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# Dietary Supplement of Vitamin E Enhanced Fertility and Hatchability of Arbor Acres Eggs in Ibadan, Nigeria

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## Abstract

Broiler breeder stocks are characterised by low egg fertility and hatchability which could be mitigated by dietary Vitamin E (VE) supplements. The optimum vitamin E requirements for efficient production by Arbor Acres plus broiler breeder hens in Ibadan were investigated in this study. Arbor Acres plus broiler breeder hens (n=180) weighing 3.85 ± 0.49 kg, aged 30 weeks were randomly allotted to diets supplemented with synthetic VE at 0, 20, 40, 60, 80, 100 IU/kg for eight weeks in a completely randomised design. Hatchability of 86.4 ± 1.9% in hens fed diets supplemented with 100IU was significantly lower ($p<0.05$) than in those on 0 (93.3 ± 1.8%), 20 (92.9 ± 2.0%), 40 (96.9+1.5%), 60 (93.2 ± 0.9%) and 80 IU (91.2 ± 1.7%). Weight of 42.9 ± 1.3 g in chicks on 40 IU was significantly higher ($p<0.05$) than 36.9 ± 1.5 g (0IU), 40.2 ± 1.3g (20IU), 39.1 ± 1.2 g (60IU), 40.0 ± 0.6 g (80IU) and 39.3 ± 0.6 g (100IU). The relationships between VE inclusions and hatchability ($R^2$=0.63); and chick weights ($R^2$=0.56) were quadratic and cubic, respectively. Optimal egg hatchability of 94.8% and chicks' weight of 41 g were achieved with diets supplemented with 37 IU/kg and 24 IU/kg supplemental VE inclusions, respectively. Thus, the dietary supplement of vitamin E enhanced the fertility and hatchability of Arbor Acres plus breeder broiler hens in Ibadan, Nigeria.

**Keywords:** Supplemental vitamin • Arbor Acres plus • Broiler breeder hens • Chickens • Fertility • Hatchability

## Introduction

Fertility and hatchability are the major reproductive indices which are both affected by genetic and non-genetic influences [1]. One non-genetic influence that affects hatchability is the nutritional requirement of the developing embryo. Inadequately stored nutrients in eggs can result in embryonic mortality which causes low hatchability. Broiler breeder diets also impact on consequent egg production and embryogenesis. Apart from the genetic component of the hen, maternal nutrition is the next most important contributor to the performance of offspring. It was observed [2,3] that nutrition affects the immuno-competence of chickens especially, trace nutrients such as fat soluble vitamins like Vitamin E (VE) which cannot be synthesized by poultry species hence, the need to have it appropriately supplemented in the diet.

The chemical structure of a compound with VE activities has been aptly documented [4], as a strong lipid soluble antioxidant which illustrates two compounds: tocopherols and tocotrienols. Tocopherols and tocotrienols have four isomers each i.e., alpha, beta, gamma and delta. All arrangements of VE contain a similar chroman ring and a side chain which determines the type of vitamin E. Tocopherols do not contain double bonds while tocotrienols are unsaturated due to double bonds along the carbon skeleton. They are synthesized by plants and are present in high quantities in dietary oils, nuts, cereals and leafy vegetables [5]. However, the synthetic forms are also available as dl-α-tocopherol which is produced by coupling trimethy-hydroquinone with isophytol.

The VE has been reported to be an exceptional inhibitor which prevents the propagation of free radicals thereby protecting cells and tissues from destruction prompted by free radicals. This can be achieved by donating $H^+$ from the hydroxy group on the ring structure to the free radical, making them unreactive. They are particularly useful in preventing the oxidation of Polyunsaturated Fatty Acids (PUFA) present in high concentrations in eggs [6]. They are usually at the water-lipid interface in the membrane structure hence their ability to scavenge for free radicals. Free radicals are Reactive Oxygen Atoms (ROS) or Nitrogen Reactive Atoms (NRS) which are positioned in the outer trajectory of an unpaired electron. They perform physiological roles which include defending the body against infectious diseases and initiation of mitogenic responses. However, when present at excessive levels, they can cause peroxidative damage to the cells [7].

