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Effects of heat stress on welfare in Karagouniko sheep breed

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Abstract. The sustainability of sheep production in the Mediterranean basin faces considerable challenges in front of the ever-escalating effects of climate change, with animal welfare seriously compromised due to heat stress and the heat stress accompanied are the most severe effects. The Karagouniko sheep is a hardy local breed very well adapted in specific environments and low input farming systems. In this article an estimation of the effects of heat stress on the welfare of Karagouniko ewes is presented, utilizing the AWIN Welfare Assessment Protocol for Sheep[®]. The Karagouniko sheep flock, where the experiment took place, is managed semi-extensively at a semi-mountainous area in Thessaly, Greece. Severe and extreme severe heat stress conditions were recorded throughout the experimental period. Despite this, ewes' welfare indices remained in their entirety within desirable levels, highlighting the importance of local farming practices.

Keywords. Welfare - Sheep - Karagouniko breed - Heat Stress.

Les effets du stress thermique sur le bien-être dans la race ovine Karagouniko

Résumé. La viabilité du secteur ovin dans le bassin méditerranéen est confrontée à des défis considérables face aux effets croissants du changement climatique. Le bien-être des animaux est en danger à cause du stress thermique et ses effets si graves. La race ovine Karagouniko est une race locale rustique très bien adaptée aux environnements spécifiques et dans des systèmes d'élevage à faibles intrants. Dans cet article on présente une évaluation préliminaire des effets du stress thermique sur le bien-être des brebis Karagouniko en utilisant le protocole de l'évaluation de bien-être des ovins, AWIN (Welfare Assessment Protocol for Sheep®). Le troupeau de la race ovine Karagouniko où l'expérimentation a eu lieu, est géré dans un système semi-extensif dans une zone semi-montagneuse en Thessalie, en Grèce. Des conditions du stress thermique sévères et extrêmement sévères ont été enregistrées tout au long de la période expérimentale. Malgré tout, les indicateurs de bien-être des brebis sont restés à des niveaux satisfaisants, soulignant l'importance des pratiques d'élevage locales.

Mots-clés. Bien-être animal – Brebis – Race ovine Karagouniko – Stress thermique.

I – Introduction

The sustainability of sheep production in the Mediterranean faces considerable challenges before the ever-escalating effects of climate change. Given that the Mediterranean basin has been identified as a highly susceptive region to climate change and accompanying systemic failures as a result of it, severe constraints to sheep productive efficiency are already observed (IPCC, 2014). Sheep welfare is seriously compromised due to the variety of climatic stresses that are imposed to them, with heat stress being accompanied with the most severe, detrimental effects (Battini et.al, 2016; Al-Dawood, 2017; Karatzia *et al.*, 2015; 2015; 2016; 2016; Sejian *et al.*, 2017; Rivas *et al.*, 2019; Silanikove, 2000).

Sheep breeding is considered one of the most dynamic sectors of the rural activity in Greece with around 8.5 million sheep raised in 83.8 thousand farms (data 2019, Hellenic Statistical Authority), making up for 14% of the total EU sheep population (Eurostat, 2020). This population is charac-

terized by a diversity of local breeds and local populations that co-exist with exotic breeds and various crossbreds raised in a variety of production environments. A major part of this population is managed in low input farming systems, capitalizing on the availability of natural pastures and the successful adaptation of the animals to their respective microclimates (Perucho *et al.*, 2018; Hatziminaoglou, 2001; Ligda *et al.* 2009).

In the region of Thessaly in Central Greece, the Karagouniko is the main local sheep breed raised on semi-extensive lowland farms. The Karagouniko sheep, predominant in the region in 1985 (Georgoudis *et al.*, 1995), shows decreasing trends and currently 4,500 ewes are under milk recording scheme, in a total estimated population of 50,000 heads (data 2019, EFABIS).

The Karagouniko sheep is highly appreciated by farmers for its milk production and ability to adapt in a variety of environments and low input farming systems (Perucho *et al.*, 2019). Recent studies in the region, showed that some farmers of Thessaly decided to reintroduce Karagouniko breeding males in highly productive crossbred flocks (involving exotic breeds) aiming to improve the flock response to some constraints of the biophysical environments, such as the sensitivity to thermal stress (Perucho *et al.*, 2019).

Although this trend is recent and observed on a limited number of farms, the future challenges associated with climate change in the Mediterranean area suggest that the use of local breeds to respond these challenges could increase in the future. In this context, there is increasing interest to study the response of the Karagouniko sheep managed in extensive or semi-extensive systems at different stressors that compromise animals' welfare.

