



Biochemical Changes and Gene Expression of Antioxidant Genes in Poultry Fed Garlic-Supplemented Diets

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Abstract

This article reviews the positive effects of adding garlic to poultry feed on biochemical changes and the gene expression of antioxidant genes. Garlic (*Allium sativum* L.) shows a remarkable ability to enhance the antioxidant status in poultry by increasing the expression of key antioxidant genes, namely superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx). These genes neutralize reactive oxygen species (ROS) and maintain oxidative balance in cells, reducing oxidative stress and limiting damage to biomolecules such as lipids and proteins.

The article reviews the possible mechanisms for this effect, which are attributed to the biologically active compounds in garlic, especially organic sulfur compounds such as allicin, which act as direct antioxidants or stimulate the body's own antioxidant defense system. It also discusses experimental studies (*in vitro* and *in vivo*) confirming that garlic-supplemented nutrition improves performance indicators, promotes gut health, and supports the immune system.

Finally, the paper provides practical recommendations regarding feed formulation and determining the optimal garlic dosage (e.g., 0.5 to 1 g/kg feed) to achieve maximum benefit without negative effects. It is concluded that incorporating garlic into poultry feed is a promising nutritional strategy for improving health and productivity by enhancing the antioxidant defense system at the genetic and biochemical levels.

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1. Introduction

The strategies for utilizing garlic (*Allium sativum* L.) as a feed supplement exhibit diversity; nevertheless, the majority of investigations employ the garlic supplementation method (Chen *et al.*, 2021) ^[12]. The inclusion of garlic in the diet elevates the expression of key antioxidant genes, specifically superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) in poultry species. Various prospective *in vivo* and *in vitro* examinations consistently reveal that the garlic-supplemented diet significantly upregulates these antioxidant genes, concomitantly enhancing the defense against oxidative stress. Subsequently, the supplementation influences synergistic biochemical processes, collectively diminishing oxidative damage to macromolecules such as lipids and proteins (M. Delles *et al.*, 2014; F. Surai *et al.*, 2019) ^[33, 16].

2. Antioxidant Genes in Poultry

Antioxidant genes play a pivotal role in maintaining redox homeostasis and mitigating oxidative stress in poultry physiological processes. Superoxide dismutase (SOD) catalyzes the dismutation of superoxide anions to oxygen and hydrogen peroxide (Bogolyubova *et al.* 2022) ^[8]. Catalase (CAT) further detoxifies hydrogen peroxide to water and oxygen, and glutathione peroxidase (GPx) reduces lipid hydroperoxides to their corresponding alcohols and also detoxifies hydrogen peroxide (F. Surai *et al.*, 2019) ^[16].

The regulation of these antioxidant enzymes at the gene-expression level is a key determinant of oxidative stress resilience. Consequently, dietary strategies that modulate the expression of *sod*, *cat*, and *gpx* genes are of considerable interest for enhancing poultry health and productivity (Gusti *et al.* 2021)^[23].

SOD constitutes a primary defense mechanism by converting highly reactive superoxide radicals, generated as metabolic by-products, into less harmful molecules, thereby preventing oxidative damage to cellular components. CAT operates predominantly in peroxisomes to rapidly reduce intracellular hydrogen peroxide levels, protecting cells from oxidative injury. GPx exhibits broader substrate specificity, targeting lipid hydroperoxides and hydrogen peroxide, thus playing a crucial role in the inhibition of lipid peroxidation and maintaining membrane integrity. Given the significance of oxidative stability in lipid-rich tissues such as egg yolk and meat, the transcriptions of *sod*, *cat*, and *gpx* genes are essential markers when assessing the antioxidant status of poultry (Goel *et al.*, 2021; Grzegorzewska *et al.*, 2024)^[20, 22].

2.1. Superoxide Dismutase (SOD)

Antioxidant enzymes play a critical role in preventing oxidative stress in birds (F. Surai *et al.*, 2019)^[16], which can impair overall well-being and reduce growth performance. Among the increasingly utilized dietary plants that enhance the antioxidant system, garlic (*Allium sativum*) is noteworthy (M. Delles *et al.*, 2014)^[33].

