الجمهورية الجزائرية الديمقراطية الشعبية PEOPLE'S DEMOCRATIC REPUBLIC OF ALGERIA Ministry of Higher Education and Scientific Research

University of Mohamed Khider, Biskra Faculty of Exact Sciences, Nature and Life Sciences

Department of Nature and Life Sciences

N° d'ordre:.....

Série :.....



Thesis submitted to the Department of Nature and Life Sciences in candidacy for the degree of Doctorate (L.M.D)

Option of the Doctorate: Valorisation and Conservation of Natural resources

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THEME

Influence of Some Blood Parameters on Reproduction Parameters in Ewes of Local Breed

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	Academic Year: 2021-2022		

Acknowledgement

I hereby would like to express sincere gratitude to my supervisor Dr. TITAOUINE Mohammed for his permanent, truthful and authentic support along this journey. I am absolutely indebted to him as well as to Pr. MEZIANE Toufik, my co-supervisor, for their insightful advice and interesting views.

Genuine and sincere gratitude goes for the respectable members of the jury who have accepted to give time to read and evaluate this humble work namely:

- Professor DEGHNOUCHE Kahramen: a professor at Biskra university.

- Doctor MAMMERI Adel: A. rank lecturer at M'sila university.

- Doctor ATTIR Badreddine: A. rank lecturer at Biskra university.

I would like to extend my gratitude to the staff at Mohamed Khider University Libraries, Teachers, Administration, especially Genetics, Biotechnology and Valorization of Bioressources Laboratory heads: Pr. LAIADI Ziane, Pr. MOUSSI Abdelhamid and Pr. MEHAOUA Mohamed Seghir and to all the Laboratory staff at (El Hadjeb campus) as well.

To all the private farms contributing to this research many thanks for your hospitality, the support you afforded meant a lot to us and added to this work.

I would like to thank all the staff at the I.T.D.A.S. -Ain Ben Noui- for opening their doors for me and the great amount of support and the assistance they provided, namely Mr MERDACI Samir; Mr HICHER Ferhat; Mme. TIBERMACINE Safia; Mme. SOUICI Soumia.

To Dr. MAZOUZI Mohamed and his wife Mme. YAAKOUB Fadjria, thank you for your efforts and help in the experimental part of this work.

To Dr. CHERFAOUI Mouloud, thank you for your help in the statistical analysis.

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ملخص

يحقق هذا البحث في وضعية تربية الأغنام في منطقة شبه صحر اوية، حيث يبحث عن مدى تأثير بعض العوامل في الدم على معاملات التكاثر عند النعاج و كذا تأثير الارتفاع عن سطح البحر على هذه الأخيرة وذلك بواسطة تقييم نسب بعض المعادن في النعاج من السلالات المحلية "سلالة اولاد جلال". يناقش البحث نسب التكاثر والخصوبة والالقاح في أربعة مواقع مختلفة الارتفاع آخذا بعين الاعتبار نسب بعض المعادن في الدم. الهدف الرئيسي هو إبراز أهمية بعض العوامل في تكاثر النعاج التي تنفذ الي معلمات الدم. وهكذا، نستكشف سلوك التكاثر في أربعة مواقع مختلفة الارتفاعات في بسكرة، الجزائر. وضع حيز التنفيذ عددا من تحاليل الدم وقياس عوامل التكاثر من أجل العثور على القيم التي تتوافق مع أفضل أداء تكاثري والقدرة على تحسينها بفعالية لضمان إنتاجية أفضل. أثبتت الدراسة وجود تأثير فعال للموقع بحيث لا تؤدي النعاج أحسن أداءً للتكاثر على ارتفاعات عالية. في حين أن النعاج التي لديها تركيزات عالية في بلازما الدم من الكالسيوم والفوسفور خلال فترة التكاثر أظهرت خصوبة أفضل في المقابل، كانت مستويات البلازما من المغنيسيوم والصوديوم والبوتاسيوم أعلى في النعاج غير المخصبة. كما أن تركيزات العوامل المعدنية التي تمت در استها أقل من المعابير الفيزيولوجية المثلي وذلك في المناطق الأربع المدروسة. وقد قمنا كذلك بدراسة تأثير نقطة الحالة البدنية على عوامل أخرى في الدم وبحثنا عن الرابط بين الأداء التكاثري فوجدنا أن كل من السكر، الكولسترول، الدهون الثلاثية، الألبومين، و هرمونى الغدة الدرقية، تتأثر بنقطة الحالة البدنية في حين أن الكرياتينين البروجسترون و البروتينات الكلية لم تكن ذات تأثير فعال على هذه الأخيرة. من ناحية أخرى، لم يكن لفرق تركيزات البلازما للبروتينات الكلية والكرياتينين واليوريا والبروجسترون تأثير فعال خلال فترة التزاوج في النعاج

الكلمات المفتاحية: معاملات التكاثر ; النعاج سلالة أولاد جلال ; الارتفاع عن سطح البحر ; عوامل الدم نقطة الحالة البدنية.

Abstract

The present research investigates the influence of some blood parameters on reproduction parameters, the effect of the altitude on reproduction parameters and mineral assessment in ewes of local breeds, taking "Ouled Djellal breed" as a sample. The research discusses the rates of fecundity, fertility, and prolificacy at four different sites in relation to some blood parameters. The main aim of the research is to reveal and to highlight the importance of some factors in ewe's reproduction which implement blood parameters. Thus, it explores the breeding conduct in four sites of different altitudes in Biskra, Algeria. The study puts into practice a number of blood analysis and measurement of reproduction parameters for the sake of finding the values corresponding to the best performances and being able to contribute in embettering them which remains a concern at both the national and the international levels. The study proved the presence of a significant site effect from which Ouled Djellal ewes do not convey their best reproduction performances at high altitudes. It was also found that fertilized ewes had high plasma concentrations of calcium and phosphorus during the mating period. In contrast, plasma levels of magnesium, sodium, and potassium were higher in nonfertilized ewes. Also, the concentrations of the mineral parameters studied are below the physiological norms in the four sites. We also studied the effect of the body condition score on other parameters in the blood and searched for the link between reproductive performance. We found that sugar, cholesterol, triglycerides, albumin, and thyroid hormones were affected by the body condition score, while creatinine, progesterone and total proteins did not have a significant influence on this latter. While total proteins, creatinine, urea and progesterone had no significant effect during the mating period.

Keywords: Reproduction parameters; Ouled Djellal breed ewes ; Altitude above sea level; Body condition score; blood parameters.

Résumé

La présente recherche étudie l'influence de quelques paramètres sanguins sur les paramètres de reproduction, l'effet de l'altitude sur les paramètres de reproduction et l'évaluation de taux des minéraux chez les brebis d'une race locale prenant comme population la «Ouled Djellal». Cette étude discute les taux de fécondité, de fertilité et de prolificité chez les ovins dans quatre sites à différentes altitudes et ce par rapport à certains paramètres sanguins. On a mis en pratique des analyses sanguines et des mesures des paramètres de reproduction dans le but de trouver les valeurs correspondantes aux meilleures performances afin d'être en mesure de les apporter sur place au niveau national. L'étude a prouvé la présence d'un effet significatif d'altitude à partir duquel les brebis Ouled Djellal n'expriment pas leurs meilleures performances de reproduction à haute altitude. On a également constaté que les brebis fécondables avaient des concentrations sanguines élevées en calcium et en phosphore dans leurs plasma pendant la période d'accouplement. En revanche, les taux plasmatiques en magnésium, sodium et potassium étaient plus élevés chez les brebis non fécondables. En outre, les concentrations sanguines en minéraux étudiés sont inférieures aux normes physiologiques et ce dans les quatre sites. Nous avons également étudié l'effet de la note d'état corporel sur d'autres paramètres dans le sang puis on a recherché le lien avec les performances de reproduction. Nous avons constaté que la glycémie, la cholestérolémie, la triglycéridémie, l'albuminémie et les hormones thyroïdiennes étaient affectés par la note d'état corporel, tandis que la créatininémie, le taux de progestérone et des protéines totales n'ont pas exercé une influence significative sur cette dernière. Tandis que la protéinémie, la créatininémie, l'urémie et la progestéronémie pendant la période d'accouplement n'avaient pas d'effet significatif.

Mots-clés: Paramètres de reproduction ; Brebis de la race Ouled Djellal ; Note d'état corporel ;Paramètres sanguins; Altitude.

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Key to Abbreviations and Acronyms

BCS :	Body Condition Score	
Ca:	Calcium	
Ca.B.P :	Calibrate Before Present	
Cm:	Centimeter	
Creat :	Creatinine	
DS:	Deviation Standard	
EDTA :	Ethylene Diamine Tetra Acetic	
F.A.O :	Food and Agriculture Organization of the United Nations	
Fec:	Fecundity	
Fert:	Fertility	
FSH :	Follicular Stimulating Hormone	
GnRH :	Gonadotropin Releasing Hormone	
GH :	Growth Hormone	
Glu	Glucose	
HPA:	Hypothalamic Pituitary Adrenal Axis	
HPG:	Hypothalamic Pituitary Gonadal Axis	
IGF-1 :	Insulin-Like Growth Factor-1	
Kg:	Kilogramm	
K :	Potassium	
Km:	Kilometer	
LH:	Luteinising Hormone	
LHCGR:	Luteinising Hormone Choriogonadotropin Receptor	
M.A.D.R :	Ministry of Agricultural and Rural Development	

Mg:	Magnesium
Na :	Sodium
O.N.S. :	Office National des Statistiques/ national office of statistics
Р:	Phosphore
pH:	Potential of Hydrogen
Prol:	Prolificacy
PGFa:	Prostaglandin F
PmSG:	Pregnant Mare Serum Gonadotropin
Т3:	Triiodothyronin
T4:	Tetraiodothyronin
TG:	Triglycerid
TP:	Total proteins

Introduction

Introduction

As we all know, because of their unique role, natural resources are the best legacy to preserve for the next generations. Their valorization and conservation is an issue to humanity even though they contribute to the real wealth of a nation.

Being a part of the animal resources, sheep population is a valuable resource that deserves preservation and better exploration. Its domestication dates from 10500 to 9000 calibrate before present (Ca.B.P) (cited in Mazinani and Rude, 2020)

World sheep population is nowadays nearly 1.18 billion head (F.A.O., 2018) where 30 % is found in Africa; this latter comes in second place after Asia 43.6 %. Algeria, as the largest African country, is among the top 5 African countries with the greatest sheep populations (Skapetas and Kalaitzidou, 2017)

In Algeria, sheep population constitutes the largest livestock; it is represented by 29 million head in 2018 according to the Ministry of Agriculture. During the period 2010-2017, sheep numbers represented 78% of the total livestock; the goat population came in second place, with 4.8 million head representing 14%, followed by the bovine species, with 1.9 million head standing for 6% of the total population.

These numbers confirm the persistent and the everlasting value of this latter. The need for sheep produces (meat, milk, wool, skin and more) is and remains, in Algeria or in all over the world, an eternal need. Nevertheless, the local number of sheep population which has increased slightly in the last decade is not considered sufficient due to the continuous demand of the market; more increase in numbers is needed indeed (Mazinani and Rude, 2020).

As an attempt to contribute in the fulfillment of local needs and international concern, study of factors that influence reproductive performances is a key step to understand how to offer better conditions to gain more numbers of heads in the herd.

Consequently, we tried to draw attention to this matter in general and this research in particular comes to remind that, the Algerian breeding system still relies on very primitive ways and techniques of rearing; moreover, operational state and private farms still use less adapted systems of rearing and rarely do introduce new techniques in their production systems and more rarely ask for help.

Thus, in this study we tried to avoid getting misled by factors that apparently influence the reproductive performance of ewes and we focused on developing a fresh perception of the state of sheep breeding in our country.

This research, as stated earlier, focuses on local breed ewes because of their availability and good qualities that may well serve our objective. Additionally, Ouled Djellal breed represents more than 60% of the local breeds and they contributed a lot in developing our comprehension of reproductive performance being the subject of several local studies, if not most of them.

Compared to other species, Ouled Djellal breed is for instance the most important Algerian sheep breed; its name stands for its historical cradle. It emerged mainly from the steppe, it is also recognized by its good reproductive qualities.

Second, and as argued above, in most of the cases some sheep breeding system breakdowns and pitfalls are the cause of most of the losses and lack of yielding. Being proficient in sheep breeding is not recommended anymore, but required. This takes us to reveal what are the possible factors that might really interfere in this process.

Therefore, one of the first things that moved us to undertake the following research was curiosity to uncover what factors other than (feeding, water, breed, rearing system, climate, body condition score, age) could interfere significantly in reproductive performances.

Interviewed farmers were wondering why would they notice different fertility, fecundity and prolificacy rates in their herds while they offer the exact same conditions of rearing like feeding (quantitatively and qualitatively), water, veterinary care, and nearly same breeding conducts to the same breed; how could it lead to different yieldings

Here by the following work we came to dig after the differences that could eventually influence reproduction performance even though indirectly and one of the hypothesis was about altitude where location of the farm and nature of earth surrounding it played a role in manipulating these reproduction parameters.

Also, we tried to examine the effect of body condition score on some blood parameters in ewes at mating period and the effect of it on reproductive performance.

3

Part 1: Literature Review

1. Overview on Sheep Rearing in Algeria

Algeria enjoys resources, both qualitatively and quantitatively, that are capable of promoting agricultural and rural progress and ensuring sustainable development. It is known for the vastness of its territory and the diversity of its environments, despite this richness these resources are not suitably exploited (Feliachi, 2003).

Historically known to be a good land for sheep breeding, Algeria is gradually losing its animal heritage that is in a sharp decline similarly to the global livestock genetic diversity which is threatened (F.A.O, 2007). This observation is alarming, there is now only the sheep of Ouled Djellal which continues to dominate in Algeria. This breed well resists the climate and the unexpected practices of Man.

Sheep rearing in Algeria is mostly performed in a traditional way. In some cases, it is performed for diversification more than specialisation as Moula, (2018) observed in his survey.

Some of the rearing system breakdowns are due to the insufficient food resources; availability of forage, for instance, is linked to climate conditions. Among the solutions adopted by the Algerian government to solve this deficit are the import of concentrate and the valorization of the agro-industrial by-products. Dates' by-products particularly are proven to be efficient on plenty of levels (Allaoui et *al.*, 2018); (Baa et *al.*, 2018); (Abaidia et *al.*, 2020).

Another feature is the uncontrolled and the archaic reproduction approaches, whether for the number of introduced rams to ewes, for the age of breeding, or even for the selection criteria (Titaouine, 2015). These practices have caused the disappearance of many breeds (Djaout et *al.*, 2017); (Moula, 2018). All of these, in addition to the poor mating practices, long periods of drought and lack of water providers (M.A.D.R., 2005) contributed to the recession of the health status of the national herd.

In Algeria, the total local breeds and their precise numbers are not yet accurately determined. This is due to the fact that they are not yet fully standardised. Even for some known breeds, genotypic or even phenotypic characters are still not fully defined. This prevents them from gaining a genuine international status (Harkat et *al.*, 2015); (Djouat et *al.*, 2017).

Moreover, the existing animal production systems in Algeria could be designated as follows (AnGR,2003);(M.A.D.R., 2005):

-Extensive system: the most dominant system. It depends mainly on traditional ways of rearing, practiced in areas with low vegetation cover. The diet offered to this type of livestock is based on grazing crop residues, rarely supported by barley or dry fodder straw; the animals could be sheltered in sheepfolds rarely. It may involve transhumance.

-Semi-extensive system: it is distinguished by a moderate use of food and veterinary products. Sheep are fed by pasture on fallow land, on crop residues and benefit from barley and hay supplements.

-Intensive system: Unlike the extensive system, this system relies on important food providing, extensive use of veterinary products as well as housing animals in equipped buildings.

Sheep production systems in Algeria, could also be divided into two systems related to mobility (cited in Harkat et *al.*, 2015):

- Sedentary or semi-sedentary: with the use of one or two seasonal pastures located not more than 50 km from the main breeding areas.

- **Transhumant**: with variable itineraries: breeders of Laghouat use western provinces such as Tiaret, Tissemsilt, Mostaganem, Relizane; breeders of Biskra move to the east "Oum Bouagui, Tébessa", while farmers of Djelfa and M'Sila use mostly central and eastern provinces "as Medea, Bouira, Bordj Bou Arreridj and Setif".

1.1 Some Existing Breeds in Algeria

In Algeria, the sheep herd is made of nearly 12 breeds according to (Djaout et *al.*, 2017). Ouled Djellal breed constitutes the largest number of the entire national sheep population, it represents nearly 63 percent of the sheep herd in Algeria (M.A.D.R., 2003) while for the remaining sheep herd it is mainly made of breeds that are in sharp decline or even endangered (Chekkal et *al.*, 2015).

