



**IJVR**

ISSN: 1728-1997 (Print)  
ISSN: 2252-0589 (Online)

**Vol. 27**

**No. 3**

**Ser. No. 96**

**2026**

# **IRANIAN JOURNAL OF VETERINARY RESEARCH**



## Review Article

# Road transportation of domestic animals: Biochemical and physiological aspects

Nazifi, S.<sup>1\*</sup> and Yaghoobpour, T.<sup>2</sup>

<sup>1</sup>Department of Clinical Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran; <sup>2</sup>Ph.D. Student in Clinical Pathology, Department of Clinical Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran

\*Correspondence: S. Nazifi, Department of Clinical Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran. E-mail: nazifi@shirazu.ac.ir



10.22099/ijvr.2026.55072.8291

(Received 2 Dec 2025; revised version 7 Jul 2026; accepted 9 Aug 2026)

This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>)

## Abstract

Transportation exposes domestic animals to multiple stressors, including environmental changes, noise, vibration, inadequate ventilation, and temporary deprivation of feed and water. The magnitude of the physiological response depends on factors such as transport duration, temperature, humidity, vehicle movement, and animal-related characteristics, including age and previous transport experience. These stressors can trigger endocrine, metabolic, and oxidative alterations that are reflected in measurable biomarkers. Evidence across species shows that transportation is associated with several physiological disturbances. Camels may exhibit increased oxidative stress, indicated by elevated malondialdehyde (MDA) levels and altered glutathione peroxidase activity. In ruminants such as sheep and calves, transport commonly increases cortisol, glucose, creatine kinase, and oxidative stress markers, while calves may also show changes in acute-phase proteins such as fibrinogen and ceruloplasmin, as well as increased intestinal permeability. In poultry, transportation is frequently associated with body weight loss, elevated blood glucose and cholesterol concentrations, and an increased heterophil-to-lymphocyte ratio. This review synthesizes current evidence on the biochemical and physiological responses of domestic animals to road transportation, focusing on key biomarkers of transport-induced stress, including cortisol, glucose, creatine kinase, acute-phase proteins, and oxidative stress indicators. The literature suggests notable species-specific differences in sensitivity to transport stress, with horses and poultry often exhibiting pronounced physiological responses. In contrast, camels and some ruminants may show variable tolerance depending on environmental and management conditions. Transport duration emerges as a major determinant of stress severity, as longer journeys are generally associated with greater metabolic disturbances, muscle damage, and oxidative imbalance. In addition, accumulating evidence highlights a close interaction between oxidative stress and inflammatory processes, whereby excessive production of reactive oxygen species may activate inflammatory pathways and contribute to tissue damage during transportation.

**Key words:** Domestic animals, Oxidative stress, Road transportation

## Introduction

Stress is a physiological response to actual or perceived threats, primarily mediated by the activation of the hypothalamic-pituitary-adrenal (HPA) axis and the subsequent release of cortisol. This mechanism facilitates short-term survival by mobilizing energy and sustaining homeostasis; however, prolonged or recurrent exposure to stressors can disturb physiological equilibrium, compromise immune function, and heighten vulnerability to disease (Tüfekci *et al.*, 2023).

In cattle production systems, transportation constitutes a major stressor affecting animal welfare. The transportation of domestic animals presents numerous challenges, such as relocation, exposure to unfamiliar environments, noise, social regrouping, vehicle

vibrations, inadequate ventilation, and lack of food and water. The severity of these stress responses may fluctuate depending on the duration of transport and the environmental and managerial circumstances encountered by the animals during transit. Unfavorable housing circumstances, veterinary interventions, animal production practices, severe weather, and hunger may induce physical and psychological stress (Padalino, 2015).

Diverse animal species demonstrate physiological and immunological reactions linked to transportation stress. These reactions encompass elevated cortisol secretion (Ruiz de la Torre *et al.*, 2001), diminished humoral immunity (Mackenzie *et al.*, 1997), and increased morbidity and mortality rates from infectious diseases in the weeks following movement (Chirase *et*

*al.*, 2004). These modifications collectively suggest that transportation stress can profoundly impact the metabolic, hormonal, and immunological status of farm animals (Fazio *et al.*, 2008).

Transporting animals to the slaughterhouse via road induces significant stress, potentially leading to hypercortisolaemia (Saeb *et al.*, 2010) and increasing free radical production in camels (Nazifi *et al.*, 2009). Physiological stress during road transportation is evidenced by alterations in blood components, including cortisol, glucose, plasma urea, and creatine kinase (Krawczel *et al.*, 2007; Fisher *et al.*, 2010). Prolonged travel has been associated with disease onset, potentially attributable to transport-related immune system suppression (Stull *et al.*, 2004).

Environmental factors might intensify transportation stress. Climatic conditions are a significant environmental stressor. In tropical areas, higher ambient temperatures and humidity exacerbate heat stress, diminishing evaporative cooling efficacy and heightening the risk of hyperthermia. This intensified thermal load not only strains cardiovascular and respiratory systems but also induces oxidative damage, increasing vulnerability to metabolic and inflammatory illnesses relative to animals in temperate climates (Kuczynski *et al.*, 2011). Prior research examining the detrimental effects of transportation on sheep for durations of 24, 48, and 72 h was predominantly carried out in temperate locations (Knowles *et al.*, 1995; Fisher *et al.*, 2010). Transport difficulties may be exacerbated in tropical climates marked by elevated temperatures and humidity (Minka and Ayo, 2012), indicating that climatic factors significantly influence the physiological reactions of animals during transit.

Despite substantial data indicating the adverse consequences of transportation, the literature often presents conflicting results concerning the extent and duration of physiological stress reactions. These differences may result from methodological heterogeneity across studies, encompassing variations in transport duration, stocking density, vehicle dynamics, and sampling techniques, thereby complicating the formulation of universal welfare standards. Furthermore, research remains fragmented; although transportation stress has been intensively investigated in several livestock species, insufficient information is available for particular animals, such as camels, and for transport conditions occurring in hot and humid areas. Consequently, there is still no consensus regarding the most reliable set of biomarkers for real-time welfare assessment across diverse transport scenarios.

Therefore, this review summarizes the physiological and behavioral responses of animals to transport-related stress and highlights key biochemical and physiological biomarkers associated with these responses. This review seeks to synthesize findings across species and transport conditions to elucidate transport-induced stress mechanisms and aid in the formulation of enhanced, welfare-focused transportation management strategies. This review's novelty derives from its comprehensive

synthesis of physiological and biochemical responses to transportation stress in many domestic animal species. This review offers a comprehensive perspective on transport-induced stress and its implications for animal welfare assessment by comparatively analyzing important stress indicators and examining the effects of transit time and environmental variables, especially in hot and humid climates.

### **Ethical statement**

This study was conducted under the supervision of the Iranian Society for the Prevention of Animal Cruelty and the Research Council of Shiraz University (IACUC No. 6387/63).

### **Common stress mechanisms (HPA axis, oxidative stress, inflammation)**

Transport stress activates a set of interconnected biological pathways that collectively shape the animal's physiological response and determine the magnitude of welfare impairment. Among these, HPA axis activation, oxidative stress, and inflammation represent the principal common mechanisms underlying stress adaptation and stress-related damage. Rather than operating independently, these pathways interact dynamically, linking neuroendocrine regulation with metabolic disturbances and immune dysregulation.

The HPA axis serves as the central neuroendocrine system responsible for maintaining physiological homeostasis during stress. Under resting conditions, hypothalamic regulation of adrenocorticotrophic hormone (ACTH) secretion by the anterior pituitary follows a circadian and pulsatile rhythm, which subsequently regulates glucocorticoid release from the adrenal cortex. However, this rhythmic pattern is highly sensitive to environmental influences, including illumination, feeding schedules, and physical activity, and is markedly disrupted during stress exposure. In acute stress conditions such as transportation, enhanced pulsatile secretion of corticotropin-releasing hormone (CRH) and arginine vasopressin (AVP) increases ACTH output and consequently elevates circulating glucocorticoid concentrations. Although CRH acts as the principal driver of pituitary-adrenal activation, AVP exerts an important synergistic effect, amplifying overall endocrine responsiveness. HPA axis activity is further modulated by additional mediators, including magnocellular AVP, angiotensin II, systemic cytokines, inflammatory lipid mediators, adrenal medullary hormones, and autonomic neural inputs. At the cellular level, glucocorticoids bind to cytoplasmic receptors, translocate to the nucleus, and regulate glucocorticoid-responsive genes through glucocorticoid response elements, while also suppressing transcription factors such as c-JUN/c-FOS and NF- $\kappa$ B. Through these integrated actions, glucocorticoids influence endocrine output, immune cell function, and inflammatory tone, while negative feedback at hypothalamic, pituitary, and

extrahypothalamic levels helps limit excessive hormonal exposure and its associated catabolic, reproductive, and immunosuppressive consequences (Tsigos *et al.*, 2002).

Alongside neuroendocrine activation, transport stress induces important metabolic changes that frequently disturb cellular redox balance. Under physiological conditions, reactive oxygen species (ROS) function as essential secondary messengers in redox-responsive signaling pathways involved in gene transcription, vascular tone regulation, and immune activation, and their steady-state concentrations are tightly controlled by endogenous antioxidant defense systems. During transport, however, the combination of physical exertion, psychological stress, and altered metabolic activity promotes increased mitochondrial ROS production and leakage. Once ROS generation exceeds the buffering capacity of intrinsic antioxidant mechanisms, the balance shifts from transient physiological redox signaling toward persistent oxidative stress, resulting in lipid peroxidation and mitochondrial dysfunction. At the same time, oxidative stress should not be interpreted as a purely linear process of injury, because its regulation is influenced by the so-called antioxidant paradox. Although oxidative damage is closely linked to cellular senescence and tissue degeneration, evidence from clinical and experimental studies suggests that indiscriminate antioxidant supplementation may not consistently provide benefits and may even interfere with adaptive mitohormetic signaling, through which moderate physiological challenges strengthen endogenous antioxidant defenses. In this context, the oxidative burden associated with transport resembles that observed during intense or unaccustomed physical exertion, where maintenance of nutritional homeostasis appears more beneficial than acute pharmacological antioxidant intervention. Thus, oxidative stress represents both a consequence of transport-related metabolic strain and a mechanistic link between neuroendocrine activation and subsequent cellular injury (Qin *et al.*, 2026).

Inflammation constitutes a third major component of the common stress response and emerges through close interaction with both HPA axis signaling and oxidative imbalance. Stress-induced activation of the HPA axis stimulates glucocorticoid secretion, and glucocorticoids are classically recognized for their anti-inflammatory and immunosuppressive effects, including suppression of lymphocyte proliferation, reduction of pro-inflammatory cytokines such as TNF- $\alpha$  and IL-6, and enhancement of anti-inflammatory mediators such as IL-10. However, under prolonged or extreme stress, glucocorticoids may exert context-dependent pro-inflammatory effects. Persistent cytokine release may reduce cytoplasmic glucocorticoid receptor expression and impair glucocorticoid receptor-mediated transcription of anti-inflammatory genes, thereby weakening negative feedback by glucocorticoid and contributing to reduced glucocorticoid responsiveness. In parallel, the sympathetic nervous system participates in inflammatory regulation through the release of norepinephrine and

neuropeptide Y, which modulate cytokine synthesis and intracellular signaling pathways in immune cells. These neuroimmune interactions promote a stress-dependent shift in inflammatory tone, whereby mild or acute stress may transiently enhance immune vigilance, whereas prolonged or severe stress results in sustained inflammatory activation and dysregulation. This state is reflected by increased concentrations of inflammatory biomarkers, including CRP, IL-6, TNF- $\alpha$ , IL-1 $\beta$ , and NF- $\kappa$ B, highlighting the role of inflammation as both a downstream consequence and an amplifying component of stress-associated physiological disruption (Liu *et al.*, 2017).

