

Journal of the Hellenic Veterinary Medical Society

Vol 73, No 2 (2022)



Grazing behaviour, intake, digestion, performance and endoparasitic infection of lambs raised at different stocking rates under low-input farming systems

Vaia Kantzoura, Ioannis Hadjigeorgiou, Christos Goulas, Georgia Zabeli, Smaragda Sotiraki, Georgios Zervas

doi: [10.12681/jhvms.26442](https://doi.org/10.12681/jhvms.26442)

Copyright © 2022, Vaia Kantzoura, Ioannis Hadjigeorgiou, Christos Goulas, Georgia Zabeli, Smaragda Sotiraki, Georgios Zervas



This work is licensed under a [Creative Commons Attribution-NonCommercial 4.0](https://creativecommons.org/licenses/by-nc/4.0/).

To cite this article:

Kantzoura, V., Hadjigeorgiou, I., Goulas, C., Zabeli, G., Sotiraki, S., & Zervas, G. (2022). Grazing behaviour, intake, digestion, performance and endoparasitic infection of lambs raised at different stocking rates under low-input farming systems. *Journal of the Hellenic Veterinary Medical Society*, 73(2), 4085–4096. <https://doi.org/10.12681/jhvms.26442>

Activities on pasture, intake, digestion, performance and endoparasitic infection of lambs raised at different stocking rates under low-input farming systems

V. Kantzoura^{A*}, I. Hadjigeorgiou^B, C. Goulas^C, G. Zabeli^D, S. Sotiraki^A, G. Zervas^B

^AVeterinary Research Institute, Hellenic Agricultural Organization-Demeter, Themi, Thessaloniki, Greece

^BDepartment of Nutrition Physiology and Feeding, Faculty of Animal Science, Agricultural University of Athens, Athens, Greece

^CDirectorate of Food Quality and Safety General Directorate of Food Ministry of Rural Development and Food, Athens, Greece

^DAnimal Genetic Resources Center Of Ioannina Ministry Of Rural Development And Food, Ioannina, Greece

ABSTRACT: Boutsiko sheep breed is a locally adapted one traditionally utilizing the mountainous areas of northern Greece under low-input farming practices. In the present study feed intake, digestibility, grazing activity, growth rate, and endoparasitic burden were evaluated on fattening lambs of this breed. Twenty-four lambs were allocated to three groups of equal average live weight (LW: 20.8 kg) and pastured at three stocking rates: SR₁, SR₂ and SR₃ (representing 60, 80 and 100 lambs/ha/60 days respectively). Animals were grazed on a semi natural sward daily and sward height in all treatments remained above 7 cm during the trial, while the herbage gross chemical composition was similar between treatments. On average, the lambs grazed 373 min, ruminated 139 min and spent 88 min on other non-feeding activities during the daily pasturing. Average herbage consumption of all lambs at the start and the finish of the trial was 714 g and 1015 g of DM/day, or 70.5 and 99.0 g/kgW^{0.75}/day respectively. Herbage DM digestibility (*in vivo*) was moderate averaging 61% and no differences were observed between the three groups of lambs. The daily live weight gain of lambs (DLWG) was different ($p < 0.05$) between SR₁ and SR₂, SR₃. Lambs on SR₁ had the highest gain (0.0950 kg LW/day) while those on SR₂ and SR₃ the lowest (0.0420 and 0.0563 kg LW/day respectively). Faecal egg counts (FEC) for gastrointestinal parasites (GIN) from lambs grazing plot SR₃ were low, while high numbers of *Moniezia* spp. eggs were present in faecal samples of all groups of lambs at the end of the experiment. It is concluded that lamb fattening on pasture can be achieved at reasonable stocking rates, without the use of anthelmintics, if a local hardy breed is used.

Keywords: extensive sheep production system; stocking rate; forage height; n-alkanes; parasites

Corresponding Author:

V. Kantzoura, Veterinary Research Institute, Agricultural Organization Demeter, Ktima Themi, 57001 Thessaloniki, Greece
E-mail address: kantzourava@yahoo.gr

Date of initial submission: 19-03-2021
Date of acceptance: 20-10-2021

INTRODUCTION

The efficient management and practices applied among small sheep farmers is the key to increase incomes in a sustainable way in the Mediterranean area. Efforts are needed to adapt existing solutions into low-cost ones for enhancing the efficiency of productivity and optimisation of natural resources use and also the income, at the scale of smallholder farmers (FAO, 2017). Furthermore, consumers and producers alike are becoming gradually more concerned about the amount of chemicals used in conventional farming. As a result, interest in traditional and low-input agriculture is growing steadily in Europe (EC, 2018). Low input farming has been established as an environmentally friendly production (Wojtkowski, 2008), while achieving high animal welfare standards (Phocas *et al.* 2016). Low-input farming systems operate in-between conventional and organic systems, trying on the one hand to minimize the use of external resources and on the other hand to optimize their cost. In this farming system a wide range of local breeds is used worldwide (Marković *et al.* 2019; Khan *et al.* 2020; Haile *et al.* 2020). The preference for local breeds is mainly due to their higher adaptability to the particular climate and to the type of vegetation found in the area, which can be used as their food (Gaughan *et al.* 2019; Perucho *et al.* 2019) and to their increased resistance to diseases (Dudu *et al.* 2020). Boutsiko breed is a small sized local breed, raised in the mountainous areas of north-western Greece, which is known for the high quality of milk and meat it produces (Zygiannidis *et al.* 1997; Kondyliet *et al.* 2011).

Low input sheep production in the Mediterranean basin, traditionally relies on grazing natural or semi-natural pastures, without the use of harvested feeds except in winter months. The level of production of these systems depends largely on the animals' ability to ingest a diet adequate to meet their nutritional requirements for maintenance and production and to overcome the adverse environmental conditions (Hodgson 1985). Grazing animals, in particular, which consume a diet of relatively low nutritional quality, require long foraging times for the harvesting of their daily ration (Herbers, 1981). It is generally accepted that ruminant's total daily foraging time is closely related to their total daily intake achieved (Newman *et al.* 1995).