The aim of the present study is to estimate the effects of heat stress on the welfare of Karagouniko sheep, taking into consideration local farming practices and climatic conditions.

II – Materials and methods

The study was carried out in a commercial dairy farm of Karagouniko sheep breed which was managed semi-extensively and was located in Avra-Kalambaka, Greece (39.714844, 21.683770) at an altitude of 220m. The flock consisted of 125 sheep, 117 of which were ewes. The animals grazed in natural and artificial pastures located in the greater area close to the farm. When necessary, supplemental nutrition of vetch, oats and corn was offered on farm. Welfare assessments, one per summer month (June-July-August), both at individual and group level were undertaken for all ewes at second lactation represented 10% of the flock. Assessments were conducted by the same researcher to ensure the validity of the data collected. All assessments were performed in accordance to the AWIN Welfare Assessment Protocol for Sheep® (2015). Ambient temperature and relevant humidity were recorded at 5-min intervals by digital sensors during grazing and on-farm (26.036 pairs of measurements) from May until September.

Thermal Heat Index was calculated according to Marai et al. (2007), by the following model:

T.H.I.= Environmental Temperature – (0.31-0.0031 * Relative Humidity) * (Environmental Temperature-14.4),

Where: T.H.I. < 22.2 indicated absence of heat stress,

22.2 ≤ T.H.I. < 23.3 indicated moderate heat stress,

23.3 ≤ T.H.I. < 25.6 indicated severe heat stress, and

T.H.I. ≥ 25.6 indicated extreme severe heat stress

Individual Thermal Heat Index was calculated by substituting Environmental Temperature with Mean Body Temperature as it was estimated by the average temperature of the forehead, the wither, the rump, the udder, the flank and the chest, using an infrared thermometer.

Additionally, rectum temperature was recorded, along with respiratory rate and body condition score, after time was allowed for the animals to rest on farm upon the return of the flock from pasture.

Behavioural observations and measurements at animal and flock level were performed (shadow-seeking, lethargic behaviour etc.), in combination with the welfare assessment protocol for sheep AWIN[®] (2015) on first and second level (Individual and farm level)

Data collected were statistically analysed using SPSS[®] v.24, at a confidence level of a = 0.05.

III – Results and discussion

Heat Stress: Environmental Monthly Thermal Heat Index was calculated on June, July and August (EMTHI) and on the specific visit days of welfare assessment (VDTHI) when the flock was assessed (from the time animals left the farm for grazing until the time they returned) (Table 1). Mean Individual Thermal Heat Index (MITHI) was calculated on visit days (one/month), using the Individual Thermal Heat Index of the 12 ewes participating at the experiment (Table 1).

Table 1. Mean values of Thermal Heat Indexes (Environmental Monthly Thermal Heat Index-EMTHI, Visit Days Thermal Heat Index-VDTHI and Mean Individual Thermal Heat Index-MITHI)

	-			,	
Month	EMTHI		VDTHI	MITHI	
	(Mean ± SE)		(Mean ± SE)	(Mean ± SE)	
June	24.56 ± 0.055 ^a	1 st Visit (June)	24.06 ± 0.385 ^a	28.76 ± 0.124 ^a	
July	26.00 ± 0.042 ^b	2 nd Visit (July)	23.55 ± 0.390 ^a	26.86 ± 0.190 ^b	
August	24.66 ± 0.056 ^a	3 rd Visit (August)	27.06 ± 0.408^{b}	29.13 ± 0.287 ^c	

Values within a column with different superscripts differ significantly at $a \le 0.05$.

Environmental Monthly Thermal Heat Index ranged between 24.56 and 26.00, indicating the prevalence of severe heat stress throughout the experimental period. A corresponding trend was recorded for Visit Day Thermal Heat Index, with 1st and 2nd visit index remaining within severe heat stress limits, while on the 3rd visit, VDTHI crossed over, increasing significantly (P≤0.05), indicating extreme severe heat stress conditions. In partial accordance to VDTHI, Mean Individual Thermal Heat Index reduced significantly (from 28.76 to 26.86, P≤0.05) at the second visit, before increasing significantly at the 3rd visit (from 26.86 to 29.13, P≤0.05) and approaching extreme severe heat stress levels.

Body Condition Score (BCS): This indicator was assessed while the ewes were restrained, by palpitation of the spine in the lumbar region, at the point after the last rib. The BCS classification was based on Morgan-Davies C. *et al*, 2007. Scores for all three visits can be seen on Table 2. Body Condition Score was evaluated as Good (>2.0, <4.0) with spine processes easily distinguished with light pressure, clear muscle, and fat cover in all ewes.