Taken together, these pathways demonstrate that transport stress is mediated by an integrated mechanistic network rather than isolated biological events. HPA axis activation provides the central neuroendocrine drive, oxidative stress reflects the metabolic cost of adaptation and redox disequilibrium, and inflammation represents the immunological dimension of prolonged physiological strain. Their convergence helps explain why transport stress is consistently associated with coordinated changes in endocrine, oxidative, and inflammatory biomarkers, and why the overall biological impact depends on the duration, intensity, and context of the stressor.

## **Impact of transport stress on physiological, hematological, and biochemical parameters in animals**

### **Sheep, cattle, and goats**

Cattle transported over long distances may become physically exhausted, dehydrated, hungry, and unable to rest or lie down. Poor transportation circumstances, such as poor ventilation, excessive temperatures, and overstocking, can severely influence animal well-being. Maximum voyage duration is an important welfare concern, particularly for unweaned calves. A positive correlation has been found between transport distance and metabolic alterations or death, particularly at long distances, often referring to travel times exceeding 8-12 h or distances greater than 500-1,000 km, depending on factors such as the conditions of travel, the type of animal, and the availability of rest stops (Buckham Sporer *et al.*, 2023). Reducing transit distances and durations may minimize mortality while also improving calves' welfare during and after transportation. Longer journeys may increase the risk of animals being unwell due to prolonged exposure to severe temperatures, more time away from feed and water, difficulty resting, and poor environmental conditions. These combined stressors can disrupt physiological homeostasis and trigger endocrine, metabolic, and immune responses that ultimately compromise animal health and welfare. Activation of the HPA axis and the sympathetic nervous system during transport plays a key role in mediating these physiological responses.

Road quality has also been shown to influence the magnitude of transport stress responses. Animals

transported on unpaved or rural secondary roads exhibit greater physiological disturbance compared with those transported on paved roads (Miranda de la Lama *et al.*, 2011). Except for hematocrit and white and red blood cell counts, all variables were significantly affected by road type. Animals transported on unpaved roads had greater levels of cortisol, lactate, glucose, and creatine kinase (CK). These changes likely reflect increased muscular exertion and metabolic demand caused by vehicle vibration and instability during transport on poor road surfaces, as well as a higher neutrophil-to-lymphocyte (N/L) ratio (Miranda de la Lama *et al.*, 2011). Elevated ambient temperatures on the farm may have caused heat stress in the non-transported sheep, leading to neutrophilia and lymphopenia. This, in turn, could explain the higher N/L ratio observed in this group, even in the absence of transport stress.

Transportation can also activate inflammatory pathways, leading to increased production of acute-phase proteins (APPs). Elevated plasma concentrations of fibrinogen, ceruloplasmin, serum amyloid A (SAA), and  $\alpha$ -acid glycoprotein have been reported in transported calves, indicating the presence of systemic inflammatory responses. This inflammation is frequently accompanied by oxidative stress, an imbalance of reactive oxygen species (ROS) produced by metabolic and inflammatory reactions, which can cause severe tissue damage, altered metabolism, and potentially impaired reproduction in bovine species (Buckham Sporer *et al.*, 2023). Sanusi *et al.* (2022) evaluated the effects of short-duration road transportation using a conventional livestock truck on three sheep breeds (Yankasa, Balami, and Sudanese Fat-

Tail). The animals were transported for approximately 3 h during the dry season, and physiological and hematological parameters were measured before departure and upon arrival. The results indicated that packed cell volume (PCV), white blood cell count (WBC), and hemoglobin (Hb) levels did not differ significantly among the transported breeds. Only the Balami breed showed a significant change in red blood cell count (RBC), whereas the Sudanese Fat-Tail and Yankasa breeds showed no significant differences. Variations may relate to differences in transport duration, handling conditions, and breed resilience. Knowles *et al.* (1995) and Hall *et al.* (1998) have also documented the use of PCV as a useful indicator to evaluate the dehydration of animals during transportation (Table 1; Figs. 1a-c).

Alterations in leukocyte distribution represent one of the most consistent hematological indicators of transport stress. Lendrawati *et al.* (2020) investigated the physiological stress responses in sheep subjected to varying durations of transportation (4, 8, and 12 h) combined with feed and water deprivation. They observed significant alterations in leukocyte profiles, including an increase in neutrophil counts, a decrease in lymphocyte counts, and a consequent rise in the N/L ratio, indicating an elevated physiological stress response with prolonged transit. This leukocyte redistribution is largely mediated by glucocorticoids released during activation of the HPA axis. This pattern was further substantiated by observations of increased N/L ratios following longer transport durations (up to 48 h), consistent with Fisher *et al.* (2010).

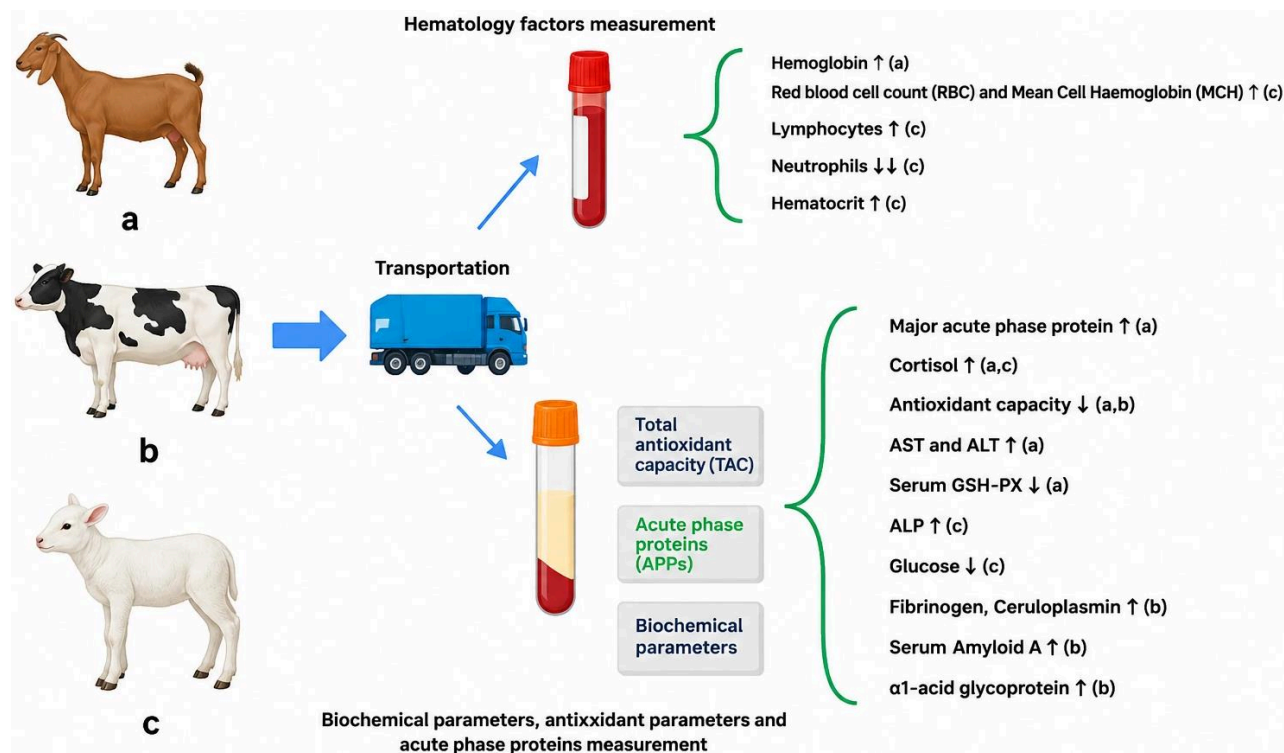


**Fig. 1:** Biochemical and physiological changes in sheep, calves, and goat induced by road transportation (a) goat, (b) cow, and (c) sheep

**Table 1:** Hematological and biochemical changes during road transportation in sheep, goats, camels, calves, horses, and poultry

| Animal  | Hematological change                                                                                                                            | Biochemical test                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           | References                                                                                                                    |
|---------|-------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------|
| Sheep   | Red blood cell count (RBC) and mean cell hemoglobin (MCH) ↑<br>Lymphocytes ↑<br>Neutrophils ↓ ↓<br>With ↑ transportation duration, hematocrit ↓ | Glucose and ALP ↑<br>With ↑ transportation duration, cortisol and glucose ↓                                                                                                                                                                                                                                                                                                                                                                                                                                                                | Lendrawati (2020), Sanusi <i>et al.</i> (2022)                                                                                |
| Goats   | Hemoglobin ↑                                                                                                                                    | IgM, IgA, IgG, and thyroxine ↓ propionic acid and epinephrine significantly ↑<br>Acetic acid, butyric acid, isobutyric acid, isovaleric acid, LPS, IL-1β, IL-6, TNFα, major acute phase proteins, and cortisol ↑↑<br>Antioxidant capacity ↓<br>AST and ALT ↑<br>Serum GSH-Px ↓<br>No significant changes in structure and composition of the rumen microbial                                                                                                                                                                               | Li <i>et al.</i> (2024)                                                                                                       |
| Camels  |                                                                                                                                                 | Cardiac-troponin-1 ↑<br>Cortisol, T4, T3 and fT4 ↑<br>Serum concentrations of glucose, NEFA, and urea nitrogen ↑<br>The mean concentration of MDA, and glutathione peroxidase ↑↑<br>Testosterone ↓ (long walk transport)<br>Cholesterol ↓ (truck vs walk/control) → within normal range<br>Liver enzymes:<br>• ALT ↑ (both transported groups)<br>• AST ↑ (higher in truck transport)<br>• ALP → within normal range (highest in control)<br>Iron ↓ (transport stress)<br>Copper ↓ (truck transport)<br>Total proteins ↓ (truck transport) | Nazifi <i>et al.</i> (2009), Saeb (2010), Mohamed <i>et al.</i> (2021), Kumar (2022)                                          |
| Calves  | In BAL fluid the number of red blood cells, white blood cells, neutrophils, macrophages, packed cell volume ↑↑ and lymphocytes ↓ ↓              | levels of cortisol, total protein, and fibrinogen ↑↑<br>Fibrinogen, ceruloplasmin, serum amyloidA, and α-acid glycoprotein ↑<br>Oxidative stress ↑<br>Haptoglobin ↓<br>Total protein ↑                                                                                                                                                                                                                                                                                                                                                     | Mohammadi <i>et al.</i> (2007), Pisoni <i>et al.</i> (2022), Buckham Sporer <i>et al.</i> (2023), Pisoni <i>et al.</i> (2023) |
| Horses  | Neutrophil counts ↑↑, lymphocytes ↓                                                                                                             | Serum albumin and plasma total antioxidant status ↑↑. Total serum protein and serum globulins ↑↑.<br>↑ Expression of 5-HT1B and 5-HT2A receptors and ↓ expression of IL-12 in PBMCs<br>Cortisol and glucose concentrations, and LDH activity ↑<br>Circulating 5-HT concentration ↓                                                                                                                                                                                                                                                         | Padalino <i>et al.</i> (2017), Bruschetta <i>et al.</i> (2024)                                                                |
| Poultry | Heterophil/lymphocyte ratio (H/L ratio) ↑                                                                                                       | Body weight losses, Serum glucose ↑ Serum cholesterol ↑<br>Creatine kinase ↑                                                                                                                                                                                                                                                                                                                                                                                                                                                               | Beski <i>et al.</i> (2011)                                                                                                    |

Interestingly, in the study by Lendrawati *et al.* (2020), the non-transported group exhibited a higher N/L ratio than the transported sheep. This finding was attributed to greater on-farm heat exposure due to increasing ambient temperature and humidity during the experimental period. Environmental measurements indicated that animals were subjected to warm ambient conditions with mild heat stress during transport, whereas a slightly higher thermal load was recorded at the farm. These results collectively suggest that the N/L ratio is a sensitive biomarker for both transport-associated stress and heat exposure, with the latter potentially exacerbating stress responses even in non-transported animals under certain farm conditions.

