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CAMEL HOMEOSTASIS AND MEAT QUALITY AFFECTED BY STRESS BEFORE SLAUGHTER- A LITERATURE REVIEW

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Abstract
The welfare of dromedaries before and at slaughter influences their homeostasis and the organoleptic characteristics, quality, and commercial shelf life of their meat. Transportation, stocking density, travel time, waiting time at the slaughterhouse, water and food deprivation, and the slaughter procedure are the most important stressors for this species. In dromedaries, these events trigger behavioral, physiological, hematological, biochemical, hormonal, and muscular responses that could be assessed using reliable and specific biomarkers, namely, heart rate, respiratory rate, rectal temperature, hemolysis, neutrophil/lymphocyte ratio, enzymes, glycolytic potential, malondialdehyde, thiols, carbonyls, catecholamines, cortisol, and thyroid hormones. The dromedary, a large and difficult to handle animal, is faced with a lack of adopted transport vehicles, equipment, resources and training of breeders, drivers, and technicians on animal welfare, and with the slaughter procedure, which favors even more stressful situations. This literature review analyzes the impact of stress induced by the different stages of handling before slaughter on homeostasis and meat composition in dromedaries, using the results provided by recent work carried out on this theme. The dromedary is subjected to more stressful handling compared to ruminant species, which begins at the farm and then at the market, and continues with loading, transportation, distance traveled, stocking density during transportation, unloading, receiving, waiting time in slaughterhouses, deprivation of food and water, and the method of slaughter. These conditions do not meet international animal welfare standards resulting in significant alterations of homeostasis and meat quality of this species. Legislation on the welfare of dromedaries at all stages of the pre-slaughter process must be developed.

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Introduction

The one-humped dromedary (*Camelus dromedarius*) is considered a fundamental element of the economy and culture of arid and semi-arid environments, capable of providing various services, as it is used for transportation, recreation, racing, and production of meat, milk, leather, and fiber [1,2]. Historically, the dromedary was widely used as the main means of transporting humans and goods between countries, hence its nickname "ship of the desert" [3]. Thanks to its physiological adaptations to heat and dehydration, this animal thrives in dry, semi-arid, and tropical climates of Africa, Asia, and Oceania [4,5]. Furthermore, this species provides additional zootechnical value to people living in these environments [6].

The phases preceding slaughter, including capture, loading, transport [7], housing at the slaughterhouse, deprivation of water and feed [8,9,10], and stocking density [11,12] are considered major stress factors for dromedary camels and can have adverse effects on their health, welfare, and other aspects determining meat quality. It is therefore not surprising that camels must also experience positive emotions to achieve their well-being of which health is an important element [13,14,15]. However, while laws have been established in developed countries to protect the welfare of farm animals at various stages of production, transport, and slaughter [16,17], in Africa and Asia, there is no legislation or standards governing and protecting their welfare.

Camels share many similarities with other ruminants, but their specific needs are distinct due to their anatomical, physiological, and behavioral differences [1]. However, they are social, calm, docile, and peaceful herd animals that maintain strong bonds within the group and preserving their welfare requires experience with them, their handling and management, as well as knowledge of animal welfare, which recommends training in animal behavior and welfare for those handling and managing dromedaries [13,14]. Padalino et al. [18] recently surveyed 61 employees at the Birqash camel market in Cairo, Egypt, and found that the understanding of animal welfare was low and its meaning seemed to be ignored. In dromedaries, various markers have been identified as strong indicators of pre-slaughter stress, including physiological, biochemical, hematological

and endocrine changes [7,8,9], leading to increased cortisol levels and lipid and protein oxidation [11,19], behavioral variations [20], changes in blood concentrations of several metabolites [21] and decreased immunity, which could have serious consequences for their welfare and immune function, as well as for the quality of their products [22,23]. Thus, early detection of stress is one of the most effective strategies to counter these disorders. In this species, the most common pre-slaughter stressors are loading, road transport, unloading, stocking density, stabling, water and food deprivation and ambient heat. However, the combination of behavioral, hematological, physiological, biochemical and hormonal measures will be of great use to reliably assess transport-induced stress in dromedaries. The present literature review discusses aspects and concerns about the impact of stress occurring before slaughter during transport, loading, unloading and lairage handling, and bleeding, on the homeostasis and meat composition of camels. This review exposes health and welfare, neurophysiology and assessment of stress, behavioral, hematological, hormonal biomarkers of stress, oxidant, thermal and pathological stress, and meat alteration by stress in camels, food safety of the consumer, legislation, conclusion and recommendations for protecting the welfare of camels during transport and at slaughterhouses.

Objects and methods

This manuscript narratively reviews studies focused on the impact of management factors during the preslaughter phases (i. e., handling, loading, unloading), and conditions of transport (i. e., stockage density in vehicle), water and food deprivation, and lairage (i. e., duration of waiting and stabling) on physiological, hematological, biochemical, hormonal, oxidant and behavioral responses and meat characteristic variation in camels. The present review includes previously published secondary data such as research articles from reputable journals, books, annual reports from national and international organizations, policy briefs, and other indexed scholarly materials related to the preslaughter stress in camels. Databases included Google Scholar, PubMed, ScienceDirect and Web of Science. After identifying the sources, the search criterion developed was that the literature must use keywords such as welfare, stress, preslaughter stress, thermal stress, heat stress, stress assessment, stress behavioral responses, stress hematological responses, stress hormonal responses, stress pathological responses, oxidant stress, meat alteration and meat safety, and availability of the full-text version. On the other hand, the review papers and articles irrelevant to the topic under study, or presenting no detailed results, were excluded from the review process. The papers were subjected to a primary screening of titles or/and abstracts performed independently by four authors. A total number of 145, 194, 98, and 125 documents were initially collected from Google Scholar, PubMed, ScienceDirect and Web of Science sites, respectively, and were screened for eligibility studies. From the first database, 130 references were related to camel following implementations of various inclusion and exclusion criteria and were selected for full review.

