

Farm animal welfare, productivity and meat quality: Interrelation with redox status regulation and antioxidant supplementation as a nutritional intervention (Review)

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Abstract. Contemporary concerns over farm animal welfare have transpired as a result of the desire of farmers to gain the benefits of the increased productivity of their farming animals. Inspired by the constant flow of scientific knowledge, technology and new production methods, the progressive exploitation of the animal biological capacity to produce economic output, has raised increasing concerns as to the extent to which the animals are being stressed. In fact, individuals worldwide depend heavily on animals for the production of meat, fat, milk and other essential products. However, in recent years, the demand for livestock products has increased tremendously and world livestock production is facing major challenges, which endanger the health of the animals. Furthermore, the utilization of proper antioxidant supplementation as an important and cost-effective treatment for beneficial effects on farm animals is also a major pillar for the manipulation of current needs in modern animal husbandry. In this comprehensive review, we aimed to summarize the knowledge regarding the interrelation of farm animal welfare, productivity and meat quality with redox status regulation and antioxidant supplementation as a promising nutritional intervention. We propose that evaluating various biomarkers may help researchers to adopt the optimal growth conditions of farm animals, a fact that is expected to protect them from redox-related pathologies, in order to enhance their productivity and to strengthen the quality of their products.

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1. Introduction

The amount of meat consumption has doubled worldwide over the past 50 years and is expected to further increase by 10% in the forthcoming decade as reported by the Food and Agricultural Organization of the United Nations (<http://www.fao.org/3/y4252e/y4252e05b.htm>). Therefore, one of the major challenges of modern society is to develop the livestock sector by improving the conditions of animal growth and adopt a new model of distribution of high-quality products. To this end, farm animal husbandry practices are intrinsically connected with animal welfare and productivity. In order to provide animal products of high quality efficiently, producers should acquire the appropriate knowledge regarding the necessary good practices that will favor the well-being of farm animals. Although over the past few years, farmers have adopted specific methods of husbandry in order to achieve a greater production of their products, it appears that these strategies are hazardous to animal health, leading to an enhanced incidence of disease. Moreover, the extreme temperatures caused by the current climate change phenomenon constitute a serious challenge for the productivity of farm animals. Therefore, producers and researchers have turned their attention to a variety of nutritional interventions that could potentially protect animals from diseases or increase their productivity and commerciality of the generated products. Specifically, recent research focuses on the administration of natural antioxidants to livestock in order to enhance the health and productivity of farm animals, mainly by protecting cellular macromolecules from detrimental oxidative modifications (1-6). Therefore, a specific need for a battery of efficient biomarkers that reflect the extend of biomolecule oxidative damage has emerged (7). The adoption of redox biomarkers, which depict the action of the antioxidant defense system and the harmful role of reactive species on biomolecules that are changed by redox altering stimuli, will enable researchers to introduce nutritional practices with beneficial antioxidant properties.

Based on the above, in this review article, we intend to highlight the interrelation of farm animal welfare, produc-

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tivity and meat quality with redox status regulation and antioxidant supplementation as a promising nutritional intervention (Fig. 1). Additionally, we stress the significance of specific redox biomarkers as research tools in order to adopt appropriate practices that will help animal husbandry to develop according to modern perspectives. On the basis of consumers' awareness and desire for healthy and qualitative animal products, we suggest that it is of utmost importance to determine redox biomarkers in farm animal blood and tissues and also to supplement proper antioxidant compounds as an important and cost-effective treatment for protecting farm animal health, enhancing their productivity and improving the quality of meat produced by livestock.

2. Regulation of redox status and farm animal welfare

The European Union (EU) has established, over the past 40 years, a broad and detailed legislative framework regarding animal welfare (Council Directive 98/58/EC: <https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX:31998L0058>). The reasons that contributed to this development are mainly humanitarian. However, economic reasons constitute key factors as well, proving that animal welfare results in the generation of improved and safer products for consumers. Currently, the EU has enacted one of the strictest legislative frameworks worldwide and, hence, the highest welfare standards protecting billions of animals reared, moved and slaughtered on the territories of its member states. Due to such persistent work by the authorities of the EU, it is now ensured that animals breed and dwell with respect to their physiological needs, whereas painful and inhumane procedures during their slaughter process have been almost fully abandoned.