Cortisol is commonly used to assess the physiological stress response in transported sheep (Krawczel *et al.*, 2007). Following a few hours of travel, the blood cortisol level in transported sheep returns to its baseline level after experiencing a rise before loading (Cockram *et al.*, 2000). In the study of Lendrawati *et al.* (2020), cortisol levels peaked after 8 h of transportation. After 12 h, cortisol levels decreased to values similar to those observed after 4 h. These findings are consistent with glucose measurements and agree with Pascual Alonso *et al.* (2017), who reported that cortisol levels doubled after 4 h of transport compared to pre-transport and were about two times higher than the control group.

These findings indicate that the interaction between

transport duration and treatment significantly affected serum glucose levels in sheep. The transport group's serum glucose rose in 4 h, reduced in 8 h, and then increased after 12 h of transport; in contrast, the non-transport sheep's serum glucose did not change over time. Indicators of an animal's metabolic reaction to stress include higher serum glucose levels (Ali *et al.*, 2006). Furthermore, as a sign of metabolic fatigue, Knowles *et al.* (1995) reported that rising plasma glucose is a reaction to early stress and will fall for up to 24 h. Miranda de la Lama *et al.* (2012) have demonstrated that increased cortisol levels during stress cause glucose to rise as a gluconeogenic impact. SAA and haptoglobin (an acute-phase protein) have been identified in feedlot beef cattle as biomarkers associated with physiological disturbances (Giannetto *et al.*, 2011).

Additionally, in the assessment of physiological stress, oxidative stress biomarkers such as reactive oxygen metabolites (dROMs), antioxidant barrier (Oxy ads), and thiol antioxidant barrier (SHp) are commonly used to evaluate the inflammatory stress response in transported animals. Increased levels of these biomarkers have been reported in ewes after transport (Piccione *et al.*, 2013; Fazio *et al.*, 2018). Transport stress can also influence metabolic responses. For instance, Zhong *et al.* (2011) reported that during prolonged transport, animals utilize glucose as a primary energy source; glycogen reserves are mobilized and may become depleted after approximately 24 h. However, the same study indicated that an 8-hour road transport did not significantly affect blood cell counts or serum cortisol concentrations, although differences were observed among sheep of different ages. The absence of major metabolic changes in that study may be related to the relatively shorter transport duration and distance.

To explain the injuries and weariness that sheep experienced during transportation, CK became an essential variable (Fisher *et al.*, 2010). Lendrawati's *et al.* (2020) findings demonstrated that when travel time increases, CK activity continued to rise. The findings of Lendrawati *et al.* (2020) also compared 4- and 8-hour periods; The 12-hour transport group had increased CK activity, while the non-transport group's CK activity tended to persist. Gregory (2005) reported that physical injuries sustained during handling, loading, and killing led to an increase in CK activity. Furthermore, Fisher *et al.* (2010) reported an increase in CK concentration upon arrival. Although this rise was also observed in the studies by Fisher *et al.* (2010) following 48-hour transports, the increase detected in the Lendrawati *et al.* (2020) investigation was comparatively minor. This suggests that CK elevation is a common response to transport stress, but its magnitude may depend on the duration and intensity of the journey.

Similar effects on blood urea nitrogen (BUN) concentration are seen in transport and starvation trials, which tend to increase with trial duration. The sheep in the study by Lendrawati *et al.* (2020) were not subjected to severe stress during the 4, 8, or 12-hour transport; however, the lack of feed and water resulted in

alterations in blood metabolites such as BUN (a marker of protein metabolism and renal function), total protein, and glucose concentration. Because of the longer transit time and greater duration of food deprivation compared with the 4- and 8-hour periods, these alterations may be related to food deprivation, which would be more noticeable during the 12 h. Feed and water deprivation during transport can stimulate protein catabolism and increase nitrogen metabolism, resulting in elevated BUN concentrations. Furthermore, Montané *et al.* (2002) reported that, in roe deer, physical activity during transport, the impact of glucocorticoids on protein catabolism, and starvation could all contribute to an increase in BUN concentrations in transported sheep.

The findings indicated that, except for alkaline phosphatase (ALP), an enzyme related to liver and bone metabolism, no discernible variations were observed in the data obtained from the analyses of any of the biochemical markers investigated in the Yankasa sheep. Except for the ALP value, which demonstrated a significant difference following transportation, there were no significant differences observed in Balami sheep for the levels of alanine aminotransferase (ALT), a liver enzyme indicating hepatic injury, aspartate aminotransferase (AST), a liver and muscle enzyme, creatinine, urea, and glucose. None of the parameters measured in the Sudanese Fat-Tail sheep differed significantly, except for glucose (Sanusi *et al.*, 2022).

According to the study by Knowles *et al.* (1995) in sheep, the cause of the decrease in plasma glucose for up to 24 h could be attributed to early stress. This would imply metabolic depletion and an initial increase in plasma glucose. Mohammadi *et al.* (2007) reported cytological, biochemical, and bacteriological examinations of bronchoalveolar lavage (BAL) fluid and hematological and biochemical examinations of blood samples in calves after 12 h of road transportation. They revealed that the number of RBC, WBCs, and neutrophils, as well as cortisol levels, PCV, total protein, and fibrinogen significantly increased. However, lymphocytes significantly decreased in the experimental (transportation) group compared with the control group on the day of transportation. In addition, regarding the BAL fluid content, the total cell count, macrophages, neutrophils, and total protein increased in the experimental group (Mohammadi *et al.*, 2007) (Table 1; Figs. 1a-c). An increase in serum APPs (fibrinogen, ceruloplasmin, SAA, and  $\alpha$ -acid glycoprotein) was reported in dairy beef calves during feed restriction, marketing, and transportation (Pisoni *et al.*, 2022; Pisoni *et al.*, 2023). According to Ishizaki and Kariya (2010), plasma cortisol levels increased significantly 4 h after calves were transported, whereas they did not change in the control group. Information on the effects of road transportation on the function of BAL fluid cells in cattle is limited (Ishizaki and Kariya, 2010).

Li *et al.* (2024) also discovered that after the goats were transported, there was a marked rise in the levels of AST and ALT. This further supports the possibility that they sustained liver injury during transportation. Clinical

indicators of renal function include urea and creatinine, which are nitrogen-containing end products of metabolism. When proteins in food and tissues are metabolized, the primary byproduct is urea. Muscle creatine catabolism results in creatinine. Further supporting the findings of Li *et al.* (2024) indicating that transportation could potentially lead to renal impairment, Dalmau *et al.* (2013) also noted a significant increase in blood urea levels in lambs following 24 h of transit. Furthermore, chronic exposure to environmental stresses can alter the body's ability to regulate the permeability and function of muscle cell membranes, allowing intracellular CK and lactate dehydrogenase (LDH, an enzyme released during tissue damage) to leak out and thereby elevating blood CK and LDH activity (Klein *et al.*, 2020).

Antioxidant enzymes play a key role in the physiological stress response. Transport stress can disrupt the balance between ROS production and antioxidant defense systems, resulting in oxidative stress. Enzymes such as superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), and catalase protect cells from ROS. SOD converts superoxide radicals into hydrogen peroxide, which is then detoxified mainly by GSH-Px and catalase. GSH-Px also reduces hydrogen and lipid peroxides, thereby protecting cellular membranes from oxidative damage (Celi, 2010). However, the activity of these enzymes, particularly GSH-Px, may vary among studies due to factors such as environmental conditions, nutrition, management practices, seasonal changes, and physiological status (Celi, 2010). Differences in experimental conditions, including transport duration and environmental temperature, may therefore explain inconsistent antioxidant responses. For example, Li *et al.* (2024) reported oxidative stress in transported goats, evidenced by a significant decrease in serum GSH-Px activity and an increase in protein carbonyl content. In contrast, Danyer *et al.* (2021) observed significantly higher serum GSH-Px and SOD activities in ewes after transport.

Variations in antioxidant responses may depend on transport duration, with short transports upregulating enzymes and longer ones depleting them. Biochemical and physiological changes in sheep, calves, and goats induced by road transportation are presented in Figs. 1a-c. It is reasonable to assume that sheep may have a compensatory increase in antioxidant enzyme activity in response to brief transit. Longer transport durations may exhaust this protective mechanism, leading to severe depletion of antioxidant defenses and, consequently, a decrease in total antioxidant capacity (Figs. 1a-c). Hematological and biochemical changes during road

transportation in sheep, goats, and calves are presented in Table 1. Overall, these findings indicate that transportation induces a complex physiological response involving endocrine activation, metabolic adjustments, inflammatory processes, and oxidative stress, the magnitude of which depends largely on transport duration, environmental conditions, and animal characteristics (Table 2).

The evidence suggests that road transportation of sheep, cattle, and goats does not yield a consistent biomarker profile, but rather a physiological pattern that is contingent on environmental conditions. Brief transport periods may result in minimal or temporary hematological alterations; however, extended transport, inadequate road conditions, lack of feed and water, and thermal stress exacerbate endocrine activation, leukocyte redistribution, muscle enzyme leakage, inflammatory signaling, and oxidative imbalance. Consequently, biomarkers including cortisol, glucose, N/L ratio, CK, BUN, APPs, and antioxidant enzymes must not be analyzed in isolation. Their diagnostic value depends on transport duration, environmental circumstances, nutritional deprivation, and breed- or species-specific resilience.

## Horse

Horses display a unique transport-stress profile marked by sympathetic activation, immunological modulation, hydration imbalance, oxidative alterations, and delayed recovery consequences. In contrast to short-term stress management, extended transportation may combine psychological stress, postural strain, limited mobility, dehydration, and modified feeding behavior, resulting in quantifiable alterations in leukocyte distribution, acute-phase responses, antioxidant capacity, and clinical indicators.

