The role of antioxidants in reproduction and fertility of poultry

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For the majority of organisms on Earth, life without oxygen is impossible. Animals, plants and many microorganisms rely on oxygen for the efficient production of energy. However, the high oxygen concentration in the atmosphere is potentially toxic for living organisms. Recently free radical research has generated valuable information for further understanding not only detrimental, but also beneficial role of free radicals in cell signaling and other physiological processes. The benefit or harm of free radicals ultimately depend on the level of their production and efficiency of antioxidant defence. During evolution, living organisms have developed specific antioxidant protective mechanisms to deal with ROS. Therefore, the presence of natural antioxidants in living organisms is the major factor that enables their survival in an oxygen-rich environment. The protective antioxidant compounds are located in organelles, subcellular compartments or the extracellular space, enabling maximum cellular protection to occur. Natural antioxidants, including vitamin E, Se and carotenoids play important roles in avian reproduction by maintaining antioxidant defences of the spermatozoa and embryonic tissues. Optimal antioxidant supplementation is shown to be important to maintain high productive and reproductive performances of commercial poultry. Se is considered to be "chief-executive of the antioxidant defence system". Indeed, it is considered to be an integral part of at least 25 selenoproteins expressed in various tissues of human and animals. It has been proven that Se participates in regulation of major physiological functions in human and animals including growth, development, spermatogenesis and embryonic development. The most fascinating part of Se-related research is coming from understanding a principal difference between various Se sources in the diet. The digestive system of animals, including birds, adapted to metabolise organic Se from plant-based feedstuffs during evolution. Therefore, inclusion of selenite or selenate in the diet is not the 'natural' situation and differences in assimilation, distribution and accumulation of Se in tissues depend on source of Se. A great body of evidence indicates that replacement of sodium selenite in the poultry diet by organic Se, in the form of Sel-Plex, can help to meet the optimal Se requirement and to increase fertility and hatchability.

Key words: antioxidants; selenium; fertility; hatchability; poultry

Introduction

The animal body is under constant attack from free radicals, formed as a natural consequence of the body's normal metabolic activity and as part of the immune system's strategy for destroying invading microorganisms. It has been calculated that about 2 $x10^{10}$ molecules of reactive oxygen species (ROS) are generated per cell per day (Chance *et al*., 1979) In stress conditions this rate is substantially increased.

During evolution, living organisms have developed specific antioxidant protective mechanisms to deal with ROS. Therefore, the presence of natural antioxidants in living organisms is the major factor that enables their survival in an oxygen-rich environment. These mechanisms are described by the

general term “antioxidant system” (Surai, 2002). It is diverse and is responsible for the protection of cells from the actions of ROS. This system includes:

- natural fat-soluble antioxidants (vitamins A, E, carotenoids, ubiquinones, etc.);
- water-soluble antioxidants (ascorbic acid, uric acid, taurine, etc.)
- antioxidant enzymes: glutathione peroxidase (GSH-Px), catalase (CAT) and superoxide dismutase (SOD).
- thiol redox system consisting of the glutathione system (glutathione/glutathione reductase/glutaredoxin/glutathione peroxidase and a thioredoxin system (thioredoxin/thioredoxin peroxidase/thioredoxin reductase).

The protective antioxidant compounds are located in organelles, subcellular compartments or the extracellular space, enabling maximum cellular protection to occur. The antioxidant system of the living cell includes three major levels of defence.

The first level is based on the activity of superoxide dismutase (SOD), glutathione peroxidase (GSH-Px) and catalase which, together with metal-binding proteins, are responsible for prevention of free radical formation and keep this process under control. The second level of antioxidant defence is based on chain-breaking antioxidants (vitamins E, C, carotenoids, etc.) and is responsible for restriction of chain formation and propagation. The third level of defence is based on the activity of specific enzymes, responsible for repairing or removal of damaged molecules from the cell.

