*	-0.07	0.40*	0.45*	0.35*	0.26*	0.17	0.36*	0.47*							
CD	0.37*	0.39*	0.22*	0.16	-0.04	0.39*	0.47*	0.25*	0.27*	0.22*	0.40*	0.31*	0.36*						
WH	**0.51***	**0.56***	0.45	0.41*	-0.09	0.20	**0.53***	**0.51***	0.41*	0.36*	**0.80***	**0.72***	**0.51***	0.45*					
FLL	0.25*	0.26*	0.07	0.21*	0.22*	**0.60***	0.40*	0.28*	0.39*	0.13	0.40*	0.39*	0.24*	0.35*	0.41*				
CC	0.25*	0.25*	0.19*	0.01	0.14	0.12	0.16	0.10	0.16	0.19*	0.08	0.14	0.25	0.17	0.14	0.19*			
CL	0.39*	0.40*	0.39*	0.36*	-0.06	0.17	0.32*	0.35*	0.18	0.18	0.35*	0.42*	0.39*	0.29*	0.41*	0.29	0.16		
CH	**0.58***	**0.60***	0.25*	0.37*	-0.03	0.33*	0.38*	0.32*	0.34*	0.19*	0.43*	**0.56***	**0.58***	0.31*	**0.56***	0.36*	0.16	0.46*	

Head length (HL); Ear length (EL); Neck length (NL); Chest width (CW); Back length (BaL); Body length (BoL); Hips width (HW); Umbilical circumference (UC); Back height (BH); Height at the rump (HR); Thoracic circumference (TC); Chest depth (CD); Withers Height (WH); Front leg length (FLL); Cannon circumference (CC); Cannon length (CL); Cannon height (CH).
* P < 0.05

Table 4. Morphometric index of the donkeys in Kabylie area, Algeria

Index	Young donkeys (≤ 5 years) (n = 13) (Mean ± SD)	Adult donkeys (> 6 - < 10 years) (n = 62) (Mean ± SD)	Aged donkeys (> 11 years) (n = 51) (Mean ± SD)	Donkeys Total (n = 126) (Mean ± SD)
BPI	0.99 ± 0.05	0.97 ± 0.05	0.97 ± 0.05	0.97 ± 0.05
PHI	0.67 ± 0.08	0.66 ± 0.03	0.66 ± 0.04	0.66 ± 0.04
DTI	0.18 ± 0.01^{a}	0.17 ± 0.02^{b}	0.18 ± 0.01^{a,b}	0.18 ± 0.01
CI	1.46 ± 0.26^{a}	1.36 ± 0.2	1.3 ± 0.16^{a}	1.34 ± 0.2
FBH	0.96 ± 0.03	0.98 ± 0.04	0.98 ± 0.04	0.98 ± 0.04
MI	1.14 ± 0.05^{a,b}	1.11 ± 0.06^{a}	1.09 ± 0.07^{b}	1.11 ± 0.06
RBI	0.89 ± 0.04^{a,b}	0.93 ± 0.07^{a}	0.95 ± 0.06^{b}	0.93 ± 0.06

Body Profile Index (BPI), Pectoral height index (PHI), Dactyl thoracic index (DTI), Compact index (CI), Front-back height (FBH), Massive index (MI), Relative Body Index (RBI).
^{a,b} Means with the same letter superscripts in each row of different ages are significantly different (P < 0.05).

The analysis of the correlation coefficients between the biometric indexes (Table 5) shows both negative and positive correlations (P < 0.001). Particularly significant positive correlation (P < 0.001) is recorded between CI and MI (r = 0.816) on one hand with significant negative correlation (P < 0.001) between CI and RBI (r = -0.71) on the other hand.

Table 5. Correlation coefficients (r) between morphometric index in donkeys

	BPI	PHI	DTI	CI	FBH	MI	RBI
BPI	-	-	-	-	-	-	-
PHI	0.038	-	-	-	-	-	-
DTI	-0.095	-0.065	-	-	-	-	-
CI	-0.001	0.034	-0.464*	-	-	-	-
FBH	0.395*	0.045	-0.011	-0.046	-	-	-
MI	0.233*	0.085	-0.469*	**0.816***	-0.329*	-	-
RBI	**-0.536***	-0.104	0.474*	**-0.71**	-0.006	**-0.69***	-

Body Profile Index (BPI), Pectoral height index (PHI), Dactyl thoracic index (DTI), Compact index (CI), Front-back height in (FBH), Massive index (MI), Relative Body Index (RBI). *P < 0.001.

DISCUSSION

Around world, and particularly in Africa, donkey is suitable in difficult regions, especially in mountainous area. They played a major role in the evolution of agriculture until the introduction mechanization that neglected this animal. Traditionally, donkeys are part of the Algerian agricultural systems used as a mean of products transport and animal draft, especially in Kabylie area. The morphobiometric characterization has been proposed as one of the strategies for analyzing and characterization of domestic populations (Bouchel *et al.*, 1997). The general objective of the current study was therefore to evaluate the morphometric variation and some biometric indexes; and to estimate the correlation coefficient between measurements of donkey in Kabylie region.

Out of a total of 126 donkeys, only two females were sampled in the present study. In Kabylie region, as in all of North Africa, donkey is certainly the most used animal in the daily life of people, especially in the village constructions. However, there are no donkey females in Kabylie area. Indeed, possession of a donkey female is not allowed for traditional reasons as breeding are located in the other regions of Algeria.

The results of survey revealed that the coat color was diversified with a predominance of brown color (46 %) following by grey color (19 %). In another survey conducted in the Tlemcen area of the East Northern Algeria, Labbaci *et al.*, (2018) reported a similar observation with the presence of five different classes of color of the studied donkeys. In Bulgaria, the coat color also varies where the more common colors are brown (57 %) and grey (Vleava *et al.*, 2016). The body coat color frequencies the Turkish donkeys are: mouse gray, white, black and brown (31.4 %, 24.7 %, 23.7 % and 20.2 %, respectively) (Yimlez and Ertuğrul, 2012). In Ethiopia, a variability of coat color in donkeys has been reported from some country localities (Kefena *et al.*, 2011). In North African region, there are two fundamental denominations of the donkey "*aγyul*" and "*ayzeḍ*", very widespread in Berber language. The word "aγyul" could be a term related to the brown color and derivative of the verb "*iγwal*" which means to be brown in the Touareg population of Southern Algeria (Camps *et al.*, 1985). Our results show that the donkey population was heterogeneous in Kabylie region. This difference of coat color could be attributed to ecological patterns and altitudinal gradients (Gizaw *et al.*, 2007).

Body weights were compared between young, adult and aged donkeys. Our results corroborate with those reported by Ebangi and Vall (1998) revealing a consistent development in body weight for estimated from 1 to 8 years with a decline thereafter. A similar donkey body weight was found in south-western Zimbabwe (Nengomasha *et al.*, 1996; 1999). In another study, the body weight average was higher than those reported by Nininahazwe *et al.* (2017) in West African and Stanišić *et al.* (2015) in Serbia. Also, this body weight is lower compared to investigation in Morocco (Boudjenane *et al.*, 2008). The differences between the average weight values can be explained by the condition of the donkeys when taking the measurements, but also by the formulas used to estimate body weight.