Camel health and welfare

According to Anses [24], animal welfare is defined as the positive mental and physical state linked to the satisfaction of its physiological and behavioral needs, as well as its expectations. This state varies depending on the animal's perception of the situation. Animal welfare has become an important topic, which must be taken into account when assessing legal requirements. Thus, good management practices and good husbandry conditions on a universal scale have stimulated significant research [25], and several studies have established the link between the welfare of farm animals during the handling stages [12,15] before slaughter and the quality and sustainability of their meat. Recently, in their work, Padalino and Faye [26] focused on health management, environmental conditions, handling, and behavioral assessment in dromedaries. Thus, the different authors agree on five freedoms guaranteeing the welfare of these animals, according to sanitary, physiological, zootechnical and psycho-behavioral criteria. The first recommends access to water and feed in appropriate quantities meeting the needs and physiological status of the animal species. For the second freedom, the breeding conditions must not induce psychological suffering to the animal. For the third, the animal must be in physical and thermal comfort. The fourth recommends the absence of pain, lesions and disease of the animal. The fifth freedom consists of the expression of normal behavior of the species. In this context, several research studies have focused on the stress of the dromedary camel under different conditions (disease management, husbandry practices, water and food deprivation, dehydration, loading and stocking density in the truck, transportation, travel time and waiting time at slaughterhouses, harsh environmental conditions and slaughter methods, revealed among the most effective factors that increase stress and, consequently, influence the quality of carcasses and meat [7–9,11,27]. The reactions of the dromedary camel to these stressful situations have been highlighted by the analysis of reliable physiological, biochemical, haematological [7–9], hormonal [10,12,21], behavioral [20], and tissue parameters [11,22]. Camels can be exposed to stress during breeding in traditional or intensive production systems, and during transport on foot or by truck. On the other hand, there is a clear and well-known link between the welfare of the dromedary and the quality of its products, and poor handling of this species could contribute to poor quality of its meat.

Neurophysiology of stress

Once stress is detected, the information is sent to the thalamus and the prefrontal cortex, which analyze the importance of the stressor [28]. Stress stimulates the sympathetic adrenal medullary system and then the

hypothalamic-pituitary-adrenal cortex (HPAC) axis to release catecholamines and corticosteroids from the adrenal glands, which is why measuring the concentrations of these hormones in the blood, especially cortisol, is commonly used as a reliable biomarker of stress [29,30]. However, under stressful events, and in order to ensure the body gets readily available energy in an emergency action, mammals produce the catecholamines released first by the sympathetic nerve endings and then by the adrenal medulla, then cortisol which remains active in the body longer than these catecholamines [31]. Thus, for the evaluation of stress, the quantification of cortisol, the most common glucocorticoid molecule (or of its metabolites), has been shown to be the most direct and reliable indicator of the physiological state of an animal, and an index of its response following activation of HPAC axis and an environmental threat of its homeostasis [30,31]. The activation of HPAC axis by stress stimulates the release of corticotrophin-releasing factor by the hypothalamus, which stimulates the adenohypophysis to secrete adrenocorticotropic hormone in blood circulation which in turn stimulates the secretion of cortisol by the adrenal gland [31].

Stress assessment in different biological matrixes

The difficulties in obtaining blood samples and the recognition of the stressful effect of blood sampling are the main drivers of the use of minimally invasive sampling media as biomarkers of adrenal cortex responses to animal stress. In the dromedary camel, biomarkers include in addition to blood [8,9,32–35], urine [12,36], feces [37,38], hair [38] and saliva [39] in different stressful situations such as road transport, dehydration and environmental heat.

Normal values for cortisol levels in serum of healthy camels ranged between 3.6 and 32.6 ng/ml with a mean ± SEM of 10.3 ± 0.6 ng/ml. Saeb et al. [35] reported a serum level between 26 and 40 nmol/L (9.4 and 14.5 ng/mL), and normal values range reported by Faye and Bengoumi [40] were between 3 to 30 ng/ml. According to Hussen and Althagafi [23], serum cortisol levels were significantly higher in female camels (10.8 ± 0.6 ng/ml) than males (7.9 ± 0.9 ng/ml), in non-pregnant (12.1 ± 1.0) than pregnant (10.2 ± 0.8) female camels, and in lactating female camels (12.5 ± 1.0 ng/ml) than non-lactating (9.2 ± 0.7 ng/ml) female camels. During other physiological adaptations, cortisol increased in camels from baseline levels of about 21.9 ± 1.0 ng/mL to over 121.6 ± 5.4 on the day of parturition or increased from 37.1 ± 1.4 ng/mL one day before weaning to 48.0 ± 1.5 and 69.5 ± 1.9 ng/mL at weaning and 3rd day after weaning, respectively [41].

The dromedary camels were defined as stressed animals when their blood cortisol levels were between 40 and 60 ng/ml and were accompanied by obvious signs of fear (restlessness, moaning, kicking, defecation) [42]. However, in addition to the levels of cortisol measured in the blood, those analyzed in the hair and feces could be useful for a reliable retrospective assessment of long-term stress [37,38]. Collection of these samples minimizes stress on the animal and it is easy to transport and store them. In fact, it has been shown in dromedaries that an intravenous injection of adrenocorticotrophic hormone (0.5 mg/animal) increased the level of cortisol in the blood and that of glucocorticoid metabolites in the feces [37]. Thus, twenty-four hours after this injection, the level of cortisol increased from 0.6–10.8 to 10.9–42.2 ng/mL in the blood, and from 286.7 to 2559.7 ng/g in the faeces [37]. Furthermore, in the camel, blood concentrations of cortisol were positively correlated with those analyzed in saliva and/or hair or feces [38]. Cortisol levels in saliva fluctuate within minutes, while the production of feces can take hours to days depending on the species. Furthermore, in the dromedary camel, cortisol levels in blood were higher in females than males and in young animals than adult, and were positively correlated with those analyzed in saliva and/or hair or feces [38,43,44].