2.2. Catalase (CAT)

Catalase (CAT) plays a crucial role in the antioxidant system by catalyzing the decomposition of hydrogen peroxide, thereby preventing oxidative damage to cells. Various stress factors—including heat, disease, and high-density rearing—promote free radical generation, leading to redox imbalance and oxidative stress that harms biological macromolecules. Such damage negatively influences growth, immune function, and overall animal health (Rasheed, 2024; Anwar *et al.* 2024)^[41, 6]. The antioxidant defense incorporates enzymes such as CAT, superoxide dismutase (SOD), and glutathione peroxidase (GSH-Px) to mitigate free radicals. In fast-growing species like yellow broilers, oxidative stress impairs intestinal integrity and general well-being, particularly under adverse environmental conditions (Eddaikra and Eddaikra 2021)^[14]. An experimental evaluation of catalase effects on growth performance, antioxidant capacity, intestinal morphology, microbial composition, and protein expression is therefore warranted (Tang *et al.*, 2022)^[49].

2.3. Glutathione Peroxidase (GPx)

Glutathione peroxidase (GPx) belongs to the family of selenium-containing enzymes. These enzymes interact with superoxide dismutase (SOD) and catalase (CAT) to provide crucial antioxidant defense within biological systems (Yang *et al.*, 2018)^[55]. Glutathione peroxidase reduces fatty acid hydroperoxides to their corresponding alcohols and catalyses the reduction of hydrogen peroxide (H₂O₂), thereby preventing lipid peroxidation and maintaining cellular integrity (Tsekhmistrenko *et al.*, 2025)^[51]. These enzymatic actions contribute substantially to the detoxification of reactive oxygen species (ROS) and the protection of organismal health. The main ROS produced in biological systems are the free radical species superoxide anion (O₂⁻), hydrogen peroxide (H₂O₂), and hydroxyl radical (OH⁻).

When created in excess, O₂⁻ is transformed to H₂O₂ by SOD, which is then reduced by CAT and GPx. SOD proteins dismutate superoxide (O₂⁻) into intermediates that CAT and GPx can quickly scavenge, preventing the generation of extremely reactive hydroxyl radicals (HO⁻) (M. Delles *et al.*, 2014)^[33].

3. Garlic as a Dietary Supplement

Garlic (*Allium sativum*) has a long history of culinary and medicinal usage, attributed to its essential vitamins, minerals, and bioactive compounds (Huang *et al.*, 2020)^[26]. The presence of active compounds such as allicin underpins its antioxidant, antibacterial, antiviral, antifungal, antitumor, and immunomodulatory activities. Furthermore, allicin exhibits protective effects against immune-mediated hepatic injury and diminishes lipid raft stability in murine models (Nadeem *et al.*, 2021)^[36]. Polyphenols in garlic restrict oxidative processes by scavenging free radicals and inhibiting decomposing enzymes (M. Delles *et al.*, 2014)^[33]. It is widely recognized that dietary antioxidant supplementation enhances the redox status across biological systems, a phenomenon documented in various studies. Supplementation of broiler diets with garlic powder elevates the expression of a range of antioxidant and immune-related genes, including GPx and CAT (Zhou *et al.*, 2022)^[57].

3.1. Nutritional Components of Garlic

Garlic (*Allium sativum* L.) is a widely consumed vegetable rich in essential nutrients, including carbohydrates, protein, vitamins, lipids, and organosulfur compounds (Škrovánková *et al.*, 2018)^[46]. Its bioactive constituents exhibit antioxidant and health-promoting properties, alongside antiatherogenic effects and antimicrobial activity (Huang *et al.*, 2020)^[26]. Polyphenols and flavonoids present in garlic contribute to its cholesterol-lowering abilities, restoration of liver enzymes, enhanced blood circulation, and hepatic anti-inflammatory effects. Traditionally, garlic has been employed to improve antioxidant capacity and reduce oxidative stress (Askari *et al.* 2021)^[7]. Variations in total polyphenol content and antioxidant activity have been reported across garlic cultivars, geographical origins, and processing methods (Škrovánková *et al.*, 2018)^[46].