According to Djaout and her collaborators (2017) the existing breeds in Algeria could be divided into two categories according to their numbers as follows:

-Breeds present in a high number of heads: Ouled Djellal, Hamra, Sidaou and Ifilene.

-Breeds present in a low number of heads: Rembi, Berber, Barbarine, D'man, Tàadmit, Tazegzawth, Srandi and Daràa.

1.2 Ouled Djellal Breed

It is the typical breed of the steppe and the high plains. The total number of heads is around 11,340,000 head, which represents 63% of the total sheep population (M.A.D.R., 2003). Being the subject of thorough studies, Ouled Djellal sheep is described by several authors as the most suitable for nomadism.

The breed is entirely white with fine wool and fine tail, high waist, and long legs suitable for walking. However, It fears the cold (Chellig, 1992), the wool covers the whole body up to the knees and to the hock for some varieties. It gradually adapted to all production systems of the country and this is the main reason behind its widespread emergence (Djaout et *al.*, 2017).



Figure.1: Photo of a ewe of Ouled Djellal Breed (Original, 2018)

According to the breeders five varieties of Ouled-Djellal breed can be defined based on morphological criteria: the Ouled-Djellal, the Mouidate, the Safra, the Baida and the Hodnia (cited in Chekkal et *al.*, 2015).

2. Ewe Reproduction

2.1. Puberty

Generally, ewe lambs reach puberty as the first ovulation and the first estrus occur, which coincide usually with a weight about 50 - 70% of their adult body weight. Dudouet (1997) defines puberty as the ability to produce efficient gamete in a way that allows the individual to reproduce efficiently, but (Edey et *al.*, 1978) believe that the first oestrus apparition does not necessarily have to coincide with the first ovulation because the two phenomena are apart.

Oestrus usually begins prior to ovulation which usually occurs 36-40 hours after the beginning of estrus. The ovum can generally be fertilized for 10-25 hours after being released while spermatozoids can survive for about 30 hours after mating. Detection of heat signs is a bit difficult to perceive especially in the sheep specy (Dudouet, 1997) The table 1 below summarizes the basic reproductive characteristics of ewes.

Phenomenon	Duration	Reference
Age at Puberty in Ouled Djellal Ewes	8-10 months	(cited in Chekkal et <i>al.</i> , 2015)
Whole Sexual Cycle Length	17 days	(Castonguay, 2018)
Oestrus	25.9 ∓6.8 hr	(Greyling et <i>al.</i> , 1993)
Life Span of Luteum Corpus	14 days	(Baird et <i>al.</i> , 1975)
Fertilizable Life Span of the Ova	10-25 hr	(Baird et <i>al.</i> , 1975)

 Table 1: Some Reproductive Characteristics of Ewes

2.2. Ewes' Sexual Cycle

In ewes, the sexual cycle is the period of time that separates two successive oestruses. Its length is 17 days on average (Castonguay, 2018); however, this period can vary according to many factors: breed, age, season, environmental stress, and the presence of the male (Jainudeen et *al.*, 2000). During estrus, the ewe seems more attractive and more receptive to ram (Abecia et *al.*, 2020).

According to Dudouet 1997, the sexual cycle of ewes comes out through some changes at the level of the ovaries; the hormones; the behaviors. It involves one ovarian and one oestrus cycle simultaneously. Oestrus is the period that lasts 36-40 hr during which ewe females accept mating. It depends mainly on the age; longer in mature ewes, shorter in lamb ewes; and on the breed, prolific breeds experience longer estrus periods (Farhat et *al.*, 1997).

The ovarian cycle is divided into two phases: follicular and luteal. The follicular phase lasts 3 to 4 days, it corresponds to the period of the cycle during which follicle growth occurs. During this period, follicles of different sizes start to grow in an accelerated rhythm under the influence of different hormones of the pituitary gland (Castonguay, 2018).



Figure 2: Hormonal Variations during Ewes' Sexual Cycle (Castonguay, 2018)

Increased secretion of hormones, the estradiol mainly will cause the onset of estral behaviour. In sheep, It was found that there could be no source of oestradiol other than Graafian follicle (Baird et *al.*, 1975).

The oestrus lasts from 24 to 72 hrs (Wagner, 1965). Its duration is generally shorter in lambs and longer in the middle of the sexual season than in the beginning or the end of it. Some studies (Cognié, 1988); (Farhat et *al.*, 1997); and (Casas, 2000) show that the duration of oestrus is longer in prolific breeds than in less-prolific breeds.

After that, comes the ovulation which corresponds to the release of ovum contained in mature follicles, 36 to 40 hours after the onset of the oestrus, or towards the end of it

(Dudouet, 1997). The follicle which ovulates turns into a structure called the corpus luteum which in its turn secretes progesterone, a hormone that blocks the secretion of the pituitary gland hormones and it is responsible for follicular growth.

There are as many lutea corpora on an ovary that there are follicles that have ovulated. So the number of lutea corpora on the ovary represents the maximum number of embryos that could have been formed in a given ovulation period (Baird et *al.*, 1975).

During the 14 days of the luteal phase, the development of follicles is slow and ovulation is impossible. If the sheep is not fertilized, the corpus degenerates to allow a recovery of the ovarian activity (follicular phase) that will lead to the ovulation from new follicles.

The estrous cycle is controlled by a complex regulatory system involving exogenous and endogenous factors including a number of hormones, as illustrated in (Figure 2 and 3).

2.2.1. Hormonal Regulation of Oestrus

The hypothalamus sends gonadotropin-releasing hormone (GnRH) to the pituitary gland, which sends luteinizing hormone (LH) and follicle-stimulating hormone (FSH) to the ovaries, prompting the ovaries to grow follicles (Cognié, 1988).

As the follicles grow, they release estradiol, which is fed back to the brain and causes the ewe to come into oestrus. The amount of estradiol being sent to the brain increases as the mature follicles get larger. When the follicles reach 0.5-1 cm in diameter, blood concentration of estradiol peaks, and the brain releases a large amount of LH, which causes ovulation.

It was found that the acquisition of the LH receptors on the granulosa took place as the follicular diameter reaches 3-3.5 mm. Ovulation could not take place before the follicular diameter reaches 6 -7 mm (Monniaux et *al.*, 2009).

After ovulation occurs, the follicle that the ovum was in collapses and forms a corpus luteum, which secretes progesterone. This increase in progesterone tells the hypothalamus to decrease production of GnRH, resulting in reduced follicular growth, causing estrus and ovulation to be suppressed as long as the progesterone level is high.

If a pregnancy is not established (which would keep the progesterone levels high), the uterus will secrete the F2a prostaglandin hormone (PGFa). This hormone causes the corpus luteum to decay and decreases progesterone secretion (Lassoued et *al.*, 1997). As a result, the hypothalamus starts producing GnRH, and the cycle begins again.



Figure 3 : Model of the reproduction regulation in ewes (Cognié, 1988)

2.2.2. Ovulation

Before its birth the lamb ewes already get their stock of ova ready. Surrounded by the follicle, this latter could exist at different maturity stages in terms of the sexual cycle phase later (Monniaux et *al.*, 2009).

Generally, ovulation occurs the 36-40 hrs that follows the onset of oestrus (Dudouet, 1997). The rate of ovulation, which corresponds to the number of ova released at ovulation, represents the maximum number of ova potentially fertilized.

It was found that the ovulation could not take place until the apparition of the LH receptors on the granulosa, Luteinising Hormone Choriogonadotropin Receptors (LHCGR) which is the sign of the complete maturity of the follicle (St Dizier, 2014).

The ovulation rate varies according to breed, nutritional level: increases with flushing (Haresign, 1981), body condition (Ducker and Boyd, 1977), age: maximum reached towards 2 to 6 years (Shorten et *al.*, 2013), individual genetic background and environmental

conditions. The ovulation rate also varies during the same sexual season, peaking towards the mid-season and then declining to cause anœstrus.



Figure 4: Ovogenesis and folliculogenesis in life of mammals, cited by (Monniaux et al.,

2009)

2.3. Fertilization

Fertilization is the point in time when the spermatozoa runs into the ova. The uterus receives the fertilized ova that has started segmentation in the uterine tube. Under the control of multiple hormones, especially ovarian, it ensures their implantation and then the nutrition of the conceptus through the placenta (Dudouet, 1997).

2.4. Gestation

It generally lasts 150+-5 days for about five months. In many references the third phase is considered the most important for both the lamb and the ewe, but each phase is important in a way. Appropriate nutrition is essential throughout gestation to optimize lamb survival.

The first phase is more important to lamb development, especially early to mid gestation (Ford et *al.*, 2007) when embryonic attachment occurs. It is compulsory to avoid stressing ewes (Henrique et *al.*, 2021) or transport. Also, nutrition has to remain available so females don't lose body condition, which raises forward stress and may lead to fetal death because it is the phase during which primary lamb organ development begins. Some studies (Ford et *al.*, 2007) went beyond this and proved that undernutrition of ewes during early to midgestation altered growth adiposity of the lambs.

The second phase is when organ development continues, performance factors like muscle tissue and lamb reproductive organs develop and grow. For instance, a female lamb's future egg production takes place. It is extremely important to provide the mother with nutrition throughout the entire pregnancy. Maintaining body condition and avoiding stress in the ewe was proved to influence the future reproductive performance of the lamb.

The third phase is when 75% of fetal growth occurs, and organ development is completed. Lambs are pulling energy from the ewe, so an increase in the amount of nutrition to the ewe to keep energy levels high is needed.

Lambs born from ewes that receive supplemental minerals throughout the pregnancy are usually born stronger, and nurse more rapidly. Supplemental nutrition can also increase the chance of multiple births. The third phase is a good time to add mineral blocks for ewes, particularly if lower body condition score is a concern.





2.5. Factors Affecting Fertility

Many factors affect conception rates in ewes: Age; stress and environment; season; ram; health; nutrition and body condition; lambing interval; genetics (Casas et *al.*, 2004);(Arbouche et *al.*, 2015);(Abecia et *al.*, 2020) Each of the following factors can have a significant effect on conception rate; in some cases, a combination of factors decreases conception rates. Out of breeding season, every factor must be managed carefully in order to have the best outcome. If conception rates are poorer than expected, we would need to consider other factors: ram management, health, nutrition, stress and environmental factors such as heat.

Lambs born from ewes that receive supplemental minerals throughout the pregnancy are usually born stronger, and nurse more rapidly. Supplemental nutrition can also increase the chance of multiple births.

2.5.1 Nutrition

Supplementation is critical as ewes enter the final 60 days of pregnancy, when 75% of fetal growth occurs. If the ewe can't get enough energy into her system during this timeframe, she can lose weight since the fetus is demanding more and more of the nutrients. (Hoversland among the first who spread the idea of flushing 1985)

As the ewe gets later into gestation, her rumen competes for space with the growing lambs. An energy-dense supplement provides the nutrition efficiently without taking much space, allowing her plenty of space for both the rumen and lambs.

Once lambs are born, maternal nutrition remains important. A higher level of dietary fat can influence milk fat content. In turn, higher milk fat helps lambs get a better start with extra energy for rapid and efficient lamb growth.

Aiming for an average body condition score of 2.5 to 3.0 on a 5 levels' scale and maintaining that level throughout the gestation is a good way to increase reproduction performances. Females at this body condition score are more likely to be healthy and not under nutritional stress.

Appropriate nutrition is key for successful estrus synchronization in sheep. If the ewes are too thin, they will not respond to the synchronization as reproduction will not replace the ewes' natural survival instinct.

The female body is programmed not to cycle or become pregnant if nutrition is inhibited, as her initial task is to survive herself. If the ewes are too fat, reproduction can also

be inhibited. Keeping ewes in a proper, consistent body condition will aid in the ewes' overall well-being and in turn increases sheep milk production capabilities.

A swinging pendulum where body condition is good at conception and reduced through gestation and lactation will not only diminish lamb colostrum quality, lamb health at birth, but could also result in reduced sheep milk production and lower weaning weights. Ewe calcium and other mineral needs are high during this time, especially with multiple lambs.

The transition from green to brown pasture is a key indicator that forage quality is on the decline. Providing the flock with supplement before forages turn yellow and throughout the autumn and winter ensures ewes receive the nutrition they need to support breeding and reproduction.

Total Reliance on grass as the sole nutrition source can have heavy repercussions on everything from ewe body condition to breeding and reproduction. Adding supplements to the diet, even in the short-term, can positively impact the flock's productivity.

2.5.2 Environment

A stressor is a stimulus that disrupts homeostasis (Smith and Dobson, 2002) and activates a variety of chemical and specific physiological processes and behavioral coping mechanisms to restore homeostasis and promote survival (Cizauskas et *al.*, 2015).

Although livestock animals, sheep for instance, tend to be well adapted to their environment, it is known that the livestock production processes subject animals to a multitude of physical and psychological stressful stimuli that have the potential to elevate the hypothalamic-pituitary-adrenal axis axis activity (HPA) (Narayan and Parisella, 2017).

Chronic stress is one of the major challenges in sheep production, as it is difficult to detect and can result in prolonged dysfunction of the HPA axis, causing downstream negative physiological effects such as immunosuppression, increased susceptibility to disease and reproductive dysfunction.

The elevation of HPA axis activity during chronic stress has been suggested as the primary neuroendocrine mechanism underlying the aetiology of reproductive dysfunction in sheep. Research in sheep has demonstrated that glucocorticoids act on the hypothalamic pituitary gonadal axis (HPG) axis at the level of the hypothalamus and hypophyseal portal

system to decrease gonadotropin secretion and at the level of the pituitary gland to reduce responsiveness and sensitivity of gonadotroph cells and their receptors to GnRH(Narayan and Parisella, 2017).

2.5.3 Body Condition Score

The rating of body condition has developed over the mid twentieth century to provide breeders and livestock partners a practical and a reliable tool that enables estimating energy reserves. Since the anatomical markers studied for the award of the score are fairly uniform, a first system for rating body condition for sheep was initially developed by Jefferis in (1961). The aim was to assess the fattening status of the sheep by palpation of the back spines, and the transverse processes of the lumbar vertebrae.

This energy balance indicator is used not only for livestock monitoring and assessment of the herd's nutritional behaviour, but also to assess its relationship with both production parameters and reproduction parameters. The awarding of such a rating would require criteria as objective as possible (Froment, 2007).



Figure 6: Body Condition Scoring (cited in Chisholm, 2021)

Ewe body condition score leading up to and during gestation plays a pivotal role in lamb development. An under-conditioned ewe won't have enough nutrition and energy to support their own maintenance needs and support lamb growth.

On the other hand, an over-conditioned ewe may be at risk for health challenges, which can negatively impact the lamb. Maintaining an average body condition score of 2.5 to 3.0 on a 5-point scale and throughout the pregnancy promotes the yieldings. Females at this body condition score are more likely to be healthy and not under nutritional stress.

Body Condition Score	Characteristic of the animal	Features during palpation
1	Emaciated	 No fat cover Sharp and prominent back lumbar spine Fingers easily pass under the muscle Transverse process is sharp
2	Thin	 Thin fat cover Smooth and prominent back lumbar spine Fingers go under the muscle with pressure Transverse process is rounded
3	Average	 Moderate fat cover Smooth and rounded back lumbar spine Fingers need hard pressure to find ends Transverse process is smooth and rounded
4	Fat	 Thick fat cover Back lumbar spine detected only as a line Muscles are full Transverse process is not felt
5	Obese	 Very thick fat cover No detectable back lumbar spine Muscles are very full Transverse process is not detectable

 Table 2: Main Features identifying the Body Condition score

2.6. Sexual Activity and Duration of Illumination (Role of Melatonin)

Sexual activity in sheep is subject to many factors which leads to certain variations in this latter; melatonin for instance, which is a natural substance secreted in the brain by the pineal gland at night, plays a crucial role in this variation which appears in the seasonality of the sexual activity of many breeds. Its duration of secretion is proportional to the duration of the dark period and it is through it that the animals perceive the duration of the illumination (Senger, 2015).

For sheep, a seasonally bred species, melatonin inhibits or triggers their sexual activity when its duration of secretion is short and long, respectively (Malpaux et *al.*, 1999). In ewes, melatonin supplementation (per implant) for 40 days triggers an activity similar to that of autumn.

It appears that, in addition to its ability to advance the mate, melatonin has beneficial effects on fertility and prolificacy (Palacin et *al.*, 2011). For lamb ewes, melatonin allows them to succeed their first mate (June-July) in rustic breeds. In practice melatonin is marketed as implants that are placed under the skin of the ears 30 to 40 days before the start of the mate (Cognié, 1988).

2.7. Some Techniques of Oestrus Induction

Grouping estrus commonly called oestrus synchronisation reduces labor over a long period of time. It also allows bringing the ewes into oestrus at an earlier time for breeding and thus getting lambs in a pre-defined period depending on forage availability and so on. Not only this but also, accelerating the rate of lambing ewes; Reducing unproductive periods; Putting the lambs into early mating. In addition, it helps ensure better monitoring because the work will be concentrated in a short period of time which simplifies rationing for instance.