Our findings revealed that young animals, adults, and aged animals do not present the same body parameters and this increases concomitantly with age for certain parameters (Table 3). There was a significant difference between the values of some variables measurements (CW, TC and CL) according to the animal age classes. This would be due to the fact that the physiological evolution according to animal age leads to an increase in weight and morphological growth. These findings corroborate with results obtained previously (Roamba, 2014; Kaboré, 2014; Nininahazwe *et al.*, 2017; Labbaci *et al.*, 2018). The size and body dimensions of donkeys in Kabylie region were similar to those reported in other parts of Africa e.g. Morocco (Pearson and Ouassat, 1996), Zimbabwe (Nengomasha *et al.*, 1999). It is reported by Wilson (1981) that there is little physical variation in donkeys found throughout Africa. Algerian donkeys have a less long body length than donkeys of Bulgaria (Barzev, 2004), Cyprus (Barzev, 2004), Turkey (Yilmaz and Ertuğru, 2012) and Martina Franca (Barzev, 2004). In Kabylie region, donkeys are used for pack transport to carry all types of merchandise e.g. during the olive picking period. The results of this study noticed that donkeys are small in size compared to the other mentioned above. This can be explained by the difficulty of living conditions such as food quantity and quality and work intensity.

From the analysis of obtained results, the correlations among 17 morphological variables observed, in general, are positive ($P < 0.05$) and similar to those reported in numerous studies (Folch and Jordana, 1997; Yilmaz and Ertuğru, 2012; Yilmaz *et al.*, 2013; Daloum *et al.*, 2015; Sobotková *et al.*, 2015).

The correlations between BW and some measurements were significant ($p < 0.05$). Regardless of the age of the donkey, the TC was the only measure highly correlated with the both results of BW formulas. Many investigations have reported a correlation coefficient of 0.90 between BW and TC (Pearson and Ouassat, 1996; Nengomasha *et al.*, 1999; Hassan *et al.*, 2013; Nininahazwe *et al.*, 2017). Furthermore, Aluja et al (2005) confirmed that the thoracic circumference was found to be an easier and more reliable measurement compared to the umbilical circumference which could be affected by other factors such as the moment of food ingestion, the food quantity and the physiological state (gestation).

In order to study deeply donkey conformation in Kabylie area, some indexes were assessed from the morphometric measurements. Our results have shown a statistically conclusive difference of biometric indexes (DTI, CI, MI and RBI) between different age groups. It is difficult to compare these results with others reported in literature because of the lack of studies on biometric indexes in donkeys.

The body profile index was 0.97. This value allows to classify the animal population as a longlinear breed (BPI < 1), meaning that its total length is substantially equal to its height. These results corroborate with those reported in donkey by Daloum *et al.* (2015) and, Folch and Jordana (1997) but seem to disagree with the results obtained in the Arabian horses Barbe and Barbe (Chabchoub *et al.*, 2004). The dactylo thoracic index shows a relationship between the mass of individuals and the members that sustain it. The DTI of the donkeys studied is defined as animals among to the category of hypermetric donkeys (DTI < 1). These results are comparable to those obtained in Spain (Folch and Jordana, 1997), Tchad (Daloum *et al.*, 2015) and Cameroun (Defeu *et al.*, 2015).

The compact index explains that the body mass of the studied donkey is greater than its size, i.e. the animal does not support its weight. In this study, the donkeys sampled have massive overload (1.34 ± 0.2 kg/cm), this corroborate with those reported by Daloum *et al.* (2015) and Defeu *et al.* (2015). The MI confirmed that donkeys studied in Kabylie area have a body overload (MI > 1). Similar characteristic is found in domestic donkeys of the Sahelian region, Tchad (Daloum *et al.*, 2015). Moreover, there was a high positive correlation between CI and MI ($r = 0.816$, $P < 0.001$).

The pectoral height index (PHI) indicated that donkeys are short-legged. The front-to-back height (FBH) suggests that donkeys have a posterior region higher than the anterior region. In this current investigation, it is revealed that donkeys are short-legged (PHI > 0.56) with a straight back (FBH ≤ 1). Our finding does not corroborate with those reported by Folch and Jordana (1997), where the height at the withers and the height at the rump are equal in the Catalan race, in other terms well balanced. The massive index indicates whether the animal supports its weight.

According to the relative body index (RBI), the obtained results (0.93 ± 0.06) confirm that donkeys are elongated in the study area (RBI > 0.90). Recently, Defeu *et al.* (2015) recorded a high RBI in the domestic donkeys of Northwest Cameroon. However, a low of RBI values has been found in different Algerian horse breed (Guedaoura *et al.*, 2011). A negative correlation was found between CI and RBI (r = -0.71, P < 0.001), i.e. the weight varies with the body length. A considerable genetic variability was observed between our results and cited studies previously. This difference could be attributed to the geographical origin of donkey genetics, which adapts to the warm and humid environment that affects the growth and development of the body.

CONCLUSION

This is a first report on the phenotypic characterization in donkeys in Kabylie area (Algeria) based on corporal measurements. Our comparative analysis of morphometric parameters; such as back length, body length, neck length; suggests that donkeys of Kabylie area are typically invariant among breeds and it has not been changed through the periods. These obtained results constitute a baseline data for a deeper understanding of the genetic diversity in equines and for using in genetic improvement. However, the molecular characterization would better identify donkey breeds in Algeria.

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RESEARCH ARTICLE

PRINCIPAL COMPONENT ANALYSIS (PCA) IN THE MORPHOSTRUCTURE OF MALE KABYLIE DONKEYS (*EQUUS ASINUS*), ALGERIA

Widya Pintaka Bayu Putra[1], Sofiane Aissanou[2], Nadir Boudjelal Dergal[3], Abdelhanine Ayad[2*]

[1]Research Center for Biotechnology, Indonesian Institute of Science, Bogor, Indonesia
[2]Department of Biological Sciences of the Environment, Faculty of Nature and Life Sciences,
University of Bejaia, Bejaia, Algeria
[3]Department of Biotechnology, Faculty of Nature and Life Sciences, University of Oran, Oran, Algeria

***Corresponding author:**
Prof. Dr. Abdelhanine Ayad
Faculty of Nature and Life Sciences,
University of Bejaia,
Bejaia/Algeria
Phone:+213772722595
ORCID: 0000-0002-9325-7889
Email:
abdelhanine.ayad@univ-bejaia.dz

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ABSTRACT

Principal component analysis (PCA) is important to describe the total phenotypic variance in livestock. Hence, the selection of livestock can be performed with the main component (PC1) of some phenotypic traits. This research was carried out to obtain the principal component (PC) in the morphostucture of male donkeys (*Equus Asinus*) at the Kabylie area of Algeria. Two different locations of Bejaia and Tizi-Ouzou province were chosen randomly. A total of 121 male donkeys (jackass) with an average age of 10.75±4.58 years were used in this study. Data of animals' morphostructure consisted of 17 body measurements and 6 body indices. The PCA analysis in body measurements and body indexes were showed 5PC's and 3PC's, respectively that explain total variance in animals' morphostructure. Besides, 5PC's of body measurements and 3PC's of body indexes had a total variance proportion of about 66.79% and 81.12%, respectively. According to Bartlett's test, both PCA's in this study were accurate (with significance of less than 0.05). According to Kaiser-Meyer-Olkin (KMO), the PCA of body indices was not accurate (KMO = 0.35). It was concluded that body measurements of chest depth, chest width, front-leg length and back length as the PC1 explain 32.16% of the total variance in animals' morphostructure. In conclusion, the PCA of body measurements in this study is accurate with a high KMO value and significance of Bartlett's test. The results of this study can be used as the basic information to develop a selection program for Algerian donkeys in the future.