3.2. Health Benefits of Garlic

Garlic, *Allium sativum*, the most widely used spice worldwide, possesses antibacterial, antifungal, lipid-lowering, cardioprotective, anticancer (Ansary *et al.*, 2020)^[5], antioxidant, and wound-healing properties (Huang *et al.*, 2020)^[26]. It is consumed as fresh garlic, aged garlic, garlic oil, or powder, either as a raw ingredient or as a dietary supplement (Sunanta *et al.* 2023)^[48].

4. Mechanisms of Action of Garlic Compounds

The antioxidant potential of garlic is attributed primarily to its organosulfur components, which modulate physiological activity in the body and promote susceptibility to lipid peroxidation (M. Delles *et al.*, 2014)^[33]. Sulfur-containing compounds can mimic endogenous antioxidants and restore glutathione (GSH) levels, thereby contributing to cellular redox balance and promoting intracellular thiol status, which is believed to protect antioxidant enzymes from oxidative degradation (Capasso, 2013)^[9]. Allicin (diallyl thiosulfinate) is a principal sulfur-containing compound responsible for the antioxidant effect of garlic. Its capacity to capture free

radicals via interactions with protein thiol groups and low-molecular-weight substances that contain sulphhydryl groups is what gives it its antioxidant properties (Seki & Hosono, 2025) ^[44]. By raising their levels, allicin has further effects on antioxidant enzymes. In particular, it protects the thiol groups of glutathione peroxidase (GPx), catalase (CAT), and superoxide dismutase (SOD) from oxidative degradation, hence enhancing their biological functions. Beyond just allicin, these beneficial actions add to the garlic components' total antioxidant capacity (Nadeem *et al.*, 2021) ^[36]. Garlic's function in poultry's antioxidant defense system is reinforced by experimental research showing that adding it to the diet increases the activity of these important antioxidant enzymes (Abd El-Ghany, 2024) ^[1].

4.1. Allicin and Antioxidant Activity

Garlic's well-researched bioactive ingredient, allicin, has several health advantages and antioxidant properties (Huang *et al.*, 2020) ^[26]. It suppresses bacterial infections, reduces immune-mediated liver damage, and has immunomodulatory effects. It has been demonstrated that the garlic organosulfide diallyl trisulfide decreases the stability of lipid rafts in macrophages and partially mediates vasodilation through hydrogen sulfide (Tang *et al.*, 2021) ^[50]. Many years ago, the antioxidant properties of allicin were identified and attributed to a number of sulfur-containing molecules, such as alliin, allyl cysteine, and allicin (Capasso, 2013) ^[9].

4.2. Other Bioactive Compounds in Garlic

The primary bioactive ingredient in *Allium sativum*, aside from allicin, is its abundance of organosulfur compounds, such as diallyl trisulfide, diallyl disulfide, diallyl sulfide, vinylthiins, and ajoene, which have strong anti-cancer and antioxidant qualities (Škrovnáková *et al.*, 2018) ^[47]. The ability of diallyl trisulfide to emit hydrogen sulfide (H₂S), a gasotransmitter that is known to mediate a number of physiological processes, emphasizes *Allium sativum*'s therapeutic significance even further. Using garlic powders that have been enzymatically digested and fermented, complementary *in vitro* studies have found seven different polyphenolic compounds, including epicatechin, catechin, and protocatechuic acid, as well as two strong antioxidant peptides, MGF and GNF, confirming the presence of a variety of bioactive substances in the plant (Škrovnáková *et al.*, 2018) ^[47]. The phytochemical profile of *Allium sativum* is further enhanced by monoterpene hydrocarbons such as γ -terpinene, α -terpinene, and myrcene; oxygenated monoterpenes such as terpinen-4-ol, α -terpineol, and linalool; and diterpene alcohols (e.g., phytol). These compounds together provide broad-spectrum biological activities, such as analgesic, antioxidant, antimicrobial, anti-inflammatory, antiviral, antitumoral, and hepatoprotective effects. In garlic extracts, more than fifty volatile compounds—mostly arranged into ketones, organosulfur derivatives, aldehydes, alcohols, and esters—have also been identified, enhancing the plant's well-known therapeutic repertoire for a variety of diseases controlled by these bioactives (Mabou & Yossa, 2021; Mołdoch *et al.* 2025) ^[34, 35].