Some obvious advantages of this approach are to organize work on the farm; to divide the work over the year by grouping lambings and raising homogeneous batches of lambs at the same physiological stage ; it makes dietary supplementation easier and more economical and most importantly sheep are more likely to be fertilized during a fixed-term period.

2.7.1. Synchronization Technique Based on Hormonal Usage

Estrus synchronization in sheep is tricking the ewes' body into believing that she is pregnant and thus preventing her from cycling normally. According to (Dudouet, 1997) it is to extend the luteal phase through a progestogen until the disparition and decay of all luteum corpuses as the sponges are removed we observe oestrus apparition and when pmsg is injected the ovulations are at their best.

Introducing progesterone or a progesterone derivative into the vaginal vault of the ewe by impregnated sponges and letting them 12-14 days prevents ewes from cycling naturally. Once the progesterone is removed, the ewe will cycle within 48 hour, $(37,3\pm5,3)$ hour in Ouled Djellal breed according to Taherti et *al.*,2016) leading to a "synchronized" estrus.

2.7.2. Ram Effect as a Natural Technique for oestrus induction

It was observed that the sudden introduction of rams into a flock of ewes will result, 17 to 25 days later, in a peak of coming in oestrus: this is the ram effect. By mastering this technique, it is sought to ensure that as many sheep as possible are protruding from the beginning of the mate thanks to the oestrus grouping (Cognié, 1988) ; (Dudouet, 1997) ; (Lassoued et *al.*, 1997) and (Thimonier et *al.*, 2000) Figure 7 illustrates the estrus lengths in anovulatory females before ram introduction.



Figure 7: Frequency distribution of the length of the estrous cycle in ewes. Sheep Production Handbook (2002).

3. Blood Parameters and Mineral Assessment in Ewes

Gestation is accompanied by a disruption in the metabolism, especially calcium and phosphorus. This is to meet the demands of metabolism and fetal growth. This disruption will have an impact in terms of food intake, nutrient absorption and hormonal regulation.

Reproductive status and seasonal variations have to be taken into consideration for a correct interpretation of the serum chemistry values of sheep. Nutritional supplements are required for sheep during certain periods to avoid a decline of their performance, which would then represent consequent economic losses (Yokus et *al.*, 2006).

Examination of the changes in blood constituents during gestation and lactation periods of some blood parameters namely: glucose, total plasma protein, albumin, urea and creatinine. During gestation all these parameters started to increase significantly during gestation, but in different stages, reaching maximum values at parturition (Elsherif and Fawzia Assad, 2001). In lactating ewes, levels of glucose, urea and creatinine returned to levels comparable to those in dry ewes.

In contrast, dry ewes showed almost stable values of the stated parameters. Urea and creatinine began to increase significantly after 10–12 weeks of gestation. The same occurred with total plasma proteins, mainly due to a sharp decrease in globulin, while albumin remained higher in dry ewes (Elsherif and Fawzia Assad, 2001).

3.1. Calcium

Calcium is 99% contained in the skeleton. The remaining percentage has many roles, such as blood clotting, enzyme activation and neuromuscular activity. This explains why chronic calcium deficiency remains clinically silent for a long time, even if it can result in bone disorders.

It is required for smooth muscle function and has a direct role in uterine contraction, so it may influence the duration of parturition. Low Ca intake influences insulin release and sensitivity (Friend et *al.*, 2020).

3.2. Phosphorus

Phosphorus is present in phosphoproteins, phospholipids, nucleic acids, ATP.etc Concentration of phosphorus in the muscle is 2 to 3g/kg while calcium is only 0.1g/kg. On

the contrary, the lack of phosphorus translates more quickly and more clearly in young animals into a poor general condition (Payne, 1983).

Phosphorus is an essential element involved not only in bone development, growth and productivity but also in most metabolic processes of the animal organism. The essential function of phosphorus from a quantitative view is the formation and bone maintenance (Suttle, 2010)

3.3. Magnesium

Low Mg intake influences insulin release and sensitivity, it results in poor glycaemic control and insulin resistance by impairing both insulin secretion and its action on peripheral tissues, also potentially altering the duration of parturition as well as risk of metabolic disease.

Magnesium is also a neuroprotectant that slows the neuronal damage during hypoxia and has been linked with thermogenesis in offspring and increased immunoglobulins in colostrum. These functions indicate potential importance in improving the ease of parturition and improved ability of the newborn lamb to thermoregulate and survive after birth (Friend et *al.*, 2020)

3.4. Sodium

Plasma Na concentrations were not affected by the level of dietary intake of Na when a low-sodium diet was given to ewes over two reproductive seasons; Na concentration in saliva and urine was significantly lower in the ewes with lower dietary intake than in the ewes with supplemented sodium intake (Vincent et *al.*, 1986).

3.5. Potassium

The active transport of mineral substances by cell membranes plays an important role in the neurohumoral regulation of reproductive processes in ewes. It is important to reveal that plasma K concentrations were not affected by the level of dietary intake, while K concentration in saliva was significantly higher (Vincent et *al.*, 1986).

The effects of hormones on K metabolism in the blood serum of ewes were studied by Hawk et *al.* (1961). Krajnicáková et *al.* (1993) who observed the dynamic changes of electrolytes in the course of the reproductive cycle in ewes. Na and K levels and their relation to ovarian hormones were determined in the systemic blood stream of ewes during oestrus synchronization and gestation.

3.6. Thyroxine

Secreted by the thyrocytes, this hormone is made up of two fractions. the triiodothyronine (T3) and tetraiodothyronine (T4). The T3 fraction is the most Active. Its action is synergistic with that of the GH in the absence of which the T3 maintains enchondrale ossification. However, the excess of this hormone accelerates the renal leakage of Ca decreased reabsorption, which explains the frequency of osteoporosis in hyperparathyroidism. Thyroid hormone would potentiate the action of vitamin D by allowing its release from its storage points.

3.7. Progesterone

It was found that analogues of progesterone could adequately improve the reproductive performance of anestrous ewes (Ahmed Amer and Maher Hazzaa, 2009). It is known that the increase of cholesterol levels is closely related to the progesterone concentration, because cholesterol is a precursor to the synthesis of steroid hormones among which the progesterone (Fall, 2008). Another study concluded that supplementation of fatty acid soaps significantly increased serum cholesterol, triglycerides, low-density lipoprotein cholesterol, glucose and progesterone (Hayat et *al.*, 2012).

3.8. Urea

The influence of dietary urea on reproductive performance in ewes was studied where the incidence and regularity of estrus and percent females lambing during one estrous period were not affected by dietary urea. The ovulation rate of ewes was not influenced by dietary urea, and the length of time ewes were fed the experimental diets prior to mating did not influence subsequent fertility and reproductive rate. The results demonstrate that high roughage diets can be supplemented with urea without reducing reproductive performance (Thompson et *al.*, 1973).

3.9. Creatinine

Creatine metabolism is an important component of cellular energy homeostasis. Mainly through the creatine kinase circuit, creatine derived from diet or synthesized endogenously provides spatial and temporal maintenance of intracellular adenosine triphosphate (ATP) production; this is particularly important for cells with high or fluctuating energy demands (Mucini et *al.*, 2021).

Yokus and his collaborators (2006) in their study indicated that creatinine varies with reproductive status but not with seasonal variations; a single reference interval for creatinine can be used for both mated and unmated sheep because no differences were found related to the reproductive status.

Some studies link perturbations in creatine metabolism to reduced fertility and poor gestation outcomes. Maternal dietary creatine supplementation during gestation as a safeguard against hypoxia-induced perinatal injury, particularly that of the brain, has also been widely studied in pre-clinical in vitro and small animal models (Mucini et *al.*, 2021).

3.10. Total Proteins

Total protein (TP) is linked with reproductive status but not seasonal variations; a one reference interval for TP can be used for both mated and unmated sheep because no differences were found in its rates following the reproductive status (Yokus et *al.*, 2006).

3.11. Albumin

Higher concentrations of albumin and cholesterol have been detected in the blood of pregnant ewes compared to the non-pregnant ones, while the opposite way has been noticed for the concentrations of total proteins (Antunovic et *al.*, 2004)

3.12. Glucose

In short term supplemented ewes, increased concentrations of glucose, insulin and leptin one day before ovulation were associated with increased numbers of follicles growing from 2 to 3 mm and with stimulation of the dominant follicle to grow for a longer period (Vinoles et *al.*, 2005).

Vinoles and his collaborators (2005) suggest that the mechanism by which short-term nutritional supplementation affects follicle development does not involve an increase in FSH concentrations, but may involve responses to increased concentrations of glucose, insulin and leptin, acting directly at the ovarian level. This effect is acute, since concentrations of all three substances decrease after reaching peak values on the third day of supplementation.

Part 2: Experimental Part
1. Details of the Study Region

The present study was carried out in the region of Biskra, located in the central-east of Algeria, at the gateway to the Algerian Sahara. It is a real buffer zone between North and South, about 400 km southeast of the capital, Algiers (approximately $35^{\circ}15'$ to $33^{\circ}30'$ N, $04^{\circ}15'$ to $6^{\circ}45'$ E).



Figure 8:Geographic Locations of the study areas at Biskra, Algeria;in green ElHadjeb (site1), in red Sidi Okba (site2), in blue Ouled Djellal (site3), and in purple Ain- Zaàtout (site4) (Original, 2021)

In general, Biskra is composed of four different geomorphological elements: the mountains, the plains, the tablelands, and the depressions (Bougherara and Lacaze, 2009; Aissaoui et *al.*, 2019).

Since the climate is the set of actions of the atmosphere (temperature, rain, wind ...), Biskra is considered to belong to the semi-arid to arid one, characterized by a cold winter and a hot dry summer with irregular rainfall.

Temperature is considered one of the most important climatic factors for both vegetation and animals; high temperatures during long periods of time accompanied with low water supply may induce heat stress though. For plants, it ensures evapotranspiration, and growth. We observed that:

- The coldest month in Biskra is January with a minimum temperature/average of 7.2/12.2°C.

- The hottest month in Biskra is July with a maximum temperature /average of 41/34.4°C (infoclimat.fr, 2022).



Figure 9: Bar graph of the monthly recorded temperatures at Biskra from 1991-2020 (infoclimat.fr, 2022)

The study area is characterized by significant diurnal thermal amplitudes. This is how the temperature, which reaches very high levels in the shade the daylong during the summer, drops to minus 50% in the evening. The variation in daily thermal amplitude is significant throughout the months of the year. The maximum temperature is reached in July: 49 °C, but in January, it oscillates around 7 °C (especially in the evening).

Rainfall is a fundamental factor in characterizing the climate of a region. We noticed that the distribution of rainfall from one season to another and from one month to another is marked by its great irregularity, the maximum rainfall is recorded during the month of November (28.9 mm), on the other hand the minimum is recorded during the month of August (4.8 mm).



Figure 10: Bar graph of the monthly recorded rainfall at Biskra from 1991-2020 (infoclimat.fr, 2022)

The relative humidity of the air varies considerably according to the season. In summer, it falls to 25% in July due to high evaporation. However, in winter, it rises up to 60% maximum.

Winds blow throughout the year. Generally, the northwest winds dominate. Southern winds are generally cold and dry in winter. They are hot and very dry in summer, that is, the sirocco. It acts by enhancing evaporation and thus increases drought. Southwest winds are hot and dry and northwest winds blow mostly in winter and bring rain to the region.

Below is a table showing the most relevant details of the study sites.

	Height above sea level	Landforms	Rainfall (mm/month)	Annual Temperature °C (mean)	Geographic Locations Latitude	Geographic Locations Longitude
Site 1	139 m	Plain	10.4 a	26.2 a	34°47′	5°36′
Site 2	54 m	Plain	10.4 a	26.2 a	34°75′	5°9′
Site 3	450 m	Tableland	12.85 b	21.05 b	34°27′	5°04′
Site 4	1000 m	Mountain	12.02 c	17.8 c	35°09′	5°50′

 Table 3 :Details on the different study sites

a Biskra meteorological station; b Ouled Djellal meteorological station; c Lotaya meteorological station

EXPERIMENT 1: Study of the Effect of the Altitude on Some Blood Minerals

Experiment 1: Study of the Effect of the Altitude on Some Blood Minerals

Materials and Methods

1. Study Region

The choice of locations was based on the geographical distribution and the characteristics of the areas (reliefs and altitudes). We selected four (4) locations (Table 1): El Hadjeb, Sidi Okba, Ouled Djellal, and Aïn Zaatout noted respectively, by site 1, site 2, site 3, and site 4 (check figure 8 in the previous chapter).

2. Choice and Distribution of Herds

The selected animals are clinically healthy and nonpregnant Ouled Djellal ewes, aged 2.5–05 years with a median body condition score of 2.8 ± 0.9 (on a scale from 1 - 5) and weighted 53.4 kg \pm 8.4. These ewes were divided into four herds as follows: in site 1 (30 ewes), in site 2 (60 ewes), in site 3 (70 ewes), and site 4 (66 ewes).

3. Livestock Management

Ewes were being reared in a semi-extensive system. Details are mentioned in the following:

3.1. Feeding Approach

The livestock of the four farms is managed to be into a semi-extensive system of rearing; in all sites herds are taken to pasture twice a day (7 a.m. to 11 a.m. and 4 p.m. to 6 p.m.). At night, all ewes were housed in a barnyard and had access to fresh clean water twice a day (one in the morning, second in the evening).

Feeding is based on complementarity between cereals and sheep grazing; However, feeding could be different in quantity from a season to another depending on circumstances. In winter, with the decrease in food resources, the herds are kept in the sheepfolds thus receiving coarse fodder (hay and straw) and concentrated feeds sometimes and if the weather is good the animals go outside for short walks.

In spring, the animals graze on the fallow lands which consists of leaving the soil for a period of time after the crop. The spontaneous vegetation that grows is used for grazing, where the fallow is mowed to produce hay. During the summer when the cereals are harvested, the stem part of cereal and crop residues are offered to feed the herds. Most of the time, animals are fed according to availability, no strict forage schedule is followed, but the deficit was marked more during winter.

In the four sites, the natural grazing land consisted of a mixture of *Stipa tenacissima*, *Ampelodesmos tenax*, and *Artemisia herba-alba*, as well as annual meadows, composed of various grasses (predominance of *Cynodon dactylon, Melilotus sulcatus, and Vicia monantha*).

3.2. Water Supply

Water is the most essential nutrient in a livestock grazing system. All of our sheep herds had access to clean and good-quality water as required, but only twice per day. Poor water quality can reduce livestock performance, it influences reproduction, and causes livestock stress that could lead to general health state decrease. That is why water should frequently be tested to ensure if it is suitable for animal health and welfare and more importantly it should be offered as the livestock needs.

3.3. Selection of Animals

In the current study all ewes are assumed to be healthy that is why a normal clinical examination on the day of sampling; and not taking any treatment during the sampling or at least the month preceding it, are some important points taken into consideration before starting. In addition to state of health, breed, age, gender and body condition scores were also taken into consideration.

3.4. Prophylaxis

All herds in this study were well supervised in terms of health care by qualified veterinarians, they received group treatments as well as individual care when necessary. The veterinarians carried out group treatment at the same time for all herds against the most encountered diseases in the region and which are of bacterial origin (Enterotoxaemia), viral origin (Sheep pox) and parasitic origin (scabies, strongylosis). The ewes were dewormed 4 times/year with anthelmintic, and they are routinely vaccinated against sheep pox, peste des petits ruminants (P.P.R.), and enterotoxaemia.

The following are some of the routinely scheduled steps in the livestock management:

• The bedding is renewed at least twice per year (from June to August and in December).

• The sheepfold walls are coated with hydrated lime simultaneously with the bedding renewal usually. (10 kg of calcium oxide "quicklime" are poured into 100 l of water)

• A separate room is devoted to keep apart the lambing ewes where bedding is renewed most frequently, and the clean out of the lambing pen is done after each lambing.

• The bathing as well as the wool shearing are done once to twice per year. In order to avoid sunburns or heat stress it is generally carried out in the beginning of summer (May) and/or in December.

• Hooves trimming is not neglected. It is performed regularly to prevent the flock from any possible issues.

4. Blood Sampling

Blood samples were taken from all the ewes of the study early in the morning. The ewes were distributed into three groups, depending on the topography of the study area: group 1 is a plain region (site 1 and site 2), group 2 is a tableland (site 3), and group 3 is a mountainous region (site 4).