Stocking rate (SR), i.e. the number of animals per unit area over a period of time, is considered the most important factor in grazing management affecting the

efficiency of pasture utilisation, since increasing SR, up to a certain limit, will lead to an increase in animal production per unit area, even despite a possible decrease in the individual animal performance (Hodgson, 1985, Binnie and Chestnut, 1994). SR may affect the individual performance of grazing sheep either directly by lowering the amount of forage available per animal or indirectly by affecting the impact of stress factors, including acquired gastrointestinal parasitic (GIN) infections. It is generally recognised that increasing SR often leads to increasing the parasitic burden of grazing animals (Celaya *et al.* 2016). GIN infections comprise an important burden to animal production from pastures (Almeida *et al.* 2020) and current control methods include the reduction of contamination through anthelmintic treatment of grazing animals and/or controlled grazing. Subsequently, avoidance of parasitic infections, mainly through the appropriate pasture management (Ruiz-Huidobro *et al.* 2019; Jaimez-Rodriguez *et al.* 2019), is of great importance for the success in low input systems (Thamsborg *et al.* 1999). Young sheep and goats are particularly susceptible to parasitic infections, because immunity is still being acquired, and growth rate can be depressed by as much as one third when infected with endoparasites (Bishop and Stear 2001; Mpofu *et al.* 2020). However, animals belonging to traditional local breeds often demonstrate resistance to parasitic infection, which allow their breeding under low input systems (Bishop and Stear 2001). In the present study feed intake, digestibility, grazing behaviour, growth rate, and endoparasitic burden were evaluated in a low-input system of raising weaned lambs, of the hardy local breed of Boutsiko, grazing a semi-natural pasture under a range of stocking rates, in order to assess the breed's capacity to produce heavy lambs under such system.

MATERIALS AND METHODS

Study area

The experimental area was part of the 40 ha semi-natural pastures of the Agricultural Research Station of Ioannina, Greece (coordinates: 39,63019 and 20,88178). The area is flat at 480 m above sea level, where the climate is characterised by cold winters (min. -10 C°) and hot summers (max. 39 C°) and annual rainfall averages 820 mm. The Station's sheep flocks graze these pastures all year round, without any fertiliser application, and when surpluses exist they are mowed for haymaking. On this area three adjacent plots, of 0.1 ha each, were established. Attention

was paid to select a homogenous area concerning the quantity and composition of pasture herbage. To assure nutritional and parasitological equivalence of the plots, the fences for the formation of plots were erected just at the start of the experiment.

Animals and experimental design

Twenty-four 105 days old male lambs of the Boutsiko breed with no previous grazing experience were divided in three groups (6, 8 and 10 lambs per group) of equal average live weight (LW: 20.8 kg). Each group was allocated to a plot so that three stocking rates (SR) were applied corresponding to 60, 80 and 100 lambs.ha⁻¹ (SR₁, SR₂ and SR₃ respectively). The lambs were allowed to graze from 8.30 to 18.30 h each day for a 60 days period (April and May) and penned overnight where only water was provided. No supplementary feed was offered to the lambs and no anthelmintics were administered either before or during the grazing period. The animals were inspected daily and examined more closely whenever indicated.

Growth rate measurements

The animals were weighed individually on the morning of the first experimental day, before turnout to pasture and then every 15 days, at the same time, until termination of the experiment. Individual animal weight data over the trial days were described by linear regression and growth variables were produced. Therefore, the following growth parameters were calculated: live weight at start (LW_i), live weight at finish (LW_f), daily live weight gain (DLWG) and total live weight gain (TLWG).

Grazing activity

Half of the animals in each experimental group (3, 4 and 5 lambs per group respectively) were conspicuously marked with different colours and their activities were monitored by direct observation, on the 19th, 33rd and 47th day of the experiment. On those days, each of the focal animals was rotationally observed for one minute sequentially by the same observer (Altman 1974), in 15 minute intervals, and the prevailing activity during this time was recorded, classified as “grazing”, “ruminating” or “other activities”.

Herbage measurements and analyses

Sward height was measured as the average of 40 individual HFRO stick recordings on each plot (Barthram 1986), randomly selected over a W-transect. Sward height was measured at the start of the experi-

ment and then every 20 days until termination.

The herbage produced under protection cages, using three cages each measuring 1.0 m × 0.35 m × 0.6 m (L×W×H) on each plot, was harvested at the start of the experiment and then every 20 days until the end. Each herbage sample was divided into two parts: one part was for the determination of botanical composition and the other part for chemical composition after drying at 60°C for 48 h. Botanical analysis classified herbage material of the first subsample into grasses, legumes, broad-leaf forbs and dead matter which was expressed on a dry weight basis after drying at 95°C for 24 hours. Herbage gross chemical composition was determined on the second subsample, after drying at 65°C for 48 hours and grinding through a 1.0 mm sieve, for dry matter, organic matter, crude protein, ether extracts and crude fiber according to the Weende procedure (AOAC 1990), while NDF, ADF, hemicellulose and cellulose were determined according to Goering and Van Soest (1970).

Herbage intake and digestibility

Herbage intake and digestibility were determined twice during the experiment, by using the “n-alkane method” (Dove and Mayes 1991). For this purpose half of the animals in each experimental group (3, 4 and 5 lambs per group respectively) were dosed with “alkane pellets”. On each month during the experiment, lambs were orally dosed, on days 1-14, with a paper pellet impregnated with 0.8 g of each C₃₂ and C₃₆ alkanes, using a standard veterinary dosing gun. Alkane dosing was always carried out at the same time each morning, before the lambs were released to the pasture, while at the same time, in the final 5 days of each dosing period, a faecal sample was collected from the rectum of the animals. Faecal samples were pooled on a per animal and period basis, kept in freezer and oven dried at 60°C for 48 h. Herbage samples were also hand-plucked in those last 5 days, representative of what sheep grazed, and pooled per SR and period basis, before storing in freezer. Dried faecal and herbage samples were saponified with 1M KOH in ethanol, heptane extracted, then the extract was purified through silica gel columns and injected to a Perkin Elmer 8310 B gas chromatograph equipped with a 6' × 2 mm glass column packed with 3% OV-1 in GROM W-HP 80/100 mesh, for the determination of alkane concentrations. A single estimate of daily herbage dry matter intake was calculated for each lamb over each 5-day period from the dosed rate of C₃₂, as well as the C₃₂ and C₃₃ concentrations in faeces and

herbage. Faecal output was determined through the dilution of the dosed C_{36} in faeces and subsequently herbage dry matter digestibility was calculated. The herbage alkane composition was measured on hand plucked herbage samples mimicking lamb grazing.

Parasitological and haematological analyses

Herbage samples were collected at the start and the finish of the trial following hand plucked herbage harvesting, mimicking lamb grazing, for parasitic larvae counting according to the method of Martin *et al.* (1990). However, since the Station's sheep flock consisted of the hardy local Boutsiko breed ewes, which were dewormed six months earlier, there were made no parasitological tests to these animals, before the start of this experiment.

Faecal samples were collected the first day of the trial and monthly from all experimental animals. Faecal egg counts (FEC) were conducted for strongyle-type eggs cumulatively and for *Nematodirus* sp., *Strongyloides* sp., *Trichuris* sp. and *Moniezia* spp. eggs separately (Thienpont *et al.* 1986). For this study, we performed the following protocol: two grams of faeces were weighed into a disposable cup and 30 mL of saturated sodium chloride solution were added. A tongue depressor was used to break up the faeces and well-homogenize the mixture before straining through two layer of sieve and the residue pressed out. We mixed the faecal solution well and using a pipet immediately filled one chamber at a time, with remixing of the solution with a tongue depressor between filling each chamber. McMaster slides were read after waiting a minimum of five minutes, and only counted eggs at least partially within the grid, including those at least partially under the outer gridlines. The number of eggs per gram of faeces was obtained by multiplying the number of eggs found per slide by 100.