Keywords: Bartlett's test, donkeys, KMO, morphostructure, PCA

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INTRODUCTION

Donkey (*Equus asinus*) is the one of livestock animals widely used for draught and meat-milk production (Pearson et al., 1999; Paul and Malcolm, 1999). Commonly, donkeys were kept as draught animals. A donkey of 250 kg of live weight has a capacity to carry a maximum load of 80 kg, or more than 25% of the animal's body weight (Orhan et al., 2012). In China, donkey meat has been found to have high nutritional value, and the Colla Corii Asini (donkey-hide glue) is popularly used as a traditional Chinese medicine (Di et al., 2017). Marchis et al. (2018) reported that donkeys had colostrum quality (g/100 mL) of fat, protein and lactose about 3.77, 2.36 and 2.35, respectively. According to FAO, the total number of donkeys in the world in the year 2006 was about 41 million heads, spread in Africa (26.9%), Pacific Asia (37.6%), Europe/Caucasus (3.7%), Latin America/ Caribbean (19.9%), Middle East (11.8%) and 0.1% in North America (Vlaeva et al., 2017). In the year 1996, about 1.70% of the African donkey's population was recorded in Algeria (Paul and Malcolm, 1999). Hence, the genetic improvement in Algerian donkeys is important to increase their performance because of the meat production in the future. The selection of donkeys is important to obtain the desirable animal's morphostructure type as the draught animal. Morphostructure of livestock can be evaluated through body measurements and body indexes with principal component analysis (PCA). The PCA has been used to obtain the main variables (components) with the highest proportion of phenotypic variance.

The PCA of body measurements and body indexes have been used to explain morphostructure in horse (Takaendengan et al., 2011), goat (Putra and Ilham, 2019) and cattle (Putra et al., 2020). Otherwise, the PCA has been performed for morphostructure evaluation in many livestock, including donkey. Recently, the study of PCA in the donkey's morphostructure has been limited. Previous studies included the work with PCA in the morphostructure of donkeys in Amiata (Italy), Tlemcen (Algeria) and Banat (Serbia) regions (Sargentini et al., 2018; Madani et al., 2018; Stanisic et al., 2020). Despite this, PCA has been used to explain the morphostructure of mules (donkey × horse) in Colombia (Mosquera et al., 2020). Furthermore, Valle et al. (2017) used PCA in the neck measurements to evaluate the fatty neck score of Italian donkeys.

In donkey, the PCA analysis of body measurements and body indices are important to get the main components for describing animal morphostructure. Hence, the selection to improve the body performance of donkey can be performed with the main (first) component of body measurements and body indices. Moreover, PCA has a high correlation with body weight of goat (Putra and Ilham, 2019) and cattle (Putra et al., 2020). However, the study of PCA to describe morphostructure in Kabylie donkey of Algeria has not been reported. This research was carried out to obtain the main component in the body measurements and body indexes of Kabylie donkeys of Algeria. The results of this study can be used as the basic information for developing a selection program for Kabylie donkeys in the future.

MATERIAL AND METHODS

Research site

The study was carried out from February to June 2018 in the Kabylie area, Algeria. Two different locations of Bejaia (36° 43' N, 5° 04' E) and Tizi-Ouzou (36° 42' N, 4° 2' E) province were chosen randomly. The topography of the Kabylie area is mostly predominated by mountains. The vegetation is mainly composed of several species of trees and natural or cultivated herbs. They constitute a part of the climate in the Mediterranean region. The maximum summer temperature is ranged from 30.3 to 36.3 °C (July) and the minimum winter temperature is ranged from 6.6 to 6.7 °C (February).

Animal and measurements

The morphostucture data were collected from 121 male donkeys (jackass) with an average age of 10.75±4.58 years. The animals' morphostructure data consisted of body measurements and body indexes. Seventeen body measurements of head length (HL), ear length (EL), neck length (NL), thoracic circumference (TC), chest depth (CD), chest width (CW), front leg length (FLL), cannon length (CL), cannon circumference (CC), cannon height (CH), withers height (WH), back length (BaL), back height (BH), hips-width (HW), body length (BoL), height at the rump (HR) and umbilical circumference (UC) were performed in this study. Thus, six body indices of body profile index (BPI=WH/BoL), pectoral height index (PHI=CD/FLL), dactyl thoracic index (DTI=CC/TC), front-back height (FBH=WH/HR), massive index (MI=TC/WH) and relative body index (RBI=BoL/TC) were calculated in this study. According to the BPI value, donkeys had three categories of long conformation (BPI > 0.90), medium conformation (0.86 < BPI < 0.88) and small conformation (BPI < 0.85) types (Mcmanus et al. 2008). According to the PHI value, donkeys had two categories of leggy (0.50 < PHI < 0.55) and leg-shorted (PHI > 0.56) types (Marcenac et al., 1980). According to the FBH and MI values, donkeys had two categories of straight back or no overload (FBH/MI ≤ 1) and the anterior region is higher than the posterior or overload (FBH/MI > 1) types (Marcenac et al., 1980; Mcmanus et al., 2008). According to the RBI value, donkeys had three categories of longilinear (RBI ≥ 0.90), mediolinear (0.84 < RBI < 0.89) and brevilinear (RBI < 0.83) types (Nick et al., 2006). The scheme of body measurements in donkey was presented in Figure 1.

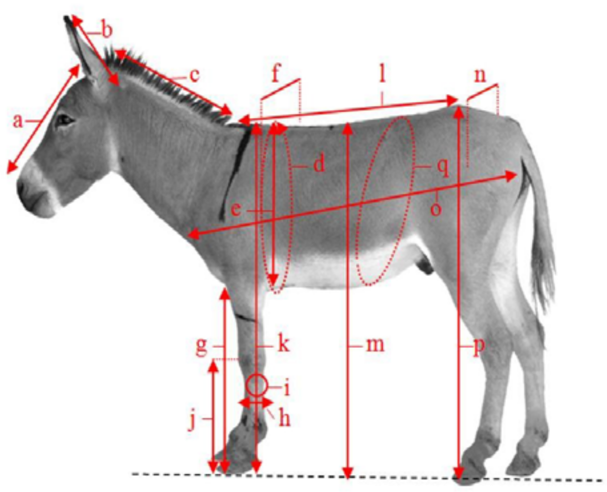

Figure 1 Scheme of the body measurements in donkey. a: head length (HL). b: ear length (EL). c: neck length (NL). d: thoracic circumference (TC). e: chest depth (CD). f: chest width (CW). g: front leg length (FLL). h: cannon length (CL). i: cannon circumference (CC). j: cannon height (CH). k: withers height (WH). l: back length (BaL). m: back height (BH). n: hips-width (HW). o: body length (BoL). p: height at the rump (HR). q: umbilical circumference (UC)