Behavioral responses

Dromedaries are generally known for their need for space to display their innate physical activity behavior [45]. After arriving at slaughterhouses following truck transport, camels showed disruptions in certain behaviors related to transport stress, including comfort (lying and standing), ingestion (feeding and rumination), elimination (defecation and urination), and body grooming (rubbing, scratching, and biting) [46]. According to Atkinson [47], animals subjected to road transport stress spent most of their time lying down during and at the end of the journey, and during the recovery period [48]. Camel exhaustion generally depends on their standing or crouching position during transport, and is related to the period and conditions of confinement in the moving means of transport [20,49]. In addition, the loading density of camels is the main concern for the animal during the journey [50], as it is a limiting factor for the animal to lie down in the truck during a long journey, and to be able to get up after a fall.

The frequency of grooming and scratching behavior is considered one of the signs of good health and vitality of animals [51]. In dromedaries, this frequency was reduced or absent during transport, which could be considered the first sign of disease and the main sign of distress, exhaustion and stress [20,52].

Feeding behavior disorders have been observed during transport and stabling and could be explained by the lack of space and social restrictions [52,53]. These disorders could be favored by the lack of watering and shelter or by dehydration induced by transport [54,55]. In addition, after being transported on foot or by trucks, dromedaries often continue to wait in markets or slaughterhouses without eating or drinking, which constitutes a situation of risk of health and behavioral problems in addition to that of transport stress, and which deserves attention to be improved and limited [12,25,56]. A decrease in feed consumption during rest periods following transport, associated with behavioral changes, was reported in dromedaries

in a study conducted in Qatar [57]. Additionally, in dromedaries, road transport was responsible for a decrease in the time spent on ingestive behavior [20], as was the case in goats [58]. This effect could be explained at least in part by the prolonged period of dehydration resulting from the long journey, especially in the absence of stops for feeding and watering [59].

Finally, behavioral responses such as slipping, falling, belching, urination, and defecation were observed in dromedaries as stress responses recorded at the end of road transport, during unloading and transport to the stabling area. The frequencies of these responses were positively and significantly correlated with serum cortisol and Malondialdehyde (MDA) levels analyzed in the same animals [7,8]. Indeed, these behavioral responses to stress could be interpreted as pain, agitation, and fear in the presence of conditions that do not respect animal welfare during and after transport and unloading [60,61]. Furthermore, a decrease in elimination behavior by defecation and/or urination during transport of dromedaries was reported by Bekele et al. [59] and Emeash et al. [20], in contrast to transported cattle, which showed an increase in elimination frequency [62,63]. According to Fraser and Broom [16], the frequency of defecation and urination increases at the beginning of the journey under stress, then gradually decreases as the animal adapts and its feces become dry and scanty due to lack of food and water, dehydration and decreased intestinal activity during the journey.

Hematological parameters

Transporting dromedaries on foot had caused a decrease in the number of red blood cells (RBC) which could result from the stress of prolonged walking [20,64,65], while an increase in this count, hematocrit and hemolysis had been noted under the effect of transport in vehicles in camels [66] and goats [67]. According to El Khasmi et al. [7], these parameters were positively correlated with the transport distance. The increase in RBC count and hematocrit observed in dromedary camels 3 hours after rest following transport was lower than that recorded after 10 hours of rest [68]. This increase could be related to body dehydration due to thermoregulation and urination and the associated water deprivation, or to sympathetic activation of the adrenal gland, responsible for an increase in the synthesis and release of catecholamines into the circulation, which stimulates contraction of the spleen, thereby releasing RBC into the circulation [69–72]. Stress induced by handling during transport was able to reduce the leukocyte count resulting in a decrease of the immunity and disruption of the homeostasis in steers [65] and camels [20,68,73]. It should also be noted that camel transportation by truck was responsible for a significant increase in neutrophil-to-lymphocyte ratio (NLR) [7,20], and the values of this parameter were positively correlated with blood cortisol, glucose, and lactate levels under transport stress [21,50,74]. According to Lemrhamed et al. [50], dromedaries showed significantly elevated NLR and hemolysis at the end of a 2-hour transport with a high load density (1.44–1.80 m^2/dromedary). Under these same conditions, this increase persisted 12 to 16 hours after the end of transport [21]. In addition to their association with leukocytosis and neutrophilia (higher NLR), in dromedary camels, blood cortisol levels have also been associated with an increase in the CD4 T cell population, a reduction in the percentage of γδ T cells, a decrease in the expression of CD172a on neutrophils and monocytes, a decrease in the expression of CD14 and CD163 on monocytes, and an increase in the expression of CD45 and MHC I on lymphocytes [23].

Hormonal parameters

According to numerous studies, in camels, high blood cortisol concentrations are a reliable indicator of physical, psychological and physiological stress [7,35,36,75–77]. The response of camels to stressful situations has been investigated in several studies. Indeed, in camels that were trucked approximately 150 km in 2 to 3 hours, serum cortisol levels were significantly higher just after transportation compared to non-transported control camels (172.1 ± 29.6 ng/mL vs. 33.5 ± 3.8 ng/mL) [20]. Other studies have reported elevated cortisol levels in the same species transported by truck [7,35,36,66]. According to El Khasmi et al. [7], highly significant elevations in blood cortisol levels (between 88.32 ± 19.4 and 152.4 ± 25.18 ng/ml) as a function of journey length (between 72 and 170 km) were noted in dromedaries. In a recent study, serum cortisol levels were analyzed in 50 male dromedaries at the slaughterhouse, and they were high (80.2–107.2 ng/ml) to very high (133.7–198.0 ng/ml) in 26, and low in the other 24 camels (13.0–67.9 ng/ml) [23]. Similarly, Sayah et al. [78] measured high to very high serum cortisol levels in male Sahrawi camels at the slaughterhouse. Furthermore, serum samples collected from camels at the slaughterhouse, directly after transport and before slaughter, showed cortisol levels ten times higher than those of serum samples collected from camels in their breeding environment (116.6 ± 15.8 ng/ml vs. 6.8 ± 0.4 ng/ml) [23]. In a recent study conducted at the Casablanca slaughterhouse in Morocco, Lemrhamed et al. [8] observed that the most stressed camels were subjected to a long waiting period before loading (> 24 h), and during loading (> 15 min), unloading (> 5 min), water and food deprivation (> 24 h), and driving to the slaughter room (> 11 min), and they all had elevated serum cortisol concentrations. However, cortisol levels recorded in different studies are highly variable and remain difficult to compare [61].