Blood samples were taken monthly by jugular venipuncture from each experimental animal. Plasma pepsinogen was determined by the method of Hirschowitz (1955) as modified by Ross *et al.* (1967). Red blood cells (RBC), haemoglobin (HGB), haematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were determined by using a semi-automated haematology analyser (Sysmex® F-820, TOA MEDICAL ELECTRONICS CO., LTD, Kobe, Japan). Percentage of eosinophils in differential leukocyte counts was determined in blood smears after staining with

Giemsa stain.

Statistical analysis

The Kolmogorov Smirnov Normality test indicated that there was a serious departure from normality ($p < 0.01$) for the variables faecal egg counts (FEC), plasma pepsinogen levels, RBC, HGB, HCT, MCH, MCHC, and proportion of eosinophils in differential leucocyte counts. Hence the nonparametric Kruskal-Wallis test was used for these variables to explore differences between the three groups of lambs. All other variables were distributed closely to normal probability function; therefore they were analysed by a GLM procedure on STATGRAPHICS statistical package to examine differences between the three groups of lambs. The level of significance was set at 5%.

RESULTS

Herbage production and herbage quality

Sward height at the start of the experiment was on average at 10.1 cm, but then showed a varied response to the different stocking rates applied (Table 1). On SR_1 sward height increased reaching 11.5 cm by the end of experiment, while on the other two stocking rates (SR_2 and SR_3) sward height decreased reaching 9.4 and 7.9 cm respectively by the end of experiment.

The gross chemical composition of the harvested herbage samples from all the three plots indicated that herbage was of good quality (Table 2). No significant differences between the chemical compositions of the harvested herbage from the three plots were observed.

The botanical composition of the harvested herbage was similar in all plots. The main plant genera composing the harvested herbage were: *Festuca*, *Agrostis*, *Bromus*, *Trifolium*, *Plantago* and *Alopecurus*. The contribution of grasses to the total DM produced was high (greater than 65%), but legumes and broad leaf forbs contributed also 7% and 7.5% respectively (Table 3). Dead herbage matter was a substantial component of the herbage harvested on all plots, reaching on average 19% of the total DM produced (Table 3).

Activities on pasture

The lambs grazed on average 373 min or 62% of their total time of daily access to pasture and additionally spent 139 min or 23.3% for rumination and 88 min or 14.7% for other non-feeding activities of which idling was the more important (Table 4), but

Table 1. Sward height (cm)±SD of pasture plots grazed by weaned lambs at three stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹ respectively) for a 60 days period

	SR ₁ Mean±SD	SR ₂ Mean±SD	SR ₃ Mean±SD
1 st day	10.5±2.69	10.0±2.64	9.8±2.10
20 th day	12.0±2.98	11.0±2.06	7.8±1.56
40 th day	11.5±2.67	11.1±2.53	7.5±1.15
60 th day	11.5±2.86	9.4±2.10	7.9±1.49

Table 2. Chemical composition of the harvested herbage from pasture plots grazed by weaned lambs at three stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹ respectively)

	SR ₁ Mean±SD	SR ₂ Mean±SD	SR ₃ Mean±SD
Dry matter %	17.45±1.10	15.95±0.81	16.95±0.96
Organic matter (%DM)	86.65±1.91	87.85±1.89	86.25±0.87
Crude protein (%DM)	21.60±0.50	21.90±0.67	23.20±0.62
Ether extracts (%DM)	2.70±0.18	2.75±0.13	2.55±0.26
Fiber (%DM)	19.05±2.76	17.95±2.93	17.85±3.57
N-free extracts (%DM)	43.30±2.65	45.25±2.00	42.65±2.99
Neutral-detergent fibre	45.66±1.97	45.30±2.60	42.90±2.80
Acid-detergent fibre	22.90±0.78	22.45±1.45	22.30±1.48
Hemicellulose (%DM)	22.75±1.70	22.85±1.74	20.60±1.41
Cellulose (%DM)	20.75±1.42	19.40±1.06	19.40±1.87

Table 3. Composition in major botanical groups (fraction of total DM) of herbage from pasture plots grazed by weaned lambs at three stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹, respectively)

	SR ₁ Mean±SD	SR ₂ Mean±SD	SR ₃ Mean±SD	Level of significance
Grasses	0.6905±0.104	0.7504±0.096	0.6645±0.071	NS
Legumes	0.1010±0.094	0.0401±0.041	0.0587±0.052	NS
Broad leaf forbs	0.0129 ^a ±0.023	0.0871 ^b ±0.069	0.1150 ^a ±0.100	*
Dead matter	0.1955±0.140	0.1222±0.096	0.1616±0.088	NS

* means within a column sharing different superscripts differ significantly (p<0.05)

Table 4. Activities time budget (min) during the daily allowance to pasturing, of the lambs grazing pasture plots at three stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹, respectively)

	SR ₁ Mean±SD	SR ₂ Mean±SD	SR ₃ Mean±SD	Level of significance
Grazing	357±13.32	397±14.40	366±17.39	NS
Ruminating	147±8.48	123±10.67	146±3.24	NS
Other activities	96±6.39	80±6.00	88±6.61	NS

p<0.05

no differences in time budgets were found between treatments. However, feeding time was distributed unevenly during their daily pasturing presenting two distinct peaks, one just after their release into pasture and the second early in the afternoon (Fig. 1).

Herbage intake, digestibility and animal performance

The average intake of herbage DM, as determined

through the “n-alkane method”, was different for all groups of lambs at the start and the finish of the experiment (averaging 714 and 1015 g DM day⁻¹ respectively), although there were no significantly different intakes between the three groups of lambs grazing at different SR on each date. Similarly, the intake expressed on a metabolic weight basis was different for all groups of lambs at the start and the finish of the trial (averaging 70.5 and 99.0 g/kgW^{0.75}/d respectively),