Blood samples were taken by puncture of the jugular vein early in the morning before food intake to limit variations related to food intake and stress. Seven milliliters of blood were collected using disposable needles in vacutainer tubes with heparin anticoagulant. The collected blood was transported in a cooler and centrifuged, before the exhaustion of 2 hr after sampling, at 3000 G at room temperature for 15 min. The plasma was divided into aliquots in microcentrifuge tubes and kept frozen at -20 °C till further analysis.

5. Analysis and Choice of Biochemical Variables

The choice of the biochemical variables to study was delicate; however, for some technical issues (availability of reagents mainly), it was necessary to determine the parameters which would be the most relevant, without forgetting the clinical interest which remains crucial.

Blood minerals: plasma calcium (Ca), plasma phosphorus (P), plasma magnesium (Mg), plasma sodium (Na), and plasma potassium (K) were estimated using commercial kits

"SPINREACT,Spain" as per standard method using the ultraviolet (UV)-visible recording spectrophotometer (UV-160A; Shimadzu Corporation, Japan)

Parameter	Analytical Method	Reference
Calcium (Ca)	Colorimetric Technique (Calcium and <i>o</i> -cresolphtalein Reaction)	1001061
Phosphorus (P)	Colorimetric Technique (Molybdate and Phosphorus Reaction)	1001150
Magnesium (Mg)	Calmagite - EGTA Colorimetric Method	1001280
Sodium (Na)	Mg-Uranyl Acetate Method	1001380
Potassium (K)	Tetraphenylboron - Na Method	1001390

Table 4: Analytical methods used to measure the studied blood's minerals rates

5.1 Calcium dosage

The measurement of calcium in the sample is based on formation of colored complex between calcium and *o*-cresolphtalein in alkaline medium

 $Ca^{2+} + o$ -cresolphtalein $^{HO+} \rightarrow Colored$ complex

The color intensity is directly proportional to the amount of calcium present in the sample that is measured by spectrophotometry at a wavelength of 570 nm.

Usually, colorimetric methods allow the measurement of total serum calcium:

Total plasma Ca = ionized Ca + bound Ca + chelated Ca

5.2 Phosphorus

Direct method for determining inorganic phosphorus. Inorganic phosphorus reacts in an acidic medium with molybdic acid, its subsequent reduction in alkaline medium originates a blue molybdenum color.

Phosphorus + Molybdate- ammonium $^{H2SO4} \rightarrow$ Phosphomolybdic complex

The intensity of the developed coloration is proportional to the concentration of phosphorus in the sample that is measured by spectrophotometry at a wavelength of 710 nm.

5.3. Magnesium

The measurement method is based on the use of the calmagite which reacts with the magnesium in an alkaline medium forming a purple colored complex.

The absorbance of the purple colored complex is measured at 520 nm and it is proportional to the magnesium concentration in the sample.

5.4. Sodium

Sodium precipitates with Mg-uranyl acetate; then the ions of uranyl remaining in the suspension form a yellow to brown complex in the presence of thioglycolic acid. The difference between reagent blank (without sodium precipitation) and the analysis is proportional to the sodium concentration.

The colored complex is determined by a spectrophotometer at a wavelength of 410 nm.

5.5. Potassium

Potassium ions in a protein-free alkaline medium react with sodium tetraphenylboron to produce a finely dispersed turbid suspension of tetraphenylboron.

The produced turbidity is proportional to the concentration of potassium. This concentration is read photometrically at 578 nm.

6. Statistical Analysis

Statistical analysis of the minerals blood concentrations was established using SPSS Inc.'s"IBM SPSS Statistics 20" software, Chicago, IL, USA. We compared calcium, magnesium, sodium, phosphorus and potassium rates for the four sites (El Hadjeb, Sidi Okba, Ouled Djellal, and Aïn Zaatout).

This same software allows the determination of the mean and standard deviation of each blood mineral parameter and the comparison of the means of each parameter between fertilized and non-fertilized ewes and between sites using the Student's test (t test). The differences were considered significant when p < 0.05.

Results

The results of plasma mineral parameters in Ouled Djellal ewes from the different sites during the mating period are reported in Tables 5, and 6 below.

 Table 5: Variations in the concentration of major plasma minerals in Ouled Djellal ewes

 according to the region in fertilized ewes

Paramete	Site1 + 2	Site 3	Site 4	<i>p</i> value	<i>p</i> value	<i>p</i> value
r	Group I	Group II	Group III	Group	Group	Group
	(M ±SD)	(M ±SD)	(M ±SD)	Ivs II	I vs III	II vs III
Ca	1.70 ± 0.28	1.58 ± 0.41	1.26 ± 0.25	NS	<i>p</i> <0.01	<i>p</i> < 0.05
(mmol/L)					1	1
Р	1.13 ± 0.42	1.02 ± 0.19	0.68 ± 0.22	NS	<i>p</i> <0.01	<i>p</i> < 0.05
(mmol/L)						
Mg	0.84 ± 0.25	0.92 ± 0.33	0.99 ± 0.2	<i>p</i> <0.05	<i>p</i> <0.05	NS
(mmol/L)						
Na	145 ± 12.55	142 ± 10.25	142 ± 10.96	NS	NS	NS
(mEq/l)						
K	4.2 ± 0.55	3.9 ± 0.49	3.8 ± 0.81	NS	NS	NS
(mEq/l)						

M: mean, SD: standard deviation, NS: non-significant

Mean plasma calcium of ewes in the plain region differed significantly (p < 0.01) with ewes in the mountain region; however, plasma calcium of ewes in the plain region did not differ significantly from the tableland ewes.

In addition, plasma calcium of ewes in the tableland region differed significantly from the mountain ewes (p < 0.05).

Plasma phosphorus levels recorded in the plain region and tableland region were significantly higher than its level in the mountainous region (p < 0.01).

Also the plasma magnesium levels recorded in the mountain region and tableland region were significantly higher than the one recorded in the plain region (p < 0.05).

Table 6 : Variations in the concentration of major plasma minerals in Ouled Djellal ewes

 according to the region in non fertilized ewes

Mineral Paramete r	Sites 1 + 2 Group I (M ± SD)	Site3 Group II (M ± SD)	Site4 Group III (M ± SD)	<i>p</i> value Group I versus group II	<i>p</i> value Group I versus group III	<i>p</i> value Group II versus group III
Ca (mmol/L)	1.42 ±0.35	1.51 ±0.58	1.08 ±0.03	NS	<i>p</i> <0.01	<i>p</i> < 0.05
P (mmol/L)	1.11 ±0.29	0.98 ±0.54	0.63 ± 0.1	NS	<i>p</i> < 0.01	<i>p</i> < 0.01
Mg (mmol/L)	0.90 ±0.18	0.98 ±0.20	1.05 ±0.31	<i>p</i> <0.05	<i>p</i> <0.05	NS
Na (mEq/l)	147 ± 11.5	145 ± 8.4	139±9.58	NS	<i>p</i> <0.05	NS
K (mEq/l)	4.7 ±0.66	4.1 ±0.74	3.9 ±0.56	NS	<i>p</i> <0.05	<i>p</i> < 0.05

M: mean, SD: standard deviation, NS: non-signification

Table 7: Comparison of means of Minerals between fertilized and non fertilized ewes

Mineral Parameter	<i>p</i> value Sites 1 + 2 Group I	<i>p</i> value Site3 Group II	<i>p</i> value Site4 Group III	
Ca (mmol/L)	Ca (mmol/L) <i>p</i> <0.05		<i>p</i> <0.01	
P (mmol/L)	Non Significant	Non Significant	Non Significant	
Mg (mmol/L)	Non Significant	Non Significant	Non Significant	
Na (mEq/l)	<i>p</i> <0.05	<i>p</i> <0.05	<i>p</i> <0.05	
K (mEq/l)	<i>p</i> <0.05	Non Significant	Non Significant	

The statistical mean of the calcemia is significantly different between fertilized and unfertilized ewes in all groups.

No significant difference was observed between fertilized and unfertilized ewes in terms of plasma phosphorus and plasma magnesium.

Plasma sodium decreased significantly (p < 0.05) in fertilized ewes as compared with non-fertilized ewes.

Plasma potassium increased significantly (p < 0.05) in non-fertilized ewes as compared with fertilized ewes in the plain region.

Discussion

Table 8: Variation in the concentration of the major plasma minerals

Parametr		Site1 + site2	Site3	Site4	<i>p</i> value	<i>p</i> value	<i>p</i> value
		GrI	GrII	GrIII	Gr I	Gr I	Gr II
		(M±SD)	(M±SD)	(M±SD)	versus	versus	versus
					Gr II	Gr III	Gr III
Ca	Fer-ewe	1.70 ± 0.28	1.58 ± 0.41	1.26 ± 0.25	NS	<i>p</i> <0.01	<i>p</i> < 0.05
(mmol/L)	Non Fer	1.42 ± 0.35	1.51 ± 0.58	1.08 ± 0.03	NS	<i>p</i> <0.01	<i>p</i> < 0.05
	<u>p value</u>	<i>p</i> <0.05	<i>p</i> <0.05	<i>p</i> <0.01			
Р	Fer-ewe	1.13 ± 0.42	1.02 ± 0.19	0.68 ± 0.22	NS	<i>p</i> <0.01	<i>p</i> < 0.05
(mmol/L)	Non Fer	1.11 ± 0.29	0.98 ± 0.54	0.63 ± 0.1	NS	<i>p</i> <0.01	<i>p</i> < 0.01
	<u>p value</u>	NS	NS	NS			
Mg	Fer-ewe	0.84 ± 0.25	0.92 ± 0.33	0.99 ± 0.2	<i>p</i> <0.05	<i>p</i> <0.05	NS
(mmol/L)	<u>NonFer</u>	0.90 ± 0.18	0.98 ± 0.20	1.05 ± 0.31	<i>p</i> <0.05	<i>p</i> <0.05	NS
	<u>p value</u>	NS	NS	NS			
Na	Fer-ewe	145 ± 12.55	142 ± 10.25	142 ± 10.96	NS	NS	NS
(mEq/l)	Non Fer	147 ± 11.5	145 ± 8.4	139±9.58	NS	<i>p</i> <0.05	NS
	<u>p value</u>	<i>p</i> <0.05	<i>p</i> <0.05	<i>p</i> <0.05			
K (mEq/l)	Fer-ewe	4.2 ± 0.55	3.9 ± 0.49	3.8 ± 0.81	NS	NS	NS
	Non Fer	4.7 ± 0.66	4.1 ± 0.74	3.9 ± 0.56	NS	<i>p</i> <0.05	<i>p</i> < 0.05
	<u><i>p</i> value</u>	<i>p</i> <0.05	NS	NS			

M: mean, SD: standard deviation, NS: non-significant, Fer-ewes: fertilized ewes, Non Fer: non-fertilized ewes

Results of ewes at plain sites (sites 1 and 2) were gathered in one group, that is, group 1, whereas the tablelands were named group 2 and 3 designated the mountain ewes. Lot of non-fertilized ewes in group 1 that is composed of the ewes at sites 1 and 2 which were 15 ewes, and this same lot (of non fertilized) is composed of 9 ewes in group 2 located in the tableland area (site 3) and 23 ewes in group 3 located in the mountains (site 4).

The plasma calcium values that we recorded in this study are below the physiological norms cited by several authors (Brugère-Picoux 2004; Kaneko et *al.*, 2008; Brouček et *al.*, 2009; Elnageeb and Abdellatif, 2010; Abarghani et *al.*, 2013).

Moreover, the majority of ewes have low plasma calcium, which indicates an insufficient dietary intake of calcium. This insufficient intake would lead to a decrease in the fertility of ewes especially for ewes reared in mountainous regions (Friot and Calvet, 1971).

In addition, the significant decrease in plasma calcium in mountain ewes compared with other regions in this study may be due to an excessive demand for muscular activity in these animals (Sejian et *al.* 2012; Titaouine and Meziane, 2015); these results disagree with those of Brouček et *al.*, (2009) who studied the effect of various factors on blood minerals in sheep and found that sheep's serum calcium is not influenced by altitude.

A significant increase (p<0.001) of plasma calcium in fertilized ewes relative to plasma calcium in non-fertilized ewes in all regions. This increase in plasma calcium is probably due to increased intestinal absorption and bone resorption of Ca due to the hormonal change during the estrous period (Takagi and Block, 1991; Abdelrahman et *al.*, 2002).

The plasma concentrations of calcium and phosphorus are controlled by the same hormone-dependent homeostatic mechanism (Hadžimusić and Krnić, 2012). The lower plasma concentration of Ca and P in this study might have been the result of decreased levels in plants of all regions.

This can be supported by the fact that when this study was carried out, the plants had dry, and turned to brown, and withered leaves and therefore had lower levels of mineral content (Khan et *al.*, 2007). The significant decrease in plasma phosphorus in mountain ewes could be a support for severe muscle exercises when walking in this region (Titaouine and Meziane, 2016).

In all groups, fertilized ewes have a high plasma phosphorus concentration compared to non-fertilized ewes, even though the statistical analysis does not show a significant difference. With the decrease of plasma phosphorus for non fertilized ewes in comparison with fertilized ewes, Jacobson et *al.*, (1972) found that plasma P concentrations are maintained by the absorption of P from the gut, and there is no specific mechanism for bone P reabsorption.

In addition, positive relationships between P dietary intake and plasma P have been observed (Abarghani et *al.*, 2013).

We observe that plasma magnesium increases with altitude. Our results are in contradiction with those of Masters et *al.*, (1993), who found no significant difference (p>0.05) in plasma magnesium related to altitude.

Differences in elevated regional plasma levels are likely due to differences in soil types, precipitation, and pasture types (Masters et *al.*, 1993); the low plasma values of this element for both classes of fertilized and non fertilized ewes in the lowlands could be attributed to high potassium intake in this region, as it has been well established that this macroelement interferes with the absorption of magnesium (Ghosh and Shashank 2008; Sowande et *al.*, 2008).

Although there is no significant difference between the two groups of ewes in plasma magnesium in all regions, it should be noted that fertilized ewes have low plasma magnesium concentration. The decrease in magnesium plasma concentrations observed in fertilized ewes can be demonstrated by low sodium (Na) content. The low level of Na in the diet or in water may suggest hypomagnesemia by decreasing the absorption of Mg by the intestinal mucosa (Schweigel and Martens, 2000).

Table 8 shows a decrease in natremia in mountain and plateau ewes compared with ewes in plain. On the other hand, the statistical analysis of the values obtained gives a significant difference (p< 0.05) only between non fertilized ewes in the plain and those in the mountains. Indeed, the adiabatic thermal gradient which suppose that the plain region will be hotter than the other regions, "we lose 1 °C every 100 m altitude"; therefore, the ewes of this region will be more dehydrated than other ewes; all authors report an increase in plasma sodium concentration if dehydration took place.

This increase is due to the decrease in glomerular filtration and tubular reabsorption of sodium. Indeed, the activation of ADH during dehydration promotes a high absorption of water and a slight reabsorption of sodium from the digestive tract under the action of aldosterone (Maloiy and Clemens, 1980).

In our study, it can be noted that fertilized ewes have lower plasma sodium and potassium concentrations than non fertilized ewes as (Elnageeb and Adelatif 2010). They attributed this situation to the increase in progesterone levels in the blood of this ewe's

category. In addition, other studies by Deghnouche et *al.*, (2013) and Laidlaw et *al.*, (1962) have described the increase in sodium excretion during progesterone administration and have suggested that the latter has an aldosterone antagonist effect on the renal tubules.

The evolution of plasma potassium in the three regions follows the same pattern as natremia. Consequently, the significant decrease (p < 0.05) of plasma potassium in the mountainous region in non fertilized ewes results from the decrease of plasma sodium in these ewes, which subsequently leads to an increase in aldosterone, which decreases the plasma potassium level maintaining the tubular elimination of potassium (Yokus et *al.*,2004).

Conclusion

The study of the influence of mineral parameters shows that fertilized ewes have high plasma concentrations of calcium and phosphorus during the mating period. In contrast, plasma levels of magnesium, sodium, and potassium were higher in non fertilized ewes.

In general, the concentrations of the mineral parameters studied are below physiological norms in the four regions. So we must add to the feeding of ewes either in the mountain, in tableland, or in the plains a mineral supplementation which will probably increase the blood levels of these minerals especially for calcium and phosphorus.

In all farms, ewes at the end of gestation or at the beginning of lactation do not receive any food supplementation. Also the breeders do not use the lick stones and do not add salt to the ration.

Taking the results together suggests that providing minerals right before the mating is a common practice, but it doesn't give enough time for the ewes body to get the most benefit from the minerals. Instead, a longer period "60" days before mating and continuing feeding throughout gestation is the ideal. A longer timeline helps to maximize ingestion, achieve optimal breeding results and correct mineral deficiencies in sheep.

EXPERIMENT 2: Study of the Influence of the Altitude on Reproduction Parameters

Experiment 2: Study of the Influence of the Altitude on Reproduction Parameters

Materials and Methods

1. Study Settings

This experiment took place during the months May - October at the level of the four sites previously mentioned in Biskra, Algeria (more details could be checked in "Details of the study area").