5. Effects of Garlic on Antioxidant Gene Expression

The antioxidant genes in poultry express a number of antioxidant enzymes that play an important physiological role in limiting the oxidative stress-induced damage. These genes include superoxide dismutase, catalase, and glutathione

peroxidase (M. Delles *et al.*, 2014) ^[33]. Garlic supplementation has long been popular due to its diverse beneficial effects. It contains various biological components that act as antioxidants and prevent oxidative species from damaging the cellular molecules. Prooxidant species induce oxidative damage to DNA bases as well as in deoxyribose, which is one of the major targets in biological systems. This change in antioxidant gene expression influences the overall health status of poultry and helps maintain it at an optimum level (Abd El-Ghany, 2024) ^[1]. During the last decade, using garlic and its extracts in poultry feed has attracted much attention. It is believed that garlic has metabolic activities and can be added successfully to the diet of poultry. In recent studies, interest has shifted toward the use of biochemical markers for the optimum health status of an animal. Plants contain many phytochemicals, some of which can act as pro and others as antioxidants. Antioxidants help prevent damage caused by oxidative stress and lipid peroxidation (Chen *et al.*, 2021; Abd El-Ghany, 2024) ^[12, 1].

5.1. Impact on SOD Expression

Superoxide dismutase (SOD) is a key antioxidant metalloenzyme responsible for the detoxification of superoxide radicals, mediating the dismutation of superoxide anions into oxygen and hydrogen peroxide (M. Delles *et al.*, 2014) ^[33]. SOD catalyzes the disproportionation of the highly reactive superoxide radical (O₂⁻) into hydrogen peroxide and oxygen, forming the first line of intracellular defense against oxidative stress, and maintaining cellular reactive oxygen species (ROS) homeostasis (Fujii *et al.*, 2022) ^[17]. Dietary antioxidant supplementation enhances lipid and protein oxidative stability of chicken broiler meat through the promotion of antioxidant enzyme activity. Consumption of high-oxidized diets reduces the activity of SOD, catalase (CAT), and glutathione peroxidase (GPx) compared to low-oxidized diets. Antioxidant-supplemented diets exhibit higher enzyme activities than basal diets (Yang *et al.* 2023) ^[54]. The presence of antioxidants in high-oxidized diets alleviates some negative effects on SOD, resulting in similar SOD activity levels between low-oxidized and antioxidant-supplemented diets. SOD and CAT activities decline under prolonged dietary stress, a change associated with oxidative tissue damage. GPx activity remains higher in antioxidant-supplemented groups compared with basal diets, with the lowest activity observed in the high-oxidized diet without antioxidants. Oxidative stress may induce a decline in these antioxidant enzymes, impacting tissue health and function (Rezar *et al.*, 2025; Rathore *et al.* 2023) ^[43, 42].

5.2. Impact on CAT Expression

Catalase (CAT) protects cells from deleterious effects of excess hydrogen peroxide by converting it to water and molecular oxygen (M. Delles *et al.*, 2014) ^[33]. The oxidative metabolism of various substrates in living systems produces superoxide (O₂^{•-}), which is primarily eliminated by converting it to hydrogen peroxide and molecular oxygen. In addition to its roles in oxidative stress defense, CAT plays several widely recognized physiological functions, including cold adaptation, thermoregulation, development, metamorphosis, and inhibition of apoptosis. When the antioxidant enzyme CAT is deprived, the cells become more sensitive to hydrogen peroxide-induced senescence, demonstrating the antioxidant role of the enzyme (Irato & Santovito, 2021; Jomova *et al.* 2024) ^[28, 30]. Consequently, an

increase in CAT expression reflects an enhancement of cellular antioxidant defenses. Experimental studies have demonstrated that dietary supplementation with garlic powder upregulates CAT gene expression in the liver (Tang *et al.*, 2022)^[49].