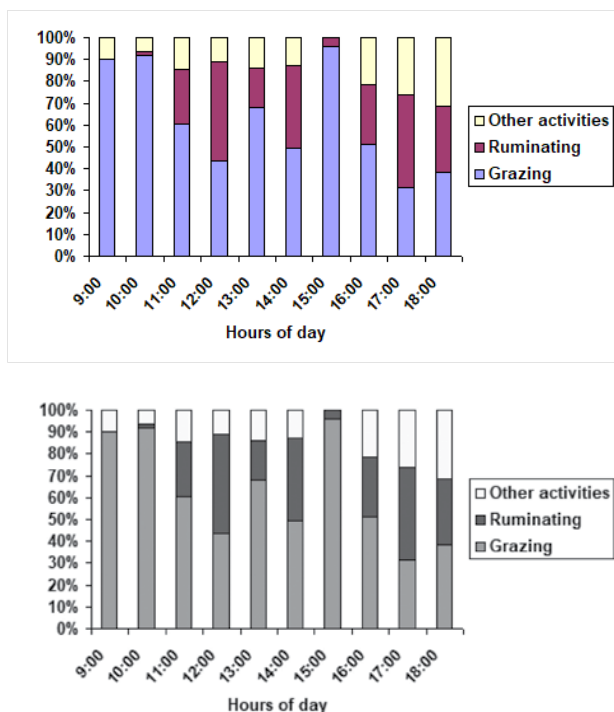


Fig. 1. Proportional allocation of time of the pastured lambs to various activities in hourly intervals

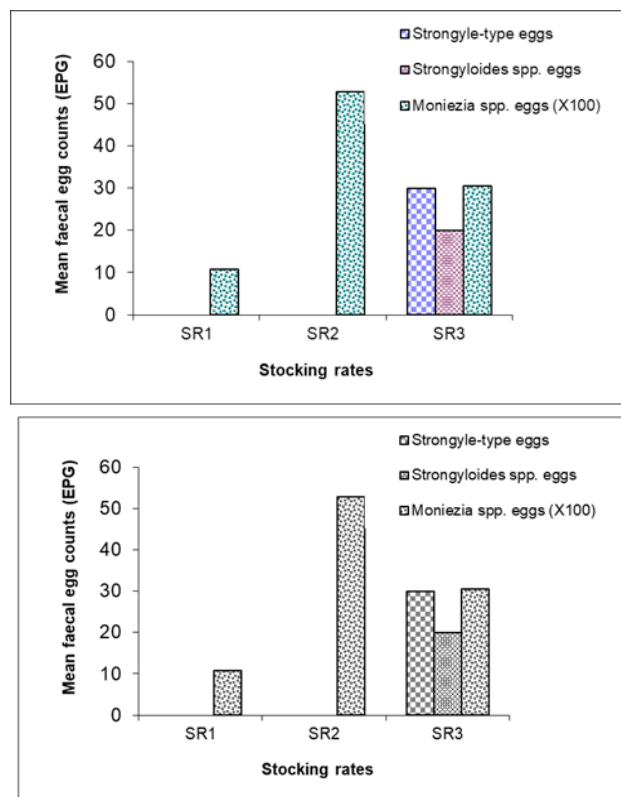


Fig. 2. Mean faecal egg counts (FEC) of parasitic eggs from lambs grazing plots receiving stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹ respectively) at the end of the experiment

but again there were no differences between the three groups of lambs (Table 5). Herbage DM digestibility measured with the n-alkanes method was moderate averaging 61% and no differences were observed between the three groups of lambs (Table 5).

Lamb growth showed a pattern of differentiation between the three treatments. The intercept of the linear equation describing lambs weight for the three groups was 20.3 kg, but then lambs on plot SR₁ had a different ($p < 0.05$) total live-weight gain (TLWG) of 5.51 kg head⁻¹ compared to the 2.43 and 3.26 kg head⁻¹ that gained the lambs on plots SR₂ and SR₃ (Table 6). This was due to different ($p < 0.05$) daily live weight gain (DLWG) for lambs on these plots (Table 6), where lambs on plot SR₁ had the highest DLWG (95 g LW/day) and lambs on both SR₂ and SR₃ plots had lower DLWG (42 and 56 g LW/day, respectively).

Parasitological and haematological findings

No infective nematode larvae were found on pasture samples either at the start or the end of the trial. GIN eggs were found only at the end of the experiment in faecal samples from lambs grazing plot SR₃, while

high numbers of *Moniezia* spp. eggs were present in faecal samples of all groups of lambs also at the end of the experiment (Fig. 2). However, no correlation was found between *Moniezia* spp. eggs and individual growth rates of lambs.

Plasma pepsinogen was traceable only at the end of the experiment in samples from lambs grazing plot SR₃. The mean plasma pepsinogen level of this group was 0.350 I.U. Tyrosine, which was within the normal limits (< 0.375 I.U. Tyrosine) (Kerboeuf 1977).

The levels of red blood cells (RBC), haemoglobin (HGB), haematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), and percentage of eosinophils in differential leukocyte counts (Table 7) were within normal range for all three SR lambs and there were no statistically significant differences between them.

DISCUSSION

Low input livestock farming brings a radical shift in thinking over the production process of animal husbandry with a greater attention to animal health and welfare, environmental conservation, food qual-

Table 5. Intake of herbage DM, the respective intake on a metabolic weight basis and DM digestibility by lambs grazing pasture plots at three stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹, respectively)

	SR ₁ Mean±SD	SR ₂ Mean±SD	SR ₃ Mean±SD	Level of significance
<i>DM intake (g/d)</i>				
April	676±29.2	812±11.0	654±18.8	NS
May	1025±109.5	1041±14.8	978±23.9	NS
<i>DM intake (g/kgW^{0.75}/d)</i>				
April	67.6±4.0	82.5±3.7	61.3±2.7	NS
May	100.2±5.3	102.2±7.6	94.5±3.1	NS
<i>DM digestibility (%)</i>				
April	58.3±1.0	64.3±1.16	58.2±1.9	NS
May	58.3±0.9	64.2±0.75	62.7±2.1	NS

p<0.05

Table 6. Live weight (LW) at the start and finish of grazing trial and daily and total live weight (LW) gain of lambs grazing pasture plots at three stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹ respectively)

	SR ₁ Mean±SD	SR ₂ Mean±SD	SR ₃ Mean±SD	Level of significance
LW at start (kg)	20.06±3.71	20.66±2.17	20.22±1.17	NS
LW at finish (kg)	25.57±5.69	23.09±2.01	23.49±2.54	NS
DLWG (kg)	0.0950 ^a ±0.039	0.0420 ^b ±0.015	0.0563 ^b ±0.034	p<0.05
TLWG (kg)	5.51 ^a ±2.29	2.43 ^b ±0.88	3.26 ^b ±1.99	p<0.05

* means within a column sharing different superscripts differ significantly (p<0.05)

Table 7. Mean values of haematological parameters of the blood samples examined from lambs grazing pasture plots at three stocking rates SR₁, SR₂ and SR₃ (60, 80 and 100 lambs.ha⁻¹, respectively) at the end of the experiment

