Heat Stress-Induced Oxidative Stress Damage to Animal Intestinal Tissue (Postprint)

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Abstract

Heat stress is a common nonspecific stressor that causes substantial losses to livestock production. Intestinal tissues readily develop ischemia and hypoxia under heat stress, and intestinal cells undergo oxidative stress, leading to apoptosis and intestinal tissue damage. As the intestine is the most important organ for nutrient absorption and pathogen barrier in animal organisms, its damage directly affects the growth, development, and health status of the animal. This article reviews recent domestic and international research progress on heat stress-induced oxidative stress in intestinal cells, oxidative stress-induced intestinal damage, and heat stress-induced apoptosis pathways.

Full Text

Oxidative Stress Induced by Heat Stress: Injury to Animal Intestinal Tissue

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Abstract: Heat stress is a common nonspecific stressor that causes significant losses to animal husbandry. Under heat stress, intestinal tissue is prone to ischemia and hypoxia, which induces oxidative stress in intestinal cells, leading to cell apoptosis and intestinal tissue damage. Since the intestine is the most important organ for nutrient absorption and pathogen defense in animals, its injury directly affects animal growth and health. This review summarizes recent domestic and international research progress on oxidative stress induced by heat stress in intestinal cells, the damage caused by oxidative stress to the intestine, and the apoptosis pathways triggered by heat stress.

Keywords: intestine; heat stress; oxidative stress; apoptosis

Classification: S852.2

Heat stress is a type of stress response that occurs when animals are exposed to high temperatures. Due to ineffective heat dissipation, animals exhibit open-mouth breathing, depression, reduced feed intake, and slowed weight gain. In severe cases, heat stress can cause death from heart failure or brain damage, while also challenging animal welfare [1]. The damage to animal tissues caused by heat stress primarily occurs through oxidative stress and inflammatory responses, which lead to cell injury and apoptosis.

The intestine performs crucial functions in nutrient absorption and pathogen barrier defense in animals. Approximately one-third of total blood flow circulates through the intestine. When animals are in high-temperature environments, blood flow is redistributed to the body surface for heat dissipation while maintaining supply to the heart and brain, leaving intestinal tissue susceptible to ischemia and hypoxia. This causes intestinal cell apoptosis and damages the intestinal mucosa, which not only impairs absorptive function but also allows pathogen invasion due to compromised mucosal integrity, potentially leading to systemic infection [2]. Consequently, animals experience poor growth from inadequate nutrition or die from infections that bypass their defense barriers. This paper summarizes recent research focusing on how heat stress-induced oxidative stress damages animal intestinal tissue.

1. Heat Stress Induces Oxidative Stress in Animal Intestinal Tissue

During normal metabolism, animals produce numerous reactive oxygen species (ROS), while maintaining an antioxidant defense system including enzymes such as superoxide dismutase (SOD). Under normal conditions, pro-oxidants and antioxidants exist in dynamic equilibrium. When cells encounter exogenous stimuli like heat stress, antioxidant levels decrease while pro-oxidants increase, disrupting this balance and resulting in oxidative stress in animal tissues and cells.

Following heat stress, the content and activity of certain biochemical factors related to oxidative stress change significantly. Heat-stressed animals show increased serum levels of corticosterone, malondialdehyde (MDA), and ROS, while SOD, glutathione peroxidase (GSH-Px) activity, and total antioxidant capacity (T-AOC) decrease [3-4], indicating a correlation between heat stress and oxidative stress. These changes are not limited to serum components; high temperature also significantly elevates ROS content in rat small intestinal epithelial cells and reduces intracellular SOD and GSH-Px activity [5], while causing severe apoptosis, necrosis, and detachment of small intestinal epithelial cells. Kikusato et al. [6] confirmed that heat stress increases mitochondrial peroxides and elevates intracellular ROS content, demonstrating that heat stress directly induces oxidative stress in cells.

Therefore, when animals experience heat stress, both serum and intestinal tissues show decreased antioxidants and increased pro-oxidants, causing oxidative stress in small intestinal cells. The resulting ROS and free radicals damage intestinal structure and function [7-9].