Road transportation can induce a wide range of physiological responses in horses, including immunological, hematological, inflammatory, and oxidative alterations. Padalino (2015) evaluated these responses in mixed breed adult horses during and after transportation compared with non-transported controls. The extended trip was associated with an acute phase reaction, evidenced by higher levels of  $\gamma$ -interferon, reduced plasma total antioxidant status (PTAS), abnormal clinical signs, neutrophilia, and decreased lymphocyte responsiveness. Specifically, a diminished lymphocyte proliferative response was observed upon unloading, supporting the theory that a horse's immune potential may decline following extended travel (Figs. 2a-b). These findings suggest that prolonged

**Table 2:** Comparative table of transport-related biomarkers across species

| Biomarker            | Cattle and sheep          | Horses                   | Camels                   | Poultry                   |
|----------------------|---------------------------|--------------------------|--------------------------|---------------------------|
| Glucocorticoids      | Primary HPA indicator     | HPA activation (varies)  | HPA/HPT axis involvement | Corticosterone (increase) |
| CK (Creatine Kinase) | High (muscle fatigue)     | High (muscle damage)     | High (muscle damage)     | High (significant marker) |
| N/L or H/L Ratio     | Highly sensitive          | Leukocyte redistribution | Less documented          | Primary welfare indicator |
| Acute phase proteins | SAA, fibrinogen (Up)      | Inflammatory response    | Moderate response        | Variable                  |
| Oxidative status     | Antioxidant depletion     | PTAS/ROMs disturbance    | MDA/Catalase increase    | Metabolic trade-offs      |
| Metabolic shift      | Gluconeogenesis (glucose) | Catecholamine-driven     | Lipid mobilization       | Catabolic (weight loss)   |

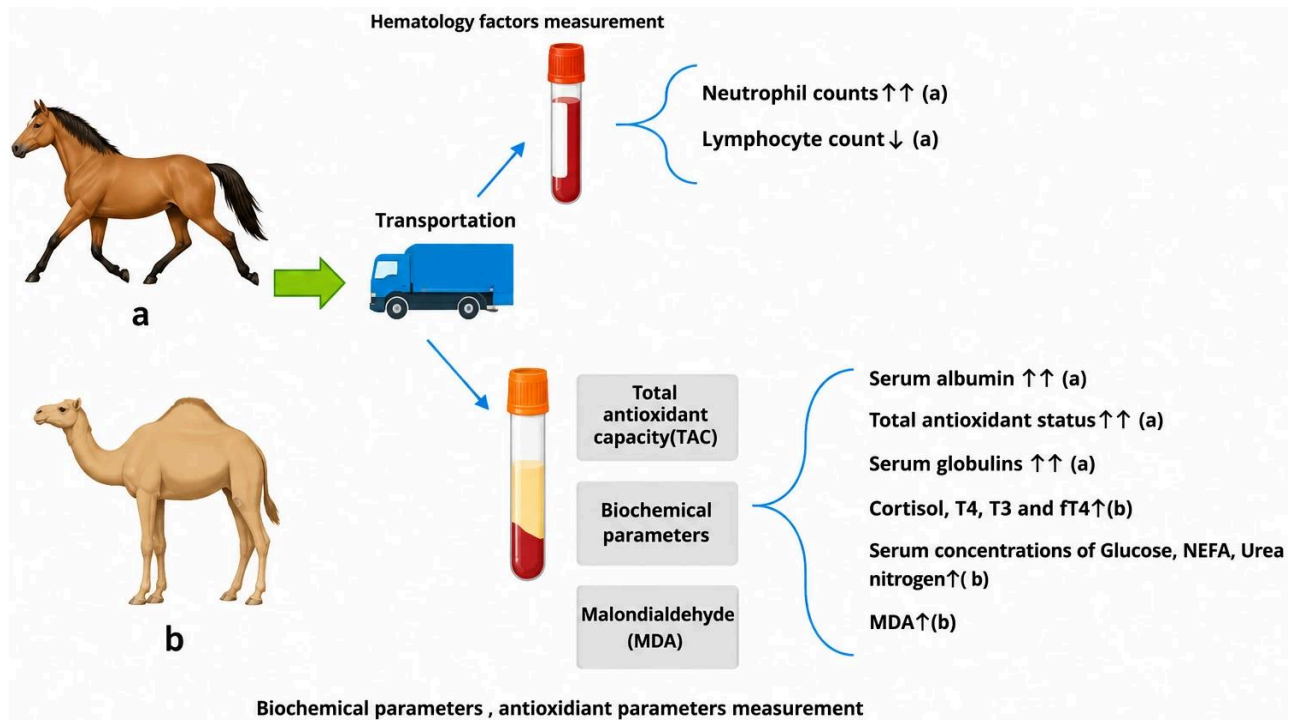


**Fig. 2:** Biochemical and physiological changes in horse and camel induced by road transportation (a) horse, and (b) camel

transportation may temporarily suppress immune competence through stress-mediated physiological pathways.

Dehydration and transport-induced catecholamine release have been attributed to increased red cell mass during shorter trips. In the Padalino *et al.* study (2017), horses were incredibly silent and less receptive during unloading, which may have resulted from catecholamine depletion or adaptation to external stimuli to help them endure the protracted trip. Catecholamine release during transport is associated with sympathetic nervous system activation, which contributes to behavioral changes, cardiovascular adjustments, and metabolic responses.

The Padalino (2015) study's observations of alterations in leukocyte counts are consistent with those of earlier investigations. Transport caused neutrophilia and lymphopenia in horses that did not become sick but did not increase the overall leukocyte count (Oikawa *et al.*, 2005; Stull *et al.*, 2008). This leukocyte redistribution is typically associated with glucocorticoid release and activation of the stress response. The transport-induced rise in cortisol has been linked to alterations in leukocyte populations and decreased leukocyte function (Stull *et al.*, 2004). However, the Padalino *et al.* (2017) investigation observed no difference in cortisol concentrations. This result conflicts with the literature because elevated cortisol has been reported in numerous other studies following brief trips (Tateo *et al.*, 2012; Fazio *et al.*, 2013), 12-hour trips (Baucus *et al.*, 1990), or 24-hour trips (Stull and Rodiek, 2000). Variations in cortisol responses among studies may be explained by differences in transport duration, animal experience, and habituation to transport, and environmental conditions.

Several clinical examination measures were above the average range, as recently documented (Stull and Rodiek, 2000; Padalino *et al.*, 2012; Padalino, 2015). The Padalino *et al.* study (2017) showed a positive correlation between these parameters and lymphocyte proliferation, indicating that the degree of insult to homeostatic processes may impact the severity of immunological impairment and the extent of the clinical reaction. These preliminary results highlight the significance of clinical assessment in determining each horse's response to transportation, even though correlation is a poor indicator of association. Therefore, clinical indicators may provide useful insight into the physiological burden imposed by transportation.

According to the Padalino *et al.* (2017) study, the best practice for identifying horses more susceptible to illness and scheduling a recuperation period is to do a clinical assessment as soon as they arrive (Hodgson *et al.*, 2013).

The transport did not affect leukocyte counts overall; however, there was considerable fluctuation in leukocyte subpopulations about transportation. Horses, in particular, showed higher neutrophil levels and reduced lymphocyte and eosinophil counts (Padalino *et al.*, 2017).

Changes in plasma protein fractions have also been reported in transported horses, reflecting inflammatory and hydration-related responses (Crisman, 2008; Cray *et al.*, 2009). The decrease in albumin concentration could be because albumin is a negative acute phase response protein or because the hydration indices had recovered from transport and the seventh day exhibited an upward trend in globulin concentration, which was correlated with an increase in acute phase proteins (Crisman *et al.*,

2008; Cray *et al.*, 2009). Dehydration or hemoconcentration may be responsible for the elevated albumin value observed during unloading (Stull and Rodiek, 2000). Transport has been shown to slow food movement through the intestines, raising the temperature, heart rate, and breathing rate in horses (Oikawa *et al.*, 2005). According to Padalino (2015), these alterations have been explained as an inevitable reaction to transportation linked to catecholamine release, muscle activity, and thermoregulation. At unloading, capillary refill time was significantly greater in the horses in the experimental group than in the control group, even though capillary refill time has been considered in previous studies to be the most trustworthy welfare indicator of adequate water intake and the absence of prolonged thirst (Dalla Costa *et al.*, 2014).

The two most frequent effects of transportation have been dehydration and weight loss. These alterations are generally associated with reduced water intake, increased sweating, and prolonged standing during transport. Weight loss attributable to transportation can range from 0.10 to 0.63% of body weight per hour of travel, with recovery often occurring within three days after arrival (Lendrawati *et al.*, 2020). There was not a severe level of dehydration because serum total proteins and albumin were still within normal limits (Reed *et al.*, 2003). Therefore, it is possible to interpret the capillary refill time and body weight changes as typical, modest responses to transit stress and hydration intake restrictions. It is therefore advised that measures be easily implemented to assess the recovery of horses after transportation, such as monitoring capillary refill time and body weight by tape after transportation. These are simple procedures that all owners and agents might perform before and after the trip.

The impact of extended travel on the immune system is intriguing since illnesses linked to transportation typically manifest clinically a week following the transportation. By day seven following the transportation, the effects of transportation were reversed in the Padalino *et al.* research (2017), and no animals had illnesses. This finding suggests that many transport-induced physiological alterations are reversible once animals are provided with adequate rest and recovery time. Stull *et al.* (2008) demonstrated that a rest period is necessary for the immune system to repair and respond to the diseases to which animals may be exposed during travel (Stull *et al.*, 2008).

Transport stress has also been associated with disturbances in oxidative balance. Onmaz *et al.* (2011) and Wessely Szponder *et al.* (2015) study suggests that changes in oxidative and antioxidative indices, such as PTAS and ROMs, can be attributed to the acute phase response caused by transport stress. This has been previously suggested by Onmaz *et al.* (2011) and Wessely Szponder *et al.* (2015). Padalino *et al.* (2017) did not observe a notable rise in ROMs, as previously reported in transported ewes (Piccione *et al.*, 2013), even though oxidative product maxima can be observed one to two days following transportation (Nazifi *et al.*, 2009;

Piccione *et al.*, 2013; Wessely Szponder *et al.*, 2015). The Padalino *et al.* (2017) study examined a group of horses that showed the highest ROM values on day 1, with a concurrent and noticeable increase in PTAS to nearly twice the normal range (Kusano *et al.*, 2016) (Table 1). The latter could be an antioxidant reaction to prevent oxidative stress by producing more free radicals (Celi, 2011; Niedźwiedz *et al.*, 2013). Such increases in antioxidant capacity may represent a compensatory mechanism aimed at counteracting increased production of reactive oxygen species. This exceptionally high PTAS level could also be connected to the experimental group horses' day-one serum albumin concentrations, which increased noticeably. Therefore, albumin is included in the assessment of PTAS (Celi, 2011), as its sulfhydryl (SH) groups are considered a vital part of the extracellular antioxidant defense system.

In 1996, Benzie and Strain found that proteins only comprise 10% of PTAS values. Uric acid, ascorbic acid, and  $\alpha$ -tocopherol comprise 60%, 5%, and 5% of the total. Thus, Padalino's *et al.* observations (2017) are in keeping with the literature suggesting that transportation increases PTAS, possibly due to the mobilization of antioxidants (Niedźwiedz *et al.*, 2013). Stressed animals are known to protect their cells from oxidative damage by releasing antioxidants, which neutralize the effects of free radicals (Sconberg *et al.*, 1993; Ralston and Stives, 2012). Specifically, Pregel suggested that PTAS could be a more accurate and sensitive indicator of stress in calves that have been transported than the measurement of individual antioxidant markers. As a result, it may be advisable to monitor PTAS and ROMs (reactive oxygen metabolites, markers of oxidative stress) before and after travel to identify calves at risk of oxidative stress and the ensuing poor health and welfare effects (Pregel *et al.*, 2005).

The Bruschetta *et al.* (2024) study revealed that transport and slaughter stress activate the serotonergic system, affecting the expression of serotonin-related receptors in peripheral blood mononuclear cells. This stress, combined with elevated glucose, cortisol, and LDH concentrations and reduced circulating 5-HT, suggests that serotonin receptor expression could serve as a key stress indicator, potentially aiding in the development of better elective protocols for transported horses (Bruschetta *et al.*, 2024) (Table 1). Cortisol concentrations did not vary significantly after either journey. Nevertheless, cortisol response magnitude is currently a useful standard indicator for stress assessment (Boucher *et al.*, 2019). Activation of the HPA axis plays a key role in restoring homeostasis, with many studies showing increased cortisol levels following transport (de Mira *et al.*, 2021; Bruschetta *et al.*, 2024). This response varies based on habituation, age, experience, transport duration, environmental conditions, and management. Foals exhibit high cortisol levels during early transport experiences, while stallions and racehorses show increases over both short and long distances (Rizzo *et al.*, 2017). Horses accustomed to transport for competitions also display significant cortisol elevation after transport

and slaughter (Bruschetta *et al.*, 2024). This supports findings that cortisol levels remain elevated throughout transport due to external stressors triggering physiological responses. Biochemical and physiological changes in horses induced by road transportation are presented in Figs. 2a-b.

