



Effect of dietary supplementation of vitamin E on production performance and some biochemical characteristics of cloacal foam in male Japanese quail



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ABSTRACT

This experiment was conducted to investigate the effects of increasing the level of dietary supplementation of vitamin E (VE) on production performance and biochemical characteristics of cloacal foam in male Japanese quail (*Coturnix coturnix japonica*).

A total of 225 male Japanese quail chicks (day old) were randomly distributed to three dietary treatments for a period of 30 weeks. Each treatment comprised of three replicates, each containing 25 chicks. The basal diet (T_1) contained 12.30 IU VE kg^{-1} and the two experimental diets were supplemented with 150 and 300 IU VE kg^{-1} (diets T_2 and T_3 , respectively). DL- α -Tocopherol acetate was used as the source of VE. All chicks were provided feed and water *ad libitum*.

Mean body weights, feed intake, feed conversion ratio (FCR) and mortality of the birds in the different treatment groups showed no significant differences ($P > 0.05$), whereas a significant ($P < 0.05$) increase (29.81 and 50.83%) in average foam weight was evident in the VE-treated groups (T_2) compared with control (T_1) and T_3 groups. The biochemical characteristics of foam, in terms of quantities of protein and nitric oxide (NO), did not differ significantly ($P > 0.05$), whereas the quantities of glucose (60.01%) and acid phosphatase (ACP, 32.46%) were significantly ($P < 0.05$) higher in the T_3 group. By contrast, the quantities of alkaline phosphatase (ALP), glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT) were significantly ($P < 0.05$) lower (48.84%, 10.38% and 22.08%, respectively) in the T_3 group and higher in the T_1 (control) and T_2 groups.

From this study, it can be concluded that dietary supplementation of VE to the basal diet has no effect on the production performance but supplementation of a higher level of VE (300 IU kg^{-1} diet) improved the biochemical characteristics of the foam and moderate levels of VE (150 IU kg^{-1} diet) improved the foam production of male Japanese quail.

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1. Introduction

The male Japanese quail (*Coturnix coturnix japonica*) produces thick foam from the proctodeal gland in the dorsal wall of the cloaca (Mohan et al., 2002). This gland is described as an 'aggregate gland' composed numerous

discrete glandular units. Both sexes of quail have the gland but functional development is seen only in sexually active male birds (Seiwert and Adkins-Regan, 1998), where the gland is enlarged and there is a high degree of correlation between gland size and testes weight and sexual activity (Mohan et al., 2002; Biswas et al., 2007). Over three decades, the physiological functions of cloacal gland foam on quail have been a subject of conflict. Mohan et al. (2002) and Fujihara et al. (1989a) reported that the foam might be involved in enhancing the

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fertilising ability of quail spermatozoa; however, fertility results were observed when semen was placed in the vagina with and without foam (Biswas et al., 2007; Ayyagari, 1982). Therefore, measurement of the cloacal gland may provide a valuable non-invasive method of predicting testicular activity and subsequently the fertilising ability of male quail. In addition, the appearance of foam in the gland indicates sexual maturity in young birds (Mohan et al., 2002). The cloacal gland secretes meringue-like foam constantly during copulation and defecation time (Fujihara, 1992). Foam is an inherent component of naturally ejaculated quail semen (Cheng et al., 1985). Foam avoids excessive loss of spermatozoa during oviposition and enables the sperm to be released slowly for better fertilisation (Mohan and Moudgal, 1996). The presence and absence (surgical removal) of the cloacal gland did not affect fertility in natural mating in quails (Singh et al., 2008). The motility of quail spermatozoa was enhanced when they came in contact with foam (Fujihara, 1992; Fujihara et al., 1989b; Cheng et al., 1989a,b). There is lack of available literature about the biochemical composition of foam. Singh et al. (2011) reported that cloacal foam extract contains some biochemical components, that is, protein, transaminase and phosphatase, which are responsible for sperm motility. Mohan and Moudgal (1996) revealed the presence of sulphated and non-sulphated mucopolysaccharide secretion of the cloacal gland during histological examination. Cloacal foam is thermostable and the protein components were almost comparable with those in blood serum (Ratnamohan, 1985). Moudgal and Mohan (1991) revealed that cloacal foam extracts can immobilise chicken spermatozoa in a reversible manner. The increased motility of sperm for longer duration when mixed with foam indicates the presence of some non-sugar component which may serve as an energy source. Furthermore, glucose, if secreted by the cloacal gland, may be completely used by the bacteria present in foam. Mohan et al. (2004) reported that *Escherichia coli* are the major bacteria present in the foam, which play an important role in foam production.

