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Application of astaxanthin in laying hens breeding

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Abstract: *Astaxanthin is a natural antioxidant, which can effectively enhance animal immunity and has coloring function. At present, astaxanthin has been widely used in the layer breeding industry as a feed additive. This review summarizes the main biological functions of astaxanthin and its effects on production performance, egg quality and nutrition, and immune performance of laying hens.*

Keyword： *Astaxanthin，Layer breeding，Yolk color，Antioxidant activity*

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Astaxanthin (3,3′-dihydroxy-β, β′-carotene-4,4′-dione) is a fat-soluble carotenoid with strong antioxidant activity，which has been widely used in the food，cosmetics and health care products industry(Zhao et al., 2022; Masaaki et al.,2009). With the prohibition of antibiotics, astaxanthin has become a potential green additive because of its natural, residue free, antioxidant and immune function (Liu et al., 2020; Allah et al.,2021).

In the animal feed industry, astaxanthin is employed as a feed additive for fish and laying hens to enhance the color of their flesh and egg yolk, respectively. Astaxanthin also has the antioxidant role of quenching singlet oxygen (Myriam et al., 2018). Its antioxidant activity is approximately 10 times higher than that of other carotenoids (e.g., zeaxanthin, lutein, tunaxanthin, cantaxanthin, and β-carotene) and 100 times greater than vitamin E (α-tocopherol). The biotechnological production of astaxanthin can be based on the utilization of waste from aquatic product processing, *Haemittococcus pluvialis,* or *Phaffia rhodozym*(Jiang et al.,2017; Yamamoto et al., 2016).

I. Main biological functions of astaxanthin

1.1 Coloring function

Astaxanthin is a dihydroxydiketone carotenoid, which has great advantages in coloring poultry egg yolks. Research has found that for the coloring of egg yolks, diketone and dihydroxycarotenoids have stronger coloring functions than monohydroxy, monoketone or epoxy carotenoids. A certain concentration of astaxanthin in the feed deepens the color of egg yolk, and the color of poultry skin, feet and beak also deepens accordingly, and the growth speed of poultry is accelerated. These have greatly improved the nutrition and commercial value of poultry eggs and meat (Son et al., 2009; Conradie et al., 2018).

1.2 Antioxidant activity

Natural astaxanthin is one of the strongest natural antioxidants, which can effectively eliminate oxygen free radicals in cells, enhance cell regeneration, maintain body balance and reduce the accumulation of aging cells, enhance body vitality and reduce stress. Excessive reactive oxygen species（ROS） are the main factor leading to oxidative damage in the body. Astaxanthin has the ability to trap reactive oxygen species, enhance the ability of cells to block oxidative stress, and play its antioxidant function by eliminating excess ROS to terminate the chain reaction. Astaxanthin has a longer conjugated system than carotene, and a longer conjugated system enables astaxanthin molecules to effectively quench highly oxidizing singlet reactive oxygen species and other free radicals in the environment(Rao et al., 2013; Zuluaga et al., 2018).

1.3 Immune function.

Astaxanthin has strong immunoregulatory activity and can stimulate the immune system to play an important role(Lee et al., 2003). Astaxanthin can promote the production of immune globulin, such as increasing the production of IgG, IgA and IgM; Enhance the functions of T cells and B cells in the body,

improve the cellular immunity and enhance the humoral immune response. Thereby reducing the incidence of poultry diseases and enhancing adaptability under stress conditions such as high temperatures(Li et al., 2019; Park et al., 2018).

II. Application of astaxanthin in layer hen breeding

Astaxanthin has a lot of applications in the field of layer feed because of its important physiological functions such as antioxidant, immune enhancement, coloring function and so on.