Data analysis

The descriptive statistics (mean, standard deviation, coefficient of variation, minimum and maximum values) and Pearson's coefficient of correlation (r) were calculated using SPSS 16.0 computer program. Thus, the principal component analysis (PCA) was performed to identify the main components to explain the proportion of the variance in animals' morphostructure. Two statistic values of Kaiser-Meyer-Olkin (KMO) and Bartlett's of the sphericity were used as the validity test (Vohra et al., 2017). The KMO is a statistic value that indicates the proportion of variance in the variables that might be caused by some factors (components). High KMO value (more than 0.50) indicated that the PCA was accurate. Bartlett's test is the hypothesis that a correlation matrix is an identity matrix, which would indicate that the variables are unrelated and, therefore, unsuitable for structure detection. Low Bartlett's test values (lower than 0.01) of the significance level indicate that the PCA is accurate. Johnson and Wichern (2002) stated that the first component (PC1) explained the largest percentage of the total phenotypic variance. In addition, the extraction communality value in PCA explains the variance in each variable accounted by the factors in the factor solution. Low values indicate variables that do not fit well with the factor solution. Hence, the main principal component was selected based on the rotated component matrix value of more than 0.50. The mathematical model in PCA, according to Karacaoren and Kadarmideen (2008), follows:

$$Y_{ij} = \sum_{k=1}^{q} a_{ik} c_{kj} + e_{ij}$$

Where Y_{ij} is the value of the i^{th} observation on the j^{th} measure; q is the number of common factor; a_{ik} is the value of the ith observation on the k^{th} common factor (factor loadings); c_{kj} is the regression coefficient of the k^{th} common factor for predicting the j^{th} measure and e_{ij} is the value of the i^{th} observation on the j^{th} communality value.

RESULTS

Animals' morphostructure

The descriptive statistics in morphostructure of Kabylie jackass was presented in Table 1. The coefficient of variation (CV) value in NL measurement was 10.22 and included a moderate category (10%<CV<20%), as presented in Table 2. Meanwhile, the CV value in other traits included a low category (CV<10%). Meanwhile, the CV value in the body indices included a low category. The high category of Pearson's coefficient of correlation (r) values (0.61<r<0.80) among body measurements in animals' study was shown in WH-BH (0.79), WH-HR (0.72) and BH-HR (0.73), as presented in Table 3. Thus, the high category of r value among body indices was shown in MI-RBI (-0.70).

Table 1 Descriptive statistic in the morphostructure of Kabylie jackass

Morphostructure	Mean	SD	CV (%)	Minimum	Maximum
Body measurements (cm)					
Head length	48.55	3.14	6.47	40.00	55.00
Ear length	24.44	1.81	7.41	20.00	28.00
Neck length	46.09	4.71	10.22	33.00	56.00
Thoracic circumference	118.56	7.24	6.11	100.00	134.00
Chest depth	49.22	1.77	3.60	45.00	53.00
Chest width	25.64	1.82	7.10	21.00	29.00
Front leg length	75.28	3.27	4.34	66.00	82.00
Cannon length	21.07	1.70	8.07	14.00	25.00
Cannon circumference	14.72	1.10	7.47	13.00	23.00
Cannon height	32.00	2.95	9.22	17.50	38.00
Withers height	107.07	5.26	4.91	94.00	118.00
Back length	63.26	2.28	3.60	60.00	72.00
Back height	107.38	5.11	4.76	92.00	120.00
Hips width	32.39	1.62	5.00	29.00	40.00
Body length	110.28	5.60	5.08	96.00	130.00
Height at the rump	109.69	4.68	4.27	97.00	118.00
Umbilical circumference	141.49	9.81	6.93	108.00	161.00
Body indexes					
Body profile index	0.97	0.05	5.15	0.86	1.09
Pectoral height index	0.66	0.03	4.55	0.58	0.76
Dactyl thoracic index	0.18	0.01	5.56	0.11	0.21
Front-back height	0.98	0.04	4.08	0.88	1.10
Massive index	1.11	0.06	5.41	0.91	1.25
Relative body index	0.93	0.07	7.53	0.77	1.14

SD: standard deviation; CV: coefficient of variation

Table 2 Pearson's coefficient of correlation among body measurements in Kabylie jackass

Body measurements	HL	EL	NL	TC	CD	CW	FLL	CL	CC	CH	WH	BaL	BH	HW	BoL	HR	UC
Head length (HL)	-	0.19	-0.42	0.49	0.07	-0.09	-0.09	0.34	0.08	0.23	0.38	0.33	0.27	0.11	0.16	0.28	0.23
Ear length (EL)		-	0.07	0.29	0.10	0.10	0.22	0.33	0.05	0.36	0.39	0.23	0.35	0.30	0.46	0.38	0.28
Neck length (NL)			-	-0.11	-0.07	0.09	0.19	-0.08	0.11	-0.04	-0.11	-0.12	0.04	0.15	0.12	-0.05	-0.12
Thoracic circumference (TC)				-	0.26	0.37	0.16	0.35	0.18	0.58	0.50	0.39	0.28	0.24	0.26	0.44	0.09
Chest depth (CD)					-	0.37	0.32	0.21	0.05	0.30	0.39	0.34	0.31	0.25	0.13	0.26	0.11
Chest width (CW)						-	0.53	0.15	0.05	0.31	0.15	0.32	0.03	0.18	-0.05	0.04	-0.22
Front leg length (FLL)							-	0.29	0.09	0.37	0.35	0.36	0.35	0.37	0.14	0.34	-0.01
Cannon length (CL)								-	-0.25	0.46	0.37	0.25	0.30	0.17	0.31	0.39	0.13
Cannon circumference (CC)									-	0.13	0.07	0.05	-0.02	0.11	-0.01	0.10	0.11
Cannon height (CH)										-	0.55	0.39	0.41	0.33	0.29	0.56	0.16
Withers height (WH)											-	0.47	0.79	0.40	0.47	0.72	0.30
Back length (BaL)												-	0.44	0.31	0.18	0.37	0.17
Back height (BH)													-	0.39	0.40	0.73	0.41
Hips width (HW)														-	0.20	0.38	0.18
Body length (BoL)															-	0.47	0.25
Height at the rump (HR)																-	0.45
Umbilical circumference (UC)																	-

Table 3 Pearson's coefficient of correlation among body indexes of Kabylie jackass

Body indexes	BPI	PHI	DTI	FBH	MI	RBI
Body profile index (BPI)	-	-0.02	-0.11	0.43	-0.22	-0.54
Pectoral height index (PHI)		-	-0.13	0.06	0.11	-0.08
Dactyl thoracic index (DTI)			-	0.00	-0.47	0.48
Front-back height (FBH)				-	-0.34	-0.03
Massive index (MI)					-	-0.70
Relative body index (RBI)						-

Principal component analysis

A total of five principal components (5 PC's) obtained from 17 body measurements in Kabylie jackass explain 66.79% of the total variance in animal morphostructure (Table 4). Moreover, KMO and Bartlett's test in PCA of body measurements were accurate. Despite this, body measurement of HW had the lowest extraction communality value (0.44) and wasn't included as a principal component. The first principal component (PC1) of body measurements included CD, CW, FLL and BL with explaining 32.16% of the total variance in animals' morphostructure. Thus, a total of three principal components (3PC's) obtained from 6 body indexes in Kabylie jackass explain 81.15% of the total variance in animals' morphostructure (Table 5). Unfortunately, the PCA in body indexes was not accurate because of the low KMO value (0.35). The PC1 of body indexes included DTI, MI and RBI with explaining 36.64% of the total variance in animals' morphostructure. The component plot in body measurement and body indexes of Kabylie jackass were presented in Figure 2.