	SR ₁ Mean±SD	SR ₂ Mean±SD	SR ₃ Mean±SD	Level of significance
RBC (x10 ⁶ /μL)	13.39±2.33	11.04±1.70	10.70±3.82	NS
HGB (g/dL)	12.72±0.58	12.01±1.65	11.59±2.81	NS
HCT (%)	55.50±7.23	50.25±8.39	49.16±14.69	NS
MCV (fL)	41.70±2.37	45.75±6.10	47.30±5.03	NS
MCH (pg)	9.67±1.30	10.95±1.17	11.51±2.27	NS
MCHC (g/dL)	23.17±2.37	24.03±1.27	24.19±2.70	NS
Eosinophils %	1.13±1.33	0.58±0.56	0.61±0.60	NS

p<0.01

ity and safety. To this direction, the use of synthetic drugs, to treat animals, needs to be minimized in small ruminants, along with environmental pollution with drug residues while the development of anthelmintic resistance in the animals of Southern Europe needs to be improved (Geurden *et al.* 2014). Therefore, grazing management and parasitic disease control are the most important practices which need to be improved for sustaining the small sized farm systems (Kumar *et al.* 2013). Furthermore, producers have the chance, in disadvantaged and remote areas, to promote their products easily through their special characteristics, achieving better market prices and thereby improving their income through the designation of local origin

products, whose quality or characteristics are essentially or exclusively due to a particular geographic environment with its inherent natural and human factors (Regulation EU1151/2012). Low-input stakeholders in Europe aim to export agro resources to improve meat and dairy quality as part of an ecological and sustainable production process (Mateo *et al.* 2017).

Feed availability is identified as one of the major constraints for the productivity of small ruminant grazing systems as well as the available pastures for the semi-extensive farming systems in Greece, which contribute to the production of high-quality milk and dairy products (Papaloukas *et al.* 2016). Sward height,

as a pasture variable, is positively related to the available herbage mass, to ingested herbage mass and subsequently to animals' performance (Binnie and Chestnutt 1994). Since sward height, on all stocking rates in this trial, was maintained above 7 cm during total duration, herbage growth and herbage intake of the lambs were not likely to be adversely affected (Penning *et al.* 1994). Several studies, in temperate areas, have shown that growth rates of weaned lambs increase only as sward height increases from 3 to 6 cm (Prache *et al.* 1990; Orr *et al.* 1990). However, growth rate recorded in this study was different between the stocking rates applied, although sward height was in all cases above 7.9 cm. Although the lambs entered the trial at a live weight typical of the breed for this age, the overall average growth rate achieved was only at 60 g/day (92, 35 and 53 g LW/day for lambs on SR₁, SR₂ and SR₃ plots respectively). This performance is considerably lower to the reported in earlier studies of grazing lambs in the area. For instance Zervas *et al.* (1999), studying crossed Boutsiko breed lambs reported a gain of 200 g/day, when fattened above 29 kg of live weight, by grazing the same field used in this trial.

No significant differences were observed between the chemical compositions of the harvested herbage in the three plots in the present study and this indicates uniformity in the quality of the available herbage of the three plots. Moreover, the botanical composition of the sampled herbage was found in general to be similar for all plots except a slightly higher content of forbs in plot SR3 which attained statistical significance mainly due to increase by the end of the trial. The main plant genera composing the harvested herbage were having a similar contribution to that previously reported in Greece (Zervas *et al.* 1999; Koidou *et al.* 2019). Temporary grazing exclusion allows recovery of species richness and productivity in a continuously grazed grassland since grazing pressure acts in favour of the less palatable species (Fedrigo *et al.* 2018).

Daily intake in this trial did not differ significantly between stocking rates in neither of the estimations through the alkanes technique. However, it is known that, unsuccessful mimicking of lambs grazing in collecting herbage samples, allows for a discrepancy between estimates and the actual daily intake, as well as the digestibility of the diet (Valiente *et al.* 2003). Daily intake achieved by herbivores has been generally assumed to be closely related to total daily foraging

time (Newman *et al.* 1995). Lambs in this study spent similar time grazing at the three stocking rates and their intakes were estimated not differing. The importance of total daily foraging time on herbage intake is supported by the observation that, when either the food requirements of herbivores increase or when the availability of food is decreased, then they compensate, up to a certain degree, by increasing their daily foraging time (Penning 1986). Grazing or rumination times, were not different for the lambs grazing plots SR₁, SR₂ and SR₃, (Table 5) indicating that sward height in all cases was above the level where animals would need to increase grazing time to compensate for low bite mass (Hodgson 1985).

The daily allowance of pasturing time in the present trial was mainly occupied by the activity of grazing, but rumination and other non-feeding activities were fitted into the intervals. Clearance of feed residues from the reticulorumen has long been recognized as the major process determining both the intake and nutritive value of forages (Ulyatt *et al.* 1984) which is assumed to depend largely on breakdown of forage particles, by teeth grinding during ingestion and rumination, to the size proper for passage through the reticulo-omasal orifice (Ulyatt *et al.* 1984) and this may explain why lambs in this study spent ruminating about a quarter of their time of daily allowance to pasture. Moreover, lambs spent a substantial 14%, of their daily allowance to pasture, in non feeding activities (mainly idling), indicating that they were satiated and probably had covered their nutritional requirements (Herbers 1981). Grazing activity was not evenly distributed within the day and two distinct peaks of this activity were observed, one at the first two hours after the release of lambs to pasture and the second early in the afternoon (Fig. 1). Similarly to grazing, rumination activity was concentrated in two peaks, one at four hours after the start of grazing and the second at the finish of the lambs' grazing allowance period. This pattern of grazing activity is in accordance with the findings of a similar study conducted at the same location and with grazing lambs of crossed Boutsiko breed (Zervas *et al.* 1999).

In the present study the lambs were allowed a limited to 10 hours pasturing in each day following a common practice of Greek sheep shepherds. Indeed shepherds follow this practice mainly for the conservation of pastures and protection of animals against predators during the night (Volanis *et al.* 2007). This time restriction might be expected to interfere with the

normal diurnal distribution of the grazing bouts of the freely grazing sheep and this would possibly lead to a reduction in grazing time and subsequently in total daily intake. However, as Iason *et al.* (1999) demonstrated, a behavioural compensation of enhanced bite rates or bite mass or the instantaneous rate of intake, which is the combination of these two, on pastures of high food availability would lead to similar herbage intakes between time restricted and freely grazing sheep. Moreover, since a connection has been found between pasturing time and meat quality in grassland systems by Zhang *et al.* (2017) this might be an advisable practice. According to these authors, a healthier meat fatty acid profile for consumers and better pasture management is achieved by limiting the grazing of lambs to 4 h per day rather than grazing over longer time periods.