Additionally, a high loading density in the transport truck, a long waiting period at the slaughterhouses after transport and the slaughter procedure induced a significant increase in blood cortisol levels in the dromedary camel [21,50,74]. Similarly, Sayah et al. [78] reported higher cortisol levels in dromedaries kept in stalls for a short period (874 ± 631 ng/ml) after arrival and before slaughter (1 to 8 h) than in animals kept for a longer period (48 to 96 h) (127 ± 39 ng/ml). The increase in

cortisol in dromedaries during the different stages of their handling before slaughter could be due to their presence in an unknown environment and situation [79]. The released cortisol will play the role of an energy regulator meeting the metabolic and physiological needs of the animals [80]. It will induce catabolic activity in peripheral tissues (glycogenolysis, proteolysis and lipolysis) and anabolic activity in the liver (gluconeogenesis and protein synthesis) necessary for stress management [81,82]. In general, an increase in blood cortisol levels is recorded after 15 minutes of exposure to stress, then returns to basal levels after approximately 3 hours [82,83]. The animal could return to normal values after a rest period, but a period of lairage that does not respect good welfare conditions for the dromedary could again increase the concentrations of these hormones [74]. More recently, Sayah et al. [78] evaluated the effect of lairage time on animal stress and meat quality in male dromedaries of the Sahraoui population. The authors found that animals subjected to short-time lairage (from 1 to 8 h) were characterized by high cortisol and plasma glucose levels, compared to those subjected to long-time lairage (from 48 to 96 h). According to Kadim et al. [84], the return to the basal physiological and behavioral state of the dromedary, under conditions of food deprivation but with access to water, was observed after a lairage period of 12 to 24 hours. In dromedaries, in addition to cortisol, other hormones have been favored as indicators of stress. Indeed, road transport [36], a long waiting period at slaughterhouses after transport and the slaughter procedure, have been able to increase blood levels of thyroid hormones [21,74]. On the other hand, the period of handling of farm animals before slaughter is generally marked by deprivation of water and food, which could cause physiological stress marked by body dehydration. Indeed, camels that were severely stressed by dehydration showed a significant increase in cortisol (9.57 ± 0.72 ng/mL vs. 1.39 ± 0.05 ng/mL), noradrenaline, and dopamine, and a significant decrease in adrenaline and serotonin [85] in order to preserve body water, maintain cardiac responsiveness, and produce energy.

Oxidant stress

Increased production of reactive oxygen species (ROS) under oxidative stress (OS) can induce the formation of lipid peroxidation products and oxidized proteins, as well as oxidative damage to DNA and RNA. These ROS are normally eliminated by enzymatic antioxidants like catalase (CAT), glutathione peroxidase (GSH-Px), superoxide dismutass (SOD), etc, and non-enzymatic antioxidants like vitamins C and E, glutathione, melatonin, etc [86]. Indeed, incubation of red blood cells with these vitamins was able to attenuate oxidative alterations of these cells by hydrogen pyroxide and reduce the production of MDA in the incubation medium [87,88].

Under the effect of transport stress, dromedaries showed an imbalance in oxidant-antioxidant status, marked by an increase in SOD activity and a decrease in GSH-Px and tissue ascorbic acid [33,89]. In addition, El Khasmi et al. [7] evaluated the OS related to road transport distance and found that plasma cortisol and MDA levels as well as catalase activity increased progressively and significantly with transport distance. Furthermore, in a survey conducted recently in the slaughterhouse of Casablanca in Morocco, Lemrhamed et al. [8] recorded the duration of the preslaughter operations and the frequency of urination in camels. The authors found that the most stressed camels were submitted to long preslaughter operations and water and food deprivation period, and they all showed a high frequency of urination. Furthermore, these animals showed high circulating levels of MDA, and low CAT and SOD activities [8].

On the other hand, in the muscle (*Musculus abdominis obliquus externus)* of camels with high circulating cortisol levels (80.29–107.21 ng/mL) after transport, MDA levels were higher and CAT activities were lower than in camels with low cortisol levels (13.07–67.9 ng/mL) [9]. Thus, analyzing circulating cortisol levels after transport just before slaughter [7,21,50,66,74], can predict meat quality and bone status in camels [9,90]. Furthermore, in the same species, Barka et al. [90] reported that MDA levels increased significantly while CAT decreased significantly in meat (*Triceps brachii*, *Musculus obliquus*, and *Diaphragma*) when the transport distance before slaughter increased (72 km vs. 160 km).

Camels severely stressed by dehydration showed a significant increase in malondialdehyde and certain antioxidants such as glutathione, retinol, thiamine, and vitamin E [85]. In fact, in this species, Ali et al. [91] investigated the effects of long-term dehydration stress on kidney cortex and medulla with respect to pro-inflammatory markers and oxidative stress, and found a significant increase in the cytokines IL-1β and IL-18 levels, MDA and GSH, and a significant decrease in SOD and CAT. Camel granulosa cells exposed to 45 °C for 2 h also showed an increase in MDA levels associated with a significant increase in the expression of heat shock proteins (HSPs), DNA repair enzymes, and glutathione S-transferase [92].

Finally, exposure of dromedaries to cold-induced stress during the winter season resulted in a significant decrease in blood levels of certain antioxidants such as vitamins A, C, and E, and glutathione associated with an increase in ROS [33,93], suggesting a low antioxidant status of these animals during the winter season [88].