Vitamin E absorption from the intestine has been reported [8] to be dependent on sufficient pancreatic function, bile secretion and micelle formation. The digestion is similar to those of dietary lipids which are by efficient emulsification, proper solubilisation by the bile salt to form micelles, uptake by enterocytes and secretion into the circulatory system *via* the lymphatic pathway. The micelles aggregate the VE molecules, solubilise and transport them through the brush border membrane of the enterocytes through diffusion. The tocopherols are then incorporated into chylomicrons within the enterocytes and released into the intracellular cavity and lymphatic system then successively into the circulatory system where they are transported by

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high and low densities lipoproteins and red blood cells to the liver. Although the routes of uptake of all tocopherol homologues in the diet are similar, $\alpha$-tocopherol forms are predominant in the blood and tissues due to the action of the binding protein that preferentially selects $\alpha$-tocopherol over others [8].

The VE is reported to be essential for the fertility and hatchability of breeder eggs [9]. A unit increase in the fertility and/or hatchability of total eggs (primary productivity criteria) converts into a great financial value. The report from a previous study ascertained that an additional concentration of VE in the diets of male poultry would elevate the $\alpha$-tocopherol composition of the semen thereby preventing lipid peroxidative damage of the spermatozoa [10]. However, the dietary supplement of VE to improve the productivity of Arbor Acres plus broiler breeder hen has not been adequately documented. Therefore, this study was aimed at assessing the effect of supplemental VE in the diets on fertility and hatchability of Arbor Acres broiler breeder plus chickens.

# Materials and Methods

## Experimental site

The research was conducted at the Poultry Unit, Teaching and Research Farm, University of Ibadan, Ibadan, Nigeria, located in the tropical rain forest zone of Nigeria within the latitude 7° 26 N and longitude 3° 54 E with a mean altitude of 277 meters above the sea level. The location has been clearly described earlier [11].

## Experimental animals and management

Arbor Acres plus broiler breeder hens (n=180) at week 30 of age, and 18 breeder cocks with standard husbandry records were procured from a reputable farm. The hens were allocated to six dietary treatments. Each treatment was replicated five times with six birds per replicate using Experimental Animal Allotment Program EAAP as described [12]. The cocks were not allotted. Both the hens and cock were confined in a 3-layer battery. Each compartment had a dimension of 50 × 45 × 40 $cm^3$ and a floor measuring 900 $cm^3$ that housed two chickens. Both the cocks and hens were weighed at the inception and weekly throughout the duration of the experiment. Regular activities such as vaccination and drug administration were followed as prescribed [13] while diets and water were provided according to the specification of the Arbor Acres plus broiler breeder production manual. The study lasted eight weeks and the experimental layout of the study is shown in Table 1. Routine care and experimental protocols used were approved by the Department of Animal Science, University of Ibadan, Nigeria, and conformed to published guidelines for the ethical conduct and reporting of animal research [14,15].

## Gross composition of experimental diets

The experimental diets of the Arbor Acres plus broiler breeders were calculated to attune with the nutritional requirement of the chickens as stated in the breeder manual of Aviagens for Arbor Acres plus broiler breeder [13]. Test ingredients were added as overage to the feed and the dietary layout of the experiment is presented in Table 2.

## Determination of egg produced, egg set, transferred, hatched, fertility and hatchability percentage

The egg produced was determined by pooling together eggs laid by the hens per replicate per week which were stored at a prescribed temperature of 16 °C [16]. The egg set was determined by physically observing the eggs to separate those that were not settable such as those with cracks, too big for the setter crates and those with too soft or hard shells. The eggs transferred were obtained by candling the eggs at day 18 of incubation by placing them in a candling machine to separate the fertile eggs from the infertile ones. The hatches were determined by separating the hatched chicks from the unhatched eggs. Fertility and hatchability were calculated according to the formulae below:

$$Fertility = \frac{Number\ of\ eggs\ transferred}{Number\ of\ egg\ set} x\ 100 \qquad (1)$$