Levels of RBC, HGB, HCT, MCV, MCH, MCHC and percentage of eosinophils in differential leukocyte counts were not different between lambs raised at different stocking rates (Schalm 1986) and were within normal ranges (Polizopoulou 2010; Mohammed *et al.* 2014). Genera of GIN typically found in the area are *Haemonchus*, *Ostertagia*, *Trichostrongylus*, *Coope-ria*, *Nematodirus*, *Oesophagostomum* and *Chabertia* (Theodoropoulos *et al.* 1998). In conventional animal farming systems chemoprophylaxis has been widely used as the main strategy to control parasitic diseases. However, the control of endoparasitic infections in small ruminants farming, can be effected through a range of alternative methods such as the breeding of animals for parasite resistance (Toscano *et al.* 2019), the use of specialty forage crops, and improved varieties of these, as well as the appropriate stocking densities (Wright *et al.* 2002). In the present study no parasitic nematode larvae were found on the grazed herbage and the faecal egg counts in general were low, without any antiparasitic drugs usage, a finding rather typical for local sheep of Greece, which often exhibit only subclinical infections (Theodoropoulos *et al.* 1998; Theodoropoulos *et al.* 2000). Resistance and resilience to infectious endoparasites appears to be an inheritable trait (Al Kalaldehy *et al.* 2019; Escribano *et al.* 2019), which allows locally adapted sheep to repel infections, as it occurs with their immune response in general (Hadjigeorgiou and Politis 2004). If any parasitic infections were developing in the lambs of the present study, these were weak enough to produce any haematological effects. Since, no effects appeared on these parameters, to indicate parasitic infestation, a strong resilience is assumed. A further observation

was the high numbers of *Moniezia* spp. eggs counted in faecal samples of all lambs at the finish of the grazing trial, but these were not significantly correlated with the individual growth rate of lambs. This finding is in agreement with earlier reports stating that the pathogenicity of *Moniezia* spp. in ruminants is low because the host does not show specific or serious symptoms (Irie *et al.* 2013).

Adaptation of animals to difficult environments, include small body size (Dumont *et al.* 2013), such as the Boutsiko sheep breed used in the present study has. Furthermore, lambs seem to be taking advantage of the available pastures and were growing at a satisfactory growth rate without any feed additives or drugs. Zygogiannis *et al.* (1997) reported for lambs of this breed that average growth rate reaches 162 g/day for the period of 9 to 36 weeks of age, while Panopoulou *et al.* (2001) reported 190 g/day for the period of 6 to 16 weeks of age. However, in both of these studies lambs were feeding a pelleted concentrate diet indoors, therefore differences in lambs management between trials make difficult a direct comparison of the findings. Nevertheless, it is clear that growth rates thrice as high to the observed, can be achieved with lambs of this breed. It is possible that the poor performance recorded in this study is related to naivety to grazing of the lambs used, in association with poor weather conditions occurring during most of the study period.

CONCLUSIONS

It is concluded that the fattening of young lambs exclusively on pasture, under a low input system, can be achieved in Greece, at stocking rates common for the region and during the herbage growth season, without depletion of the sward herbage mass. However, although there is a strong risk of parasitic infestation, since noanthelmintics are administered, the use of a traditional local breed, adapted to its natural environment, can offer serious advantages.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest. All authors have read and approved this research article.

ACKNOWLEDGEMENTS

The authors wish to thank the late Prof Georgios Theodoropoulos for his critical advice on parasitological examination and an earlier draft. This research did not receive any specific funding.