2.1 The impact on the production performance of laying hens

In many studies, the effects of different concentrations of astaxanthin added to the diets of laying hens on the performance of laying hens were discussed. 43-day-old Jinghong hens were selected and randomly divided into five groups. They were fed the basal diet added with 0.6，1. 2，2.4 and 3.6 g /kg astaxanthin compound additive，respectively．The results showed that the addition of astaxanthin compound additive had no significant effects on average daily feed intake， laying rate and feed /egg of laying hens (P＞0.05) (Wang et al., 2018)．

350-day-old healthy Hy-Line Brown laying hens were e fed diets containing different levels of natural astaxanthin(0, 25,50, 100mg/kg). The addition of natural astaxanthin had no significant ellects on production perlormance of laying hens comparedto the control group (P > 0.05)（Li et al., 2019）. 60-week-old Lohmann Brown laying hens were randomly assigned to four groups. The astaxanthin content in the diet was 0.96 mg/kg, 1.44 mg/kg and 1.92 mg/kg respectively. Results showed that dietary PR supplementation tended to increase daily feed intake (p=0.0512) （Zhu et al., 2021）.

Some studies have also shown that the addition of astaxanthin to the diet of laying hens can improve the feed to egg ratio or daily average feed intake and other indicators. Average daily feed intake is one of the important indicators for evaluating the production performance of laying hens, and also an important indicator for measuring breeding costs(Ahmad, 2015). The results of Xue indicate that throughout the entire experimental period (1-5 weeks), the average daily feed intake of each experimental group was lower than that of the control group (P<0.05), and the experimental group I had the best effect. It showed that adding astaxanthin could reduce the average daily intake（Xue et al.，2015）.

The feed to egg ratio is one of the many indicators for evaluating the production performance of laying hens, and it is also an important indicator for measuring the cost of breeding. Adding astaxanthin can reduce the feed to egg ratio, and the feed to egg ratio of each test group is significantly different from that of the control group (P<0.05)(Bai, 2021). Because astaxanthin has the function of maintaining and promoting the normal growth of animals and improving animal immunity, it can reduce the feed intake and increase the egg weight, indirectly reducing the feed to egg ratio.

The difference in the results of adding astaxanthin to the diets of laying hens on the production performance of laying hens may be related to the factors such as the composition of the diet（Akiba, 2008; Moreno, 2020）, the content of astaxanthin and the way of addition（Honda, 2021）, the type and age of laying hens(Yang et al., 2019), the breeding environment. Further research is needed in this area in the future.

2.2 The impact on egg quality

Although many reports indicate that astaxanthin has little effect on egg quality, it is very significant in improving yolk color. Generally, with the increase of astaxanthin, the color of egg yolk increases. Yang et al (2006) showed that dietary astaxanthin had no significant effect on the performance of laying hens; The yolk color increased linearly with the increase of dietary astaxanthin level, and decreased significantly with the increase of storage time. However, Dansou et al (2021) showed that there was a decrease in concentration and coloration efficacy of astaxanthin at high dose supplementation (213.4 mg/kg) compared to moderate doses (21.3 and 42.6 mg/kg).

Takahashi et al. (2023) studied the effects of *Paracoccus carotinifaciens* supplementation containing high astaxanthin levels on egg production performance and taste of astaxanthin-rich egg yolk. The results showed that ASX supplementation did not affect production performance or egg quality. ASX levels in the egg yolk became saturated after seven days of 16 ppm supplementation and decreased to less than one-tenth of the saturated levels seven days after supplementation cessation. Supplementation with 16 ppm ASX for 28 d did not affect egg yolk taste. Experiments conducted by Zhu et al. (2022) have indicated that the level of astaxanthin in laying hens' diet of 10~30 mg/kg can improve egg quality, yolk color and eggshell thickness, and 30 mg/kg astaxanthin can significantly increase the content of astaxanthin in yolk.

2.3 The impact on egg nutrition

The level of consumers' satisfaction with the quality of chicken eggs is determined, in particular, by the attractive appearance of the yolks and their content of biologically active substances that have functional properties.