5.3. Impact on GPx Expression

Several studies report increased glutathione peroxidase (GPx) activity in broilers after garlic-supplemented diets. One report attributes this response to S-allyl cysteine, which up-regulated GPx mRNA expression (Griela *et al.*, 2021)^[21]. Increased enzyme activity positively affects biochemical indices, notably reductions in malondialdehyde (MDA) and thiobarbituric acid reactive substances (TBARS) content (Carmo de Carvalho e Martins..., 2022)^[10]. One explanation for this effect is garlic's high antioxidant capacity, with phenolic compounds exhibiting radical scavenging, metal chelation, and enzyme inhibition, including allicin, which increases as fresh garlic is crushed and exposure continues (M. Delles *et al.*, 2014)^[33]. This multi-mechanistic action generates a powerful synergistic effect able to significantly boost GPx cellular expression, thus enhancing the organism's overall antioxidant defenses (Rampengan *et al.* 2025)^[40].

6. Biochemical Changes in Poultry

Oxidative stress results from an imbalance between the formation and neutralization of reactive oxygen species (ROS) that in turn leads to damage of proteins, lipids, carbohydrates and DNA, and contributes to many diseases (M. Delles *et al.*, 2014)^[33]. Many physicochemical, biochemical and economic changes influence the maintenance of optimum nutrient levels and productivity of poultry. Overproduction of free radicals is considered the initiator and propagator of lipid peroxidation, resulting in damage to biomolecules and hence cell injury. Extensive research has been carried out with the use of single or mixed antioxidants to protect poultry from free radical damage and thus minimize biochemical changes and oxidative stress in birds (Nawaz and Zhang 2021; Abdulrahman *et al.* 2024)^[38, 2]. Feed formulation encompasses the process of taking various raw materials or ingredients and accurately combining and mixing these materials into a finished feed particular to specific needs in terms of age, production and species. Optimal nutrient utilization and thus efficient poultry production systems can be achieved through a thorough knowledge of prerequisite nutritional requirements and a balanced accurate feed formulation (George and George 2023; Gao *et al.*, 2025)^[19, 18]. Dietary antioxidants such as vitamins E and C, flaxseed; vitamin E and selenium; vitamin E, selenium and their possible interactions have been reported to improve the oxidative stability of poultry products by increasing antioxidant enzyme activities (Desbruslais & Wealleans, 2022)^[13].

6.1. Oxidative Stress and Antioxidant Defense

Oxidative stress occurs when cellular redox balance favors reactive oxygen species (ROS) formation over their removal. Excessive ROS can alter biomolecules, damage cells and tissues, and dysregulate signaling pathways. Endogenous enzymatic antioxidants (superoxide dismutase [SOD], catalase [CAT], glutathione peroxidase [GPx]) and non-enzymatic antioxidants (e.g., glutathione) protect cells from oxidative damage. The first line of defense, SOD catalyzes

conversion of superoxide anions to hydrogen peroxide; second-line enzymes CAT and GPx process hydrogen peroxide into less harmful molecules (M. Delles *et al.*, 2014)^[33]. Antioxidant enzyme levels indicate cellular antioxidant capacity and 'redox state' (F. Surai *et al.*, 2019)^[16]. To overcome oxidative stress, organisms either increase antioxidant enzyme production or decrease ROS production (Huchzermeyer *et al.*, 2022)^[27].

6.2. Effects of Garlic on Lipid Peroxidation

Garlic supplementation in animal diets has led to an improvement in antioxidant status by maintaining normal levels of antioxidant enzymes and reducing lipoperoxidation. Lipoperoxidation occurs when free radicals attack the double bonds of polyunsaturated fatty acids in organism tissues, generating superoxide radicals (M. Delles *et al.*, 2014)^[33]. Free radicals have the potential to degrade lipids, proteins, and DNA, thus causing damage at the subcellular structural level. Indeed, lipid peroxidation propagates the process of oxidative damage and is thought to contribute to the deterioration of the quality of foods during storage; antioxidant molecules alleviate this process (Engwa & Nweke..., 2022; Tumilaar *et al.* 2024)^[15, 52]. Allicin (diallyl thiosulfinate), the primary bioactive component of freshly crushed garlic, exerts antioxidant effects in two ways: (1) an indirect effect related to the ability of allicin to react with thiol groups, forming compounds that could regenerate endogenous antioxidants and (2) a direct free-radical quenching effect. The free-radical scavenging activity of allicin allows it to reduce the damage of oxidative stress by preventing the formation of lipid peroxides in the blood and organs of various animals (Halliwell, 2024)^[24].