Aragona *et al.* (2024) studied the effects of transport distance and time of day on body temperatures in athletic horses. No significant differences were found in the left and right sides, inner thigh, or rectal temperatures across distances or times. However, jugular, shoulder, and croup temperatures were higher in the afternoon than in the morning for both distances. The study suggests that the time of day influences ocular temperature as much as transport distance.

Transportation to slaughterhouses can also result in several physiological and physical stress indicators in horses. After transport, some horses had superficial bleeding wounds, an increased respiratory rate, and elevated blood lactate concentration in both adults and foals. Blood glucose decreased in some horses, while plasma total protein increased. Bruising was observed in both adults and foals. Although the effects on physiology and bruising were minor, handling, transport, and lairage still caused injuries and stress, indicating suboptimal management (Roy *et al.*, 2015).

The adrenocortical response is characterized by an increase in cortisol levels previously recorded in both stallions and geldings in response to excitement, transportation, and stress (Villani *et al.*, 2006; Fazio *et al.*, 2013). It is thus reasonable to hypothesize that the horse's physical component of transport and slaughter stress stimuli may combine with the psychic one to modify the adrenocortical responses (Figs. 2a-b; Table 2). Overall, these findings indicate that road transportation triggers complex physiological responses in horses involving endocrine activation, immune modulation, metabolic alterations, and oxidative stress. Hematological and biochemical changes during road transportation in horses are presented in Table 1.

## Camel

In camels, transport-induced stress responses must be understood in the context of their remarkable adaptation to dry conditions and extended resource scarcity. Despite camels' physiological resilience, transportation can nevertheless trigger endocrine, metabolic, oxidative, hepatic, and mineral-related changes. In comparison to other livestock species, their reaction seems to be significantly influenced by energy conservation, lipid mobilization, thyroid-related metabolic regulation, and the maintenance of water balance.

Camels are well adapted to harsh environmental conditions, yet transportation can still impose physiological stress that triggers a range of metabolic and endocrine responses (Saeb *et al.*, 2010). Transport stress activates the hypothalamic-pituitary-thyroid (HPT) axis, leading to increased secretion of thyroid hormones to meet elevated energy demands (Hyyppä, 2005; Saeb *et al.*, 2010). Activation of this axis helps increase

metabolic rate and energy mobilization to meet the elevated physiological demands associated with transport stress. However, hydration status remained stable, as serum total protein and albumin levels did not show significant changes.

Cardiac troponin-1 serves as a sensitive biomarker of cardiac muscle damage (Tharwat *et al.*, 2013a). Tharwat *et al.* (2013b) found that its levels increased after endurance exercise, suggesting elevated cardiac stress under hypoxic conditions. These findings indicate that prolonged physical exertion during transport or endurance exercise may temporarily increase cardiac workload in camels. Though levels normalized after seven days, chronic exercise-induced stress may contribute to cardiac damage, indicating the need for adequate recovery time. Increased concentrations of cortisol,  $T_4$ ,  $T_3$ ,  $fT_4$ , glucose, NEFA, and urea nitrogen were reported in the serum of one-humped camels following endurance load exercise (Kumar *et al.*, 2022).

Metabolic biomarkers are frequently used to evaluate the physiological impact of transport stress and the load-carrying capacity of camels. Most biochemical parameters remained unchanged, except for a notable increase in triglycerides, likely due to lipid mobilization for energy (Aragon and Younossi, 2010). This response reflects the camel's well-known ability to mobilize fat reserves from the hump to meet increased energetic demands during periods of stress or limited feed availability. Oxidative stress-related parameters, including MDA and catalase activity, increased with transit distance, suggesting free radical production and erythrocyte membrane damage (Ramnath *et al.*, 2008; Nazifi *et al.*, 2009). Greater attention has been given to adaptive characteristics that help animals survive in challenging circumstances, in addition to generic physiological reactions. Specifically, elevated thyroid hormone activity, which aids in mobilizing energy reserves in arid and resource-constrained environments, is linked to their metabolic resilience. This hormonal change helps to maintain metabolic function and enhances resistance to environmental stresses (Taff *et al.*, 2024).

Under stress, the sympathetic-adrenal medullary system releases catecholamines, causing spleen contraction and erythrocyte release (Montane *et al.*, 2002). These findings highlight the physiological impacts of transport stress on camels and their adaptive responses. Testosterone, an anabolic steroid, decreased in camels subjected to long-walk travel, reflecting metabolic adaptation to prolonged feed and water deprivation through a reduction in metabolic rate (Baghshani *et al.*, 2010). Reductions in testosterone during prolonged stress may reflect a shift in physiological priorities from reproductive functions toward energy conservation and survival. Although truck-transported camels showed lower cholesterol than walking or control groups, their values remained within the normal range for dromedaries (Mohammed *et al.*, 2007; Eltahir *et al.*, 2016). The reduction in testosterone along with elevated cholesterol in walking camels may

indicate mobilization of hump fat to maintain cholesterol homeostasis after more than 30-40 days of desert travel. Similar to the current findings, road transport did not significantly affect testosterone release in stallions (Deichsel *et al.*, 2015).

All transported camels exhibited increased liver enzyme activities, suggesting hepatic stress. ALT concentrations were elevated in both transported groups and were comparable to the upper limits previously reported in non-transported young camels (Eltahir *et al.*, 2016). AST levels were higher in truck-transported camels, consistent with observations in horses transported for long durations (>20 h) (Padalino *et al.*, 2017). ALP was highest in control animals but remained within normal physiological ranges (Eltahir *et al.*, 2016).

Transport stress also reduced serum iron, similar to decreases observed after surgical castration or vaccination (Greunz *et al.*, 2018). Transport stress has also been associated with alterations in mineral metabolism. Despite this decline, iron values remained several-fold higher than reference levels for young female camels, likely due to differences in sex, breed, and diet. Copper concentrations, which typically rise as a positive acute-phase response (Gruys *et al.*, 1999), decreased in truck-transported camels, likely reflecting feed restriction. Even the lowest copper values recorded exceeded previously reported reference ranges for dromedaries (Baghshani *et al.*, 2010; Eltahir *et al.*, 2016). Overall, the available evidence suggests that road transportation induces complex physiological responses in camels involving endocrine activation, metabolic adjustments, oxidative stress, and alterations in mineral and liver metabolism. Hematological and biochemical changes during road transportation in camels are presented in Table 1. Biochemical and physiological changes in the camel induced by road transportation are presented in Figs. 2a-b.

Total protein levels decreased significantly in truck-transported camels, aligning with reductions previously reported in young bulls transported by truck (Padalino *et al.*, 2017) and in animals undergoing prolonged dehydration (Kataria *et al.*, 2007). However, other studies have documented increased total protein shortly after transport (Earley *et al.*, 2010; Saeb *et al.*, 2010; Earley *et al.*, 2011; Cywinska *et al.*, 2012; Tharwat *et al.*, 2013; Emeash *et al.*, 2016), likely reflecting transient dehydration or acute-phase responses (Niedzwiedz *et al.*, 2012; Mohamed *et al.*, 2021). These discrepancies may result from species- and context-specific adaptation to transport-related and environmental stressors. Differences in transport duration, hydration status, feeding conditions, and environmental temperature may also contribute to variability among studies (Figs. 2a-b; Table 1; Table 2).

## Poultry

Transportation is a multifaceted stressor for broilers, impacting both body mass and immune status. Beski *et al.* (2011) demonstrated that transport duration is

positively correlated with body weight loss and the heterophil-to-lymphocyte (H/L) ratio. The heterophil-to-lymphocyte ratio rises due to glucocorticoid-mediated immune shifts, a process that can compromise host defenses and increase disease susceptibility following transport. The rise in the H/L ratio serves as a reliable marker of HPA axis activation, reflecting a systemic shift in leukocyte distribution mediated by corticosterone. The group that was transported for 3 h had the highest percentage of body weight loss and H/L ratio (Beski *et al.*, 2011).

However, compared to the control, the impact of transit on PCV% and hemoglobin concentration was not statistically significant. Furthermore, Ghareeb and Böhm (2009) found an increase in heterophils in broilers after 24 h of transit. However, Voslarova found no evidence of a transportation-related impact on the patient's H/L ratio in *Phasianus colchicus* (Voslarova *et al.*, 2006).

Beyond the economic implications of body weight loss, these physiological markers highlight the significant welfare burden imposed on birds during transit. According to Altan *et al.* (2003), a rise in the H/L ratio is a physiological sign of stress in birds. Plasma corticosterone levels increase following road transportation (Satterlee *et al.*, 1989).

According to Beski *et al.* (2011), there were no statistically significant differences between transported broilers and the control group in terms of serum total protein and triglyceride levels. However, Minka and Ayo (2008) reported that serum total protein levels significantly decreased in Shika Brown pullets following transport. Similarly, Huff *et al.* (2008) observed a decrease in serum triglyceride levels in transported turkeys. These variations may be attributed to differences in transportation conditions, travel duration, or dietary composition. Transported chickens showed a significant increase in serum CK compared with the control group. Birds transported for 3 h exhibited the highest CK levels. This finding is consistent with the study by Mitchell *et al.* (1992), which reported a significant increase in plasma creatine kinase levels in broilers after 216 min of transportation. Similarly, Scope *et al.* (2002) reported a significant increase in blood creatine kinase levels in pigeons following transportation. Elevated levels of intracellular muscle enzymes such as creatine kinase in plasma may indicate tissue damage and muscle dysfunction induced by transport stress (Mitchell *et al.*, 1992) (Table 1; Fig. 3). The leakage of CK into the circulation is indicative of skeletal muscle membrane permeability changes, likely resulting from physical exertion, and postural instability during transit, and potentially, localized ischemia.