It was found by Honchar et al (2022）that the addition of astaxanthin to the diet of laying hens reduced and stabilized the ratio of ω3/ω6 PUFA in yolks during egg storage to a greater extent than the addition of lycopene. Storage of astaxanthin-enriched edible chicken eggs at 4 ±0.5 ℃ and 12 ±0.5 ℃ for 30 days can be used to correct the fatty acid profile of yolk lipids. Shevchenko （2020） conducted the study with 45 High Line W36 chickens at the age of 24 weeks . It was found that astaxanthin at a dose of 10 mg/kg of feed reduced the content of palmitoleic acid by increasing the proportion of cis-10-heptadecenoic acid in the lipids of eggs.

2.4 The effect on the antioxidant performance of laying hens

Astaxanthin is widely known for its antioxidant activities, and it possesses a particularly high antioxidant capacity, as it has more conjugated double bonds than many other carotenoids. Its molecular structure characteristics determine the active Electronic effect of astaxanthin, which can provide electrons to free radicals or attract electrons from inactive free radicals, so it can play a role in scavenging free radicals and anti-oxidation.(Yu et al., 2019; Liu et al., 2016; Zhu et al.,2022)

The results by Gao et al showed that the total antioxidant capacity, superoxide dismutase level, and glutathione peroxidase level in the plasma, livers, and egg yolks were significantly increased in the ASTA groups compared with those of the control group ($P < 0.05$), whereas the content of malondialdehyde linearly decreased ($P < 0.05$). The plasma levels of high-density and very-low-density lipoprotein cholesterol in the ASTA groups were significantly higher than those in the control group ($P < 0.05$).

He et al（2023） showed that Astaxanthin increased the expression of nuclear factor e2-related factor 2 (NRF2) in the ovary ($P < 0.05$), enhanced the antioxidant capacity of aged laying hens ($P < 0.05$), and reduced cellular apoptosis ($P < 0.05$). In addition, astaxanthin improved serum reproductive hormone levels (follicle-stimulating hormone, luteinizing hormone, and progesterone) ($P < 0.05$) with a maximum value observed in Ax60. Therefore, astaxanthin prevented ovarian aging by improving the antioxidant capacity of laying hens and promoting the production of reproductive hormones.

2.5 Effects on immune function of laying hens

Astaxanthin can protect the integrity of immune cells, ensure the normal immune response process, increase the quality of immune organs, and promote the secretion of cytokines (Kumar et al., 2022; Xu et al., 2022; Chen et al., 2020; Ihab, 2021)

It was found by Shevchenko et al(2021) that the addition of lycopene (20 mg/kg) and astaxanthin (10 mg/kg) for 30 days did not affect the hematological profile of laying hens. Increasing the content of lycopene to 40 and 60 mg/kg or astaxanthin to 20 or 30 mg/kg of feed for 30 days reduced the number of leukocytes and hemoglobin in the blood compared to the control.

In the study of Bai (2020), 120 one-day-age Hyline brown laying hens were randomly divided into four groups, and they were fed with 0.5%, 1.0%, and 1.5% diet of astaxanthin from the age of 6 days. The result showed that compared with the control group, the antibody level (humoral immune index), the percentage of T lymphocytes (cellular immune index) and the index of immune organs were significantly improved in the astaxanthin group ($P<0.05$). Astaxanthin can regulate the cellular and humoral immunity of the body and enhance the specific immune function.

III. Summary and Outlook

Astaxanthin plays a crucial role in the poultry industry due to its health-promoting effects, antioxidant properties, and distinctive pigmentation property. There are many studies on the application of astaxanthin in layer breeding, and most of them have shown positive effects. However, further research is needed on the mechanism of astaxanthin isomer accumulation in vivo, the biological activity evaluation of astaxanthin from different sources, and the deposition efficiency and mechanism of astaxanthin in egg yolk, which will favor more efficient utilization of astaxanthin in commercial feed for layer hens.

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