Thermal stress

When faced with environmental heat stress, the dromedary camel showed variations of physiological, biochemical, hematological and hormonal parameters. High ambient temperature increases the ruminant's effort to dissipate body heat, which leads to an increase in heart rate, respiratory rate, core body temperature and water consumption, while food consumption decreases [94–97]. In camels, a decrease in blood glucose in hot climates has been noted in an attempt to reduce their basal metabolic rate to a minimum level. This decrease in glucose utilization could be

attributed to glucose consumption by respiratory muscles, decreased food intake, and hyperinsulinemia [98,99]. The increase in heart rate could be an attempt to improve cardiac output, thereby increasing blood flow to the peripheral circulation and facilitating greater heat loss through increased water evaporation at the body surface or the airway mucosa, which helps prevent hyperthermia [100,101].

Regarding the impact of summer heat stress on blood cortisol levels, in dromedaries, a higher cortisolemia (ng/mL) in summer than in winter was observed by Baraka [43] (38.6 ± 5.3 *vs* 28.5 ± 4.8) and Elias and Weil [102] (45.0 ± 11.9 *vs* 8.0 ± 1.3). A similar trend was reported in the same species for corticosteronemia by Zia-ur-Rahman et al. [34]. These authors explained this hyperactivity of the adrenal cortex observed in summer by the stress induced by the external heat load and the dehydration of the body following intense sweat losses, as a major thermolytic pathway. However, other studies, analyzed the values of cortisol in serum, hair and faeces at two distinct periods in 20 male dromedaries aged 3 to 8 years, from semi-extensive farms in the region of Essaouira and intended for slaughter at municipal slaughterhouses in Casablanca, Morocco. These studies reported that cortisol levels in serum (ng/mL), hair and feces (ng/g) were significantly higher in winter than in summer (66.01 ± 13.19 *vs* 25.71 ± 6.71; 0.93 ± 0.26 *vs* 0.61 ± 0.08 and 2.74 ± 0.14 *vs* 1.42 ± 0.35, respectively; $p < 0.05$) [38,103].

Concerning total vitamin D and 25-hydroxyvitamin D, in dromedary, serum levels were significantly higher during the summer season compared to the winter season and showed no significant variation with the season, in the liver, kidney, meat and hump [11,103,104]. According to Shany et al. [105], in dromedary, serum levels of 25-hydroxyvitamin-D (ng/mL) in summer and winter were 443 ± 96 and 276 ± 13, respectively. The increase in the levels of vitamin D and its metabolites in summer could be explained by the strong sunshine stimulating their biosynthesis. Other studies have shown that circulating vitamin D levels in llamas, alpacas and camels [106,107] are not influenced by age, but vary seasonally. These levels were highest during the period from February to July [107]. Variations in circulating vitamin D concentrations in camelids could also be impacted by coat color, month of birth, light intensity and physiological stage [106–109].

During transport, heat stress within the transport vehicle space is recognized as the main threat to animal welfare and health [110]. In fact, in the dromedary camel, a 2-hour road transport by truck during the hot season resulted in a significant increase in rectal temperature (°C), heart rate (beats/min) and respiratory rate (cycles/min) compared to the values measured before transport (40.2 ± 0.4 vs. 38.1 ± 0.3; 58 ± 4 vs. 46 ± 3 and 17 ± 2 vs. 11 ± 2, respectively) [66]. In addition, by comparing the effect of two loading densities in trucks on these physiological parameters, Lemrhamed et al. [50] found that heart rate and respiratory rate were significantly higher at high stocking density (1 camel/1.44–1.80 m^2) than those observed at low stocking density (1 camel/2–3.6 m^2) (62 ± 4 vs. 47 ± 4 and 20 ± 2 vs. 12 ± 1, respectively) which could incriminate thermal stress due to the reduction of surface area for animals' bodies and lack of ventilation. Rectal temperature and heart and respiratory rates, measured just after the camel was transported, approached basal values after 3 hours of rest and returned to their basal values after a 12–16 hours rest period [21,68].

In addition, camels transported by truck in a hot environment showed a significant increase in plasma MDA concentrations up to 24 hours after the end of transport, hyperthermia and increased hematocrit due to water loss through thermolysis, and hemolysis [19,66] suggesting oxidative alterations of the red blood cell plasma membrane [7].

Furthermore, Moussahil et al. [11] recently studied the *antemortem* effect of heat stress associated with high stocking density during transport, on the chemical composition of meat at the 24-hour post-mortem stage in the dromedary camel. The authors observed a significant decrease in pH and catalase activity, and a significant increase in exudate and cooking losses, electrical conductivity, and malondialdehyde and carbonyl contents in meat from animals transported at 29–35 °C with a stocking density of 1 animal/1.74–2.13 m^2 compared to animals transported under moderate temperature and stocking density conditions (21–23 °C and stocking density of 1 animal/3.12–4.31 m^2). Furthermore, dromedary meat exposed to heat (80 °C) was more oxidized than that exposed to 18 °C, showing high contents of malondialdehyde, carbonyls and peroxides, and lower activities of enzymatic antioxidants (CAT, SOD and GSHPx) [27].

Pathological stress

Livestock are exposed to environmental stresses during transport, such as noise, heat, cold, humidity, movement, and unusual social groupings. Furthermore, handling, loading and storage conditions in the vehicle, unloading, and confinement are responsible for distress, injury, and mortality, particularly when transport is not properly managed [111].