Table 2. Mean values of Body Condition Score	and their classification
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	BCS (Mean ± SE)	Maximum	Minimum	Classification
1 st Visit (June)	3.20 ± 0.071	3.5	2.75	Good
2 nd Visit (July)	3.35 ± 0.066	3.5	3	Good
3 rd Visit (August)	3.2 ± 0.072	3.5	3	Good

The increase in BCS that was recorded at the 2nd visit, along with its reduction at the 3rd visit could be possibly attributed to the short-term rainy period that was observed during early July and resulted in rapid growth of vegetation in pastures, which allowed for the flock to have access to high quality and increased quantity grass.

Water availability: Animals had access to communal water troughs on pasture which utilized natural water sources, functioning, clean, unpolluted, and accessible. On a course of 7.4 kilometres on average, that sheep covered at each grazing, 2-3 communal water troughs were available (depending on the route they chose). On farm, water was available in 3 metal sheet troughs 5 meters long, where animals had access to fresh water upon their return from pasture and at nighttime. The troughs on farm were also fully functioning and cleaned regularly.

Fleece cleanliness: Most sheep were clean and dry with no signs of dirt or contamination on their fleece. It should be noted that the animals were sheared right before the onset of the experiment as is common practice in Greece, to help alleviate animals of the effects of heat stress and maintain feed conversion efficiency. 86.7% of sheep were scored as 0, and 13.3% of sheep were scored as one, which is attributed to the fact that during the second visit on the farm the weather was rainy for 5 days and animals got slightly damp during grazing.

Panting: This indicator was assessed before sheep were handled for other measurements and while they were in the pens. No animals with open panting were observed. On average, 33.3% exhibited normal respiration with an average of 27 ± 2.9 breaths per minute with the mouth closed, while 66.6% of the sheep showed signs of mild heat stress with a respiration rate of 34 ± 3.6 breaths per minute, where respiration occurred with the mouth closed and so, was not scored as panting.

Respiration Quality: After panting assessment and observation for hampered or audible breathing and persistent coughing and while ewes were handled, breathing was evaluated. No respiratory issues were detected in any animals. Breathing was normal with no obvious efforts to draw breath, no audible noise accompanying breathing, no coughing, and no nasal discharge.

Access to shade and shelter: While on pasture or during the grazing route followed by the flock daily, multiple sights with trees and shrubs functioning as shade/shelter were available, often in the vicinity of water troughs. The sheep utilized these sights by resting there when heat was severe or when it was raining heavily. The shrubs there also served as sources of feed.

Stocking Density: The indicator was assessed on farm, where adult ewes without lambs were estimated to have approximately 1.4 m^2 of pen area per animal at their disposal. Feeding throughs were located both in the pen and at the yard, to minimize overcrowding when supplementary feed was offered.

Hoof Overgrowth/ Lameness: All animals assessed showed no signs of hoof overgrowth. As the flock walked long distances twice daily for grazing, natural wear of the hoofs occurred, and no lameness issues were recorded (Score 0-Not lame). As the flock approached the farm after grazing, the movement of the ewes was assessed as smooth, with weight equally borne on all four feet with no shortening of stride. Some minor head nodding which is acceptable was evident, as animals were walking on uneven ground.

Body and Head Lesions/ Leg Injuries: No red skin patches, scabs, skin lesions, or wounds (current and healed) were observed at any area of the body (head and neck, ears, eyes, body) of the assessed sheep. At the first visit, a minor heat rash was recorded in two animals, at the flank area, due to the shearing that had been performed two days prior to the first evaluation. The rashes in both ewes had healed by the second visit and caused no distress or appetite decrease. Additionally, a complete absence of swellings, hairless patches, callus, lesions, or scabbed areas on the joints of the legs, which would indicate arthritis, injuries or trauma, or prolonged lying on hard surfaces were recorded.

Faecal Soiling: Very light soiling (Score 1), described as a small quantity of faecal matter in the wool around the anus was assessed in 66.6% of the ewes. Light soiling and dags (Score 2), specified as some soiling around the anus and dags (matted areas of faecal matter adhering to the wool) in this area only, was recorded in 33.3% of the animals. As the breed has a long semi-fat tail, that occasionally can even reach the ground, soiling can be observed often.

Mucosa Colour: The colour of the conjunctiva of the bottom eyelid of the ewes was inspected and evaluated as not anaemic in all animals. On average of the three visits, the 16.7% was classified by Score 0 (deep red mucosa), while the rest by Score1 (bright ted mucosa).