REFERENCES

- Almeida FA, Albuquerque AC, Bassetto CC, Starling RZC, Lins JG, Amarante AF (2020) Long spelling periods are required for pasture to become free of contamination by infective larvae of *Haemonchus contortus* in a humid subtropical climate of São Paulo state, Brazil. *Veterinary Parasitology*279:109060. doi: 10.1016/j.vetpar.2020.109060
- Al Kalaldehy M, Gibson J, Lee SH, Gondro C, van der Werf JHJ (2019) Detection of genomic regions underlying resistance to gastrointestinal parasites in Australian sheep. *Genetic Selection Evolution*51:37. doi: 10.1186/s12711-019-0479-1.
- Altman J (1974) Observational study of the behaviour: sampling methods. *Behaviour*49:227-267. <https://www.jstor.org/stable/4533591?seq=1>
- A.O.A.C. (Association of Official Analytical Chemists International), 1990. 15th Edition. Official Methods of Analysis, Vol.1. AOAC, Arlington, VA.
- Barthram GT (1986) Experimental techniques: the HFRO sward stick. In: HFRO Biennial Report 1984-85, pp. 29-30
- Binnie RC and Chestnutt DMB (1994). Effect of continuous stocking by sheep at four sward heights on herbage mass, herbage quality and tissue turnover on grass/clover and nitrogen-fertilized grass swards. *Grass and Forage Science*49 (2):192-202.
- Bishop SC, Stear MJ (2001) Inheritance of faecal egg counts during early lactation in Scottish Blackface ewes facing mixed, natural nematode infections. *Animal Science*73:389-395. <https://doi.org/10.1017/S1357729800058355>
- Celaya R, Moreno-Gonzalo J, López López C, Ferreira LM, García U, Ferre I, Osoro K (2016) Productive responses of breeding Cashmere goats and their kids to different stocking rates on improved upland pastures. *Journal of Animal Science* 94:1276-86. doi: 10.2527/jas.2015-0053. DOI: 10.2527/jas.2015-0053
- Dove H, Mayes RW (1991) The use of plant wax alkanes as marker substances in studies of the nutrition of herbivores: A review. *Australian Journal of Agricultural Research*42:913-952. <https://agris.fao.org/agris-search/search.do?recordID=AU9200258>
- Dudu A, Popa GO, Ghiță E, Pelmuș R, Lazăr C, Costache M, Georgescu SE (2020) Assessment of genetic diversity in main local sheep breeds from Romania using microsatellite markers. *Archives Animal Breeding*63:53-59. doi: 10.5194/aab-63-53-2020.
- Dumont B, Fortun-Lamothe L, Jouven M, Thomas M, Tichit M (2013) Prospects from agroecology and industrial ecology for animal production in the 21st century. *Animal: An International Journal of Animal Bioscience*7 (6):1028-1043. <https://doi.org/10.1017/S175173112002418>
- Escribano C, Saravia A, Costa M, Castells D, Ciappesoni G, Riet-Correa F, Freire T (2019) Resistance to *Haemonchus contortus* in Corriedale sheep is associated to high parasite-specific IgA titer and a systemic Th2 immune response. *Scientific Reports*9:19579. doi: 10.1038/s41598-019-55447-6.
- EC (2018) EU agricultural outlook for markets and income, 2018-2030. European Commission, DG Agriculture and Rural Development, Brussels. <https://ec.europa.eu/info/food-farming-fisheries/farming/facts-and-figures/markets/outlook/medium-term>
- EU (2012) Regulation EU 1151/2012 of the 21/12/2012 on quality schemes for agricultural products and foodstuffs. <https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX%3A32012R1151>
- FAO (2007) The State Of The World's Animal Genetic Resources For Food And Agriculture, Commission On Genetic Resources For Food And Agriculture Food And Agriculture Organization Of The United Nations, Rome, 2007. <http://www.fao.org/docrep/pdf/010/a1250e/a1250e05.pdf>
- Fedriço JK, Ataide PF, Filho JA, Oliveira LV, Jaurena M, Laca EA, Overbeck GE, Nabinger C (2018) Temporary grazing exclusion promotes rapid recovery of species richness and productivity in a long-term overgrazed Campos grassland. *Restoration Ecology*26 (4):677-685. <https://doi.org/10.1111/rec.12635>
- Gaughan JB, Sejian V, Mader TL, Dunshea FR (2019) Adaptation strategies: ruminants. *Animal Frontiers*9:47-53. <https://doi.org/10.1093/af/vfy029>.
- Geurden T, Hoste H, Jacquiet P, Traversa D, Sotiraki S, Frangipane di Regalbono A, Tzanidakis N, Kostopoulou D, Gaillac C, Privat S, Giangaspero A, Zanardello C, Noé L, Vanimisetti B, Bartram D (2014) Anthelmintic resistance and multidrug resistance in sheep gastrointestinal nematodes in France, Greece and Italy. *Veterinary Parasitology*201:59-66. doi: 10.1016/j.vetpar.2014.01.016.
- Goering HK, Van Soest P (1970) In: Forage Fiber Analysis. Agriculture Handbook, Vol. No 379, U.S. Department of Agriculture, Washington, DC. p. 20.
- Hadjigeorgiou I and Politis I (2004) Seasonal variation in non-specific immunity in relation to management and feeding practices in a semi-extensive dairy sheep farm in Greece. *Small Ruminant Research*53 (1-2):53-60.
- Haile A, Getachew T, Mirkena T, Duguma G, Gizaw S, Wurzinger M, Soelkner J, Mwai O, Dessie T, Abebe A, Abate Z, Jembere T, Rekkik M, Lobo RNB, Mwacharo JM, Terfa ZG, Kassie GT, Mueller JP, Rischkowsky B (2020) Community-based sheep breeding programs generated substantial genetic gains and socioeconomic benefits. *Animal*1-9. doi: 10.1017/S1751731120000269.
- Herbers JM (1981) Time resources and laziness in animals. *Oecologia (Berl)*49:252-262. <https://link.springer.com/article/10.1007/BF00349198>
- Hirschowitz BI (1955) Pepsinogen in the blood. *Journal of Laboratory and Clinical Medicine*46:568-579. <https://www.ncbi.nlm.nih.gov/pubmed/13263737>
- Hodgson J (1985) The control of herbage intake in the grazing ruminant. *Proceedings of the Nutrition Society*44:339-346. <https://doi.org/10.1079/PNS19850054>
- Irie T, Sakaguchi K, Ota-Tomita A, Tanida M, Hidaka K, Kirino Y, Nonaka N, Horii Y (2013) Continuous *Moniezia benedeni* infection in confined cattle possibly maintained by an intermediate host on the farm. *The Journal of Veterinary Medical Science*75 (12):1585-1589. <https://doi.org/10.1292/jvms.13-0250>
- Iason GR, Mantecon AR, Sim DA, Gonzalez J, Foreman E, Bermudez FF, Elston DA (1999) Can grazing sheep compensate for a daily foraging time constraint? *Journal of Animal Ecology*68:87-93. doi/pdf/10.1046/j.1365-2656.1999.00264.x
- Jaimez-RodríguezPR, González-Pech PG, Ventura-Cordero Brito JDRB, Costa-Júnior LM, Sandoval-Castro CA, Torres-Acosta JFJ (2019) The worm burden of tracer kids and lambs browsing heterogeneous vegetation is influenced by strata harvested and not total dry matter intake or plant life form. *Tropical Animal Health and Production*51:2243-2251 <https://doi.org/10.1007/s11250-019-01928-9>
- Khan S, Khan IU, Khan AZ, Zaman S, Majid A, Rehman AU, Khan MA, Maris H, Ullah R, Qureshi S (2020) Evaluating fertility and growth rate potential of indigenous sheep breeds submitted to heat stress under different management systems. *Journal of Advanced Veterinary and Animal Research*7:170-176. <https://doi.org/10.5455/javar.2020>.