7. Experimental Studies

In vivo and *in vitro* studies have tested garlic as a potential dietary supplement capable of improving antioxidant defense. Chicken embryo fibroblast (CEF) cells treated with 1.0–10 mg/mL of garlic extract displayed increased viability. Moreover, broilers fed various levels (0, 0.5, 1, 1.5, 2, 4, and 6 g/kg) of garlic extract for four weeks exhibited a significant enhancement in the antioxidant system (Abd El-Ghany, 2024; Liao *et al.* 2022)^[1, 32]. Combined with improved performance, these results suggest that garlic supplementation, through the promotion of antioxidant gene expression, can ameliorate oxidative damage and support immune system development (M. Delles *et al.*, 2014)^[33].

7.1. Studies on Poultry

Studies on the capacity of organic garlic powder as a nutritional supplement to improve antioxidant capacity in broilers have been performed (M. Delles *et al.*, 2014)^[33]. The antioxidant system represents a protective defense mechanism that involves several antioxidant enzymes and molecules. For instance, the primary active antioxidant enzymes, including superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase (CAT), play significant roles in ameliorating oxidative stress. Upregulation of the gene expression levels of these enzymes increases resistance against oxidative damage (Cecerska-Heryć *et al.* 2021)^[11]. Hence, degradation of harmful free radicals by these enzymes suppresses ROS formation and enhances the antioxidant defense system of the bird (Abdulrahman *et al.* 2024)^[2].

7.2. Studies on Antioxidant Activity

Food analysis and free radical damage studies have shown that the antioxidant activity of the *Allium* plant extracts correlate very well with the flavonoid content. Reduced glutathione (GSH) plays a central role in the cellular scavenging of reactive oxygen species and is essential for the reduction of peroxides. It has been suggested that carotenoids have a protective effect against cardiovascular disease and oxidant induced lung damage (M. Delles *et al.*, 2014) [33]. Garlic displays a variety of antioxidant types of activity that correlate to its phytochemical profiles. Garlic contains polyphenol components attributed to the total antioxidant capacity (Akullo *et al.*, 2023; Skoczylas *et al.* 2023) [4, 45].

8. Nutritional Implications

The incorporation of garlic into poultry diets as an additive or supplement has emerged as a promising nutritional strategy for modulating the expression of antioxidant genes and associated biochemical parameters. Several principal considerations shape the formulation of such diets to optimize alkaline extract use as a feed supplement (Abd El-Ghany, 2024) [1].

Dosing is a critical concern. One study found that doses of 3% or more of an alkaline extract of *Allium mongolicum*, a wild relative of garlic, in the diet of Linwu ducks led to significant up-regulation of antioxidant genes encoding superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) (Wójcik *et al.*, 2023) [53]. Lower doses produced inconsistent effects, suggesting a threshold level for efficacy. Similar observations were made with *Agaricus blazei* aqueous extract in broilers, where 150 or 300 mg/kg stimulated antioxidant gene expression more strongly than 30 or 50 mg/kg (Najafi *et al.*, 2025) [37].

The potential for antagonistic interactions is another key factor. For instance, excessive intake of natural or synthetic antioxidants can impair liver function by disrupting the cytochrome P450 (CYP450) enzyme system. It is therefore advisable to moderate garlic-based supplementation or alternate it with other agents to avoid adverse effects (M. Delles *et al.*, 2014) [33].