According to a recent study, a high incidence of respiratory problems, such as pneumonia, was observed in camels transported long distances in poor conditions [8,112]. This incidence could be explained by the presence of several well-known predisposing risk factors, including inappropriate handling, transportation, mixing, and overcrowding [3]. Transportation remains the main factor leading to stressful conditions and decreased welfare in dromedaries [35], which influences their physiology and behavior [20,55]. Transportation has been mentioned as the main cause of respiratory problems, diarrhea, and colic [18]. Therefore, the resulting pain causes camels suffering from these problems to exhibit abnormal behaviors at the slaughterhouse, associated with higher serum cortisol levels capable of modulating the immune system of these animals [15,23,113,114]. On the other hand, there is a high risk of spreading infectious diseases through various

handling of livestock before and during transport [115]. Finally, when animals are exposed to heat stress for a long time, they could experience health problems, infertility, decreased growth, production and immune defense, and cellular and mitochondrial oxidative damage [116]. Thus, it is recommended to improve the transport conditions of these animals in order to preserve their health and welfare during and after transport, not only for them, but also for the welfare of people likely to consume their meat [12]. Table 1 summarizes the mean values of different stress and oxidative stress indicators measured before slaughter in the blood and after slaughter in the meat of camels.

Meat alteration

Different stress conditions before and/or during the slaughter of farm animals significantly influence the postmortem biochemical transformation of their muscle into meat, by modifying the evolution of its ultimate pH (24 h postmortem stage) (Figure 1).

The camel meat is more prone to quick drip loss and *post-mortem* variations in pH, lipid oxidation rates, and oxidative metabolism due to high myoglobin and polyunsaturated fatty acids concentrations [117–120], thus, it would be more likely to be influenced by the various antemortem manipulations of the dromedary.

Table 1. Mean values of different stress and oxidative stress indicators analyzed before slaughter at the blood level and after slaughter at the meat level in camels

Stressors	Samples	Stress biomarkers	Non stressed	Stressed	References
Transport	Blood	Cortisol (ng/mL)	28.58±0.52	152.4±25.18	[44]
		Thyroxine (nM)	154.3±12.6	216.7±24.3	[36]
		Glucose (mM)	5.07±0.28	7.08±0.21	[7]
		Lactate (mM)	9.97±0.31	12.99±0.16	
		Hematocrit (%)	39.17±1.17	43.17±0.98	[66]
		MDA (nM)	1.58±0.38	3.88±0.20	[7]
		CAT (kU/L)	79.13±3.84	60.08±3.18	
	Meat	Glycogen (mmol/kg)	39.4–42.6	34.5–37.5	[36]
		24h *postmortem* pH	5.60±0.11	6.60±0.11	[9]
		Carbonyls (nmol/mg)	1.15±0.11	1.87±0.13	[22]
		Haem pigment (µg/g)	10.18±1.06	13.39±1.18	[9]
		MDA (nmol/kg)	112.74±15.53	157.23±18.41	[22]
		CAT (U/kg)	364.13±19.34	301.53±17.48	
		SOD (µmol/min/ mg)	8.67±0.14	8.67±0.14	
		Exsudate (%)	0.47±0.12	0.76±0.13	
		Drip loss (%)	9.23±1.12	12.56±1.45	[9]
		Cooking loss (%)	21.36±2.64	30.23±3.51	
Loading density	Blood	Hematocrit (%)	33.59±2.18	35.15±3.32	[50]
		NLR	0.91±0.01	1.31±0.11	
		Glucose (mM)	6.50±0.03	7.20±0.02	
		Cortisol (ng/mL)	25.21±0.67	53.33±5.33	
		T_3 (nM)	1.426±0.174	3.569±1.25	
		Thyroxine (nM)	66.50±5.86	110.8±19.93	
	Physiology	Cooking loss (%)	37.7±0.4	40.2±0.4	
		HR (beats/min)	47±4	62±4	
		RR (cycles/min)	12±1	20±2	
Heat	Meat	MDA (nmol/kg)	145±2	281±2	[11]
		Carbonyls (nmol/mg)	1,47±0,01	1,83±0,02	
		CAT (µmol/h/mg)	10,80±0,61	9,00±0,56	
		SOD (µmol/min/ mg)	9,23±0,14	7,86±0,11	
		Water retention (mg/g)	161,46±23,24	112,13±21,33	
		Exsudate (%)	0.43±0.04	0.72±0.06	
		Drip loss (%)	22,63±2,15	28,54±2,34	
		Conductivity (µS/ cm/g)	41,59±3,53	52,45±4,37	
Waiting duration in abattoir	Blood	NLR	0.91±0.1	1.31±0.1	[21]
		Cortisol (ng/mL)	25.21±2.67	54.33±5.33	
		T_3 (nM)	1.42±0.17	3.56±0.25	
		Thyroxine (nM)	66.50±6.86	117.8±16.93	
		Glucose (mM)	7.28±0.03	8.14±0.02	

MDA — malondialdehyde, SOD — superoxide dismutase, CAT — catalase, GSHPx — glutathion peroxidase, HR — heart rate, RR — respiratory rate, NLR — neutrophile lymphocyte ratio, T_3 — tri-iodothyronine.

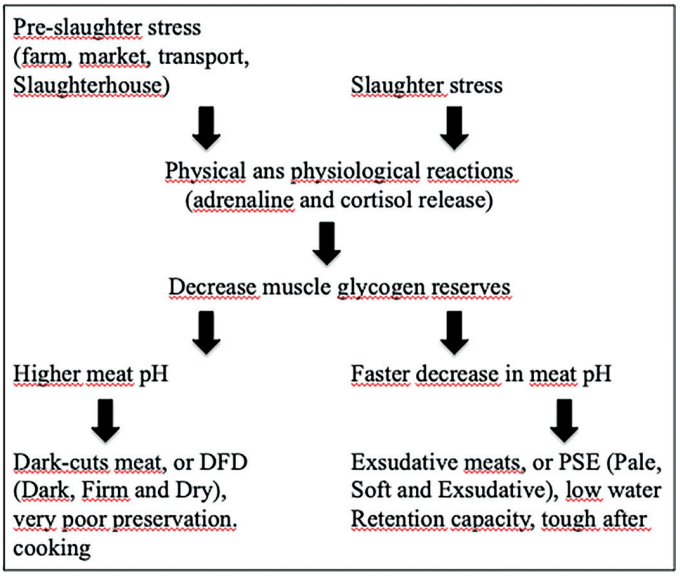

Figure 1. Impact of preslaughter and slaughter stress on the technological and sensory qualities of meat