2. Choice and Distribution of Herds

The selected animals are clinically healthy and nonpregnant Ouled Djellal ewes, aged 2.5 - 05 years. All the ewes had successfully lambed by the previous year, and the interval between the last lambing and the beginning of the experiment was \geq 70 days.

Rams are totally isolated from the females and are introduced into the herd only during mating period.

Locations	Landform	Altitude (m above sea level)	Number of Studied Ewes (heads)
site 1: El hadjeb	Plain	139	30
site 2: Sidi Okba	Plain	54	60
site 3: Ouled Djellal	Tableland	450	70
site 4: Ain Zaatout	Mountain	1000	66

Table 9: Some details on the study sites and herds' distribution

3. Livestock management

The study was based on 226 healthy ewes of the Ouled Djellal breed, aged between 2 and 5 years. The selected animals are clinically healthy and nonpregnant. They were divided into four herds as shown in Table 9 above. All ewes were identified through ear loop tags which facilitated our task a way more. Basically the same conditions were maintained as in the previous experiment (For more details about livestock management check out Materials and methods in experiment 1).

4. Synchronization of Oestrus and Detection of Gestation

Oestrus synchronization is realized by the use of vaginal sponges impregnated with 20 mg of fluogestone acetate FGA (Chronogest ®), and kept in for a duration of 14 days (09 th to 23 rd of May). After each introduction, the applicator is soaked in a bucket containing a solution of Potassium Permanganate's disinfectant to prevent the transmission of germs from one ewe to another.

14 days later, The sponges were removed and (PMSG) Pregnant Mare Serum Gonadotropin (Folligon \mathbb{R} at 1000 IU/UI: lyophilisate injection + solvent) was injected at 400 IU / ewe. The injection is performed by an intramuscular syringe; the needle of the syringe was changed at each injection to avoid any possible contamination. 24 hr later the ewes were put to natural mate.

Gestation was detected on the basis of non-return on oestrus after two observation periods (19 to 23 th and 40 to 44 days after mating period) as reported by Ouedraogo et *al.* (2008), where gestation was determined 30 - 60 days following the mate by a simple abdominal palpation (Ahmed et *al.*, 1998).

The studied reproduction parameters were fertility, prolificacy, and fecundity.

Fertility rate

Number of lambing ewes/Number of ewes put to mate

Prolificacy rate

Number of born lambs/Number of lambing ewes

Fecundity rate

Number of born lambs/Number of ewes put to mate

All these parameters were calculated according to Charray et al. (1992).

5. Statistical Analysis

Statistical analysis of the reproductive variables was established using SPSS Inc.'s"IBM SPSS Statistics 20"software, Chicago, IL, USA. We compared fertility, fecundity, and prolificacy rates for the four sites (El Hadjeb, Sidi Okba, Ouled Djellal, and Aïn Zaatout).

We used χ^2 test to verify the link between the measured rates and the four sites, in addition to multiple comparisons that revealed sites with significant differences.

For the interpretation of the parameters of reproduction, we considered the following formulas:

<u>Fertility:</u>	Yes : fertile ewe ; N	No: infertile ewe
Fecundity:	0: zero born lamb;	1: only one born lamb ; 2: two born lambs
Prolificacy:	1: simple birth ;	2: double birth

The differences were considered significant when p < 0.05.

Results

Results of the reproduction parameters of the different sites are gathered in Tables 10 and 11, and illustrated in a bar chart in Figure 11.

	Ewes	Lambing	Infertile	Lambs	Simple	Double
	number	ewes	ewes	number	gestation	gestation
					number	number
Site 1	30	21	9	25	17	4
Site 2	60	54	6	64	44	10
Site 3	70	61	9	75	47	14
Site 4	66	43	23	47	39	4
Total	226	179	47	211	147	32

Table 10: The numbers registered in the studied sites

In the light of these findings, we observe that the average fertility rate of ewes was 78%; that is to say, there are 78 lambing ewes and 22 non-lambing (unfertilized), the average prolificacy rate of the studied ewes was 117% yielding 17 more lambs for 100 lambs, and the average fecundity rate was 92%.

Fertility averages that are below the overall average were recorded in the herds of site 1 (plain region) and site 4 (mountain region), which were 0.7 and 0.65, respectively, but the averages for herds at sites 2 and 3 were above the overall average and were, respectively, 0.9 and 0.87.

Concerning the third reproduction parameter, we observe that the average fecundity is below the general average; it was recorded in the flocks of sites 1 and 4 which were 0.83 and 0.71, respectively. On the other hand, the herd averages for sites 2 and 3 were higher than the general average, which were 1.06 and 1.07, respectively. Site 3 showed the best fecundity rate.

The results recorded at the four studied sites are gathered in the table below. These results clearly show and gather the calculated rates of the three reproduction performances' parameters as well as the results of the statistical analysis.

_	Fertility	Prolificacy	Fecundity
Site1	0.7	1.19	0.83
Site2	0.9	1.18	1.06
Site3	0.87	1.23	1.07
Site 4	0.65	1.09	0.71
Mean ± standard	0.78±0.12	1.17±0.06	0.92±0.18
deviation			
Pearson's	22.829	6933	29.963
chi-square			
P value	0.001	0.074	0.0001
Site 1 vs 2 vs 3 vs 4			
P value	0.016	/	0.056
Site1 vs site 2			
P value	0.041	/	0.116
Site 1 vs site 3			
P value	0.589	/	0.341
Site 1 vs site 4			
P value	0.611	/	0.741
Site 2 vs site 3			
P value	0.0001	/	0.0001
Site 2 vs site 4			
P value	0.0001	/	0.0001
Site 3 vs site 4			

Table 11 : Averages of fertility, prolificacy and fecundity of Ouled Djellal ewes

Herds in site 2 (tableland region) showed the best fertility rate, and the statistical study showed that the region influences fertility. Comparisons two to two of fertility rates show that the difference is significant between site 1 and site 2 and between site 1 and site 3 and very highly significant between site 2 and site 4 and between site 3 and site 4.

Prolificacy was found in the herds of sites 1, 2, and 3 higher than the general average and higher than the average recorded in the herd of site 4. The ewes of site 3 have the best prolificacy rate, and no significant difference was found between herds relating to the region.

Like fertility, the statistical study showed that the region significantly influences fecundity: highly significant difference between site 2 and site 4 and between site 3 and site 4 and no significant difference between the fecundity rates between site 1 and site 3, site 1 and site 4, and the site 2 with site 3.

The bar chart below illustrates the rates of the three studied parameters of reproduction fertility, prolificacy, and fecundity in the four sites.





It is apparent that the rates of fertility rose slightly in sites 2 and 3 which correspond to the plain and tableland regions respectively(it oscillates between 87 % and 90 %), whilst fertility rates fell slightly in mountain region site 4 and plain region site 1.

Also it can be seen that the recorded rates of prolificacy remained close to 120 % in the sites 1, 2 and 3 both plain and tabeland regions, but it can be noticed that the prolificacy decreased to 109 % in the mountainous region.

Fecundity rates have dropped to 71 % in the mountainous regions (site 4) compared to plain and tableland regions where the recorded rates reached 107 % in site 3 in tableland and between (83 % and 106 %) in plain regions (sites 1 and 2).

Overall the best reproduction performances were recorded in tableland region (site 3) good performances were also recorded in plain regions (sites 1 and 2) the lowest performances were recorded in the mountainous region (site 4).

Discussion

1. Fertility

Discussing the findings of fertility of the present study, we found lower rates of fertility compared with those recorded in Ouled Djellal breed (100%) cited by (Chellig, 1992), with a difference of 22%, and the rates recorded in Ouled Djellal breed described by (Dekhili, 2004) in Setif region, where the average was 92% and those of (Safsaf and Tlidjane, 2010) which was 91% with a difference of 14% and 13% respectively.

The bar chart below (Figure 12) compares the rates of fertility as a parameter of reproduction in the four sites.



Figure 12: Bar chart showing the recorded fertility rates in the four sites.

If we look at the four studied sites we can say that good rates of fertility were recorded in general, they are all above 65%. These rates go from 65 % in site 4 to 90 % in site 2. The rate of fertility nearly remained the same in sites 2 and 3. Fertility rates are average among the three parameters.

Also it can be seen that the recorded rates of fertility remained close to 90 % in site 2 and 3, but it can be noticed that the fertility decreased to 65 % in the mountainous region.

Overall the lowest performances were recorded in the mountainous region (site 4) and the highest reproduction performances were recorded in plain region (site 2).

Among certain Moroccan breeds described by Chikhi and Boujenane (2003) as the Boujaâd, Sardi, Timahdite, and Beni Guil breeds, which varied on average by 98%, 85 to 92%, 77 to 95%, and 82 to 87%, respectively, with differences of 20%, 7 to 14%, 1 to 17%, and 4 to 9%, respectively, pointed out that the effect of rearing conduct particularly food supply during mating period and the absence of flushing technique are of an elementary role and are mainly behind this drop, a thing that is early revealed by authors like Abbas et *al.* (2004) and Deghnouche (2011) who reported that good quality feeding improves ewes' fertility.

Another reason could also interfere which is drought-related dietary conditions in the study area. Thus, the fertility rate is influenced by the season and the month of mating (Dekhili and Aggoun, 2004).

The best performances of fertility were obtained when fertilization took place, while the food supply was in large quantities and of a good quality.

2. Prolificacy

The average prolificacy of Ouled Djellal ewes was 117%, and our results are lower than the rates recorded in Ouled Djellal ewes described by Dekhili (2004) and Deghnouche (2011) which were 130% and 147% with a difference of 13% and 30%, respectively, and lower than the rates recorded in the D'Man breed ewes described by Chellig (1992) which varied from 185 to 200% with differences from 68 to 83%.





The bar chart above (Figure 13) compares the rates of prolificacy as a parameter of reproduction in the four sites.

If we look at the three studied parameters of reproduction we can say that good rates of prolificacy were recorded in the four sites, they are all above 109%. These rates go from 109 % in site 4 to 123 % in site 3. The rate of prolificacy remained the same in sites 1 and 2 of the plain region. Prolificacy rates are the highest among the three parameters.

Overall the lowest performances were recorded in the mountainous region (site 4) and the highest reproduction performances were recorded in tableland region (site 3).

However, our results are similar to the rates recorded in Hamra breed described by Chekkal et *al.* (2015) and the Moroccan Sardi breed described by Chikhi and Boujenane (2003) which varied respectively from 110 to 120% and from 107 to 121%, and our findings exceeded the rates recorded by Dekhili and Aggoun (2007) in Ouled Djellal breed in Sétif region which was 109%.

Prolificacy rate is influenced by the feeding of ewes during flushing. Chentouf et *al.* (2003) report that providing supplements to ewes before and during the mate period have led to an improvement in the ovulation rate and consequently the rate of prolificacy. Clement et *al.* (1997) report that sufficient and good quality feeding during the mate could favor double births.

3. Fecundity

The average fecundity of ewes was 92%, and this rate was lower than the rates recorded for Ouled Djellal, Rembi, and Barbarine breeds described by Chellig (1992) that was 95%, with a difference of 3%, and lower than the rates cited by Deghnouche (2011) which varied between 110 and 125%, and higher than the rate recorded by Merghem (2009) which was 70% for Ouled Djellal type Hodna breed in the region of Sétif.

The bar chart below (Figure 14) compares the rates of fecundity as a parameter of reproduction in the four sites.



Figure 14: Bar chart showing the recorded fecundity rates in the four sites.

Fecundity rates have dropped to 71 % in the mountainous regions (site 4) compared to plain and tableland regions where the recorded rates reached 107 % in site 3 in tableland and between (83 % and 106 %) in plain regions (sites 1 and 2). Overall the lowest performances were recorded in the mountainous region (site 4) and the highest reproduction performances were recorded in tableland region (site 3).

The differences between the results obtained in the present study and those recorded by the different authors can be explained by racial differences, the mode of rearing, and the mode of feeding (feeding of ewes during the mate periods).

In this study, flocks were extensively reared with inadequate feeding which barely covered maintenance requirements. This undernourishment, especially during the mate period, negatively affects the reproductive potential of the studied ewes.

As conclusion, the results of this study indicate clearly the presence of differences in reproductive performances between the four herds. We consider the herd located in plain region of Sidi Okba (site 2) as the most fertile and the herd located in tableland region of Ouled Djellal (site 3) as the most prolific and most fecundable, while the herd 4 located in mountain region of Aïn Zaatout (site 4) is the least fertile herd, the least fecundable, and the least prolific.

It is known that the performance of an animal is determined by the expression of its genotype, the influence of the environment in which it evolves, and the interaction between these two factors (Dekhili and Aggoun, 2006).

Therefore, the difference in the reproductive performance between the four flocks from the four sites is the first explanatory element of this reaction of Ouled Djellal ewes from one site to another, and the inferiority of the fertility, prolificacy, and fecundity rates at site 4 (Aïn Zaatout) can be explained by the decrease in progesterone levels in ewes at high altitudes (Barnett et *al.*, 1978).

The same authors report that high levels of progesterone in the blood can improve ovulation rates, embryo survival, and nesting of fertilized eggs in the uterus and consequently reproduction rates.

The control of livestock management and the improvement of food supply, as well as the availability which is uncertain in arid zones, can lead to a significant improvement in breeding parameters.

Conclusion

The table below compares the studied parameters of reproduction fertility, and fecundity in the four sites.

If we look at the three studied parameters of reproduction we can say that good rates of prolificacy were recorded in the four sites, they are all above 109%. These rates go from 109 % in site 4 to 123 % in site 3. The rate of prolificacy remained the same in sites 1 and 2 of the plain region. Prolificacy rates are the highest among the three parameters, yet no significant difference was found between sites of the study.

Fertility	Site 1 vs.	Site 2 vs.	Site 3 vs.	Site 4 vs.
Site 1	-	7	7	\leftrightarrow
Site 2	\checkmark	-	\leftrightarrow	$\downarrow\downarrow$
Site 3	7	\leftrightarrow	-	$\downarrow\downarrow$
Site 4	\leftrightarrow	↑ ↑	↑ ↑	-
Fecundity	Site 1 vs.	Site 2 vs.	Site 3 vs.	Site 4 vs.
Site 1	-	7	\leftrightarrow	\leftrightarrow
Site 2	7	-	\leftrightarrow	$\downarrow \downarrow$
Site 3	\leftrightarrow	\leftrightarrow	-	$\downarrow\downarrow$
Site 4	\leftrightarrow	↑ ↑	↑ ↑	-
very signific:	antly lower	↑↑ very significa	antly higher	

Table12 : Comparison of the studied reproduction parameters according to the site of study

↓ very significantly lower
 ↑↑ very significantly higher
 >> significantly lower
 >> significantly higher

 \leftrightarrow no significant difference

N.B. The table must be read from upper cases to lower ones; i.e. site 1 showed significantly lower fecundity rates comparing to site 2

Also it can be seen that the recorded rates of fertility remained close to 90 % in site 2 and 3, but it can be noticed that the fertility decreased to 65 % in the mountainous region. Fecundity rates have dropped to 71 % in the mountainous regions (site 4) compared to plain and tableland regions where the recorded rates reached 107 % in site 3 in tableland and between (83 % and 106 %) in plain regions (sites 1 and 2).

Overall the lowest performances were recorded in the mountainous region (site 4) and the highest reproduction performances were recorded in tableland region (site 3).

In conclusion, the study proved the presence of a characteristic site effect from which Ouled Djellal ewes do not show their best reproduction performances at high altitudes as illustrated in the figure 14 reporting all the findings.

On the other hand, rehydrating the ewes through distribution of water during grazing or digging wells in the pastures, to combat the dehydration of ewes, which is in this study a limiting factor for the ewes to be fertilized.



Experiment 3: Study of the Influence of Body Condition Score on Some Blood Parameters and on Reproduction Parameters

Materials and Methods

1. Study Region

This study was carried out on ewes belonging to a state farm: The technical institute for the development of Saharan agriculture I.T.D.A.S., located in Ain Ben Noui, Ehadjeb to the west of Biskra (northeast of Algeria). The farm is located on the 46 th national road towards Tolga, about 7 km to the west of the town of Biskra. It occupies an area of 83 hectares, the dedicated area for sheep grazing is approximately 900 m² (Altitude: 80 Latitude: 34 °, 30 N Longitude: 05 °, 38 E). It is built on a plain area, and it is watered naturally.(more details on the study area could be found in the previous chapter about details of the study area)



Figure 8: view of satellite of the location of the study region (Google Earth 2021 ITDAS)

2. Choice and Distribution of Herds

At the beginning of April, thirty clinically healthy, non-lactating, non-pregnant Ouled Djellel ewes, aged 2.5 to 5 years, having an average body condition score of 2.8 ± 0.9 and weighing 54.83 kg \pm 9.4 were assigned into two groups according to their body condition score. (Group I: BCS < 2.50 units; n = 17, Group II: BCS > 2.50 units; n = 13) using a scale from 1 (emaciated status) to 5 (extremely fat), assessed by the same well-experienced person, each of the groups was housed in a large barnyard. Animals were fed different quantities of

the same diet. All the ewes had successfully lambed the previous year, also at least 70 days separated the date of lambing from the day of the beginning of the experiment. Rams are totally isolated from the females and are introduced into the herd only during periods of mate.