Table 4 Rotated component matrix scores, extraction communality values, initial eigenvalues, KMO and Bartlett's test for the body measurements of Kabylie jackass

Body measurement	PC1	PC2	PC3	PC4	PC5	EC
Head length	-0.03	0.18	0.32	0.77*	0.10	0.74
Ear length	0.03	0.27	0.69*	-0.09	0.04	0.55
Neck length	0.03	-0.06	0.20	-0.80*	0.16	0.70
Thoracic circumference	0.38	-0.01	0.59*	0.44	0.29	0.77
Chest depth	0.66*	0.25	-0.07	0.12	-0.06	0.53
Chest width	0.80*	-0.33	0.14	-0.07	0.06	0.77
Front leg length	0.73*	0.14	0.18	-0.31	-0.03	0.67
Cannon length	0.24	0.07	0.62*	0.22	-0.45	0.69
Cannon circumference	0.06	0.07	0.01	-0.01	0.92*	0.85
Cannon height	0.46	0.18	0.58*	0.17	0.12	0.63
Withers height	0.40	0.63*	0.40	0.21	-0.01	0.76

Body measurement	PC1	PC2	PC3	PC4	PC5	EC
Back length	0.58*	0.32	0.13	0.30	0.04	0.54
Back height	0.31	0.78*	0.27	0.04	-0.10	0.79
Hips width	0.40	0.41	0.22	-0.19	0.17	0.44
Body length	-0.07	0.39	0.65*	-0.12	-0.08	0.60
Height at the rump	0.25	0.68*	0.44	0.11	0.03	0.74
Umbilical circumference	-0.16	0.72*	0.07	0.15	0.10	0.58
Total initial eigenvalues	5.47	1.99	1.57	1.25	1.08	-
Variance (%)	32.16	11.68	9.22	7.35	6.37	-
Cumulative (%)	32.16	43.84	53.06	60.41	66.79	-
Kaiser-Meiyer-Olkin (KMO)			0.79			
Bartlett's test			**			

PC: principal component; EC: extraction communality; *main component. **(P<0.01)

Table 5 Rotated component matrix scores, extraction communality values and initial eigenvalues in the body indices of Kabylie jackass

Body indexes	PC1	PC2	PC3	EC
Body profile index	-0.25	0.88*	-0.15	0.86
Pectoral height index	-0.10	0.03	0.97*	0.94
Dactyl thoracic index	0.73*	-0.04	-0.17	0.56
Front-back height	0.17	0.80*	0.18	0.69
Massive index	-0.85*	-0.40	0.04	0.89
Relative body index	0.91*	-0.30	0.08	0.93
Total initial eigenvalues	2.20	1.66	1.01	-
Variance (%)	36.64	27.65	16.83	-
Cumulative (%)	36.64	64.29	81.12	-
Kaiser-Meiyer-Olkin (KMO)		0.35		
Bartlett's test		**		

PC: principal component; EC: extraction communality; *main component; **(P<0.01)

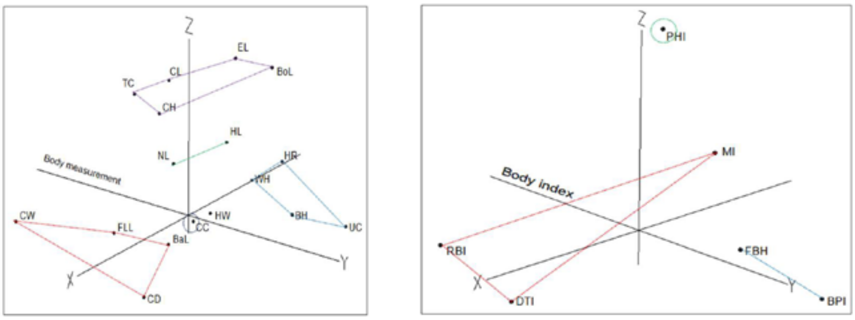

Figure 2 Component plot of body measurements and body indices of Kabylie jackass

DISCUSSION AND CONCLUSION

Sargentini et al. (2018) obtained EL (29.90±2.40 cm), NL (62.00±4.60 cm), TC (144.80±6.60 cm), CW (28.80±2.40 cm), FLL (71.80±2.40 cm), CC (16.80±1.10 cm), WH (127.80±3.50 cm), BL (55.30±5.50 cm), HW (40.50±3.10 cm), BoL (137.30±7.80 cm) and HR (131.30±3.70 cm) in adult Amiata female donkeys (Janets) reared at a coastal area. Kostukova et al. (2012) obtained TC (127.50 cm), WH (102.44 cm) and HR (105.32 cm) in Czech donkeys. Madani et al. (2018) obtained HL (53.15±3.22 cm), EL (30.05±2.26 cm), CW (27.20±2.60 cm), CC (17.77±1.81 cm) and WH (116.45±6.28 cm) in jackass at Tlemcen of Algeria that are higher than in Kabylie jackass. In our investigation, EL and HL of Kabylie donkeys were higher than those reported in the recent studies (John et al., 2017; Mustefa et al., 2020). Quaresma et al. (2019) obtained TC (143.80±23.10 cm), WH (127.40±14.70 cm) and BoL (131.40±25.30 cm) in Miranda donkeys that are higher than in Kabylie jackass in this study. Thus, Nininahazwe et al. (2017) obtained EL (25.70±2.21 cm), NL (30.90±4.55 cm), WH (98.90±4.61 cm), TC (106.90±6.89 cm) and BoL (103.10±7.66 cm) in the West African donkeys. Also, male mules had TC, WH and BoL about 152.58±1.45 cm; 135.28±1.05 cm and 135.14±0.99 cm, respectively (Mosquera et al., 2020) and are higher than in Kabylie jackass. According to the previous study, FLL in Kabylie jackass was higher than in Italian jennets. The body measurement of TC in Czech donkeys was higher than in Kabylie jackass. Thus, body measurements of NL, TC, BoL and WH in Kabylie jackass were higher than in the West African jackass.

According to BPI, PHI, FBH, MI and RBI values, most of Kabyle jackass included long conformation, longilinear and leggy types of the animal with overload status. Sargentini et al. (2018) obtained DTI (0.12±0.05), MI (1.13±0.05) and RBI (0.95±0.03) in Amiata jennets. Mosquera et al. (2020) obtained BPI (1.00±0.07) and RBI (0.89±0.08) in male mules that included long conformation and longilinear types of animals. According to the previous study, Amiata jennets and Kabylie jackass had a similar type of animal. Mosquera et al. (2020) obtained a moderate r value (0.40<r<0.60) in WH-BL (0.65) and WH-TC (0.64) in mules, higher than in Kabylie jackass. Hence, the r-value of BPI-RBI in Kabylie jackass in this study was (-0.54) and close to mules (-0.59), as reported by Mosquera et al. (2020). The difference in these results compared to previous studies can be caused by variation in age, sex, breeds (genetic), management and geographical area.