Ocular Discharge: Due to irritation caused by little twigs on gorse plants (*Pistacia lentiscus*), ocular discharge was observed in 2 animals at one visit (5.55%) and was treated by the farmer after consulting a veterinarian.

Mastitis and Udder Lesions: The indicators were recorded as the ewes were gently restrained in a standing posture and the udder was inspected from behind for colour and symmetry, palpated on both sides feeling for lumps, hardness and fibroids. No lesions to the udder or teats were found in any animals, as well as no mastitis or lesions were present. Udders in all animals were soft and piable at palpation, without any redness or hardness.

Fleece Quality: As previously mentioned, the flock was sheared two days prior to the first assessment and as a result fleece quality was not be evaluated. No bald patches or shed areas were found on any ewes.

Tail Length: The indicator was observed both in unhandled animals and verified in handled ewes. Karagouniko sheep have semi-fat long tails and common practice in commercial farms is to avoid docking lambs' tails. Undocked tails extended approximately below the hocks in all animals assessed.

Social Withdrawal/Stereotypy: The undisturbed flock was observed for 20 minutes, upon their return on farm from pasture. No single animals were clearly apart from the rest of the social group, standing at the back of a pen, standing or lying apart from main body of the flock and not engaged in any maintenance activity and unresponsive to activity occurring around them. Stereotypy assessment was also performed after the flock was left undisturbed for 20 minutes. Repetitive pacing or circling where the animal follows the same route back and forth or around the pen; repeatedly curving the head back over the shoulders and looking upwards; repeatedly pulling as well as biting or plucking the wool along the back of another ewe, were not observed. Surprisingly, a characteristic stillness behaviour (Budging) was observed. Budging was spontaneously manifested upon return from grazing, when sheep would hoard at the darkest area of the building and remained still and in immediate contact with each other. This behaviour could originate from a visceral reaction to avoid sunlight in order to achieve heat stress alleviation.

Excessive Itching: The undisturbed flock was observed for 20 minutes, upon their return on farm from pasture. No animals showed signs of excessive itching: repeated or prolonged rubbing or scratching, against pen or paddock fixtures or with the hooves. Ewes did not roll the head backwards over the shoulders attempting to scratch with the horns.

Quality Behaviour Assessment: Four observation points were selected in the pens with five minutes of observation per observation point. All ewes were alert (observant and vigilant), relaxed (at ease, free from anxiety, agitation or tension and they appeared to be unthreatened), content (satisfied and at peace, with their needs met), sociable (seeking and interacting with other sheep, appeared to be enjoying/taking comfort from their contact and chose to be part of a flock and not fully isolate themselves), calm (placid and sedate), bright (alert, lively and aware of environment) and assertive (displaying confidence or determination).

Familiar Human Approach Test: The farmer was asked to approach the sheep in the normal manner on foot, as if he was inspecting sheep. The purpose of this assessment was to gauge whether the farmer can feasibly approach his stock to carry out an inspection. Record the closest possible distance of approach before a flight response is elicited. No flight response was triggered (sheep remained motionless at human approach) and the ewes actively moved towards (sheep walked directly towards the farmer) and interacted (sniffing, nosing) with the farmer.

IV – Conclusions

Farms on semi-mountainous/mountainous areas are confronted with Heat Stress effects at an escalating intensity as semi-extensive management practices offer little protection to sheep from the adverse effects of climate change and microclimate both on farm and pasture is of integral importance for the protection of the flock's welfare. In spite of the challenges encountered on a daily basis, the production system applied in the semi-intensive farm studied, allowed for the management of sheep more as individuals rather than as a flock and for the daily adaptation of grazing schedule and duration by the farmer, according to the flock's needs. This fact provided sheep with the flexibility of adjusting their grazing and resting habits according to weather conditions and Heat Stress effects, thus protecting their welfare. In support of the above, Mean Individual Thermal Heat Index was significantly increased - and decreased - in accordance to Visit Day Thermal Heat Index, indicating that ewes have the ability to quickly adapt to the surrounding environmental conditions and de-escalate the effects of heat stress. The manifestation of budging behaviour on flock level upon return from grazing is a characteristic example of behavioural adaptation to environmental conditions, as an effort to achieve metabolic homeostasis, originating from a visceral reaction. This collective animal behaviour as a form of social reaction involving the coordinated action of the whole flock accentuates the capability of Karagouniko sheep to engage strategies that protect their welfare. Despite the prevalence of individual extreme severe heat stress, ewes' welfare indices remained in their entirety within desirable levels, highlighting the importance of local farming practices (such as shearing at the beginning of the summer and tail docking avoidance).

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