Despite the presence of the aforementioned analyzed strict legislation, the problems that stem from trespassing, negatively affect welfare of productive animals. To this end, the majority of the animals in the current production systems are farmed under non-appropriate conditions and the dynamics of disease transmission emerges as a serious issue in comparison to the animal populations that are farmed in open range traditional systems. This mainly occurs as animals are more closely in contact with each other and disease transmission may occur more readily. Therefore, the transmission of an infectious agent between animals can be sustained for longer time periods. Severe infectious diseases in farm animals have been recorded in a plethora of publications and are mainly associated with oxidative stress, a major health-related issue in modern livestock units (8). In particular, the pathogenesis of common diseases, such as pneumonia (9), enteritis (10) and sepsis (11,12) is associated with disorders in animal redox status and, more specifically, with oxidative modifications leading to the damage of biomolecules (i.e., proteins, lipids and DNA). The oxidation of macromolecules generates various end-products that can be measured and characterized as redox biomarkers in order to assess oxidative stress *in vivo* (7). Towards this direction, in order to clarify the impact of oxidative stress on farm animal welfare, a wide range of biomarkers in biological fluids and tissues has been adopted.

It has been recently suggested that protein oxidation is an important indicator of infections induced by parasites, bacteria and viruses that leads to a variety of pathological conditions,

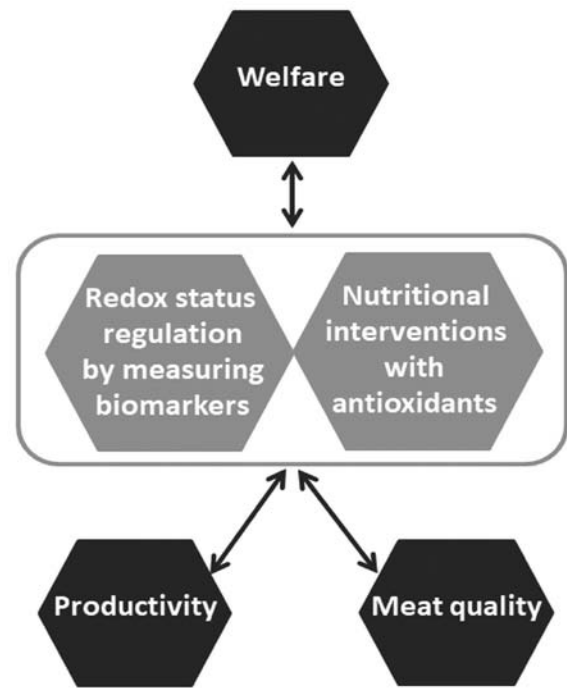


Figure 1. The interrelation of farm animal welfare, productivity and meat quality with redox status regulation and antioxidant supplementation as a promising nutritional intervention.

such as increased immunogenicity (13). This fact suggests that protein oxidation is an important phenomenon, depicted by specific biomarkers, that putatively reflects the presence of specific diseases, although there is not yet a known correlation between certain pathologies and redox biomarkers. However, it seems that limiting protein oxidation is an important pillar for protecting farm animals. One way to do this is to administer natural antioxidants contained in plant extracts, a practice that also contributes to prolonging life of animals (14). There are several large clinical trials that have been conducted to investigate the hypothesized benefit of antioxidant supplements in disease prevention (15-17). Similar results have been produced by other relevant studies in sheep (18) and cattle (19), providing added value to the livestock products, namely meat or milk. Concomitantly, further studies from our group have shown that the administration of feed, including bioactive compounds from winery by-products improves redox profile of farm animals. Specifically, feed supplemented with grape pomace has been shown to decrease oxidative stress-induced toxic effects and improve the chicken redox status, and thus, it may also improve their wellness (20). The results of that study demonstrated that the aforementioned feed protected the animals from protein oxidation and lipid peroxidation, and increased reduced glutathione (GSH) in blood and several tissues of vital chicken organs. Similar results were observed in another study following the administration of the same feed to lambs, while showing an enhanced growth of probiotic bacteria and inhibited growth of pathogen populations, such as Enterobacteriaceae and *E. Coli*, suggesting the improvement of gut microbiome function (21). Moreover, the supplementation of this innovative feedstuff to piglets has been shown to result in an increased GSH concentration, H₂O₂ decomposition rate and total antioxidant capacity, implying improved antioxidant