Madani et al. (2018) obtained 2 PC's from 11 body measurements in donkeys reared at Tlemcen (Algeria), which explain 62.61% of the total variance in animal morphostructure. Takaendengan et al. (2011) obtained 3 PC's from 12 body measurements in Minahasa stallions of Indonesia that explain 80.20% of the total variance in animal morphostructure. Paksoy and Ünal (2019) obtained 4 PC's from 14 body measurements of Thoroughbred horses that explains 87.28% of the total variance in animal morphostructure. In addition, Hosseini et al. (2016) obtained 3 PC's from 23 body measurements in Iranian horses that explains 76.00% of the total variance in animal morphostructure.

The body measurements included in PC 1 (CD, CW, FLL and BL) can be used as a selection criterion to improve the performance traits of Kabylie jackass. Besides, body measurements of CD, CW and BL included in PC1 in Thoroughbred horses can be affected in race performance traits (Paksoy and Ünal, 2019).

The PCA in body measurements and body indexes of Kabylie jackass revealed 5 PC's (66.79% of total variance) and 3 PC's (81.12% of total variance), respectively. The PCA of the body measurements in this study is accurate with high KMO value and significance of Bartlett's test. Hence, the selection to improve the performance of Kabylie jackass can be performed effectively with four body measurements of CD, CW, FLL and BL.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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ANALIZA GLAVNIH KOMPONENTI (PCA) U MORFOSTRUKTURI MUŽJAKA KABYLIE MAGARACA (*EQUUS ASINUS*) IZ ALŽIRA

SAŽETAK

Analiza glavnih komponenti (PCA) je značajna za opis ukupne fenotipske varijanse stoke. Stoga se odabir stoke može vršiti upotrebom glavne komponente (PC1) pojedinih fenotipskih karakteristika. Cilj našeg istraživanja jeste odrediti glavnu komponentu (PC) u morfostrukturi mužjaka magaraca (*Equus asinus*) u području Kabylie u Alžiru. U provincijama Bejaia i Tizi-Ouzou su metodom slučajnog uzorka odabrane dvije lokacije. U istraživanju su korištena 121 mužjaka magarca prosječne starosti 10.75±4.58 godina. Podaci o morfostrukturi životinja su obuhvatili 17 tjelesnih mjera i 6 tjelesnih indeksa. PCA analiza je pokazala 5 PC tjelesnih mjera i 3 PC tjelesnih indeksa koji objašnjavaju ukupnu varijansu u morfostrukturi životinja. Osim toga, 5 PC tjelesnih mjera i 3 PC tjelesna indeksa su pokazali udjele u ukupnoj varijansi od 66.79% i 81.12%. Prema Bartlettovom testu, obje PCA u našem istraživanju su bile precizne sa signifikantnošću manjom od 0.05. Prema Kaiser-Meyer-Olkinu (KMO), PCA tjelesnih indeksa nije bila precizna (KMO = 0.35). Zaključeno je da tjelesne mjere dubine grudnog koša, širine grudnog koša, dužine prednje noge i dužine leđa kao PC1 objašnjavaju 32.16% ukupne varijanse životinjske morfostrukture. U zaključku, PCA tjelesnih mjera u našem istraživanju je precizna sa visokom KMO vrijednošću i signifikantnošću dokazanom Bartlettovim testom. Rezultati našeg istraživanja se mogu koristiti kao osnovne informacije za razvoj programa selekcije za alžirske magarce u budućnosti.

Ključne riječi: Bartlettov test, magarci, KMO, morfostruktura, PCA genes

Communication

Investigation of Cerebellar Abiotrophy (CA), Lavender Foal Syndrome (LFS), and Severe Combined Immunodeficiency (SCID) Variants in a Cohort of Three MENA Region Horse Breeds

Abdelhanine Ayad [1,†], Saria Almarzook [2,†], Omar Besseboua [3], Sofiane Aissanou [1], Katarzyna Piórkowska [4], Adrianna D. Musiał [4], Monika Stefaniuk-Szmukier [4] and Katarzyna Ropka-Molik [4,*]

1 Department of Environment and Biological Sciences, Faculty of Nature and Life Sciences, University of Bejaia, Bejaia 06000, Algeria; abdelhanine.ayad@univ-bejaia.dz (A.A.); sofiane.aissanou@univ-bejaia.dz (S.A.)
2 Faculty of Applied Sciences, University of Applied Sciences Europe, 10963 Berlin, Germany; saria.almarzook@ue-germany.de
3 Department of Agronomic and Biotechnological Sciences, Faculty of Nature and Life Sciences, University H. Benbouali, Chlef 02000, Algeria; besseboua.omar@gmail.com
4 Department of Animal Molecular Biology, National Research Institute of Animal Production, Krakowska 1, 32-083 Balice, Poland; katarzyna.piorkowska@iz.edu.pl (K.P.); adrianna.musial@iz.edu.pl (A.D.M.); m.k.stefaniuk@gmail.com (M.S.-S.)
* Correspondence: katarzyna.ropka@iz.edu.pl
† These authors contributed equally to this work.

Abstract: Genetic disorders in horses are mostly fatal or usually cause significant economic losses for breeders and owners. Here we studied a total of 177 Arabian, Barb and Arab-Barb horses from the Middle East and North Africa (MENA) using Sanger Sequencing and PCR-ACRS (polymerase chain reaction—artificially created restriction site) approaches to examine the genetic disorders in the studied horse breeds. We identified the genetic variations related to Cerebellar Abiotrophy (CA), Severe Combined Immunodeficiency (SCID) occurrence, and the studied population was free of the mutant allele determined Lavender Foal Syndrome (LFS). Overall, presented data showed that 15 of the studied horses are carriers of two genetic disorders; the investigated horse population showed that five Arabian horses were heterozygous for the CA-associated SNP (rs397160943). The SCID-deletion TCTCA within *PRKDC* was detected in ten horses (nine Arabian horses and one Arab-Barb horse). This investigation shows the importance of testing these breeds for genetic disorders to avoid further spread of deleterious variants

Keywords: genetic disorders; Arabian horses; Barb horses; Arab-Barb horses; CA; LFS; SCID; molecular biology

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1. Introduction

The earliest evidence of horse domestication is described in Botai culture 5500 years before present in central Asian steppe. This process may have taken place in different places at different times [1] such as Anatolia, the Pontic Caspian Steppes, and Iberian Peninsula regions [2]. Recent genomic research indicates a cultural and geographic context in which the modern horse lineage emerged [3]. The spread of the horses from Mesopotamia to North-Eastern Arabia, later reaching South Arabia in what is known today as the MENA region (the Middle East and North Africa) is based on archaeological evidence of horse remains appearing on archaeological sites in the Levant during early historical periods [4].

Arabian (Figure 1), Barb (Figure 2) and Arab-Barb (Figure 3) horses are the original breeds in the MENA region. The three breeds have been bred geographically close for hundreds of years. The Arabian and Barb horses hold a prominent place in the MENA region and are recognised as the noblest and most influential breeds due to their contribution in the development of many other horse breeds worldwide. A recent analysis of Y chromosome diversity revealed that Barb horses cluster with barouqe, sorraia, and Spanish breeds [5]. Barbs were bred with Arabian horses resulting in the foundation of Arab-Barb horses, known for their high economic value due to their stamina and courage, maintenance and efficient feeding [6,7].