2. Damage of Heat Stress-Induced Oxidative Stress to Animal Intestine

Morphological studies of heat stress effects on animal intestine have revealed decreased villus height, increased villus width, reduced epithelial cell area, significantly diminished mucosal thickness, intestinal wall thickness, and crypt depth, and notably fewer goblet cells. Hematoxylin-eosin (HE) staining clearly shows severe damage to the intestinal villi. These histomorphological changes indicate serious impairment of both absorptive and barrier functions [10-14].

The intestinal epithelial secretory cell lineage comprises Paneth cells, goblet cells, and enteroendocrine cells, which protect the intestine from harmful pathogens and regulate intestinal function through hormone secretion. Mature small intestinal epithelial cells typically survive 4-5 days before shedding into the intestinal lumen, with new epithelial cells differentiating from stem cells at the crypt base and migrating to the villus tips to replace lost cells. This proliferation and death balance is strictly maintained to preserve intestinal barrier and absorptive functions [15]. Epidermal growth factor is a known mitogen with broad biological activity in animals, including promoting intestinal development and repairing damaged mucosa [16]. Liu et al. [17] found that heat stress downregulates epidermal growth factor and its receptor expression in porcine jejunum, slowing epithelial cell proliferation and compromising mucosal integrity.

Acute heat stress increases serum corticosterone and mortality in broilers, reduces feed intake and daily weight gain, and decreases feed conversion efficiency —changes potentially related to multifocal acute enteritis induced by acute heat stress [18]. Examination of jejunal tissue in heat-stressed animals shows significantly increased mRNA expression of pro-inflammatory cytokines TNF-α, IFN-γ, and IL-β [19], elevated myeloperoxidase (MPO) activity, and increased blood endotoxin levels [20], confirming that heat stress causes intestinal inflammation.

ROS and MDA produced during heat stress are important factors damaging intestinal tissue. ROS accumulation can damage DNA and induce apoptosis, while MDA, a product of lipid peroxidation, damages cell membranes and also triggers apoptosis [21-22]. In summary, heat stress damages intestinal cells or induces apoptosis through multiple pathways including oxidative stress and inflammatory responses, thereby destroying intestinal mucosal integrity—findings consistent with Cui et al.' s [23] proteomic study of heat stress effects on porcine small intestine.

3. Apoptosis Pathways Induced by Heat Stress

Persistent heat stress severely damages intestinal tissue, with apoptosis being the primary mechanism. Heat stress induces oxidative stress in intestinal cells, and excessive ROS production causes DNA damage, protein and lipid peroxidation, ultimately leading to apoptosis [24-25]. Recent research indicates that heat stress-induced apoptosis pathways are complex, including mitochondrial, death receptor, and endoplasmic reticulum stress pathways.

3.1 Mitochondrial Apoptosis Pathway

Transmission electron microscopy of rat jejunal ultrastructure reveals that heat stress causes mitochondrial swelling and vacuolation [19], demonstrating a direct link between heat stress-induced apoptosis and mitochondria. In the mitochondrial pathway, cytochrome C forms an apoptosome with Caspase-9 and apoptotic protease-activating factor 1 (APAF1), activating the apoptosis executor Caspase-3. Xu et al. [26] induced oxidative stress in cultured rat intestinal epithelial cells (IEC-6) with hydrogen peroxide and measured apoptosis-related genes and mitochondrial membrane potential, finding consistent changes: significantly increased ROS, decreased SOD activity, increased MDA, elevated cytoplasmic cytochrome C, decreased mitochondrial membrane potential, and increased Caspase-9 and Caspase-3 levels. Genes related to the mitochondrial pathway, Bax (pro-apoptotic) and Bcl-2 (anti-apoptotic), and apoptosis rates increased significantly, indicating that oxidative stress induces apoptosis via the mitochondrial pathway. However, Bcl-2 overexpression may represent cellular self-defense against oxidative stress. Jia et al. [27] reported that heat stress upregulates Caspase-3, Caspase-8, Caspase-9, and Bax gene expression while also upregulating Bcl-2 expression in porcine small intestinal epithelial cells, suggesting that heat stress induces apoptosis through both mitochondrial and death receptor pathways.