Vitamin E (VE) is beneficial to avian species and often necessary for optimum production, reproduction cell integrity and immunity. A delicate antioxidant/pro-oxidant balance in the body is an important determinant of chicken health, sperm quality and reproductive characteristics. VE is widely used in poultry diets and the levels of its supplementation have been increased several fold during the last few years (Etches, 1996). Antioxidants increase the foam quality and fertilising ability of spermatozoa by protecting them against oxidative reactions (Biswas et al., 2009; Surai and Ionov, 1992). VE is considered very important for regulation of foam quality in poultry (Surai, 1999). High contents of VE, well above the standard recommendations, are needed to inhibit lipid peroxidation in spermatozoa (Surai, 1991). There is no available literature regarding the exact role of VE in the cloacal foam and its biochemical characteristics.

The studies of Biswas et al. (2007, 2008, 2009) in which quails and chickens received dietary supplementation of VE well above Nutritional Research Council (NRC, 1994) requirements indicate that greater levels of certain vitamins may be required during critical periods of

embryonic development to enhance embryo survival. Thus, the objective of this study, conducted at the physiology and reproduction division of Central Avian Research Institute, Izatnagar, India, was to examine the effect of two higher levels of dietary supplementation of VE in diets on production performance and biochemical characteristics of cloacal foam in the male Japanese quail.

2. Materials and methods

2.1. Housing and rearing of birds

A total of 225 day-old male Japanese quail chicks were randomly divided into nine groups of 25 chickens each (three dietary treatments $\times$ three replicates). The experiment had a randomised design (Snedecor and Cochran, 1994). Chicks were placed in electrically heated battery brooders with a wire mesh floor and reared under uniform husbandry conditions (14 h light/day and 25–32 °C) and provided with a standard basal diet. The same technicians provided feed and water and collected data from the birds during the course of the experiment (up to 30 weeks). The experiment followed the guidelines of 'Institutional Animal Ethics Committee' (IAEC, CARI, Izatnagar).

2.2. Formulation of experimental diets

The basal diet (T_1) was formulated according to the NRC requirements for Japanese quail (Table 1). Two experimental diets T_2 and T_3 were formulated to contain an additional dietary supplementation of 150 and 300 IU VE kg^{-1} diet, respectively. DL- α -Tocopherol acetate was used as the source of VE.

2.3. Production performance

The body weight of each bird, feed intake of each group (10 birds/replicate, i.e., 30 birds/treated group), feed conversion ratio (FCR) and mortality were recorded weekly up till the end of the experiment.

2.4. Collection of foam and extraction

Foam was collected from 25 male birds from each treated group up to the 30th week (from each treatment group) by gently squeezing the cloacal gland on either side with fingers and the thumb. The amount of foam production from each bird was measured in the morning (10.00 h) and night (22.00 h) at 6-week intervals in different dietary groups. After the collection, the foam was stored in air-tight glass bottles to prevent evaporation (Mohan et al., 2002). Subsequently, the foam produced by each group of male birds was pooled and its weight was determined immediately using the electronic analytical balance to the nearest 0.01 mg. After weighing the foam, an extract was prepared using the following procedure. Prepare 20% (w/v) homogenate of foam using normal saline (0.89%). Then, centrifuge the contents at 35,000 rpm in an ultracentrifuge (Himac CP80 β) for 45 min. Collect the supernatant and store at –80 °C until further biochemical examination.

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