mechanisms in almost all tissues (22). Furthermore, the piglets exhibited decreased oxidative stress-induced damage to lipids and proteins, as well as an improved fecal microbiota, suggesting a beneficial effect on piglet welfare (22). Similar results have been reported following the administration of feed generated by pioneering techniques, namely enrichment with olive oil mill wastewater bioactive compounds. Specifically, the supplementation of this feed to broiler chickens could be used to reduce oxidative damage to proteins and lipids and to increase of antioxidant mechanisms through molecules, such as GSH and catalase and also by upregulating the total antioxidant capacity (23). Young piglets that also consumed this enriched feed were found to have a decreased $\omega 6/\omega 3$ fatty acid and PUFA/MUFA ratio compared to the control group, suggesting that polyphenols contained in this feed protect the animals from the manifestation of pathological conditions (24). In addition, the administration of olive oil mill wastewater enriched feed has been shown to improve antioxidant mechanisms in lambs, mainly by increasing GSH levels and reducing the detrimental effects of oxidative damage to basal macromolecules (25). Additionally, a promising supplement for farm animals potentially protecting their welfare is vitamin E. This is due to the fact that it is considered the most important chain-breaking, lipid-soluble antioxidant (26) and, therefore, it lowers oxidative stress levels and improves the health of dairy cows (10). All these observations highlight the importance of the administration of plant-derived compounds with valuable properties for the improvement of the farm animal redox status, promoting the well-being of the animals. The evaluation of oxidative stress in livestock is significant for the understanding of the fundamental procedures involved in diseases and metabolic disorders. Therefore, in order to avoid the high mortality rates of farm animals from the aforementioned diseases and to protect their tissues from the harmful oxidation process, it is considered necessary to analyze and study their redox molecular background by measuring specific redox biomarkers (27,28).

3. Regulation of redox status and farm animal productivity

Farm animal productivity is an issue of utmost importance concerning the producers and the professionals of the animal agriculture sector. It is an indisputable fact that specific strategies regarding productive animal farming can be detrimental for their healthy development. More specifically, animal dwelling in close restraint cages, lifetime enclosure in indoor sheds, housing under inappropriate conditions causing discomfort and injuries, restriction or prevention of normal bouts of exercise, absence of daylight or fresh air and poor air quality and overcrowding of healthy or even sick animals, can seriously compromise their productivity. As we have already mentioned, farm animals can be exposed, during the production cycle, to environmental stressors that may result in oxidative stress and, consequently, in severe health abnormalities. The efficacy of animal production is based principally on balanced nutritional systems that meet the individual needs of farm animals. Animal growth can be influenced or even regulated by the supplementation of antioxidants in animal feed (29); thus, it is likely to be affected by the overall redox status of a productive animal.