Effect of natural antioxidants on fertility

Chicken spermatozoa are unique in their structure and chemical composition. The most important feature of lipid composition of the avian semen is the extremely high proportions of long–chain polyunsaturated fatty acids (PUFAs) in the phospholipid fraction of spermatozoa. On the one hand, the high PUFA proportion is a necessity in order to maintain specific membrane properties (fluidity, flexibility, etc). On the other hand, spermatozoa became very susceptible to lipid peroxidation and, therefore, the antioxidant defence is considered to be a key element in maintaining semen quality. Vitamin E was discovered as a “vitamin of reproduction” in 1922. In recent years it has been shown that vitamin E is located in spermatozoa and provides antioxidant protection, especially in stress-conditions of in vitro semen manipulation, including dilution, storage and deep freezing of spermatozoa (see Surai, 2002 for a review). Furthermore, it was shown that vitamin E provides additional protection in the case of fatty acid manipulation of the semen (Surai et al., 2000; Zinini et al., 2003; Cerolini et al., 2005). However, in some studies the vitamin E dose-response in cockerels was shown to be non-linear (Lin et al., 2005).

Se supplementation is known to affect the antioxidant defenses of chicken semen (Surai et al., 1998). Furthermore, Edens (2002) showed that, when cockerels were fed on a basal diet containing 0.28 ppm Se without additional dietary supplementation of this trace element, the percentage of normal spermatozoa was only 57.9% and two major abnormalities seen were bent midpiece (18.7%) and corkscrew head (15.4%). When this diet was supplemented with an additional 0.2 ppm Se in the form of selenite, the percentage of normal spermatozoa increased to 89.4% and abnormalities in the form of bent midpiece and corkscrew head were decreased down to 6.2 and 1.8% respectively. However, when organic Se was included in the cockerel’s diet in the same amount, semen quality was further improved and those abnormalities decreased down to 0.7 and 0.2% and the percentage of normal spermatozoa increased up to 98.7%. These results clearly showed that the form of dietary Se supplementation is a crucial factor of its efficiency, with organic Se being much more effective in comparison to selenite. Therefore Se deficiency is associated with midpiece damage to spermatozoa It is clear that the midpiece of spermatozoa of the Se-deficient male is broken. In such conditions, sperm motility and fertilizing capacity would be compromised. Organic selenium can also improve fertility and, more importantly, increase the duration of fertility (Agate *et al*., 2000). Preliminary observations in female chickens have also revealed the effectiveness of dietary supplementation with vitamin E, organic Se or both, to sustain fertility in aging flocks (Breque *et al*., 2003). Thus avian spermatozoa might be expected to have systems which will maintain stability throughout this period. Indeed, recent results have confirmed the existence of a complex antioxidant system in the utero-

vaginal portion of the fowl oviduct (Breque and Brillard, 2002). In particular GSH-Px activity in the utero-vaginal junction was 12-fold higher than in the liver.

Effect of natural antioxidants on hatchability

The hatching process is considered to be a time of oxidative stress. Therefore, improved antioxidant defences during embryonic development potentially could increase hatchability. It was shown that vitamin E, carotenoids and Se can be transferred from the diet to the egg and consequently to the developing embryo (Surai, 2002). There are species-specific differences in vitamin E transfer from feed to the egg and, therefore, to the developing embryo. In particular, when chicken, turkey, goose and duck diets were supplemented with the same amount of vitamin E, the highest alpha-tocopherol concentration was found in chicken eggs and chicken embryonic tissues (Surai et al., 1998). Increased vitamin E concentration in the chicken embryonic tissues was associated with decreased tissue susceptibility to lipid peroxidation (Surai et al., 1999). Similarly, increased carotenoid concentration in the chicken embryo decreased the susceptibility of the tissues to lipid peroxidation (Surai and Speake, 1998; Surai et al., 2003). It seems likely that protective effect of vitamin E can be seen more clearly in stress condition, for example, in laying hens consuming T-2 toxin (Tobias et al., 1992) or in birds that consume vicine (from faba beans) (Muduili et al., 1982).