Long transport distances are stressful factors that could affect the ultimate pH (pHu) (pH at 24 h postmortem) of camel meat. According to Barka et al. [90], the pHu values of muscles (*Triceps brachii*, *Musculus obliquus* and *Diaphragma*) had been higher in camels transported for 160 km compared with camels transported for 72 km only (6.4 ± 0.2 *vs* 5.7 ± 0.2; 6.5 ± 0.2 *vs* 5.6 ± 0.1 and 6.3 ± 0.1 *vs* 5.6 ± 0.2, respectively). High pHu values in camel meat had been associated with high circulating levels of cortisol at slaughter and low post-mortem levels of glycogen in meat [36]. The glycogen levels in camel muscles (*Triceps brachii*, *Musculus obliquus* and *Diaphragma*) (mg/100g) decreased significantly at high distance compared with short one (170 ± 20 *vs* 226 ± 25; 191 ± 21 *vs* 241 ± 27 and 180 ± 23 *vs* 237 ± 25, respectively) [90]. The low glycogen levels might be explained by the duration of the fast [121]. In the dromedary camel, the *antemortem* circulating levels of cortisol were positively correlated with the values of *postmortem* pH, drip loss, cooking loss, dimensional shrinkage and total haem pigment in meat [9]. However, in the same species, although blood levels of 25-hydroxyvitamin D were not influenced by transport stress, the content of this vitamin D metabolite in meat was negatively correlated with dripping loss and cooking loss [122], suggesting a possible role of this vitamin in dromedary meat quality.

At the end of transport, animals' inadequate waiting conditions at the slaughterhouses showed negative effects on the quality of the camel meat. More recently, Sayah et al. [78] evaluated the effect of lairage time on meat quality of male dromedaries of the Sahrawi population. The authors found that animals subjected to long-time lairage (from 48 to 96 h) were characterized by meat with high pH, mineral content, and moisture at 24 h postmortem, compared to those subjected to short-time lairage (from 1 to 8 h), probably due to depletion of glycogen reserves and lactic acid accumulation in the pre- and postslaughter period, and animal fatigue [117,123]. Additionally, Moussahil et al. [22] studied the impact of the durations of different stages prior to slaughter on the physicochemical and biochemical parameters of dromedary meat. The authors found that high duration of transportation (10–11 h), unloading (11–20 min), driving to the slaughterhouse (11–20 min), slaughter (11–20 min) and bleeding (8–9 min) were associated with significantly high exudate content, cooking loss, carbonyl and MDA contents, and significantly low catalase and superoxide dismutase activity in meat. Interrelationship of stress factors occuring before slaughter and their biomarkers and meat quality is presented in Figure 2.

Food safety

The World Organization for Animal Health regards pre-slaughter treatments and handling as important for animal welfare and the nutritional quality of their products [124]. However, handling of camels that does not comply with ethical rules or animal welfare protection laws alters organoleptic, sensory, and technological parameters, which could impact consumer health [84,117]. Moreover, particularly in camel-rearing countries where many diseases are prevalent, animals that have been stressed before

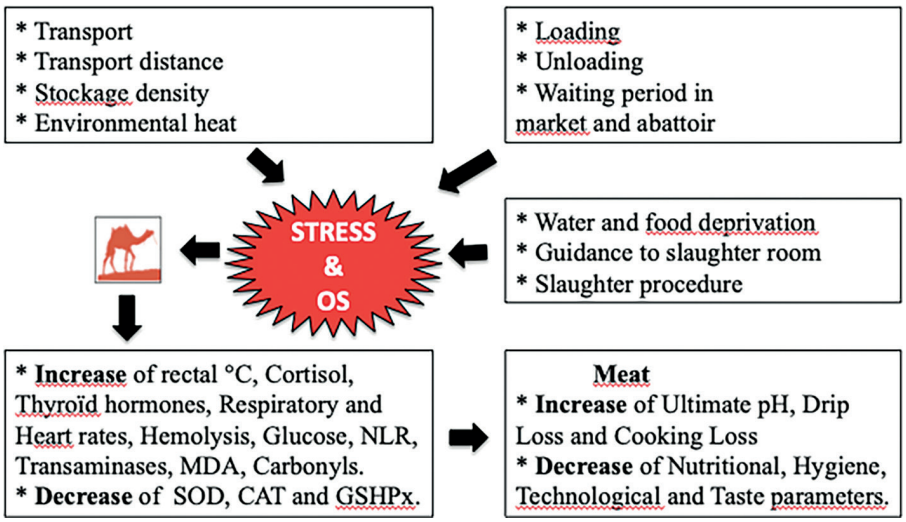

Figure 2. Interrelationship of pre-slaughter stress factors and biomarkers and meat quality (°C — temperature, NLR — neutrophile lymphocyte ratio, MDA — malondialdehyde, SOD — superoxide dismutase, CAT — catalase, GSHPx — glutathion peroxidase)

and during slaughter have pathogen levels ten times higher than normal, and produce poor-quality meat [124]. The sympathetic activity of the adrenal medulla and the adrenocorticotropic axis during stress affects the body's normal functioning, leading to decreased productivity and resistance to infections. Furthermore, the activity of these glands under stress reduces muscle glycogen concentrations, thus significantly reducing meat quality and the shelf life of meat products [61,84]. It should be added that exposing animals to pre-slaughter stress (transport, dehydration, hunger, overcrowding, mental stress, slippery floors, poorly designed premises, poor equipment, excessive noise, and darkness) further weakens their immunity and their ability to resist pathogens, and causes health problems for slaughterhouse employees and consumers [124].

Other factors can affect the nutritional and hygienic quality of meat and, consequently, the health of consumers. Camel slaughter is carried out without stunning, with the legs tied with a rope, in the presence of pain and injuries and without restraint. Camels are slaughtered in a crouching position with their heads held in a caudal position [74], which makes the animals excited and agitated, and reduces the likelihood of having good quality meat [61]. Slaughterers are not trained in animal welfare and food safety and do not have the technical knowledge or experience necessary to carry out their tasks [124].