Indeed, it has been suggested that the high metabolic rate of growing tissues generates abnormally large amounts of free radicals (30). In order to avoid the formation of free radicals that usually lead to oxidative stress and other redox-related pathologies, they must be safely scavenged or removed via the administration of antioxidant compounds. On this basis, numerous studies over the past few years have examined the hypothesis that the supplementation of feed that is enriched with antioxidants to farm animals will provide extra nutritional value to the meat product and will act protectively for their welfare. For instance, the supplementation of poultry with selenium, a known potent antioxidant, appears to play a key role in sustaining the health and productive performance of the animals (31). Similarly, whey protein has also been proven to be beneficial on pig productivity, since, as previously demonstrated, there was a differentiation in the weight of the animals as those that received food enriched with whey exhibited significantly higher body weight compared to the control group (32). The fact that liquid whey supplement leads to enhanced productivity, thus accelerating the slaughter process, but luckily without affecting meat quality or carcass traits, is worthy of mention. In addition, the use of fulvic acid, an organic acid that is formed naturally during the decomposition of organic matter known as humus or humic compounds, as a supplement, has been shown to improve performance and the immune system of broiler chickens, whereas it does not affect meat quality and safety (33). Previous publications have associated the supplementation of vitamin E in ruminants with the enhancement of productive performance (34,35). It has also been suggested that vitamin E supplementation leads to beneficial alterations of redox status in blood, liver and mammary gland (36). Specifically, vitamin E supplementation reduces oxidative damage in liver, but has no obvious effect on mammary gland and blood MDA concentrations (36).

Another environmental factor that affects animal productivity is heat stress or a hot environment (37). Specifically, it appears that heat stress alters several farm animal traits, such as growth, meat and milk yield and quality, productive performance, metabolic and health status (38). It has also been suggested that hot conditions capable of producing moderate or severe heat stress can result in oxidative stress in dairy cows (39,40). Furthermore, it has been reported that acute heat stress (i.e., 32°C for 6 h) induces oxidative stress in 5-week old broiler chickens (41). The impaired farm animal productivity induced by the aforementioned factors can potentially cause devastating economic consequences to producers and to the sector of global animal agriculture, in general (42). As a consequence, the health status of farm animals is highly affected by enhanced oxidative stress and disturbed redox equilibrium in blood and tissues under exposure to heat stress and other non-appropriate farming conditions (39). Previous studies have suggested that antioxidants, such as selenium and vitamin E can probably ameliorate these negative outcomes (43-45). Therefore, as Celi and Chauhan (37) have suggested, the manipulation of oxidative stress during the transition period by the enrichment of the farm animal diet with antioxidants may prove to be promising towards the improvement of their health and productivity. Hence, well-designed and applied research that will outline further

opportunities for vitamin E and selenium in this field, and subsequently for other recommended antioxidants is required.

4. Regulation of redox status and farm animal meat quality

Meat quality is a predominant factor contributing to the consumer selection and acceptability of products generated from specific producers. As regards meat quality, parameters, such as color, water-holding capacity and the oxidative stability of lipids and proteins are usually referred to. It has been shown that the successful management of oxidative stress and the optimization of blood and tissue redox status are key factors for the improvement of the quality of meat and milk produced by livestock (19,46). The disruption of the redox equilibrium resulting in oxidative damage to macromolecules in productive animals equals to unpleasant economic situations due to the negative impact on meat quality. As long as the priorities of producers and consumers are to avoid or delay the development of rancid products, an important goal should be the high oxidative stability of meat-based foods. The enhanced antioxidant status of the living animal and a subsequent increased oxidative stability of the meat product is regarded particularly helpful and desirable for both the consumer and the processing industry (47).

Hereby, we discuss several factors that can affect the quality of meat produced by farm animals. Firstly, studies have associated meat quality with nutritional management, since feeding and developmental conditions seem to be very influential on the oxidative stability of the generated meat. Over the past century, it is widely acknowledged that human nutrition has been directed towards a healthier motif. Therefore, there is an urgent need to return to a balanced fatty acid diet by decreasing the intake of cholesterol and saturated fats (48). It has been found that the oxidative modification of lipids, which are susceptible to peroxidation, degrades animal productivity and meat quality (49,50). Towards this direction, a herbal extract mixture administered to animals has been shown to significantly increase the content of polyunsaturated fatty acids (PUFAs), creating meat with beneficial health properties which is less susceptible to oxidation (51). Another relevant study suggested that it is possible to modify the fatty acid composition of the carcass fat of lambs by manipulating post-weaning nutrition, highlighting the importance of antioxidant administration to these animals (52). Additionally, a mixture of ascorbic acid- α -tocopherol, as well as oregano supplemented to chickens, has been shown to protect against lipid peroxidation by preventing the increase in muscle thiobarbituric acid reactive substances (TBARS) generated by the environmental stress imposed on the animals during the transport and slaughter process (47). In that study, ascorbic acid- α -tocopherol and oregano supplementation increased erythrocyte stability, reflecting an increased antioxidant status in chickens, and this seemed to provide protection against environmental stress that could potentially induce lipid peroxidation and detrimental oxidative modifications of biomolecules (47). Erythrocyte stability can be affected by several factors related to nutritional interventions, such as membrane lipid composition, intracellular GSH concentrations and vitamin E content (53-55). Several other studies have reported similar results about the beneficial actions of