Se transfer from the diet to the egg and then on to the developing embryo has, so far, received only limited attention. However, it is proven that it is only organic Se that is effectively transferred, since the major form of Se in the egg is SeMet, but the chicken cannot synthesise this compound. Our data indicate that Se in the maternal diet affects Se concentration in tissues of the postnatal quail. Indeed, when newly hatched quail from Se-enriched eggs and normal quail eggs were placed on low Se-diet (0.1 ppm), the Se concentration in tissues dropped dramatically for the first 2 weeks posthatch (Surai et al., 2006). This finding suggests that Se in the liver of newly hatched quail is rapidly diluted by the post hatch growth of the tissue unless the diet of the neonate is supplemented with this element.. It is possible to suggest that Se absorption from the diet is not optimal during the first few days of life and the chick must rely on reserves of the element accumulated during embryogenesis. However, the difference in tissue Se concentrations between the control and experimental groups was still significant at 2 weeks posthatch. These results clearly indicate that the maternal diet affects not only newly hatched quails, but also has a sustained effect on the chicks during postnatal development.

Recently a new experiment was conducted at SAC to address this question (Pappas et al. 2005). The maternal diet was supplemented with 0.4 ppm organic Se in the form of Sel-Plex and comparison was made with a basic diet containing 0.1 ppm feed-derived Se. As a result of dietary Se supplementation the Se concentration in the egg yolk, albumin, shell, shell membrane and perivitelline membrane was significantly increased. The newly hatched chicks were placed on a basal diet (0.1 ppm) without Se supplementation for the next 4 weeks posthatch. After hatching, chickens fed diets low in Se (0.1 ppm), but originating from parents fed diets high in Se (0.5 ppm), had, up to 4 weeks post-hatch, significantly higher blood Se levels than those that originated from parents fed diets low in Se (0.1 ppm). It seems likely that Se inadequacy is more often observed in commercial breeders, since Se supplementation in the organic form is shown to positively affect hatchability (Sefton and Edens, 2004; Edens and Sefton, 2003; Sefton and Edens, 2004a; Renema and Sefton, 2004; Renema, 2004; Renema. 2003).

Conclusions

Natural antioxidants, including vitamin E, Se and carotenoids play important roles in avian reproduction by maintaining antioxidant defenses of the spermatozoa and embryonic tissues. Optimal antioxidant supplementation is shown to be important to maintain high productive and reproductive performances of commercial poultry. Replacement of sodium selenite in the poultry diet by organic Se, in the form of Sel-Plex, can help to meet the optimal Se requirement and to increase fertility and hatchability.

Data are actively accumulating to indicate the possibility of maternal programming at early stages of foetal development (Maloney and Rees, 2005). Although there are still controversial areas, there is at present sufficient scientific evidence for foetal programming to be regarded as an additional risk factor for chronic disease, in interaction with genetic and lifestyle risk factors (Haimov-Kochman, 2005). The available evidence suggests that nutrient sensing regulatory systems are present in many tissues during early development (Maloney and Rees, 2005). Programming agents seem to include growth factors, cytokines and hormones, all of which can be altered by stress. As a consequence, such 'stress-modified' systems of the offspring are more susceptible to environmental influences during later life, e.g. the development of atopic diseases upon exposure to antigens (Knackstedt *et al*., 2005). Therefore, oxidative stress is involved in programming and oxidative stress may be a common link underlying the superficial "programming" associations between adverse foetal growth or preterm birth and elevated risks of certain chronic diseases. The mechanisms of oxidative stress programming may be through the direct modulating of gene expression or, indirectly, through the effects of certain oxidized molecules (Luo *et al*., 2006; Langley-Evans and Sculley, 2005). Taking into account that Se can affect concentration of the above-mentioned programming agents, it could well be that Se plays an important role in the foetal programming. Whether maternal programming exists in birds is not known at present but it is reasonable to suggest that egg composition, including Se, vitamin E and carotenoid concentrations, could potentially affect gene expression in the developing embryo and thereby be a base for maternal programming. This could result in long-term consequences for the developing chicken. Clearly, this area needs further investigations.

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