3.2 Death Receptor Pathway

The death receptor pathway, also called the extrinsic apoptosis pathway, is triggered when tumor necrosis factor receptor (TNFR) family members bind their ligands. For example, Fas binding to its ligand (FasL), with assistance from Fas-associated death domain protein (FADD), recruits and accumulates Caspase-8 precursors to form the death-inducing signaling complex (DISC), activating Caspase-8. Activated Caspase-8 initiates a caspase cascade, further activating Caspase-3 and ultimately causing apoptosis [24,28-29]. Heat stress causes overexpression of Fas, FasL, Caspase-8, and Caspase-3 genes and significantly increases apoptosis rates in cultured cells, demonstrating that heat stress induces apoptosis through the death receptor pathway [30-31].

3.3 Endoplasmic Reticulum Stress Pathway

The endoplasmic reticulum (ER) is a vital organelle in all eukaryotic cells with protein processing and modification functions. Cellular stimuli such as hypoxia, hypoglycemia, hyperglycemia, and acidosis cause accumulation of unfolded and misfolded proteins in the ER, triggering ER stress (ERS). Cells protect themselves from ERS damage through a highly conserved stress-adaptive response called the unfolded protein response (UPR), but prolonged adverse stimulation or insufficient UPR leads to apoptosis [32-33].

In animal cells, Caspase-12 is a specific mediator of ERS-induced apoptosis that is not activated in death receptor or mitochondrial pathways [34-35]. Yin et al. [36] studied rat small intestine injury mechanisms under simulated transport-induced heat and vibration stress. Transmission electron microscopy revealed severe ER swelling in jejunal epithelial cells, indicating ER stress. KEGG pathway analysis of stressed rat jejunum showed that apoptosis and autophagy, MAPK signaling, P53 signaling, and mTOR signaling pathways were involved in heat- and vibration-induced cell injury.

4. Measures to Enhance Intestinal Heat Stress Resistance

Heat stress is a nonspecific stressor whose damage can be alleviated at the whole-animal level through improved ventilation and cooling in animal housing. Since heat stress-induced intestinal damage is primarily caused by oxidative stress, protecting and improving intestinal injury should focus on antioxidant approaches and timely anti-inflammatory treatment for the inflammatory response occurring during heat stress.

Common antioxidants include selenium, vitamin C, and traditional Chinese medicine components such as astragalus polysaccharides. Liu et al. [37] demonstrated that selenium and vitamin C can alleviate the adverse effects of heat stress in pigs and protect intestinal barrier integrity. Chinese researchers have studied antioxidant traditional Chinese medicine formulations, such as modified Baihu Decoction, which effectively reduces heat stress damage in dairy cows and promotes heat dissipation [38]. Taurine possesses multiple biological functions including antioxidant and anti-apoptotic effects, alleviating oxidative stress damage to intestinal mucosa and inhibiting intestinal epithelial cell apoptosis [39]. Probiotics play important roles in intestinal health; feeding probiotic mixtures effectively alleviates intestinal barrier dysfunction caused by heat stress [40], and supplementing with Lactobacillus strains improves broiler performance under heat stress conditions [41].

Comprehensive research on heat stress effects on animal intestine shows that intestinal tissue is vulnerable to heat stress damage. When animals remain in high-temperature environments without relief, intestinal mucosal integrity is compromised, and intestinal cells undergo oxidative stress with massive apoptosis. Current domestic and international research primarily focuses on oxidative stress damage and apoptosis in mammalian small intestinal epithelial cells, particularly regarding death receptor and mitochondrial pathways, with less research on avian species. Future studies should examine relationships between apoptosis pathways induced by heat stress and explore methods to enhance intestinal heat stress resistance. Heat stress impairs nutrient absorption and facilitates pathogen invasion, ultimately causing growth retardation or death from infection. This highlights the importance of preventing heat stress in animal production. Improving ventilation and cooling in hot regions can effectively reduce heat stress losses, and investigating mechanisms of intestinal damage while seeking methods to improve intestinal heat stress resistance are key to solving this problem.

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