plant antioxidants on reducing lipid oxidation (56-58). As the above-mentioned studies have reported, limiting lipid peroxidation is an important pillar for improving meat quality (59).

Apart from lipid peroxidation, comprehending the biological mechanisms of protein oxidation will enable producers to generate meat with a better appearance, quality and nutritional value (60). To date, little is known about the relation between protein oxidation and meat quality deterioration. However, it is known that the modifications caused by reactive oxygen species (ROS) in muscle (the main origin of meat that humans consume) proteins could be implicated in the loss of their functionality and, therefore, in the impairment of meat quality (61). The consequences of protein oxidation have been thus far mainly identified as the deterioration of meat tenderness and juiciness (60,62,63). It has been confirmed that the oxidation of proteins causes their polymerization and aggregation, thus affecting their digestibility, a fact that inevitably leads to a marked reduction in the nutritional value of meat (64). In addition, when myofibrillar protein isolate is incubated with oxidizing agents, the total amounts of amino acids (e.g., cysteine, tyrosine, methionine, histidine and alanine) decrease, demonstrating a putative mechanism for the reduction of the meat nutritional value (65). Towards dealing with the protection of muscle protein oxidation and as a result of the avoidance of compromising meat quality, the supplementation of antioxidants to productive animals may prove to be a rather promising practice. Indicatively, a study on chickens fed with a high antioxidant diet containing apples and broccoli revealed a lower level of protein carbonyl content compared to chickens that were fed a poor antioxidant diet (66). However, extra experimental evidence is required to reveal the full extent of the consequences of protein oxidation in meat quality.

Another factor that, according to the literature, affects the quality of meat is the animal breed. It has been reported that the quality of lamb meat is significantly dependent on the genetic background of the animals, although it is feasible to be affected and specifically further improved by changes in the post-weaning nutrition (67). It has been indicated that the effect of breed is apparent and among other factors that have been studied, breed seems to be the most crucial regarding the effects on carcass fat and its cholesterol content (68). The above-mentioned study was conducted in three independent experiments, where different factors, such as breed, sex, the degree of maturity at slaughter process and slaughter weight were compared in order to clarify which factor affects meat quality to a greater extent. As the degree of maturity and nutritional management changes, breed seems to be the main factor that affects the cholesterol content (68). This fact strengthens the view of the superiority of the genetic factor. In agreement with these results, another study evaluated the effects of breed on fatty acid composition and, subsequently, on meat quality and reported that three different breeds of lambs affected most of the fatty acids studied in this experiment (53). The fatty acid composition of carcass fat of meat producing animals has been recognized to be of utmost importance regarding several characteristics of overall meat quality (69-71). The above-mentioned evidence can serve as important guidelines for producers so that they can select the breed of interest according to its physical characteristics in order to improve

the quality of the carcasses they produce in terms of fatty acid composition. Moreover, as mentioned above, a study by our team indicated that feed containing polyphenolic additives from byproducts of processed mill wastewater with recognized antioxidant properties was beneficial for the lipid ratio in the meat of piglets, affecting not only the health of livestock, but also the quality of the produced meat (24). Further, the administration of grape pomace to weaned piglets as a feed additive, has been shown to improve the antioxidant status and affect the fatty acid composition. Specifically, it increased n-3 fatty acids and decreased n-6/n-3 ratio suggesting beneficial impact on meat quality (22). Therefore the evaluation of fatty acid composition is crucial for meat quality.