g407

- Kerboeuf D (1977) Changes in pepsinogen, proteins and lipids in the serum during experimental haemonchosis in sheep. *Annales de Recherches Vétérinaires. Annals of veterinary*8:257-266. <https://www.ncbi.nlm.nih.gov/pubmed/606141>
- Koidou M, Mountousis I, Dotas V, Zagorakis K, Yiakoulaki M (2019) Temporal variations of herbage production and nutritive value of three grasslands at different elevation zones regarding grazing needs and welfare of ruminants. *Archives Animal Breeding*62 (1):215-226. <https://doi.org/10.5194/aab-62-215-2019>
- Kondyli E, Svarnas C, Samelis J, Katsiari MC (2012) Chemical composition and microbiological quality of ewe and goat milk of native Greek breeds. *Small Ruminant Research*103 (2-3):194-199. <https://doi.org/10.1016/j.smallrumres.2011.09.043>
- Kumar N, Rao TK, Varghese A, Rathor VS (2013) Internal parasite management in grazing livestock. *Journal of Parasitic Diseases*37 (2):151-157. <https://doi.org/10.1007/s12639-012-0215-z>
- Marković B, Dovč P, Marković M, Radonjić D, Adakalić M, Simčič M (2019) Differentiation of some Pramenka sheep breeds based on morphometric characteristics. *Archives Animal Breeding*62:393-402. doi: 10.5194/aab-62-393-2019.
- Martin RR, Beveridge I, Pullman AL, Brown TH (1990) A modified technique for the estimation of the number of infective nematode larvae present on pasture, and its application in the field under South Australian conditions. *Veterinary Parasitology*37:133-143. [https://doi.org/10.1016/0304-4017\(90\)90068-M](https://doi.org/10.1016/0304-4017(90)90068-M)
- Mateo L, Delgado P, Ortuno J, Banon S (2018) Maternal grazing on stubble and Mediterranean shrubland improves meat lipid profile in light lambs fed on concentrates. *Animal*12:1547-1554. doi: 10.1017/S1751731117003263
- Mpofu T, Nephawe KA, Mtšeni B (2020) Prevalence of gastrointestinal parasites in communal goats from different agro-ecological zones of South Africa. *Veterinary World*13 (1):26-32. DOI: 10.14202/vet-world.2020.26-32
- Mohammed A, Campbell M, Youssef FG (2014) Serum copper and haematological values of sheep of different physiological stages in the dry and wet seasons of central Trinidad. *Veterinary Medicine International* 972074. <https://doi.org/10.1155/2014/972074>
- Newman JA, Parsons AJ, Thornley JHM, Penning PD, Krebs JR (1995) Optimal diet selection by a generalist grazing herbivore. *Functional Ecology*9:255-268. <https://www.jstor.org/stable/2390572>
- Orr RJ, Parsons AJ, Penning PD, Treacher TT (1990) Sward composition, animal performance and the potential production of grass/white clover swards continuously stocked with sheep. *Grass and Forage Science*45:325-336. <https://doi.org/10.1111/j.1365-2494.1990.tb01957.x>
- Panopoulou E, Koutsotolis K, Charismiadiou M, Zervas G (2001) The effect of feeding methods on carcass and adipose tissue characteristics of Boutsiko breed lambs. *Animal Science Reviews*28:57-71 (in Greek, with English abstract).
- Papaloukas L, Sinapis E, Arsenos G, Kyriakou G, Basdagianni Z (2016) Effect of season on fatty acid and terpene profiles of milk from Greek sheep raised under a semi-extensive production system. *Journal of Dairy Research*83 (3):375-382. doi:10.1017/S0022029916000327
- Penning PD, Parsons AJ, Orr RJ, Hooper GE (1994) Intake and behaviour responses by sheep to changes in sward characteristics under rotational grazing. *Grass and Forage Science*49 (4):476-486.
- Perucho L, Hadjigeorgiou I, Lauvie A, Moulin CH, Paoli JC, Ligda C (2019) Challenges for local breed management in Mediterranean dairy sheep farming: insights from Central Greece. *Tropical Animal Health and Production*51:329-338. <https://doi.org/10.1007/s11250-018-1688-2>
- Phocas F, Belloc C, Bidanel J, Delaby L, Dourmad JY, Dumont B, Ezan- no P, Fortun-Lamothe L, Foucras G, Frappat B, González-García E, Hazard D, Larzul C, Lubac S, Mignon-Grasteau S, Moreno CR, Tixer-Boichard M, Brochard M. (2016) Review: Towards the agroecological management of ruminants, pigs and poultry through the development of sustainable breeding programmes: I-selection goals and criteria. *Animal*.10:1749-1759. doi:10.1017/S1751731116000926
- Polizopoulou ZS (2010) Haematological tests in sheep health management. *Small Ruminant Research* 92:88-91. <https://doi.org/10.1016/j.smallrumres.2010.04.015>
- Prache S, Bechet G, Thériez M (1990) Effects of concentrate supplementation and herbage allowance on the performance of grazing suckling lambs. *Grass and Forage Science*45 (4):423-429. <https://doi.org/10.1111/j.1365-2494.1990.tb01967.x>
- Ross JG, Purcell DA, Dow C, Todd JR (1967) Experimental infections of calves with *Trichostrongylus axei*; the course and development of infection and lesions in low level infections. *Research in Veterinary Science*8:201-206.
- Ruiz-Huidobro C, Sagot L, Lugagne S, Huang Y, Milhes M, Bordes L, Prévot F, Grisez C, Gautier D, Valadier C, Sautier M, Jacquet P (2019) Cell grazing and *Haemonchus contortus* control in sheep: lessons from a two-year study in temperate Western Europe. *Scientific Reports*9:12699. doi: 10.1038/s41598-019-49034-y.
- Schalm OW, Jain NC, Carroll EJ (1986) *Veterinary Haematology*. 3rd edition. Lea and Febiger, Philadelphia, pp. 209-213.
- Thamsborg SM, Roepstorff A, Larsen M (1999) Integrated and biological control of parasites in organic and conventional production systems. *Veterinary Parasitology*84:169-186. DOI:10.1016/s0304-4017(99)00035-7
- Theodoropoulos G, Koutsotolis K, Nikolaou E, Kalogiannis D, Petrakos G (1998) Seasonal variation of gastrointestinal nematodes of sheep in the region of Joannina, Greece. *International Journal for Parasitology*28:1287-1292. DOI:10.1016/s0020-7519(98)00100-3
- Theodoropoulos G, Zervas G, Kouneli A, Martínez-Gonzales B, Petrakos G, Kostopoulos J (2000) Seasonal patterns of strongyle infections in grazing sheep under the traditional production system in the region of Trikala, Greece. *Veterinary Parasitology*89:327-335. [https://doi.org/10.1016/S0304-4017\(00\)00215-6](https://doi.org/10.1016/S0304-4017(00)00215-6)
- Thienpont D, Rochette F, Vanparijs OFJ (1986) Diagnosing Helminthiasis by Coprological Examination. *Jansen Research Foundation* pp. 39-41.
- Toscano J, Okino CH, Dos Santos IB, Giraldele LA, von Haehling MB, Esteves SN, de Souza Chagas AC (2019) Innate Immune Responses Associated with Resistance against *Haemonchus contortus* in Morada Nova Sheep. *Journal of Immunology Research* 3562672. <https://doi.org/10.1155/2019/3562672>
- Ulyatt MJ, Dellow DW, John A, Reid CSW, Waghorn GC (1984) Contribution of chewing during eating and rumination to the clearance of digesta from the ruminoreticulum. In: Milligan, L.P., Grovum, W.L., Dobson, A. (Eds), *Control of digestion and metabolism in the ruminants*. Prentice-Hall, Englewood Cliffs, New Jersey 07632, pp. 498-515.
- Valiente OL, Delgado P, De Vega A, Guada, JA (2003) Validation of the n-alkane technique to estimate intake, digestibility, and diet composition in sheep consuming mixed grain: roughage diets. *Australian Journal of Agricultural Research*54 (7):693-702.
- Volanis M, Stefanakis A, Hadjigeorgiou I, Zoiopoulos P (2007) Supporting the extensive dairy sheep smallholders of the semi-arid region of Crete through technical intervention. *Tropical Animal Health and Production*39 (5):325-334. DOI:10.1007/s11250-007-9019-z
- Wojtkowski PA (2008) Agroecological Economics Sustainability and Biodiversity. *Agroecological Economics* 1-12 <https://doi.org/10.1016/B978-012374117-2.50003-0>

- Wright IA, Zervas G, Louloudis L (2002) The development of sustainable farming systems and the challenges that face producers in the EU. In: Kyriazakis, I., Zervas, G. (Eds) Organic meat and milk from ruminants. EAAP publication 106:27-37.
- Zervas G, Hadjigeorgiou I, Zabeli G, Koutsotolis K, Tziola C (1999) Comparison of a grazing with an indoor-system of lamb fattening in Greece. *Livestock Production Science* 61:245-251. <https://doi.org/10.1016/S0301>
- Zhang XQ, Jin YM, Badgery WB (2017) Diet selection and n-3 polyunsaturated fatty acid deposition in lambs as affected by restricted time at pasture. *Scientific Reports* 7 (1):15641. <https://doi.org/10.1038/s41598-017-15875-8>
- Zygiogiannis D, Kyriazakis I, Stamataris C, Friggens NC, Katsaounis N (1997) The growth and development of nine European sheep breeds. 2. Greek breeds: Boutsko, Serres and Karagouniko. *Animal Science* 65:427-440. <https://doi.org/10.1017/S1357729800008626>