Alternative growth-promoting supplements may also influence the choice of garlic dosage. Though some additives exhibit significant anticoccidial activity, others exert only marginal or even detrimental effects. Compatibility considerations arise because substances with similar chemical structures can compete, reducing each other's effectiveness. Consequently, optimal supplementation often involves tailored combinations (Hayat *et al.*, 2022) [25].

The available evidence highlights the potential of dietary supplementation with garlic or related extracts to enhance the antioxidant defense system by modulating gene expression of SOD, CAT, and GPx. Identifying optimal dosing regimens and combination strategies remains the focus of ongoing investigation (Jahanbakhshi *et al.* 2022) [29].

8.1. Diet Formulation Strategies

Commercially promoted for its nutritional benefits, garlic is a lucrative product in the food processing sector. Antioxidant genes including glutathione peroxidase (GPx), catalase (CAT), and superoxide dismutase (SOD) are expressed when it is added to chicken diets at concentrations of 4 g/kg feed or more (Abd El-Ghany, 2024) [1]. Enzymatic antioxidants, trace minerals, organic compounds, and certain proteins all influence the overall antioxidant state of chicken. Even under

healthy physiological settings, reactive oxygen species (ROS) are constantly being produced (Zandi & Schnug, 2022) [56]. The metalloenzyme SOD is in charge of dismutating extremely reactive superoxide radicals into molecular oxygen (O₂) and hydrogen peroxide (H₂O₂). A hemoprotein called CAT, which is found in mitochondria, lysosomes, and peroxisomes, speeds up the breakdown of H₂O₂, shielding tissues and cells from harmful effects (Karmakar *et al.*, 2022) [31]. As the last stage of the enzymatic detoxification of peroxides, GPx consists of a group of isozymes that contain selenium and catalyze the reduction of H₂O₂ to H₂O. By drastically reducing the production of hydroxyl radicals, the conversion of superoxide ions by SOD, CAT, and GPx prevents oxidative damage to proteins, DNA, and lipids (M. Delles *et al.*, 2014) [33].

8.2. Optimal Garlic Dosage in Poultry Diets

Garlic, alongside other phytochemical substances, is frequently utilized in poultry diets due to its noteworthy health benefits. Determining the precise quantities needed to achieve desired outcomes is essential, with excessive additive intake potentially leading to adverse effects such as increased fat accumulation (Petričević *et al.*, 2019) [39]. Comprehensive research efforts have sought to establish the optimal garlic dosage that maximizes developmental advantages in poultry production. (Htet Htet Kyaw Kyaw San Win Khin Khin Lay *et al.*, 2017).

Identified concentrations between 0.5 and 1.0 g/kg of diet as optimal for enhancing the performance of broiler chickens. These authors also indicated that supplementation exceeding 1 g/kg does not confer additional benefits, and hypothesized that higher dosages could result in diminished growth rates. Recognizing the need to avoid the deleterious consequences of excessive supplementation, the present analysis recommends incorporating garlic into poultry diets at levels below 1 g/kg to achieve beneficial effects without incurring negative outcomes (Chen *et al.*, 2021; Adjei-Mensah *et al.*, 2022) [12, 3].

9. Conclusion

Aryl hydrocarbon receptor (AhR) agonists such as 3-methylcholanthrene (3-MC) or 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) markedly suppressed expression of superoxide dismutase (SOD) 1, SOD 2, catalase (CAT), and glutathione peroxidase (GPX) 1 mRNAs in the chick embryo hepatocyte model, in a time- and dose-dependent manner. The inhibition of SOD 1, SOD 2, CAT, and GPX 1 gene expression was also observed in the liver of AhR agonist-treated chicken. Among the redox-related genes examined thereafter, these four genes were found to be dominantly downregulated by AhR agonist in a chicken hepatic cell line. Further, the inhibitory effects of AhR agonist were relieved by AhR antagonists, α -naphthoflavone and resveratrol. These results demonstrate that the antioxidant enzymes SOD, CAT, and GPX are primary targets of AhR-mediated transcriptional regulation in birds.

The results suggest that dietary garlic supplementation improves antioxidant status and expression of SOD, CAT, and GPX gene from poultry fed garlic-supplemented diets.

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