Figure 1. Arab horse in the desert (photo credit: Glenn Jacobs).

Figure 2. Arab-Barb horse (photo credit: Paula Da Silva).

Figure 3. Grey Barb stallion (photo credit: Paula Da Silva).

The Arabian, Barb and Arab-Barb horses are selectively bred to have the preferred traits that improve the phenotypic features or the aesthetic appeal and enhance athletic performance. Considering the origin of Arabian and Barb horses and their great influence on other breeds worldwide, this study aimed at screening a cohort of Arabian, Barb, Arab-Barb horses for the three genetic disorders as the Cerebellar Abiotrophy (CA), Lavender Foal Syndrome (LFS) and Severe Combined Immunodeficiency (SCID), in MENA horse populations. The obtained information can shed new light on the carriers of the studied diseases.

2. Materials and Methods

The Scientific Council approved the research of the Faculty of Nature and Life Sciences (Report of Faculty Scientific Council #05 dated 11 November 2020), University of Bejaia, Algeria). Concerning the ethical aspects, the experimental procedure was performed according to good veterinary practice under farm conditions.

2.1. Sampling

A total of 177 horses were chosen randomly and investigated for each of the three genetic disorders. Follicle hair samples from purebred Arabian (n = 80), Barb (n = 41) and Arab-Barb horses (n = 56) were used in this study. Thirty hairs 7 cm long were pulled out at the root of the neck or tail skin. The samples collected were preserved in an individually labelled paper envelope stored at room temperature until use for genetic disorders screen. Horses were checked clinically by a veterinarian. The neurologic examination evaluates (1) the cranial nerves, (2) the gait, or walk, (3) the neck and front legs, and (4) the torso, hind legs, anus, and tail. None of the horse showed signs of lowering reflexes of the head deterioration, constant pacing, seizures, a head turn or circling in one direction or other unusual head movements. None of the horses showed any signs of dysfunction within gait evaluation like circling, weakness or complete paralysis of any limbs, falling, stumbling, rolling, or loss of coordination. For evaluation of neck and front legs there were no evidence of pain, loss of muscle tone or cramp in the neck. None of the horses showed signs of loss of feeling or hypersensitivity to light touch or pinpricking, and loss of muscle mass. Therefore, the veterinarian claims all animals involved in study as healthy. Without suspicion of disease, there was no reason to order blood tests.

2.2. DNA Isolation and Genotyping

Genomic DNA was extracted from all samples using the Sherlock AX kit (A&A Biotechnology, Gdańsk, Poland) according to protocol. The DNA quality and concentration were measured with NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA).

2.2.1. Cerebellar Abiotrophy (CA)

The PCR-ACRS method was designed to identify the single nucleotide polymorphism that determined the CA occurrence (rs397160943, NC_009145.3:g.13122415C>T; ENSECAT 00000024892.2:c.284G>A, ENSECAP00000020698.1:p.Arg95His). The primers were designed using Primer3 Input (version 0.4.0) and *TOE1* gene (ENSECAG00000023204) reference (Table 1), and due to the modified primer sequence, the artificial restriction site for *HpyCH4*III (BioLabs, New England, Ipswich, MA, USA) was created. The PCR was obtained using 2xPhanta Max Master Mix (Vazyme, Polgen, Łódź, Poland), and after 16 h digestion, the products were obtained as follows: C allele-111, 22 bp and T allele-133bp.

Table 1. The primers used for detection of each polymorphism.

Gene	Primers	Method Used for Genotyping
TOE1	F: GGATCTCAACCCTCCTCTCC R: CGTGTGTCATGCTGCCAGGAaCC	PCR-ACRS
PRKDC	F: GGTAGCTTTGTGTTCCTGTTG R: TTCTCTCATTGCCAGAAGCA	Sanger sequencing
MYO5A	F: CAGAGCCTGAAGGAGGAGAA R: GTCAGCCGGGTGATCTCAT	Sanger sequencing

2.2.2. Severe Combined Immunodeficiency (SCID)

The five base pair deletion (TCTCA) in *PRKDC* gene was detected using Sanger sequencing. The primers spine mutation site was designed using Primer3 Input (version 0.4.0) based on ENSECAG00000020168 reference (Table 1). The PCR product was obtained using Ampli Tag 360 DNA Polymerase (Applied Biosystems, Thermo Fisher Scientific, Waltham, MA, USA) according to protocol, and purified using EPPIC (A&A Biotechnology, Gdańsk, Poland). Next sequencing was performed with BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, Thermo Fisher Scientific, Waltham, MA, USA), Big Dye Xterminator Purification Kit (Applied Biosystems, Thermo Fisher Scientific, Waltham, MA, USA) and 3500XL Genetic Analyser platform (Applied Biosystems, Thermo Fisher Scientific, Waltham, MA, USA).

2.2.3. Lavender Foal Syndrome (LFS)

The LFS genetic background mutation (*MYO5A* gene; g.138235715del) was detected using the Sanger sequencing method according to the same procedure described for SCID disorder. The primers used for *MYO5A* gene were designed based on ENSECAG00000021742 reference (Table 1).

For all three disorders, the negative and positive samples (heterozygote horses for each disorder) were included in every analysis.

3. Results and Discussion

Genetic disorders can have a dramatic impact on horse's breeding. These disorders are mainly responsible for foal losses and significant economic losses due to the treatments and maintenance costs during pregnancy and breeding. Cerebellar Abiotrophy (CA), is a progressive neurological disease characterised by the degeneration of cerebellar Purkinje cells, and it is likely caused by an intrinsic metabolic disorder [8], which affects many animal species [9]. It has been demonstrated that CA is recognised almost exclusively in the Arabian horse breed [10]. However, the probability of CA mutation occurrence is present in other Arabian-cross horse breeds [11]. CA clinical symptoms usually are developed dramatically between the age of 6 weeks and 4 months, represented mainly by ataxia, hypermetria, intention head tremors, and the absence of a menace response. CA has an autosomal recessive mode of inheritance. The genetic mutation responsible for the disorder would be a single nucleotide polymorphism (SNP) located in exon 4 of *TOE1* gene (Target Of Early Growth Response 1) on equine chromosome 2, resulting in the incorporation of arginine instead of histidine at this position [12]. Therefore, crossing two carriers results in 25% affected foals in the population [12]. Reveal the number of horses in each genotype group and the percentage of each genotype/allele, 2.8% of the investigated horses (5 Arabian horses) were heterozygous for the rs397160943 SNP in *TOE1* gene at ECA2 with allele frequency of 0.1%, and no homozygous-affected horses for the mutation were identified (Table 2).

Table 2. The genotypes and alleles frequency distribution of rs397160943 SNP in *TOE1* gene related with CA (Cerebellar Abiotrophy) occurrence.