3. Livestock Management

Ewes were being reared in a semi-extensive system as described in the following:

3.1. Feeding Approach

The main objective of the ITDAS farm is the production of seeds of major cereal crops (soft wheat, durum wheat, barley); edible legumes (faba beans, lentils and chickpeas) and forages. Intending to meet the food needs of the livestock throughout the year through cultivating of phoenicultural areas and recycling of fodder crops to produce green throughout the year. Alfalfa is the staple crop, the winter off-peak is fulfilled by green barley and forage rapeseed. At the end of June, sorghum and sesbania culture are produced until mid-October.

Basically similar conditions were maintained as described in the previous experiments

4. Blood Sampling

This study was carried out during 1 breeding campaign the samples were taken from the ewes clinically healthy by puncture of the jugular vein on morning before food intake. Blood was collected in vacutainer dry tubes. To limit the consumption of blood glucose by red blood cells and degradation of metabolites due to enzymatic action, after clotting, samples of blood were centrifuged at 2500rpm, for 10 minutes within an hour and a half from sampling. Indeed, if the sample is left at room temperature without being centrifuged, the glucose concentration drops 5-10% per hour. The corresponding sera were divided into aliquots and stored at -20 ° C till analysis. Blood samples were taken from all the ewes of the study early in the morning. The ewes were distributed into two groups:

-Group I: BCS < 2.50 units; n = 17,

-Group II: BCS > 2.50 units; n = 13.

-D0 corresponds to the day of insertion of the intra vaginal sponges. (May 9th)

-The First sampling: D 14 corresponds to the day of removal of the sponges and PMSG injection.(May 23rd)

5. Synchronization of Oestrus

Oestrus cycles of the thirty ewes were synchronized during the first week of May (day 1), by the insertion of an intravaginal sponge containing 20 mg fluogestone acetate (CHRONOGEST®, MSD Santé Animale)and kept in for 14 days. After this period, at the day of sponge removal (day 14), ewes received an i.m. injection of 400 IU of PMSG (Folligon ®, MSD Santé Animale) to synchronize ovulation. The following day (day15), the fertile ram will be introduced in each of the groups for natural mate (Thimonier et *al.*, 2000). Then the ram was withdrawn 48 h after its first introduction (day 17).

6. Scoring of Body Condition and Weighing of Ewes

The scoring method used is that of lumbar palpation by the same person who assigns marks according to a scoring chart ranging from 0 to 5 following (Russell et *al.*, 1969) method. In the end only ewes of adequate score were subject to the study (2-4). The ewes were then classified according to their body condition scores. Weighings were also carried out on ewes using (automatic scales) offered for this purpose performed by the farmers in the site of the study and after 12 hours of fasting in order to get more accurate weighings following the F.A.O., 2013 recommendations.

7. Analysis and Choice of Biochemical Variables

Blood concentrations of different metabolites have been determined by spectrophotometry using dosage kits of (SPINREACT) Spain, glucose, cholesterol, triglycerides, creatinine, urea and albumin as well as the dosage of the total proteins was carried out by the same kits (SPINREACT) Spain, and read on the spectrophotometer (UV-2005 . SELECTA).

The choice of the biochemical variables to study was delicate; however, for some technical issues (availability of reagents mainly), it was necessary to determine the parameters which would be the most relevant, without forgetting the clinical interest which remains crucial.

Hormonal parameters T3, T4, and progesterone were estimated by radioimmunoassay (RIA) using a gamma counter (PC-RIA MAS; Stratec, Germany) employing RIA kits supplied by Tecan IBL International GmbH, Hamburg, Germany.

Parameter	Analytical Method	Reference
Glucose	Trinder. GOD-POD	1001191
Triglycerids	GPO-POD. Enzymatic Colorimetric	1001311
Cholesterol	CHOD-POD. Enzymatic Colorimetric	1001091
Albumin	Bromocresol green. Colorimetric	1001020
Total Proteins	Biuret. Colorimetric	1001291
Creatinine	Modified JAFFE Method	-
Urea	Bertholet. Enzymatic Colorimetric	-

Table 13: Analytical methods used to measure the studied blood parameters rates

8. Statistical analysis

Statistical analysis of the blood and hormonal variables was established using "IBM SPSS Statistics 20" software from SPSS Inc. Chicago, Illinois, USA. This software allows the determination of the mean and standard deviation of each parameter, the comparison of two means of each parameter between group I and group II using the Student's test (t-test). The differences were considered significant when p < 0.05.

Results and Discussion

The results of the effect of BCS on blood parameters and hormonal profiles in Ouled Djellal ewes are reported in table 14 below.

 Table 14: Variation of biochemical parameters and hormonal profiles according to the BCS

 during mating period in Ouled Djellel ewes

Parameters	Group I	Group II	<i>p</i> value
	(M±SD)	(M±SD)	Group I Vs. Group II
Glucose (g/l)	0.49±0.36	0.42±0.25	<i>p</i> < 0.05
Cholesterol (g/L)	0.68±0.25	0.49±0.19	<i>p</i> < 0.05
Triglycerides (g/L)	0.27±0.1	0.35±0.03	<i>p</i> < 0.01
Total Proteins (g/L)	58.74±3.74	64.89±4.85	NS
Albumin (g/L)	25.71±2.31	28.89±4.57	<i>p</i> < 0.05
Creatinine (mg/L)	11.14±1.9	12.4±2.3	NS
Urea (g/l)	0.28±0.6	0.25±0.09	NS
T3 Triiodothyronine (nmol/l)	1.90± 0.21	2.20±0.15	<i>p</i> < 0.01
T4 Thyroxine (nmol/l)	148.0±11.39	160.4±8.71	<i>p</i> < 0.01
Progesterone (ng/mL)	5.3±0.96	4.8±0.8	NS

Results are expressed as Mean (M) ± standard deviation (SD). NS=Non-significant

1. Glucose

Glucose is essential for vital organ functions, fetal growth and milk production. Thus, any deficiency in this metabolite leads to a decrease in fetal growth, a reduction at birth weight and an increase in perinatal mortality (Moallem et *al.*, 2012).

In the present study, the results for both groups fall within the range of international results (0.4 - 0.7 g / L). However, our results were superior to those obtained by (Yagoubi and Atti, 2020) (0.11 - 0.15 g/L), and lower than those obtained by (Carlos et *al.*, 2015) (0.60 - 0.63 g/L) in ewes of Morada Nova breed.

In the present study, plasma glucose concentrations were significantly (p < 0.05) affected by BCS similar to the results of (Moeini et *al.*, 2014) and (Sitaresmi et *al.*, 2020), unlike (Jalilian and Moeini, 2013) who found that ewes with different BCS did not affect glucose levels.
However, (Caldeira et *al.*, 2007) recorded a different metabolic status for ewes with different BCS with lower glycaemia for lean animals (BCS between 1 and 2) than for fatty animals (BCS between 3 and 4).

In our study, Glycaemia in group I was higher than that in group II, this hyperglycemia is probably due to the low insulinemia in group I, because their needs are higher than those provided by the ration, particular as regards to energy during the mating period, animals will use their body reserves to compensate for the deficit, unlike the ewes of group II, which did not mobilize their reserves sufficiently.

Moreover, animals with higher plasma glucose levels had more energy, which could be used for improving reproductive performance, as shown in previous studies (Yagoubi and Atti, 2020) (Stefańska et *al.*, 2016).

2. Cholesterol

Cholesterol has an essential role that is acting as a precursor of steroid hormones and bile acids, as well as in the composition of cell membranes. The cholesterol concentrations in Ouled Djellal ewes are within the physiological standards reported by (Macías-Cruz et *al.*, 2017) (0.63g /L), but it appeared relatively low compared to the data cited by (Jalilian and Moeini, 2013) (0.93 - 0.99 g / L) and (Carlos, et *al.*, 2015) (0.71 - 0.74g / L).

There is a significant increase (p < 0.05) in cholesterol level in ewes with BCS < 2.5 in comparison with ewes with BCS > 2.5, this is consistent with the results obtained by (Özpinar and Firat, 2003). Thus, the increase in the value of cholesterol in group I, consequently leads to an increase in the concentrations of progesterone during the luteal phase compared to group II (Deghnouche et *al.*, 2019).

In addition, the nutritional status of ewes around the mating period affects the ovarian activity in several breeds of sheep; the energy deficit leads to the mobilization of lipid reserves and causes an increase in the concentration of cholesterol in group I.

On the other hand, the attribution of these changes to variations in thyroid activity. Thyroid hormones stimulate cholesterol synthesis as well as the hepatic mechanisms that remove cholesterol from the circulation. The decrease in plasma cholesterol because the level of the last process exceeds that of the first. Serum cholesterol concentration may be considered the most reliable indicator of dietary variation and indicates that blood cholesterol is highly correlated with various dietary intakes (Boudebza, et *al.*, 2016).

3. Triglyceride

The highest serum TG concentrations were observed in group II which can be attributed to an increased rate of TG synthesis in the intestinal mucosa, due to increased availability of substrates (Caldeira, et *al*, 2007).

In addition, (Mazur et *al.*, 2009) showed lower values for plasma triglycerides in undernourished ewes, while (Pesántez-Pacheco et *al.*, 2019) reported that ewes with a higher BCS during the postpartum had a higher triglyceride concentration than sheep with a lower BCS. Thus, the low plasma triglyceride level in group I could be linked to a lack of glucose in the ration compared to group II (Herdt, 2000).

4. Total Proteins

Total protein measured in this study remains close to physiological standards cited by (Carlos et *al.*, 2015) (60.2 - 68.4 g / L). However, they are higher than values reported by (Jalilian and Moeini, 2013) (47 – 55 g/L).

From the results of the changes in proteinemia, no significant difference was recorded between the two groups. This is the same conclusion found by (Caldeira et *al.*, 2007) (Sitaresmi et *al.*, 2020), total proteins were not affected by BCS in ewes.

The level of total proteins varies in the same direction as the evolution of the body condition; lower in animals with a low body condition score, than in animals with a high BCS (Caldeira et *al.*, 2007).

The nutritional level and particularly the protein consumption is closely related to the BCS. Thus, this is probably related to the nutritional or health status of the animal, since the level of plasma proteins may reflect the nutritional or health status of the animal. All of these proteins, synthesized by the liver, also serve as hepatic markers.

Indeed, the plasma concentration varies with the food intake; an increase in protein ingestion usually leads to an increase in protein synthesis at the body level, not to mention the protein synthesis, which is, reduced in (Group I) due to anabolic depression related to a hormonal upheaval.

5. Albumin

Albumin is a protein synthesized in the liver. It serves to maintain oncotic pressure, and transport thyroid hormones, fat-soluble hormones, free fatty acids, calcium, unconjugated bilirubin, and buffer the pH.

In group II, the level of albuminemia is higher than that of group I, this significant difference (p < 0.05) is probably related to the increase in calcium level due to the increase in parathyroid hormone (PTH) caused by estradiol, through hormonal activity, which requires high levels of albumin.

(Schenck and Chew, 2010) have also found that hypoalbuminemia can cause a decrease in the amount of protein bound calcium and therefore possible total hypocalcemia and vice versa.

In addition, this effect can be explained by the proportional availability of amino acids for albumin synthesis and by the role of albumin as a supplier of amino acids to the peripheral tissues when a nutritional shortage of protein occurs (Caldeira et *al.*, 2007).

6 Creatinine

Creatinine is formed by irreversible dehydration of creatine phosphate for the release of energy in skeletal muscles, and it is increased by the body's content of creatine which is directly related to muscle mass and therefore to the BCS (Samra and Abcar, 2012).

The creatinine concentration was high in group II, these results agree with the results reported by (Yagoubi and Atti, 2020) and (Widiyono et *al.*, 2020). Therefore, lower creatinine levels in group I were likely justified by low muscle mass and low protein turnover.

7. Uremia

Uremia is a good indicator of nitrogen intake and assesses the level of nitrogen feed in ruminants (Titaouine and Meziane, 2015). The indicated values of uremia are high in group I compared to group II. These values recorded in the two groups are in the reference intervals cited by (Kaneko et *al.*, 2008) (0.17-0.43g / l).

Comparison of means did not show significant differences between the two experimental groups. This increase of uremia in Group 1, which are undernourished ewes compared to Group II, can be explained by mechanisms adopted to conserve as much nitrogen as possible, increasing urea recycling in the rumen and decreasing its urinary excretion (Marini and Amburgh, 2003).

8. Progesterone

Progesterone is the main hormone involved in the maintenance of gestation and may play an important role in mediating the effect of nutrition on embryo development and that the high level of serum progesterone over the luteal phase of the estrous cycle is indicator to high conception rates (Safsaf et *al.*, 2012).

In addition, Progesterone is a hormone, which regulates various reproductive functions. Progesterone plays a key role in regulating the length of estrous cycle and implantation of blastocysts.

(Barnett et *al.*, 1978) report that high levels of progesterone in the blood can improve ovulation rates, embryo survival and nesting of fertilized eggs in the uterus and consequently reproduction rates. Progesterone concentrations of ewes in this study are illustrated in table 12. Ewes of group II had a decreased concentration of Progesterone compared to ewes of group I.

At the time of sponge withdrawal in ewes of group I, Progesterone concentrations averaged 5.3 ± 0.93 ng/ml, at same time in ewes of Group II, Progesterone concentrations averaged 4.8 ± 0.8 ng/ml. Similarly, a previous study demonstrated that Progesterone concentrations at the time of sponge removal averaged 5.1 ± 0.3 ng/ml (Van Cleeff et *al.*, 1998).

In fact, Progesterone exercises a negative feedback effect on the hypothalamus and pituitary to regulate gonadotropin release, mainly directed towards luteinizing hormone (LH) release (Dias et *al.*, 2015).

In group II, One possible explanation for this fact, the decrease in mean progesterone at the time of sponge removal would not be sufficient to suppress the hypothalamic-pituitary axis, leading to an estrus response (Moonmanee and Yammuen-art, 2015) (Blaschi et *al.*, 2014).

Hence, gestation rates in the group II was 10 among 17 ewes were pregnant unlike in the group I this rate was 11 among 13 ewes. Also, there was higher plasma progesterone concentration in group I than for Group II. This suggests that BCS around the mating period only slightly affects the circulating progesterone concentrations, but not significantly.

Thus, it is possible that progesterone levels in blood were a little increased with underfeeding before mating (Abecia et *al.*, 2006) (Macías-Cruz et *al.*, 2017). The reduction in gestation rates of ewes with peripheral progesterone concentrations above 5ng/ml was probably due to direct effects of undernutrition rather than elevated progesterone.

On the other hand, the increases in serum concentrations of progesterone are noted in response to feed restriction because feed restriction may have altered the metabolic clearance of progesterone (Safsaf et *al.*, 2012)

9. Triiodothyronine and thyroxine T3 and T4

Both T4 and T3 followed a similar direction in the present study. Their level decreased in group I. In addition, blood thyroid hormones are considered to be good indicators of the nutritional status of an animal (Sejian et *al.*, 2010).

Following feed restriction or food deprivation, plasma thyroid hormone concentrations were reduced in sheep (Rae et *al.*, 2002). These effects suggest that energy balance could play a major role in affecting the decrease in plasma thyroid hormone levels.

The decrease in thyroid hormones could be a response to negative energy balance. Nutrition plays a primary role in thyroid gland activity and in blood thyroid hormone concentrations (Stefańska et *al.*, 2016) (Sejian et *al.*, 2010) (Todini, 2007). Hence, nutrition restriction could have elicited a reduction in thyroid hormone concentration in group I.

Conclusion

The analysis of the results showed that the body condition score of the ewes has an influence on glucose, cholesterol, triglyceride, albumin and thyroid hormones (T3 and T4).

On the other hand the plasma concentrations of the total proteins, creatinine, urea, and progesterone have no significant difference during the mating period in ewes.

These findings are gathered and illustrated in one table 15 below. It is clear that very significant differences were found between ewes of two groups in the following parameters: Triglycerides, T3 and T4 where the lowest levels were recorded in ewes of smaller body score (group 1). Same with albumin where a significant drop was noticed in the group 1.

A slight but not significant increase in progesterone in group1 was observed too. The remaining parameters: Urea, Creatinine, Total proteins, did not differ between the groups.Unlike the glucose and cholesterol which were higher in group 1.