Transport-related mortality is not merely a function of travel duration, but is strongly modulated by seasonal environmental conditions and species-specific thermoregulatory capacities. End-of-lay hens and broiler chickens showed the highest winter mortality, whereas turkeys and ducks were more affected during the summer months. The heightened vulnerability of end-of-lay hens

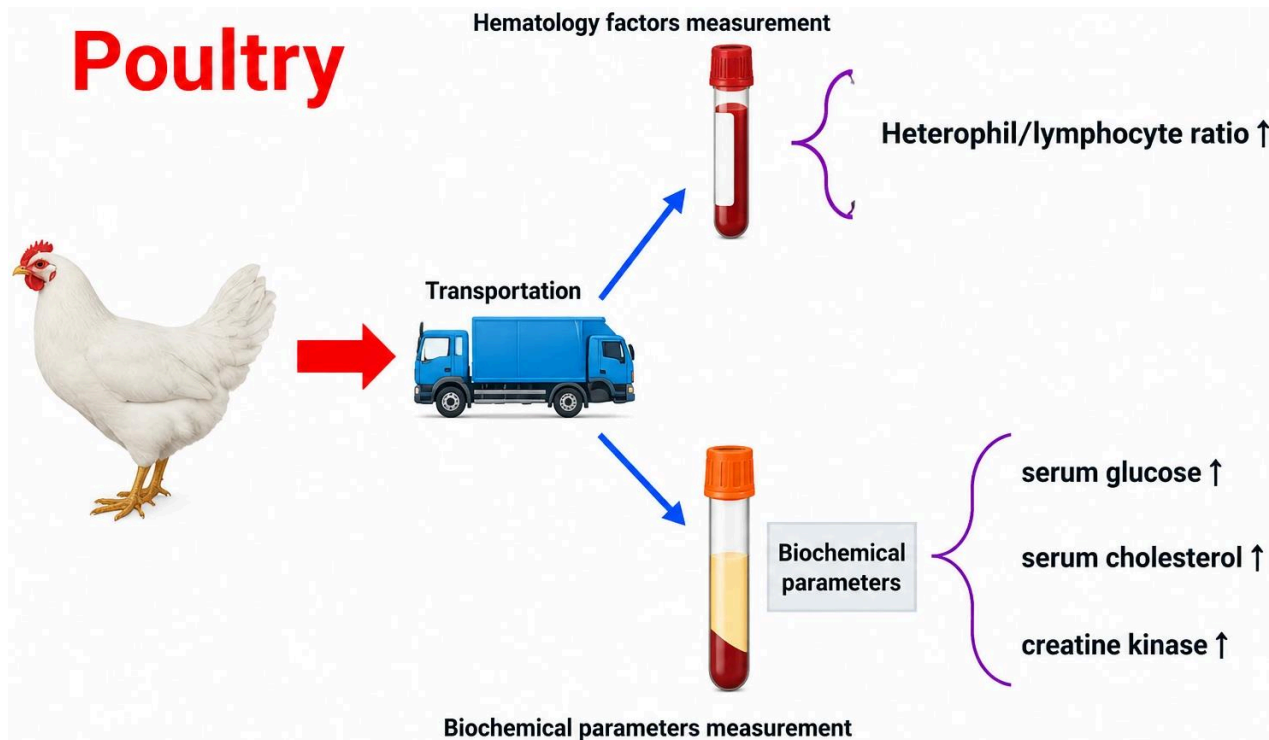


**Fig. 3:** Biochemical and physiological changes in poultry induced by road transportation

is attributable to inadequate feather insulation, low body energy reserves, and overall metabolic depletion (Nielsen *et al.*, 2022; Richards *et al.*, 2012). Microclimatic assessments further demonstrated that birds located in peripheral transport positions were exposed to substantial cold stress when temperatures fell below 10°C (Richards *et al.*, 2012). Elevated winter mortality in broilers suggests that factors such as heterogeneous thermal distribution, humidity, wet litter, and suboptimal ventilation may override their capacity for endogenous heat production. This implies that when environmental conditions exceed the bird's thermoneutral zone, their ability to maintain homeostasis is severely compromised, leading to increased mortality risk.

Although summer heat stress in broilers is extensively documented, cold stress remains comparatively understudied despite evidence of peak mortality in winter (Grilli *et al.*, 2018; Teke *et al.*, 2019). These findings reinforce the role of seasonal conditions and transport duration as key determinants of transport-related mortality (Cockram *et al.*, 2019; Justova *et al.*, 2025). In summary, the physiological response of poultry to transportation involves a complex interplay of endocrine activation, metabolic depletion, and physical fatigue, with outcomes heavily influenced by thermal environment and individual bird resilience (Table 2). Hematological and biochemical changes during road transportation in poultry are presented in Table 1. Biochemical and physiological changes in poultry induced by road transportation are presented in Fig. 3.

### Comparative interpretation across species

Across species, transportation triggers a conserved

stress-response network that interacts with the hypothalamic-pituitary-adrenal axis, sympathetic nervous system, metabolic substrate mobilization, inflammatory signaling, and oxidative stress. Nevertheless, the manifestation of these reactions varies significantly among taxa. In ruminants, the transport duration, road conditions, feed and water deprivation, and breed robustness predominantly influence changes in cortisol, glucose, N/L ratio, CK, BUN, and acute-phase proteins. Prolonged travel in horses is closely linked to immune regulation, alterations in hydration status, oxidative imbalance, and the need for recovery upon arrival. In camels, the response is influenced by metabolic resilience, thyroid-mediated energy regulation, lipid mobilization, and adaptability to arid environments. In chickens, transport-related responses are significantly influenced by environmental temperature, with the H/L ratio, CK activity, body weight loss, and mortality serving as critical welfare indicators. Consequently, transport-related biomarkers must be understood within the context of a cohesive physiological network rather than as standalone indicators.

### Conclusion

Effective management of livestock transportation relies on a multifaceted approach, incorporating optimal environmental control via sufficient ventilation and suitable stocking density, as well as genetic factors related to stress resilience. Alleviating transport-related physiological and metabolic stress is essential for animal welfare, health, and productivity. Future research and species-specific solutions, particularly in response to the

challenges associated with climate change and long-distance travel, are crucial for improving animal welfare during transit.

## Acknowledgement

The authors gratefully acknowledge Shiraz University, Shiraz, Iran, for supporting this study.

## Conflict of interest

The authors declare that they have no conflict of interest. Furthermore, the authors declare that the research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

## Research funding

No funding (review article).

## Use of generative AI and AI-assisted technologies statements

The authors did not use the assistance of any Artificial Intelligence Generated Content (AIGC) tools to develop any portion of the manuscript.

## References

- Ali, BH; Al Qarawi, AA and Mousa, HM (2006). Stress associated with road transportation in desert sheep and goats, and the effect of pretreatment with xylazine or sodium betaine. *Res. Vet. Sci.*, 80: 343-348. <https://doi.org/10.1016/j.rvsc.2005.07.012>.
- Altan, O; Pabuçcuoğlu, A; Altan, A; Konyalıoğlu, S and Bayraktar, H (2003). Effect of heat stress on oxidative stress, lipid peroxidation and some stress parameters in broilers. *Br. Poult. Sci.*, 44: 545-550. <https://doi.org/10.1080/00071660310001618334>.
- Aragon, G and Younossi, ZM (2010). Evaluating mildly elevated liver enzymes. *Cleve. Clin. J. Med.*, 77: 195-204. <https://doi.org/10.3949/ccjm.77a.09064>.
- Aragona, F; Arfuso, F; Rizzo, M; Fazio, F; Acri, G; Piccione, G and Giannetto, C (2024). Infrared thermography for assessing thermal homeostasis during road transport in athletic horses. *J. Equine Vet. Sci.*, 138: 102-105. <https://doi.org/10.1016/j.jevs.2024.105102>.
- Baghshani, H; Nazifi, S; Saeb, M and Saeb, S (2010). Influence of road transportation on plasma concentrations of acute-phase proteins, including fibrinogen, haptoglobin, serum amyloid A, and ceruloplasmin, in dromedary camels (*Camelus dromedarius*). *Comp. Clin. Pathol.*, 19: 193-198.
- Baucus, KL; Squires, EL; Ralston, SL; McKinnon, AO and Nett, TM (1990). Effect of transportation on the estrous cycle and concentrations of hormones in mares. *J. Anim. Sci.*, 68: 419-426. <https://doi.org/10.2527/1990.682419x>.
- Benzie, IF and Strain, JJ (1996). The ferric reducing ability of plasma (FRAP) as a measure of antioxidant power: The FRAP assay. *Anal. Biochem.*, 239: 70-76. <https://doi.org/10.1006/abio.1996.0292>.
- Beski, SS; Bamerni, AO and Shamdeen, ROR (2011). Effects of road transportation on some hematobiochemical and biochemical parameters of broiler chickens. *J. Dohuk Univ.*, 14: 52-56.
- Boucher, P and Plusquellec, P (2019). Acute stress assessment from excess cortisol secretion: Fundamentals and perspectives. *Front. Endocrinol.*, 10: 749. <https://doi.org/10.3389/fendo.2019.00749>.
- Bruschetta, G; Zanghì, G; Giunta, RP; Ferlazzo, AM; Satuè, K; D'Ascola, A and Fazio, E (2024). Short road transport and slaughter stress affect the expression profile of serotonin receptors and adrenocortical and hematochemical responses in horses. *Vet. Sci.*, 11: 113. <https://doi.org/10.3390/vetsci11030113>.
- Buckham Sporer, K; Earley, B and Marti, S (2023). Current knowledge on the transportation by road of cattle, including unweaned calves. *Animals*. 13: 3393. <https://doi.org/10.3390/ani13213393>.
- Celi, P (2010). Biomarkers of oxidative stress in ruminant medicine. *Immunopharmacol. Immunotoxicol.*, 32: 540-551. <https://doi.org/10.3109/08923971003616330>.
- Celi, P (2011). Biomarkers of oxidative stress in ruminant medicine. *Immunopharmacol. Immunotoxicol.*, 33: 233-240. <https://doi.org/10.3109/08923973.2010.514917>.
- Chirase, NK; Greene, LW; Purdy, CW; Loan, RW; Auvermann, BW; Parker, DB; Walborg Jr, EF; Stevenson, DE; Xu, Y and Klaunig, JE (2004). Effect of transport stress on respiratory disease, serum antioxidant status, and serum concentrations of lipid peroxidation biomarkers in beef cattle. *Am. J. Vet. Res.*, 65: 860-864. <https://doi.org/10.2460/ajvr.2004.65.860>.
- Cockram, MS; Dulal, KJ; Mohamed, RA and Revie, CW (2019). Risk factors for bruising and mortality of broilers during manual handling, module loading, transport, and lairage. *Can. J. Anim. Sci.*, 99: 50-65.
- Cockram, MS; Kent, JE; Goddard, PJ; Waran, NK; Jackson, RE; McGilp, IM; Southall, EL; Amory, JR; McConnell, TI; O'Riordan, T and Wilkins, BS (2000). Behavioural and physiological responses of sheep to 16 h transport and a novel environment post transport. *Vet. J.*, 159: 139-146. <https://doi.org/10.1053/tvjl.1999.0412>.
- Cray, C; Zaia, J and Altman, NH (2009). Acute phase response in animals: A review. *Comp. Med.*, 59: 517-526.
- Crisman, MV; Scarratt, WK and Zimmerman, KL (2008). Blood proteins and inflammation in the horse. *Vet. Clin. North Am. Equine Pract.*, 24: 285-vi. <https://doi.org/10.1016/j.cveq.2008.03.004>.
- Cywinska, A; Szarska, E; Gorecka, RL; Witkowski, RM; Hecold, M; Bereznowski, A; Schollenberger, A and Winnicka, A (2012). Acute phase protein concentrations after limited distance and long distance endurance rides in horses. *Res. Vet. Sci.*, 93: 1402-1406.
- Dalla Costa, E; Murray, L; Dai, F; Canali, E and Minero, M (2014). Equine on farm welfare assessment: A review of animal based indicators. *Anim. Welf.*, 23: 323-341. <https://doi.org/10.7120/09627286.23.3.323>.
- Dalmau, A; Di Nardo, A; Realini, CE; Rodríguez, P; Llonch, P; Temple, D; Velarde, A; Giansante, D; Messori, S and Villa, PD (2013). Effect of the duration of road transport on the physiology and meat quality of lambs. *Anim. Prod. Sci.*, 54: 179-186. <https://doi.org/10.1071/AN13024>.
- Danyer, E; Bilal, T; Altiner, A; Aytekin, İ and Atalay, H (2021). Effect of vitamin E treatment on selected immune and oxidative parameters in Kivircik ewes suffering from transport stress. *J. Anim. Physiol. Anim. Nutr.*, 105: 34-41.