	Genotypes (N/%)			Alleles	
Breed	**CC**	**CT**	**TT**	**C**	**T**
Barb	41 100%	-	-	1	
Arab Barb	56 100%	-	-	1	
Arabian	75 93.7%	5 6.3%	-	0.94	0.06
Total	172 97.2%	5 2.8%	-	0.99	0.01

P The data presents the numbers of horses in each genotype group and the percentage of each genotype/allele; N—number of horses detected; '-'—the lack of detected animals with given genotype; C—the reference allele; T—allele related with Cerebellar Abiotrophy disorder.

The CA is commonly recognised in Arabian horses, and studies showed that other breeds are at risk for the disease if Arabian founders were used as breeding stock [11]. The CA appears to be transmitted with a low frequency to other breeds descended from Arabian founders like the six heterozygous horses reported by Brault et al. [12], comprising 2 Trakehners, one Welsh pony, and 3 Bashkir Curly horses. Recent studies investigated the carrier's frequency in 808 purebred Arabian horses, and its results confirmed that CA-related mutations tend to show a high allele frequency in Arabian horses [13].

Lavender Foal Syndrome (LFS) is a rare autosomal recessive lethal genetic coat colour-associated disorder combined with severe neurological symptoms. Arabian horse with heterozygous genes represents a carrier case, while that the homozygous genes individuals are mostly dying within a few hours or days [14]. Regarding the Lavender Foal Syndrome (LFS), our investigated horses were clear of this lethal disorder. Results recently obtained by Bugno-Poniewierska et al. [13] confirmed the absence of LFS in the studied group of purebred Arabian horses in Poland, indicating that the lethal allele was not introduced yet in some populations. To date, reports from South Africa, Croatia, and Egypt reported the incidence of LFS carriers [15–17]. Therefore, the occurrence of mutated alleles in horse population from the MENA regions is not inconceivable.

Severe Combined Immunodeficiency (SCID) is an autosomal recessive genetic disease, first described in two Arabian foals, full siblings 70 years ago [18]. Later studies confirmed that Equine SCID primarily affects Arabian horses and their crossbreeds [19]. SCID is characterized mainly by the inability of foals to produce antigen-specific immune responses due to a lack of functional B and T lymphocytes. The genetic basis of Equine SCID is the deletion of 5 base pairs (TCTCA) in the DNA-protein kinase catalytic subunit (DNA-PKcs) located in the short arm of chromosome 9 (ECA9p12) [20,21]. The obtained results confirmed the presence of the five nucleotides deletion within *PRKDC* associated with

SCID disorder in the studied horses. The data shows that 5.7% of the studied horses (10 horses) are SCID carriers with a total SCID-associated allele frequency of 0.03. The ten carriers were nine Arabian and one Arab-Barb horses (Table 3).

Table 3. The genotypes and alleles distribution of deletion within *PRKDC* gene related with SCID occurrence in different horse breeds.

	Genotypes (N/%)			Alleles	
Breed	**WT/WT**	**WT/SCID**	**SCID/SCID**	**WT**	**SCID**
Barb	41 100%	-	-	1	
Arab Barb	55 98.2%	1 1.3%	-	0.99	0.01
Arabian	71 88.7%	9 11.3%	-	0.94	0.06
Total	167 94.3%	10 5.7%	-	0.97	0.03

The data presents the numbers of horses in each genotype group and the percentage of each genotype/allele; N—number of horses detected; '-'—the lack of detected animals with given genotype; WT—wild allele (reference); SCID—the deletion of TCTCA motif within *PRKDC*.

In other investigated horse populations, researchers determined the frequency of SCID among 21 Arabian and Arabian crossbred horses in Morocco, where SCID carriers were representing 14 (7%) Arabian horses and 6 (4%) Arab-Barb horses [22]. While in the genetic screening of Arabian horses in Turkey, no SCID carrier cases were identified [23]. Moreover, a recent report confirmed introducing SCID-associated allele in three heterozygous Polish Arabian horses with a low frequency that did not exceed the threshold of 0.004 [13].

4. Conclusions

This study allowed us to identify the alleles responsible for both equine genetic diseases CA and SCID among Arabian horses in the MENA region. Investigated genetic disorders are autosomal, recessive, hereditary diseases occurring in the horse populations, and according to literature, a carrier of the disease-causing allele shows no clinical symptoms. With the arising of revolutionary research technologies and investigating methods, breeders became more aware of the best procedures to enhance their production and minimise the risks of genetic disorders that might negatively affect their profits. Therefore, our study strongly supports the need for genetic testing in Arabian and Arabian-crossed horses in the MENA region.

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Résumé

La présente étude est la première à décrire le statut reproductif chez l'âne commun d'Algérie (*Equus asinus*) et à évaluer les variations saisonnières des indices biométrique et histophotométrique des testicules ainsi que les concentrations de Testostérone et des paramètres biochimiques du plasma sanguin chez les ânes locaux algériens. Nos résultats suggèrent que l'activité sexuelle a lieu durant la période des jours courts correspondante en saisons d'hiver et d'automne. En outre, nos résultats d'histophotométrie et de biométrie testiculaire ainsi que les niveaux de Cholestérol et de Triglycéride sont significativement corrélés avec les données photopériodiques des différentes saisons, ce qui peut être une bonne indication de la performance de reproduction chez les ânes. De plus, l’effet du poids corporel et de l’âge sur les paramètres testiculaires, scrotal et épididymaire a bien été analysé.

Mots clés : *Equus asinus*, Reproduction du mâle, Saisonnalité, Biométrie testiculaire, Testostérone

Abstract

The present study is the first to describe the reproductive status of the Algerian common donkey (*Equus asinus*) and to evaluate seasonal variations in biometric and histophotometric indices of the testes as well as concentrations of testosterone and biochemical parameters of blood plasma in local Algerian donkeys. Our results suggest that sexual activity takes place during the corresponding short day period in winter and autumn seasons. Furthermore, our histophotometry and testicular biometry results as well as Cholesterol and Triglyceride levels are significantly correlated with photoperiodic data of different seasons, which may be a good indication of reproductive performance in donkeys. In addition, the effect of body weight and age on testicular, scrotal and epididymal parameters was well investigated.

Keywords: *Equus asinus*, Male reproduction, Seasonality, Testis biometry, Testosterone

الملخص

الدراسة الحالية هي الأولى التي تصف الحالة الإنجابية للحمار الجزائري المشترك ولتقييم التغيرات الموسمية لمؤشرات القياسات الحيوية والنسيجية للخصيتين بالإضافة إلى تركيزات التستوستيرون والإعدادات البيوكيميائية لبلازما الدم في حمير المنطقة المحلية الجزائرية. تشير نتائجنا إلى أن النشاط الجنسي يحدث خلال فترة الأيام القصيرة المقابلة في فصلي الشتاء والخريف. علاوة على ذلك ، ترتبط نتائج قياس بيانات الفترة الضوئية والقياسات الحيوية للخصيتين وكذلك مستويات الكوليسترول والدهون الثلاثية ارتباطًا كبيرًا ببيانات الفترة الضوئية للفصول الأربعة ، مما قد يكون مؤشرًا جيدًا على الأداء الإنجابي للحمير. بالإضافة إلى ذلك ، تم تحليل تأثير وزن الجسم والعمر على إعدادات الخصية والصفن بشكل جيد

الكلمات المفتاحية: تكاثر الذكور ، التموسم ، القياسات الحيوية للخصية ، التستوستيرون