Examples of guidelines for protecting the animal welfare

Unlike developed countries, very little information is available on camel welfare in relation to mishandling at slaughterhouses in underdeveloped countries. Indeed, measures relating to health controls, animal welfare protection and slaughter procedures have been regulated in Europe (Regulations EC853/2004, 854/2004 and 1099/2009 of the European Commission), Canada (Canadian Food Inspection Agency), the United Kingdom (Department of Environment, Food and Rural Affairs), the United States (United States Department of Agriculture) and New Zealand (National Advisory Committee on Animal Welfare, within the Ministry of Primary Industries) [124]. As an example of guidelines for protecting the welfare of dromedaries during slaughter, "The Australian Animal Welfare Standards and Guidelines" (www.animalwelfarestandards.net.au) is aimed at minimizing their stress, pain, and suffering. These animals are given 24 hours to move around and explore their new environment to acclimatize, reduce stress, and facilitate handling. Feeders and waterers are required for those confined for more than 24 hours. When loading, short, straight alleys and low-incline ramps must be used. Within 24 hours before slaughter, animals are examined by meat safety inspectors to ensure their good health and the quality of their meat, which is fit for human consumption. Just before slaughter, animals are led through a passageway to the slaughterhouse, where they enter a stunning cage and are then slaughtered by severing the main vessels in the neck. The slaughterhouse must meet these standards to obtain Animal Welfare Certification.

Conclusion

The welfare of farm animals and its impact on the quality of their products has long been of interest to scientific researchers in developed countries. However, this topic remains largely unexplored in developing countries, especially in relation to dromedaries, so, no country has conducted any tests or procedures to monitor or control the welfare of camels during transport, sale or slaughter, either through official veterinary services or an animal protection association. Like other domestic animals, dromedaries cannot escape stressful conditions that begin on the farm, at the breeding site, and at the market, and continue with loading, transportation, distance traveled, stocking density during transport, unloading, reception, waiting time, deprivation of water and food, and the slaughter process. This species is more susceptible to these various stress factors and oxidative stress, which can alter camel homeostasis and meat quality, potentially endangering animal health and, consequently, public health and food safety. In Morocco, like in other countries of Africa and the Middle East, the animal welfare standards of the World Organization for Animal Health are often not respected, and are almost non-existent when it comes to dromedaries. Indeed, their transportation and slaughter are not sufficiently regulated by law and are not subject to any official welfare controls. Similarly, since most countries in these regions do not have animal welfare laws, the development of specific laws and regulations addressing this issue remains mandatory, particularly with regard to the transportation, handling, deprivation of water and food, stabling, and slaughter of dromedaries. Furthermore, the vehicles are multipurpose and are neither adapted nor designed for the transport of dromedaries, nor equipped with loading and unloading devices, and offer no protection against harsh thermohygrometric conditions. The animals are also handled roughly by untrained and unqualified operators. Reduction of the time spent on various camel handling operations prior to slaughter, transport means and loading/unloading equipment appropriate for this species are required. Finally, to our knowledge, gaps in current research still exist regarding the topic of pre-slaughter stress/camel welfare/product quality/consumer safety, and no country has conducted tests or procedures to monitor and control camel welfare during transport or slaughter, or to analyze organoleptic and sensory quality under stressful conditions, either through official veterinary services or an animal protection association.

Recommendations

With the aim of addressing the behavioral needs of camels while adopting appropriate and sustainable technologies and practices that respect their welfare, we make the following immediate, medium-term, and long-term recommendations:

Immediate recommendations

- Provide camels with access to a clean water source and feed before slaughter to avoid prolonged periods of hunger and thirst.
- Keep camels in groups to demonstrate their social behaviors.
- Prevent camels from becoming injured, stressed, or overexcited during transport to the slaughterhouse and before slaughter.
- Prevent rough handling of camels by untrained and unskilled operators.
- Avoid prolonged periods of time spent on various camel handling operations before slaughter, such as transport, loading, unloading, and stabling.
- Unload camels as soon as possible after arrival at the slaughterhouse.
- At the end of the transport, unload and move the camels to the stabling area and then to the slaughterhouse, after separating sick or injured ones.
- Never slaughter camels in front of other camels.
- Never leave sharp knives in front of the camels to be slaughtered.
- In the slaughterhouse, thoroughly clean the bleeding trap before slaughtering each camel.
- Encourage respect for camel welfare, their inspection at slaughterhouses and the inspection of their meat by veterinary services to protect consumer health.

Medium-term recommendations:

- Camels must express their feeding behavior, have access to pasture, and be free to walk.
- Camels must have a dedicated area for their handling and observation.
- V-shaped circulation corridors must be used to guide camels after unloading in good conditions.
- Comfortable areas for movement, comfortable rest, and protection from pain, distress, and suffering must be provided, while avoiding poorly designed facilities and allowing animals to recover properly after the transport stress.
- Veterinarians must be regularly present at markets and slaughterhouses to care for camels and ensure their welfare during transport and slaughter.
- Persons handling or transporting camels must have practical experience and/or sufficient training in animal welfare.

Long-term recommendations:

- Slaughterhouse staff must be trained and public awareness about camel welfare and consumer health must be raised.
- Camel transport drivers must be trained and certified to obtain a mandatory license.
- Facilities that allow for the expression of social behaviors, other behaviors, and good human-animal relations must be provided.
- Guidelines and procedures for unloading, holding, and moving camels at markets and then to slaughterhouses must be developed.
- Vehicles must be suitable and designed for transporting dromedaries, equipped with suitable loading and unloading equipment, and protected against harsh thermohygrometric conditions.
- Procedures for stunning dromedaries before bleeding that comply with halal requirements must be developed.
- Finally, national legislation on the welfare of dromedaries at all stages of the process prior to slaughter, in accordance with international standards, must be developed.

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Completely prepared the manuscript and is responsible for plagiarism.

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