- <https://doi.org/10.1111/jpn.13560>.
- Deichsel, K; Pasing, S; Erber, R; Ille, N; Palme, R; Aurich, J and Aurich, C** (2015). Increased cortisol release and transport stress do not influence semen quality and testosterone release in pony stallions. *Theriogenology*. 84: 70-75.
- de Mira, MC; Lamy, E; Santos, R; Williams, J; Pinto, MV; Martins, PS; Rodrigues, P and Marlin, D** (2021). Salivary cortisol and eye temperature changes during endurance competitions. *BMC Vet. Res.*, 17: 329. doi: 10.1186/s12917-021-02985-9.
- Earley, B; McDonnell, B; Murray, M; Prendiville, DJ and Crowe, MA** (2011). The effect of sea transport from Ireland to the Lebanon on inflammatory, adrenocortical, metabolic and behavioural responses of bulls. *Res. Vet. Sci.*, 91: 454-464.
- Earley, B; Murray, M and Prendiville, DJ** (2010). Effect of road transport for up to 24 hours followed by twenty four hour recovery on live weight and physiological responses of bulls. *BMC Vet. Res.*, 6: 38. doi: 10.1186/1746-6148-6-38.
- Eltahir, YE; Ali, HM; Hago, BE and Mahgoub, O** (2016). Serum biochemistry parameters in the Omani racing Arabian camels (*Camelus dromedarius*). *J. Agric. Mar. Sci.*, 21: 64-75.
- Emeash, HH; Mostafa, AS; Karmy, M; Khalil, F and Elhussiny, MZ** (2016). Assessment of transportation stress in dromedary camels (*Camelus dromedarius*) using behavioural and physiological measures. *J. Appl. Vet. Sci.*, 1: 28-36.
- Fazio, F; Arfuso, F; Rizzo, M; Giannetto, C; Giudice, E; Zanghi, E and Piccione, G** (2018). Livestock handling and road transport influence some oxidative stress parameters in ewes. *J. Vet. Behav.*, 26: 5-10. <https://doi.org/10.1016/j.jveb.2018.02.001>.
- Fazio, E; Medica, P; Aronica, V; Grasso, L and Ferlazzo, A** (2008). Circulating  $\beta$  endorphin, adrenocorticotrophic hormone and cortisol levels of stallions before and after short road transport: Stress effect of different distances. *Acta Vet. Scand.*, 50: 6. <https://doi.org/10.1186/1751-0147-50-6>.
- Fazio, E; Medica, P; Cravana, C; Aveni, F and Ferlazzo, A** (2013). Comparative endocrinological responses to short transportation of Equidae (*Equus asinus* and *Equus caballus*). *Anim. Sci. J.*, 84: 258-263. <https://doi.org/10.1111/j.1740-0929.2012.01065.x>.
- Fisher, AD; Niemeyer, DO; Lea, JM; Lee, C; Paull, DR; Reed, MT and Ferguson, DM** (2010). Effects of 12, 30 or 48 h of road transport on physiological and behavioral responses of sheep. *J. Anim. Sci.*, 88: 2144-2152. <https://doi.org/10.2527/jas.2008-1674>.
- Ghareeb, K and Böhm, J** (2009). Stress indicators to pre slaughter transportation of broiler chickens fed diets supplemented with a synbiotic. *Int. J. Poult. Sci.*, 8: 621-625. <https://doi.org/10.3923/ijps.2009.621.625>.
- Giannetto, C; Fazio, F; Casella, S; Marafioti, S; Giudice, E and Piccione, G** (2011). Acute phase protein response during road transportation and lairage at a slaughterhouse in feedlot beef cattle. *J. Vet. Med. Sci.*, 73: 1531-1534. <https://doi.org/10.1292/jvms.10-0520>.
- Gregory, NG** (2005). Recent concerns about stunning and slaughter. *Meat Sci.*, 70: 481-491. <https://doi.org/10.1016/j.meatsci.2004.06.026>.
- Greunz, EM; Krogh, AKH; Pieters, W; Ruiz, OA; Bohner, J; Reckendorf, A; Monaco, D and Bertelsen, MF** (2018). The acute phase and hemostatic response in dromedary camels (*Camelus dromedarius*). *J. Zoo Wildl. Med.*, 49: 361-370.
- Grilli, C; Stocchi, R; Loschi, AR; Conti, F and Rea, S** (2018). Survey on broiler pre slaughter mortality in a commercial abattoir of central Italy. *Ital. J. Food Saf.*, 7: 5878. doi: 10.4081/ijfs.2018.5878.
- Gruys, E; Toussaint, MJM; Landman, WJM; Tivapasi, M; Chamanza, R and van Veen, L** (1999). Infection, inflammation and stress inhibit growth: mechanisms and nonspecific assessment of the processes by acute phase proteins. In: Wensing, T (Ed.), *Production diseases in farm animals*. 10th International Conference, Wageningen Press. PP: 72-87.
- Hall, S; Broom, D and Kiddy, G** (1998). Effect of transportation on plasma cortisol and packed cell volume in different genotypes of sheep. *Small Rumin. Res.*, 29: 233-237. [https://doi.org/10.1016/S0921-4488\(97\)00114-4](https://doi.org/10.1016/S0921-4488(97)00114-4).
- Hodgson, DR; McGowan, CM and McKeever, KH** (2013). *The Athletic horse: Principles and practice of equine sports medicine*. 2nd Edn., USA, Saunders Company. PP: 207-251.
- Huff, GR; Huff, WE; Rath, NC; Anthony, NB and Nestor, KE** (2008). Effects of *Escherichia coli* challenge and transport stress on hematology and serum chemistry values of three genetic lines of turkeys. *Poult. Sci.*, 87: 2234-2241. <https://doi.org/10.3382/ps.2008-00128>.
- Hyypä, S** (2005). Endocrinal responses in exercising horses. *Livest. Prod. Sci.*, 92: 113-121. <https://doi.org/10.1016/j.livprodsci.2004.11.014>.
- Ishizaki, H and Kariya, Y** (2010). Road transportation stress promptly increases bovine peripheral blood absolute NK cell counts and cortisol levels. *J. Vet. Med. Sci.*, 72: 747-753. <https://doi.org/10.1292/jvms.09-0441>.
- Justova, E; Vecerek, V; Semerad, Z; Vucinic, M; Valkova, L and Voslarova, E** (2025). The synergistic effect of transport distance and season on poultry mortality during transport to slaughterhouses. *Poult. Sci.*, 104: 105447. <https://doi.org/10.1016/j.psj.2025.105447>.
- Kataria, N; Kataria, AK; Agarwal, VK; Garg, SL and Sahn, MS** (2007). Solute loads and transfer function of kidney in dromedary camel during dehydration and rehydration in winter and summer. *Vet. Arh.*, 77: 237-246.
- Klein, R; Nagy, O; Tóthová, C and Chovanová, F** (2020). Clinical and diagnostic significance of lactate dehydrogenase and its isoenzymes in animals. *Vet. Med. Int.*, 2020: 5346483. <https://doi.org/10.1155/2020/5346483>.
- Knowles, TG; Brown, SN; Warriss, PD; Phillips, AJ; Dolan, SK; Hunt, P; Ford, JE; Edwards, JE and Watkins, PE** (1995). Effects on sheep of transport by road for up to 24 h. *Vet. Rec.*, 136: 431-438. <https://doi.org/10.1136/vr.136.17.431>.
- Krawczel, PD** (2007). Effect of duration of transport on indicators of stress in lambs. Doctoral Dissertation, Texas A&M University. PP: 1-112.
- Kuczynski, T; Blanes-Vidal, V; Li, B; Gates, RS; de Alencar Naas, I; Moura, DJ and Banhazi, TM** (2011). Impact of global climate change on the health, welfare and productivity of intensively housed livestock. *Int. J. Agric. Biol. Eng.*, 4: 1-22.
- Kumar, S; Lamo, D; Gahlawat, G; Bharti, VK and Kumar, K** (2022). Effect of endurance load exercise on physio biochemical and hormonal parameters of single humped camels (*Camelus dromedarius*) at high altitude. *Indian J. Anim. Sci.*, 92: 837-842. <https://doi.org/10.56093/ijans.v92i7.115253>.
- Kusano, K; Yamazaki, M; Kiuchi, M; Kaneko, K and Koyama, K** (2016). Reference range of blood biomarkers for oxidative stress in Thoroughbred racehorses (2-5 years