This experiment attempted to contribute in setting reproductive standards and to enrich the knowledge on the effect of body condition scores on reproductive performance and on hormonal and biochemical parameters in Ouled Djellal ewes, further studies should be carried out to promote these findings. **Table 15:** Comparison of the studied parameters according to the BCS during mating
 period in Ouled Djellel ewe

	Group 1 Vs. 2 Observation						
Glucose	7	Hyperglycemia is due to the low insulinemia in group I, due to their needs that are higher than those provided by the ration.					
Cholesterol	7	Thyroid hormones stimulate cholesterol synthesis as well as the hepatic mechanisms that remove cholesterol from the circulation. The decrease in plasma cholesterol because the level of the removal process exceeds that of the synthesis and vice versa.					
Triglycerides	$\downarrow\downarrow$	The low plasma triglyceride level in group I could be linked to a lack of glucose in the ration compared to group II.					
Total Proteins	\leftrightarrow	-					
Albumin	Ń	The increase in calcium level due to the increase in parathyroid hormone (PTH) caused by estradiol, through hormonal activity, which requires high levels of albumin.					
Creatinine	\leftrightarrow	-					
Urea	\leftrightarrow	-					
Т3	$\downarrow\downarrow$	The decrease in thyroid hormones is considered as a response to negative energy balance. Nutrition					
T4 Thyroxine	$\downarrow\downarrow$	plays a primary role in thyroid gland activity					
Progesterone	\leftrightarrow	_					
↓ very significant	-	ery significantly higher					

 \mathbf{Y}

significantly higher 7

significantly lower no significant difference \leftrightarrow

Conclusion

Conclusion

The importance of sheep production keeps increasing in all fields as it is a solid means of rural development and environmental sustainability. In Algeria, sheep produces have been a tool for ensuring food security mainly, even though it is a resource that is not properly exploited.

The first part of this research attempts to shed light on ewe reproduction and sheep rearing in Algeria as well as to some blood parameters assessment in ewes' body in order to provide a theoretical framework that helps the readers understand the field of study that has been discussed. It tackles the current situation of sheep breeding in Algeria and the development of some adopted techniques and methods in the field as a provider for both national and international markets. In addition, it presents a very brief overview on some blood parameters assessment in ewes.

The second part was first devoted to the geographical framework and background; it dealt mainly with details of the study area.

Within the experimental part, we have described the conducted study on sheep rearing, its background, sub-fields and its occurrence in the studied areas. It is mainly devoted to the research situation and methodology. It presents the beginning of the fieldwork general procedure and blood analysis.

This chapter presents and discusses the findings of the conducted research. Divided into two experiments, the first experiment studied the effect of altitude on some blood parameters; the second examines the influence of altitude on reproduction parameters.

The first two experiments were conducted in 4 sites of different altitudes revealing the characteristics of breeding in the arid environment at Biskra (Eastern Algeria). The study dealt with 226 Ouled Djellal ewes, divided into 4 herds, where the aim was evaluating the performance of reproduction in ewes and highlighting the values of some blood parameters, including analyzing the influence of the region on these parameters in Ouled Djellal sheep.

One of the main concerns of this research is the implementation of the measurement of blood parameters within the development of analyzing the general state of the ewes and adjusting a more adapted conduct of rearing. This is why we undertook the third experiment; in order to reveal the effect of the body condition score of ewes, on some blood parameters that were measured in relation to reproduction parameters.

This work is devoted to the study of both dependent (reproduction parameters) and independent (altitudes) variables. It relies on the reproduction parameters: fecundity, fertility and prolificity.

In short, the study proved the presence of a characteristic site effect from which Ouled Djellal ewes do not show their best reproduction performances at high altitudes. The study of the influence of mineral parameters shows that fertilized ewes have high plasma concentrations of calcium and phosphorus during the mating period.

In contrast, plasma levels of magnesium, sodium, and potassium were higher in nonfertilized ewes. In general, the concentrations of the mineral parameters studied are below the physiological norms in the four regions. So it is a must to add to the feeding of ewes either in the mountain, in tableland, or in the plain mineral supplementation which will probably increase the blood levels of these minerals especially for calcium and phosphorus.

The analysis of the results showed that the body condition score of the ewes has an influence on glucose, cholesterol, triglyceride, albumin and thyroid hormones (T3 and T4). On the other hand the plasma concentrations of the total protein, creatinine, urea, and progesterone have no significant differences at the mating period.

We noticed that in all farms, ewes at the end of gestation and at the beginning of lactation do not receive any food supplementation. Also the breeders do not use the lick stones and do not add salt to the ration.

Another condition is omitted too, rehydrating the ewes through distribution of water during grazing or digging wells in the pastures, to combat the dehydrating, which is in this study a limiting factor for the ewes to be fertilized.

We are proposing a new understanding of the situation of rearing in the country and this is how we lay the ground to future researchers to look deeply into the influence of the environment on the inner side of the living organism. We opened the door to future researchers to investigate the deep connection between altitude and blood profile. Future researchers in this field could focus on exploring this link in other breeds. Another aspect is neglected which is the enormous capacity of this breed not only as a meat provider but also as a good milk provider par excellence. We suggest focusing on this produce and sheep produces in general are witnessing waste and lack of valorization.

Taking the results together suggests that Algerian farmers need to take serious steps to develop their rearing conducts. However, the efforts made up until now are not sufficient and more researchers need to investigate this area of study in the country.

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REGULAR ARTICLES



Effect of different altitude on reproductive performances and mineral assessment in Ouled Djellal ewes during the mating period

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Received: 23 January 2020 / Accepted: 21 July 2020 / Published online: 25 July 2020 \odot Springer Nature B.V. 2020

Abstract

The aim of this study is to evaluate reproductive performances and to reveal the influence of mineral profile on Ouled Djellal ewes reared in different sites during their mating period. Two hundred twenty-six clinically healthy and nonpregnant Ouled Djellal ewes have been used and assigned to four groups according to altitude factor (two plain regions, one tableland region, and one mountain region). In all groups, oestrus was synchronized with ram affect method, and the ewes were naturally inseminated; it was conducted during the month of May for all herds. After 5 months of gestation, reproductive parameters were calculated. Concerning the determination of plasma mineral parameters and its influence on reproduction parameters, blood samples were taken from all ewes on the day of introduction of entire rams into all herds. The analyzed variables were fertility, prolificacy, and fecundity, and their overall averages were 78%, 117%, and 92%, respectively, which are well below the averages in flocks reared in intensive mode. However, the lowest rates are recorded at mountain region (Aïn Zaatout) with 65%, 109%, and 71%, respectively. Plasma sodium and potassium were significantly lower in fecundable ewes than in non-fecundable ewes, whereas plasma calcium levels were significantly increased. The region showed in this study a significant influence on all plasma levels of major minerals during mating period. This study made it possible to describe the evolution of some mineral elements in ewes according to whether they are fecundable or not and according to the region (mountain, tableland, plain) during the mating period. This gave us useful information on the impact of plasma minerals levels on reproductive parameters.

Keywords Altitude · Reproduction parameters · Ouled Djellal ewes · Mineral profile · Mating period

Introduction

In Algeria, the sheep herd represents the largest animal resource; it is estimated at more than 26 million heads of which

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80% are of breeding ewes (M.A.D.R. 2016). Despite its economic and social importance, sheep farming is poorly managed in terms of the technical organization and the function of its production systems (Benyounes et al. 2013a), as well as the environmental conditions and the methods of conduct adopted that are at the origin of low productivity. Improving the productivity of sheep farming goes hand in hand with the control of reproduction and the improvement of its performances. It allows choosing the period of lambing, reducing the unproductive periods and optimizing the size of the embryo (Chemineau 1996). Some authors (Bodin et al. 1999) advocate the genetic pathway and argue that it can be obtained by selection or by crossing. Other studies highlight feeding status and recommend the allowance of the nutritional level before, during, and just after the mating as at the end of gestation (Benyounes et al. 2013b).

Several authors report that blood mineral levels can be used as indicators of prepartum and/or postpartum problems (Khatun et al. 2011; Khaled and Illek 2012). In view of the fact that minerals play an important role in the regulation of physiological functions before, during, and after the puerperal period, their concentrations in blood represent homeostatic mechanisms that are closely related to neurohumoral regulation (Piccione et al. 2009). Deficiencies and mineral imbalances are often cited as causes of poor reproduction. Moreover, some studies show that minerals have a critical role in reproduction performance of ewes (Gabryszuk and Klewiec 1997). This may affect the ovulation rate (Marais 2011) and the embryo survival and increase the number of weaned lambs per ewe per year (Benyounes et al. 2013b). To this extent, in Biskra, sheep farming is one of the main activities of the district, and the fact that Ouled Djellal breed constitutes the majority of the herds in the region, Ouled Djellal sheep has been the subject of several studies relating to production and reproduction performances. The latter is perfectly adapted to the extreme conditions of arid environment. Thus, the objective of this study is to update Ouled Djellal ewe's reproduction performances in four sites of Biskra governorate, as well as to compare and to evaluate the influence of some blood mineral levels during the mating period.

Materials and methods

Study region

The present study was carried out in the region of Biskra, located in the central-east of Algeria, at the gateway to the Algerian Sahara. It is a real buffer zone between North and South, about 400 km southeast the capital, Algiers (approximately 35°15' to 33°30' N, 04°15' to 6°45' E). In general, Biskra is composed of four different geomorphological elements: the mountains, the plains, the tablelands, and the depressions (Bougherara and Lacaze 2009; Aissaoui et al. 2019). The study area is characterized by significant diurnal thermal amplitudes. This is how the temperature, which reaches very high levels in shade the daylong during the summer, drops to minus 50% in the evening. The variation in daily thermal amplitude is significant throughout the months of the year. The maximum temperature is reached in July, 49 °C, but in January, it oscillates around 5 °C (especially in the evening). Rainfall is low and irregular from month to month depending on the year. The rain rate mostly increases in autumn and winter. The relative humidity of the air varies considerably according to the season. In summer, it falls to 25% in July due to high evaporation. However, in winter, it raises up to 60% maximum. Winds blow throughout the year. Generally, the northwest winds dominate. Southern winds are generally cold and dry in winter. They are hot and very dry in summer, that is, the sirocco. It acts by enhancing evaporation and thus increases drought.

Choice and distribution of herds

The choice of locations was based on the geographical distribution and the characteristics of the areas (reliefs and altitudes). We selected four (4) locations (Table 1): El Hadjeb, Sidi Okba, Ouled Djellal, and Aïn Zaatout, noted, respectively, by site 1, site 2, site 3, and site 4.

The selected animals are clinically healthy and nonpregnant Ouled Djellal ewes, aged 2.5–05 years and with a median body condition score of 2.8 ± 0.9 (on scale 1–5) and weighted 53.4 kg ± 5.4. All the ewes had successfully lambed by of the previous year, and in the interval lambing, the beginning of experiment \geq 70 days, this ewes were divided into four herds, in site 1 (30 ewes), in site 2 (60 ewes), in site 3 (70 ewes), and site 4 (66ewes).

Rams are totally isolated from the females and are introduced into the herd only during mating period.

Livestock management

Ewes were being reared in semi-extensive system; the ewes during our experiment and in all sites are taken to pasture twice a day (7 a.m. to 11 a.m. and 4 p.m. to 6 p.m.).

At night, all ewes were housed in a barnyard and had access to fresh clean water twice a day (one in the morning, second in evening).

The ewes were dewormed 4 times/year with anthelmintic, and they are routinely vaccinated for sheep pox, peste des petits ruminants (P.P.R.), and enterotoxaemia.

In the four sites, the natural grazing land consisted of a mixture of *Stipa tenacissima*, *Ampelodesmos tenax*, and *Artemisia herba-alba*, as well as annual meadows, composed of various grasses (predominance of *Cynodon dactylon*, *Melilotus sulcata*, and *Vicia monantha*). All ewes received a same daily supplementation of 200–300 g of commercial cereal concentrate and 300–400 g of good quality hay at the barnyard twice a day (at 12 p.m. and 8 p.m.). The amount of concentrate fed differed according to body condition score.

Synchronization of oestrus and detection of pregnancy

The ewes at the four sites were isolated from any contact with rams (sound, sight, and odor) at least for 4 full weeks (De St Jorre et al. 2012; Mayorga et al. 2019). Seven adult rams aged of 3 to 5 years were vasectomized and isolated in another area far from all herds. These males were used to induce the "ram effect" (Rosa and Bryant 2002; Fabre-Nys et al. 2016). During the month of May, the vasectomized rams were introduced (day 0) into flocks at the rate of 1 ram for 30 to 40 ewes (Castonguay 2018) (1 ram in site 1 and 2 rams for sites 2, 3 and 4). The vasectomized rams were removed from the herd on the evening of the 24th day; the following day (day 25), the

Table 1Details on the differentstudied sites

	Height above sea level	Landforms	Rainfall (mm/	Annual temperature °C	Geographic locations	
	sea level		month)	(mean)	Latitude	Longitude
Site 1	139 m	Plain	10.4 ^a	26.2 ^a	34°47′	5°36′
Site 2	54 m	Plain	10.4 ^a	26.2 ^a	34°75′	5°9′
Site 3	450 m	Tableland	12.85 ^b	21.05 ^b	34°27′	5°04′
Site 4	1000 m	Mountain	12.02 ^c	17.8 ^c	35°09′	5°50′

^a Biskra meteorological station

^b Ouled Djellal meteorological station

^c Lotaya meteorological station

fertile rams will be introduced into all flocks with a ratio of 1 male to 10 females for natural mate (Thimonier et al. 2000) (Castonguay 2018). The rams were withdrawn 48 h after introduction (day 27).

Pregnancy was detected on the basis of non-return on oestrus after two observation periods (19 to 23 and 40 to 44 days after mating period) as reported by Ouedraogo et al. (2008), where gestation was determined 30–60 days following the mate by a simple abdominal palpation (Ahmed et al. 1998).

The studied reproduction performances were fertility, prolificacy, and fecundity.

Fertility rate

- = (Number of lambing ewes/Number of ewes put to mate)
 - $\times 100$

Prolificacy rate

= (Number of born lambs/Number of lambing ewes)

 $\times 100$

Fecundity rate

= (Number of born lambs/Number of ewes put to mate) $\times 100$

All these parameters were calculated according to Charray et al. (1992).

Blood samples

On the day of the introduction of the fertile rams into the herds, blood samples were taken from all the ewes of the study.

The ewes were distributed into three groups, depending on the topography of the study area: group 1 is a plain region (site 1 and site 2), group 2 is a tableland (site 3), and group 3 is a mountainous region (site 4).

Blood samples were taken by puncture of the jugular vein early in the morning before food intake to limit variations related to food intake and stress. Seven milliliters of blood were collected using disposable needles in vacutainer tubes with heparin anticoagulant. The collected blood was transported in a cooler and centrifuged, before the exhaustion of 2 h after sampling, at 3000 G at room temperature for 15 min. The plasma was divided into aliquots in microcentrifuge tubes and kept frozen at -20 °C till further analysis.

Analysis method

Blood minerals, plasma calcium (Ca), plasma phosphorus (P), plasma magnesium (Mg), plasma sodium (Na), and plasma potassium (K) were estimated using commercial kits "SPINREACT," Spain, as per standard method using the ultraviolet (UV)-visible recording spectrophotometer (UV-160A; Shimadzu Corporation, Japan).

Statistical analysis

Statistical analysis of the reproductive variables was established using SPSS Inc.'s "IBM SPSS Statistics 20" software, Chicago, IL, USA.

We compared fertility, fecundity, and prolificacy rates for the four sites (El Hadjeb, Sidi Okba, Ouled Djellal, and Aïn Zaatout). We used $\chi 2$ test to verify the link between the measured rates and the four sites, in addition to multiple comparisons that revealed sites with significant differences. For the interpretation of the parameters of reproduction, we considered the following formulas:

Fertility : yes = ewe fertile, no = ewe infertile

= two born lambs

Prolificacy : 1 = simple birth, 2 = double birth

This same software allows the determination of the mean and standard deviation of each blood mineral parameter and the comparison of the means of each parameter between fecundable and non-fecundable ewes and between sites using the Student's test (t test). The differences were considered significant when p < 0.05.

Results

Reproduction parameters

Results of the reproduction parameters of the different sites are gathered in Tables 2 and 3.

In the light of these findings, we observe that the average fertility rate of ewes was 78%; that is to say, there are 78 lambing ewes and 22 non-lambing (infertile), the average prolificacy rate of the studied ewes was 117% yielding 17 more lambs for 100 lambs, and the average fecundity rate was 92%.