5. Conclusions

The study of the pathophysiological effects caused by the oxidative stress of farm animals has been of great interest over the past decades. As we have already mentioned above, a number of research articles have indicated that this situation is affected by a variety of environmental and nutritional factors. The examination of these factors and of the mechanisms involved is crucial for the sustainable development of the livestock industry. Furthermore, a priority should be the safekeeping of animal welfare based on all available scientific evidence, a fact that will lead to enhanced animal productivity and, subsequently, to the improved quality and commerciality of animal products. From now on, emphasis should be given to the scientific understanding of animal physiology and how this is affected by housing practices, health care measures, diseases, transport and nutritional management procedures. All these fields of research will provide useful information and guidelines for the manipulation of healthy animal development. Future research in animal nutrition, as well as nutritional manipulation should be based on modern research tools and methods. Over the past few years, there is a growing demand for omics-based approaches in animal science (72). Such directions will enlighten differential issues on animal management that need to be clarified. The elucidation of the mechanisms of action, the discovery of new functions, the quantification of efficient biomarkers and the nutritional impact on the -omics level should be a priority for future research projects. As regards the use of different nutritional interventions on animal diet, researchers should focus on deepening the knowledge of the interaction between antioxidants and animal physiology in order to provide substantial information on the response of animals to the consumption of feed enriched with antioxidants.

Therefore, we propose herein a new approach that investigates the biological effects of the supplemented antioxidants and, subsequently, reveals the correlation between complex systems, such as proteome, genome and metabolome with the redox status in animal models (Fig. 2). Moreover, another aspect considering the increasing awareness among consumers of the benefits of antioxidant enriched foods is that livestock industries have the potential to improve the production of antioxidant enriched products as functional foods. The proposed approach will create the background and the prerequisites for a substantial future partnership of breeders, consumers and companies in order to optimize

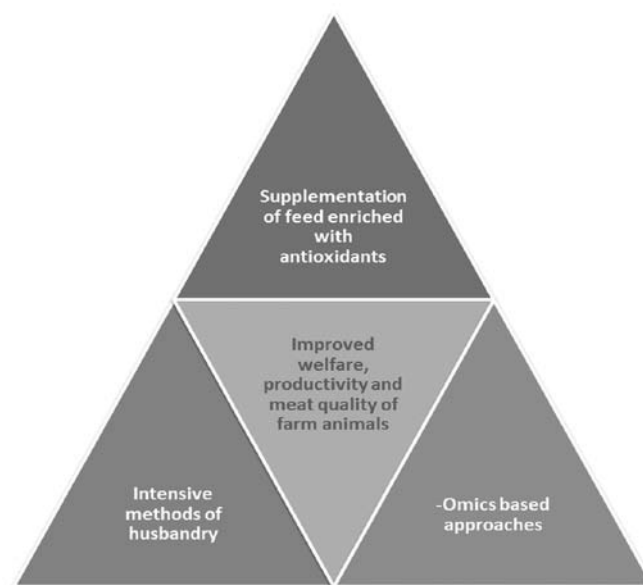


Figure 2. The novel approaches highlighted hereby with respect to the improvement of farm animal welfare, productivity and meat quality by adopting supplementation of feed enriched with antioxidants, intensive methods of husbandry and -omics-based research methodology.

animal husbandry and commercialization of the produced meat. Incorporating innovative technologies and scientific knowledge in practice will help increase the added value of meat, ensure the sustainability of farms and promote the future development of the livestock sector. The adoption of such strategies will provide a comprehensive frame of the optimum conditions for animal growth and nutrition contributing decisively to better service of producer and consumer response.

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Availability of data and materials

Not applicable.

Author's contributions

ZS performed the literature search, and collected the data used in this review article. ASV and DK were involved in the conception and design, and editing of this review article. ZS and ASV were involved in the preparation of the figures. All authors have read and approved the final manuscript.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

All the authors declare that there are no competing interests.

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