- old). *J. Equine Sci.*, 27: 125-129. <https://doi.org/10.1294/jes.27.125>.
- Lendrawati, L; Priyanto, R; Jayanegara, A; Manalu, W and Desrial, D** (2020). Effect of different transportation period on body weight loss and hematological and biochemical stress responses of sheep. *J. Indonesian Trop. Anim. Agric.*, 45: 115-123. <https://doi.org/10.14710/jitaa.45.2.115-123>.
- Li, R; Wang, L; Chen, B; Zhang, Y and Qi, P** (2024). Effects of transportation on blood indices, oxidative stress, rumen fermentation parameters and rumen microbiota in goats. *Animals*, 14: 1616. <https://doi.org/10.3390/ani14111616>.
- Liu, YZ; Wang, YX and Jiang, CL** (2017). Inflammation: the common pathway of stress-related diseases. *Front. Hum. Neurosci.*, 11: 316. doi: 10.3389/fnhum.2017.00316.
- Mackenzie, AM; Drennan, M; Rowan, TG; Dixon, JB and Carter, SD** (1997). Effect of transportation and weaning on humoral immune responses of calves. *Res. Vet. Sci.*, 63: 227-230. doi: 10.1016/s0034-5288(97)90025-4.
- Minka, NS and Ayo, JO** (2008). Haematology and behaviour of pullets transported by road and administered with ascorbic acid during the hot dry season. *Res. Vet. Sci.*, 85: 389-393. <https://doi.org/10.1016/j.rvsc.2007.11.009>.
- Minka, NS and Ayo, JO** (2012). Assessment of thermal load on transported goats administered with ascorbic acid during hot dry conditions. *Int. J. Biometeorol.*, 56: 333-341. <https://doi.org/10.1007/s00484-011-0430-2>.
- Miranda de la Lama, GC; Monge, P; Villarroel, M; Olleta, JL; García Belenguer, S and María, GA** (2011). Effects of road type during transport on lamb welfare and meat quality in dry hot climates. *Trop. Anim. Health Prod.*, 43: 915-922. <https://doi.org/10.1007/s11250-011-9783-7>.
- Miranda de la Lama, GC; Salazar Sotelo, MI; Pérez Linares, C; Figueroa Saavedra, F; Villarroel, M; Sañudo, C and María, GA** (2012). Effects of two transport systems on lamb welfare and meat quality. *Meat Sci.*, 92: 554-561. <https://doi.org/10.1016/j.meatsci.2012.05.026>.
- Mitchell, MA; Kettlewell, PJ and Maxwell, MH** (1992). Indicators of physiological stress in broiler chickens during road transportation. *Anim. Welf.* 1: 91-103.
- Mohamed, RH; El Maaty, AMA; Abd El Hameed, AR and Ali, AH** (2021). Impact of travel by walk and road on testicular hormones, oxidants, trace minerals, and acute phase response biomarkers of dromedary camels. *Heliyon*, 7: e06879. doi: 10.1016/j.heliyon.2021.e06879.
- Mohammadi, GR; Nazifi, S; Rezakhani, A and Esmailnejad, Z** (2007). Effect of transportation stress on blood and bronchoalveolar lavage fluid components in calves. *Comp. Clin. Pathol.*, 16: 85-95. <https://doi.org/10.1007/s00580-007-0673-3>.
- Mohammed, AK; Sackey, AKB; Tekdek, LB; Gefu, JO and Mohammed, AK** (2007). Serum biochemical values of healthy adult one humped camel (*Camelus dromedarius*) introduced into a sub humid climate in Shika Zaria, Nigeria. *J. Anim. Vet. Adv.*, 6: 597-600.
- Montané, J; Marco, I; López Olvera, J; Manteca, X and Lavín, S** (2002). Transport stress in roe deer (*Capreolus capreolus*): Effect of a short acting antipsychotic. *Anim. Welf.*, 11: 405-417.
- Nazifi, S; Saeb, M; Baghshani, H and Saeb, S** (2009). Influence of road transportation during hot summer conditions on oxidative status biomarkers in Iranian dromedary camels (*Camelus dromedarius*). *Afr. J. Biochem. Res.*, 3: 282-287.
- Niedzwiedz, A; Kubiak, K and Nicpoń, J** (2013). Plasma total antioxidant status in horses after 8 h of road transportation. *Acta Vet. Scand.*, 1: 55-58. <https://doi.org/10.1186/1751-0147-55-58>.
- Niedzwiedz, A; Zawadzki, M; Filipowski, H and Nicpon, J** (2012). Influence of 8 hour road transportation on selected physiological parameters in horses. *Bull. Vet. Inst. Pulawy*, 56: 193-197.
- Nielsen, SS; Alvarez, J; Bicout, DJ; Calistri, P; Canali, E; Drewe, JA; Bastuji, BG; Rojas, JLG; Schmidt, CG; Herskin, M; Michel, V; Chueca, MAM; Padalino, B and Roberts, HC** (2022). Welfare of domestic birds and rabbits transported in containers. *EFSA J.*, 20: 56.
- Oikawa, M; Hobo, S; Oyamada, T and Yoshikawa, H** (2005). Effects of orientation, intermittent rest and vehicle cleaning during transport on development of transport related respiratory disease in horses. *J. Comp. Pathol.*, 132: 153-168. <https://doi.org/10.1016/j.jcpa.2004.09.006>.
- Onmaz, A; Van Den Hoven, R; Gunes, V; Cinar, M and Kucuk, O** (2011). Oxidative stress in horses after a 12 hours transport period. *Rev. Med. Vet.*, 162: 213-217.
- Padalino, B** (2015). Effects of the different transport phases on equine health status, behavior, and welfare: A review. *J. Vet. Behav.*, 10: 272-282. <https://doi.org/10.1016/j.jveb.2015.02.002>.
- Padalino, B; Maggolino, A; Boccaccio, M and Tateo, A** (2012). Effects of different positions during transport on physiological and behavioral changes of horses. *J. Vet. Behav.*, 7: 135-141.
- Padalino, B; Raidal, SL; Carter, N; Celi, P; Muscatello, G; Jeffcott, L and De Silva, K** (2017). Immunological, clinical, haematological and oxidative responses to long distance transportation in horses. *Res. Vet. Sci.*, 115: 78-87. <https://doi.org/10.1016/j.rvsc.2017.01.024>.
- Pascual Alonso, M; Miranda de la Lama, GC; Aguayo Ulloa, L; Villarroel, M; Mitchell, M and María, GA** (2017). Thermophysiological, haematological, biochemical and behavioural stress responses of sheep transported on road. *J. Anim. Physiol. Anim. Nutr.*, 101: 541-551. <https://doi.org/10.1111/jpn.12455>.
- Piccione, G; Casella, S; Giannetto, C; Bazzano, M; Giudice, E and Fazio, F** (2013). Oxidative stress associated with road transportation in ewes. *Small Rumin. Res.*, 112: 235-238. <https://doi.org/10.1016/j.smallrumres.2012.11.001>.
- Pisoni, L; Devant, M; Bassols, AM; Saco, Y; Pato, R; Pujols, J and Marti, S** (2023). Effects of colostrum consumption and feed restriction during marketing and transportation on nutritional status and recovery in male dairy calves. *J. Dairy Sci.*, 106: 9304-9322. <https://doi.org/10.3168/jds.2023.23374>.
- Pisoni, L; Devant, M; Blanch, M; Pastor, JJ and Marti, S** (2022). Simulation of feed restriction and fasting: Effects on animal recovery and gastrointestinal permeability in unweaned Angus Holstein calves. *J. Dairy Sci.*, 105: 2572-2586. <https://doi.org/10.3168/jds.2021.20878>.
- Pregel, P; Bollo, E; Cannizzo, FT; Biolatti, B; Contato, E and Biolatti, PG** (2005). Antioxidant capacity as a reliable marker of stress in dairy calves transported by road. *Vet. Rec.*, 156: 53-54. <https://doi.org/10.1136/vr.156.2.53>.
- Qin, Y; Qian, C; Li, W; Wang, Q; Sheng, Q; Chen, Z; Zhang, W; Li, W; Ge, G; Yan, Z and Geng, D** (2026). Oxidative stress: Molecular mechanisms, diseases, and therapeutic targets. *MedComm.*, 7: e70600. <https://doi.org/10.1002/mco2.70600>.
- Ralston, S and Stives, M** (2012). Supplementation of ascorbic acid in weanling horses following prolonged transportation. *Animals*, 2: 184-194. <https://doi.org/10.3390/ani2020184>.
- Ramnath, V; Rekha, PS and Sujatha, KS** (2008).

- Amelioration of heat stress induced disturbances of antioxidant defense in chickens by Brahma Rasayana. *Evid. Based Complement. Alternat. Med.*, 5: 77-84. <https://doi.org/10.1093/ecam/nel116>.
- Reed, SM; Bayly, WM and Sellon, DC** (2003). *Equine internal medicine*. 2nd Edn., St. Louis, Missouri, Elsevier. PP: 117-126.
- Richards, GJ; Wilkins, LJ; Weeks, CA; Knowles, TG and Brown, SN** (2012). Evaluation of the microclimate in poultry transport module drawers during marketing of end of lay hens. *Vet. Rec.*, 171: 474.
- Rizzo, M; Arfuso, F; Giannetto, C; Giudice, E; Longo, F; Di Pietro, S and Piccione, G** (2017). Cortisol levels and leukocyte profiles in transported and exercised horses after acupuncture stimulation. *J. Vet. Behav.*, 18: 56-61.
- Roy, RC; Cockram, MS; Dohoo, IR and Ragnarsson, S** (2015). Transport of horses for slaughter in Iceland. *Anim. Welf.*, 24: 485-495.
- Ruiz de la Torre, JL; Velarde, A; Diestre, A; Gispert, M; Hall, SJ; Broom, DM and Manteca, X** (2001). Effects of vehicle movements during transport on stress responses and meat quality of sheep. *Vet. Rec.*, 148: 227-229. <https://doi.org/10.1136/vr.148.8.227>.
- Saeb, M; Baghshani, H; Nazifi, S and Saeb, S** (2010). Physiological responses of dromedary camels to road transportation relative to cortisol, thyroid hormones and biochemical parameters. *Trop. Anim. Health Prod.*, 42: 55-63. <https://doi.org/10.1007/s11250-009-9385-9>.
- Sanusi, M; Yusuf, S and Komolafe, PL** (2022). Effect of short distance travel on haematology, physiology, and behaviour of sheep transported in the savanna region of Northern Nigeria. *Niger. J. Anim. Prod.*, 49: 89-96.
- Satterlee, DG; Aguilera Quintana, I; Munn, BJ and Krautmann, BA** (1989). Vitamin C amelioration of adrenal stress response in broilers prepared for slaughter. *Comp. Biochem. Physiol. A*, 94: 569-574. [https://doi.org/10.1016/0300-9629\(89\)90595-1](https://doi.org/10.1016/0300-9629(89)90595-1).
- Sconberg, S; Nockels, CF; Bennett, BW; Bruyninckx, W; Blancquaert, AM and Craig, AM** (1993). Effects of shipping, handling, ACTH and epinephrine on  $\alpha$  tocopherol in bovine blood. *Am. J. Vet. Res.*, 54: 1287-1293.
- Scope, A; Filip, T; Gabler, C and Resch, F** (2002). Influence of transport and handling stress on hematologic and clinical chemistry parameters in racing pigeons. *Avian Dis.*, 46: 224-229.
- Stull, CL; Morrow, J; Aldridge, BA; Stott, JL and McGlone, JJ** (2008). Immunophysiological responses of horses to a 12 hour rest during 24 hours of road transport. *Vet. Rec.*, 162: 609-614. <https://doi.org/10.1136/vr.162.19.609>.
- Stull, CL and Rodiek, AV** (2000). Physiological responses of horses to 24 hours of transportation using a commercial van during summer conditions. *J. Anim. Sci.*, 78: 1458-1466. <https://doi.org/10.2527/2000.7861458x>.
- Stull, CL; Spier, SJ; Aldridge, BM; Blanchard, M and Stott, JL** (2004). Immunological response to long term transport stress in mature horses and effects of adaptogenic dietary supplementation as an immunomodulator. *Equine Vet. J.*, 36: 583-589. <https://doi.org/10.2746/0425164044864589>.
- Taff, CC; Baldan, D; Mentasana, L; Ouyang, JQ; Vitousek, MN and Hau, M** (2024). Endocrine flexibility can facilitate or constrain the ability to cope with global change. *Philos. Trans. R. Soc. B*, 379: 20220502. <https://doi.org/10.1098/rstb.2022.0502>.
- Tateo, A; Padalino, B; Boccaccio, M; Maggiolino, A and Centoducati, P** (2012). Transport stress in horses: Effects of two different distances. *J. Vet. Behav.*, 7: 33-42.
- Teke, B; Akin, PD; Akdag, F; Ugurlu, M and Ekiz, B** (2019). Effect of season on dead on arrival rate and meat quality of broiler chickens transported under commercial slaughter conditions. *Large Anim. Rev.*, 25: 147-151.
- Tharwat, M; Al Sobayil, F and Buczinski, S** (2013a). Effect of racing on the serum concentrations of cardiac troponin I and creatine kinase myocardial band in racing camels (*Camelus dromedarius*). *Vet. Res. Commun.*, 37: 139-144. <https://doi.org/10.1007/s11259-013-9556-z>.
- Tharwat, M; Al Sobayil, F and Buczinski, S** (2013b). Cardiac biomarker changes in camels (*Camelus dromedarius*) secondary to road transportation. *J. Vet. Cardiol.*, 15: 15-22. <https://doi.org/10.1016/j.jvc.2012.08.004>.
- Tsigos, C and Chrousos, GP** (2002). Hypothalamic-pituitary-adrenal axis, neuroendocrine factors and stress. *J. Psychosom. Res.*, 53: 865-871.
- Tüfekci, H and Sejian, V** (2023). Stress factors and their effects on productivity in sheep. *Animals*, 13: 2769. <https://doi.org/10.3390/ani13172769>.
- Villani, M; Cairoli, F; Kindahl, H; Galeati, G; Faustini, M; Carluccio, A and Veronesi, MC** (2006). Effects of mating on plasma concentrations of testosterone, cortisol, oestrone sulphate and 15 ketodihydro PGF $_{2\alpha}$  in stallions. *Reprod. Domest. Anim.*, 41: 544-548. <https://doi.org/10.1111/j.1439-0531.2006.00711.x>.
- Voslarova, E; Bedanova, I; Vecerek, V; Pistekova, V; Chloupek, P and Suchy, P** (2006). Changes in haematological profile of common pheasant (*Phasianus colchicus*) induced by transit to pheasantry. *Dtsch. Tierarztl. Wochenschr.*, 113: 375-378.
- Wessely Szponder, J; Belkot, Z; Bobowiec, R; Kosior Korzecka, U and Wójcik, M** (2015). Transport induced inflammatory responses in horses. *Pol. J. Vet. Sci.*, 18: 407-413. <https://doi.org/10.1515/pjvs-2015-0052>.
- Zhong, RZ; Liu, HW; Zhou, DW; Sun, HX and Zhao, CS** (2011). The effects of road transportation on physiological responses and meat quality in sheep differing in age. *J. Anim. Sci.*, 89: 3742-3751. <https://doi.org/10.2527/jas.2010-3693>.