Fertility averages that are below the overall average were recorded in the herds of site 1 (plain region) and site 4 (mountain region), which were 0.7 and 0.65, respectively, but the averages for herds at sites 2 and 3 were above the overall average and were, respectively, 0.9 and 0.87. Herd in site 2 (tableland region) showed the best fertility rate, and the statistical study showed that the region influences fertility. Comparisons two to two of fertility rates show that the difference is significant between site 1 and site 2 and between site 1 and site 3 and very highly significant between site 2 and site 4 and between site 3 and site 4.

Prolificacy was found in the herds of sites 1, 2, and 3 higher than the general average and higher than the average recorded in the herd of site 4. The ewes of site 3 have the best prolificacy rate, and no significant difference was found between herds relating region.

Concerning the third reproduction parameter, we observe that the average fecundity is below the general average; it was

Table 3 Averages of fertility, prolificacy and fecundity of Ouled Djellal ewes

		Fertility	Prolificacy	Fecundity
Site1		0.7	1.19	0.83
Site2		0.9	1.18	1.06
Site3		0.87	1.23	1.07
Site 4		0.65	1.09	0.71
Mean \pm standard deviation		0.78 ± 0.12	1.17 ± 0.06	0.92 ± 0.18
Pearson's chi-square		22.829	6933	29.963
p value	Site 1 vs 2 vs 3 vs 4	0.001	0.074	0.0001
	Site1 vs site 2	0.016	/	0.056
	Site 1 vs site 3	0.041	/	0.116
	Site 1 vs site 4	0.589	/	0.341
	Site 2 vs site 3	0.611	/	0.741
	Site 2 vs site 4	0.0001	/	0.0001
	Site 3 vs site 4	0.0001	/	0.0001

recorded in the flocks of sites 1 and 4 which were 0.83 and 0.71, respectively. On the other hand, the herd averages for sites 2 and 3 were higher than the general average, which were 1.06 and 1.07, respectively. Site 3 showed the best fecundity rate. Like fertility, the statistical study showed that the region significantly influences fecundity: highly significant difference between site 2 and site 4 and between site 3 and site 4 and no significant difference between the fecundity rates between site 1 and site 3, site 1 and site 4, and the site 2 with site 3.

Plasma minerals concentrations

The results of plasma mineral parameters in Ouled Djellal ewes from the different sites during mating period are reported in the Table 4.

Mean plasma calcium of ewes in plain region differed significantly (p < 0.01) with ewes in mountain region; however, plasma calcium of ewes at plain region did not differ significantly from the tableland ewes. In addition, plasma calcium of ewes in tableland region differed significantly from the mountain ewes (p < 0.05). The statistical mean of the calcemia is

Table 2 The numbers registered in studied sites		Ewes number	Lambing ewes	Infertile ewes	Lambs number	Simple pregnancy number	Double pregnancy number
	Site 1	30	21	9	25	17	4
	Site 2	60	54	6	64	44	10
	Site 3	70	61	9	75	47	14
	Site 4	66	43	23	47	39	4
	Total	226	179	47	211	147	32

Table

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Table 4 Variation in the concentration of major plasma minerals in Ouled Djellal ewes according to the region and to being fecundable or not

Parameters		Site1 + site2 Group I (M ± SD)	Site3 Group II (M ± SD)	Site4	<i>p</i> value			
				Group III (M ± SD)	Group I versus group II	Group I versus group III	Group II versus group III	
Ca (mmol/L)	Fec-ewes	1.70 ± 0.28	1.58 ± 0.41	1.26 ± 0.25	NS	<i>p</i> < 0.01	<i>p</i> < 0.05	
	Non Fec-ews	1.42 ± 0.35	1.51 ± 0.58	1.08 ± 0.03	NS	<i>p</i> < 0.01	p < 0.05	
	p value	<i>p</i> < 0.05	<i>p</i> < 0.05	<i>p</i> < 0.01				
P (mmol/L)	Fec-ewes	1.13 ± 0.42	1.02 ± 0.19	0.68 ± 0.22	NS	<i>p</i> < 0.01	<i>p</i> < 0.05	
	Non Fec-ews	1.11 ± 0.29	0.98 ± 0.54	0.63 ± 0.1	NS	<i>p</i> < 0.01	<i>p</i> < 0.01	
	p value	NS	NS	NS				
Mg (mmol/L)	Fec-ewes	0.84 ± 0.25	0.92 ± 0.33	0.99 ± 0.2	<i>p</i> < 0.05	<i>p</i> < 0.05	NS	
	Non Fec-ews	0.90 ± 0.18	0.98 ± 0.20	1.05 ± 0.31	<i>p</i> < 0.05	<i>p</i> < 0.05	NS	
	p value	NS	NS	NS				
Na (mEq/l)	Fec-ewes	145 ± 12.55	142 ± 10.25	142 ± 10.96	NS	NS	NS	
	Non Fec-ews	147 ± 11.5	145 ± 8.4	139 ± 9.58	NS	<i>p</i> < 0.05	NS	
	p value	<i>p</i> < 0.05	<i>p</i> < 0.05	<i>p</i> < 0.05				
K (mEq/l)	Fec-ewes	4.2 ± 0.55	3.9 ± 0.49	3.8 ± 0.81	NS	NS	NS	
	Non Fec-ews	4.7 ± 0.66	4.1 ± 0.74	3.9 ± 0.56	NS	<i>p</i> < 0.05	<i>p</i> < 0.05	
	p value	<i>p</i> < 0.05	NS	NS				

M mean, SD standard deviation, NS non-signification, Fec-ewes fecundable ewes, Non Fec-ews non-fecundable ewes

significantly different between fecundable and nonfecundable ewes in all groups. Plasma phosphorus levels recorded in plain region and tableland region were significantly higher than its level in mountain region (p < 0.01), by cons; the plasma magnesium levels recorded in mountain region and tableland region were significantly higher than the one recorded in plain region (p < 0.05). No significant difference was observed between fecundable and non-fecundable ewes in terms of plasma phosphorus and plasma magnesium. Plasma sodium decreased significantly (p < 0.05) in fecundable ewes as compared with non-fecundable ewes. Plasma potassium increased significantly (p < 0.05) in non-fecundable ewes as compared with fecundable ewes in plain region.

Discussion

Reproductive performance

Fertility

Discussing the findings of the present study, lower rates of fertility compared with that recorded in Ouled Djellal breed (100%) cited by (Chellig 1992), with a difference of 22%, and the rates recorded in Ouled Djellal breed described by (Dekhili 2004) in Setif region, where the average was 92% and those of (Safsaf and Tlidjane 2010) which was 91% with a difference of 14% and 13% respectively. Among certain

Moroccan breeds described by Chikhi and Boujenane (2003) as the Boujaâd, Sardi, Timahdite, and Beni Guil breeds, which varied on average by 98%, 85 to 92%, 77 to 95%, and 82 to 87%, respectively, with differences of 20%, 7 to 14%, 1 to 17%, and 4 to 9%, respectively, pointed out that the effect of breeding method particularly food supply during mate period and the absence of flushing technique are of an elementary role and are mainly behind this drop, a thing that is early revealed by authors such Abbas et al. (2004) and Deghnouche (2011) who reported that good quality feeding improves ewes' fertility. Another reason could also interfere which is drought-related dietary conditions in the study area. Thus, the fertility rate is influenced by the season and the month of mating (Dekhili and Aggoun 2004). The best performances of fertility were obtained when fertilization took place, while the food supply was in large quantities and of a good quality.

Prolificacy

The average prolificacy of Ouled Djellal ewes was 117%, and our results are lower than the rates recorded for Ouled Djellal ewes described by Dekhili (2004) and Deghnouche (2011) which were 130% and 147% with a difference of 13% and 30%, respectively, and lower than the rates recorded in the D'Man breed ewes described by Chellig (1992) which varied from 185 to 200% with differences from 68 to 83%. However, our results are similar to the rates recorded in Hamra breed described by Chekkal et al. (2015) and the Moroccan Sardi breed described by Chikhi and Boujenane (2003) which varied respectively from 110 to 120% and from 107 to 121%, and our findings exceeded the rates recorded by Dekhili and Aggoun (2007) in Ouled Djellal breed in Sétif region which was 109%.

Prolificacy rate is influenced by the feeding of ewes during flushing. Chentouf et al. (2003) report that providing supplements to ewes before and during the mate period have led to an improvement in the ovulation rate and consequently the rate of prolificacy.

Clement et al. (1997) report that sufficient and good quality feeding during the mate could favor double births.

Fecundity

The average fecundity of ewes was 92%, and this rate was lower than the rates recorded for Ouled Djellal, Rembi, and Barbarine breeds described by Chellig (1992) which was 95%, with a difference of 3%, and at the rates quoted by Deghnouche (2011) which varied between 110 and 125%, higher than the rate recorded by Merghem (2009) which was 70% for Ouled Djellal type Hodna breed in the region of Sétif.

The differences between the results obtained in the present study and those recorded by the different authors can be explained by racial differences, the mode of rearing, and the mode of feeding (feeding of ewes during the mate periods). In this study, flocks were extensively driven with inadequate feeding and covered only maintenance requirements. This undernourishment, especially during the mate period, negatively affects the reproductive potential of the ewes studied.

As conclusion, the results of this study indicate clearly the presence of differences in reproductive performances between the four herds. We consider the herd located in plain region of Sidi Okba (site 2) as the most fertile and the herd located in tableland region of Ouled Djellal (site 3) as the most prolific and most fecundable, while the herd 4 located in mountain region of Aïn Zaatout (site 4) is the least fertile herd, the least fecundable, and the least prolific.

It is recognized that the performance of an animal is determined by the expression of its genotype, the influence of the environment in which it evolves, and the interaction between these two factors (Dekhili and Aggoun 2006). Therefore, the difference in the reproductive performance between the four flocks from the four sites is the first explanatory elements of this reaction of Ouled Djellal ewes' from one site to another, and the inferiority of the fertility, prolificacy, and fecundity rates at site 4 (Aïn Zaatout) can be explained by the decrease in progesterone levels in ewes at high altitudes (Barnett et al. 1978). The same authors report that high levels of progesterone in the blood can improve ovulation rates, embryo survival, and nesting of fertilized eggs in the uterus and consequently reproduction rates. The control of livestock management and the improvement of food supply, as well as the availability which is uncertain in arid zones, can lead to a significant improvement in breeding parameters.

Plasma minerals concentrations

Results of ewes at plain (sites 1 and 2) were gathered in one group, that is, group 1, whereas the tablelands were named group 2 and 3, respectively. Lot of non-fecundable ewes in group 1 is composed of the ewes at sites 1 and 2 which was 15 ewes, and this same lot is composed of 9 ewes in group 2 located in the tableland area (site 3) and 23 ewes in group 3 located in the mountains (site 4).

The plasma calcium values that we recorded in this study are below the physiological norms cited by several authors (Brugère-Picoux 2004; Kaneko et al. 2008; Brouček et al. 2009; Elnageeb and Adelatif 2010; Abarghani et al. 2013). Moreover, the majority of ewes have low plasma calcium, which indicates an insufficient dietary intake of calcium. This insufficient intake would lead to a decrease in the fertility of ewes especially for mountain ewes (Friot and Calvet 1971). In addition, the significant decrease in plasma calcium in mountain ewes compared with other regions in this study may be due to an excessive demand for muscular activity in these animals (Sejian et al. 2012; Titaouine and Meziane 2015); these results disagree with those of Brouček et al. (2009) which study the effect of various factors on blood minerals in sheep and found that sheep's serum calcium is not influenced by altitude. A significant increase (p < 0.001)of plasma calcium in fecundable ewes relative to plasma calcium in non-fecundable ewes in all regions. This increase in plasma calcium is probably due to increased intestinal absorption and bone resorption of Ca due to the hormonal change during the estrous period (Takagi and Block 1991; Abdelrahman et al. 2002).

The plasma concentrations of calcium and phosphorus are controlled by the same hormone-dependent homeostatic mechanism (Hadžimusić and Krnić 2012).

The lower plasma concentration of Ca and P in this study might have been as a result of decreased levels in plants of all regions. This can be supported by the fact that when this study was carried out, the plants had dry, brown, and withered leaves and therefore had lower levels of mineral content (Khan et al. 2007). The significant decrease in plasma phosphorus in mountain ewes could be a support for severe muscle exercises when walking in this region (Titaouine and Meziane 2016).

In all groups, fecundable ewes have a high plasma phosphorus concentration compared with non-fecundable ewes, but the statistical analysis does not show a significant difference. With the decrease of plasma phosphorus for nonfecundable ewes in comparison with fecundable ewes, Jacobson et al. (1972) found that plasma P concentrations are maintained by the absorption of P from the gut, and there is no specific mechanism for bone P reabsorption. In addition, positive relationships between dietary P intake and plasma P have been observed (Abarghani et al. 2013).

We observe that plasma magnesium increases with altitude. Our results are in contradiction with those of Masters et al. (1993), who found no significant difference (p > 0.05) in plasma magnesium as a function of altitude. Differences in elevated regional plasma levels are likely due to differences in soil types, precipitation, and pasture types (Masters et al. 1993); the low plasma values of this element for both classes of fecundable and non-fecundable ewes in the lowlands could be attributed to high potassium intake in this region, as it has been well established that this macroelement interferes with the absorption of magnesium (Ghosh and Shashank 2008; Sowande et al. 2008).

Although there is no significant difference between the two groups of ewes in plasma magnesium in all regions, it should be noted that fecundable ewes have low plasma magnesium concentration. The decrease in magnesium plasma concentrations observed in fecundable ewes can be demonstrated by low sodium (Na) content. The low level of Na in the diet or in water may suggest hypomagnesemia by decreasing the absorption of Mg by the intestinal mucosa (Schweigel and Martens 2000).

Table 4 shows a decrease in natremia in mountain and plateau ewes compared with ewes in plain. On the other hand, the statistical analysis of the values obtained gives only a significant difference (p < 0.05) between non-fecundable ewes in the plain and those in the mountains. Indeed, the thermal gradient adiabatic assumes that the plain region will be hotter than the other regions, "we lose 1 °C every 100-m altitude"; therefore, the ewes of this region will be more dehydrated than other ewes; all authors report an increase in plasma sodium concentration in the event of dehydration. This increase is due to the decrease in glomerular filtration and tubular reabsorption of sodium. Indeed, the activation of ADH during dehydration promotes a high absorption of water and a slight reabsorption of sodium from the digestive tract under the action of aldosterone (Maloiy and Clemens 1980).

In our study, it can be noted that fecundable ewes have lower plasma sodium and potassium concentrations than non-fecundable ewes (Elnageeb and Adelatif 2010). They attributed this situation to the increase in progesterone levels in the blood of this ewe's category. In addition, other studies by Deghnouche et al. (2013) and Laidlaw et al. (1962) have described the increase in sodium excretion during progesterone administration and have suggested that the latter has an aldosterone antagonist effect on the renal tubules.

The evolution of plasma potassium in the three regions follows the same pattern as natremia. Consequently, the significant decrease (p < 0.05) of plasma potassium in the mountainous region in non-fecundable ewes results from the

decrease of plasma sodium in these ewes, which subsequently leads to an increase in aldosterone, which decreases the plasma potassium level maintaining the tubular elimination of potassium (Yokus et al. 2004).

Conclusion

In conclusion, the study proved the presence of a characteristic site effect from which Ouled Djellal ewes do not externalize their best reproduction performances at high altitudes.

The study of the influence of mineral parameters shows that fecundable ewes have high plasma concentrations of calcium and phosphorus during the mating period. In contrast, plasma levels of magnesium, sodium, and potassium were higher in non-fecundable ewes.

In general, the concentrations of the mineral parameters studied are below physiological norms in the four regions. So we must, on the one hand, add to the feeding of ewes either in the mountain, in tableland, or in the plain mineral supplementation which will probably increase the blood levels of these minerals especially for calcium and phosphorus. On the other hand, rehydrating the ewes through distribution of water during grazing or digging wells in the pastures, to combat the dehydrating of ewes, which is in this study a limiting factor for the ewes to be fertilized?

Further work would be needed to complete this study, including determining the effects of ewe's age, season, rearing, feeding, physical activity, and variables as body temperature, respiratory frequency, and heartbeat, in order to understand the body metabolism in different environment, without forgetting other climatic data on reproduction for the purposes of development sheep flock in Algeria.

Acknowledgments The authors would like to thank the University of Biskra for availing the data which was used in this research. The authors also wish to sincerely thank the DEDSPAZA laboratory, and we thank the Technical Institute for the Development of Saharan Agronomy, "ITDAS," Biskra, for the use of their animals.

Authors' contributions All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by Titaouine Mohammed, Makhlouf Asma, and Yakoub Fedjria. The first draft of the manuscript was written by Titaouine Mohammed, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Statement of animal rights All procedures performed in this research were approved by the ethics committee on the use of animals from the Genetic, Biotechnology and Valorization of Bioressources Laboratory - University of Biskra.

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