**Table 1.** Layout of broiler breeder hens fed varying dietary inclusion of vitamin E.

| Diet | Inclusion Level of $\alpha$-tocopherol (IU/kg) |
|---|---|
| Treatment 1 | Basal diet |
| Treatment 2 | Basal diet+20 |
| Treatment 3 | Basal diet+40 |
| Treatment 4 | Basal diet+60 |
| Treatment 5 | Basal diet+80 |
| Treatment 6 | Basal diet+100 |

**Table 2.** Gross composition (g/100g) of diets fed to Arbor Acres broiler breeder hen.

| Ingredient | 0 IU/kg | 20 IU/kg | 40 IU/kg | 60 IU/kg | 80 IU/kg | 100 IU/kg |
|---|---|---|---|---|---|---|
| Corn | 65.42 | 65.42 | 65.42 | 65.42 | 65.42 | 65.42 |
| SBM | 21.15 | 21.15 | 21.15 | 21.15 | 21.15 | 21.15 |
| Wheat offal | 5.63 | 5.63 | 5.63 | 5.63 | 5.63 | 5.63 |
| Limestone | 8.22 | 8.22 | 8.22 | 8.22 | 8.22 | 8.22 |
| DCP | 1.64 | 1.64 | 1.64 | 1.64 | 1.64 | 1.64 |
| Salt | 0.33 | 0.33 | 0.33 | 0.33 | 0.33 | 0.33 |
| vitamin-mineral premix | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| Methionine | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 |
| Lysine | 0.007 | 0.007 | 0.007 | 0.007 | 0.007 | 0.007 |
| $\alpha$-Tocopherol ($10^{-3}$) | - | 1.8 | 3.6 | 5.4 | 7.2 | 9 |
| Metabolisable energy (kcal/kg) | 2828 | 2828 | 2828 | 2828 | 2828 | 2828 |
| Crude protein (%) | 15.48 | 15.48 | 15.48 | 15.48 | 15.48 | 15.48 |
| Phosphorus (%) | 0.4 | 0.4 | 0.4 | 0.4 | 0.4 | 0.4 |
| Calcium (%) | 3.2 | 3.2 | 3.2 | 3.2 | 3.2 | 3.2 |

SBM: Soya Bean Meal, DCP: Di Calcium Phosphate means with different superscripts are significantly different (P<0.05) SEM: Standard Error of Means, VIT A 10 000IU, VIT D3 2500IU, VIT E 50 IU, VIT k3 2.5 mg, VIT B1 2.5 mg, VIT B2 8 mg, VIT B6 0.015 mg, Nicotinic Acid 45 mg, Pantothenic acid 15 mg Choline 1400 mg Biotin 0.2 mg, Folic acid 1.5 mg, Cu 8 mg, Fe 80 mg, Zn 70 mg, Mn 95 mg, Se 0.18 mg, I 0.35 mg

$$Hatchability = \frac{Number\ of\ hatched\ chicks}{Number\ of\ egg\ transferred} x\ 100 \qquad (2)$$

## Determination of chicks' weight and relative organ weights of chicks

On day 18 of egg incubation using Chicksmaster incubator model 99, the eggs were candled, and the fertile ones were separated. Subsequently, eggs were transferred to the Chicksmaster model 90 L hatcher for 3-days till hatching at day 21. Hatched chicks were counted, and weighed. One chick per replicate per week was selected, sacrificed and properly dissected to determine chicks' organ weights. The heart, liver and yolk sac weights were determined using a sensitive digital scale, (Weigh-gram, model number 8541976147).

$$Relative\ organ\ weight = \frac{weight\ of\ organ}{Chick\ weight} x\ 100 \ldots \qquad (3)$$

## Experimental design and dietary layout

The experimental design was a completely randomised design with details presented in Tables 1 and 2.

**Statistical model**

$$Yij = \mu + \tau i + \in ij \qquad (4)$$

Where

μ= common effect of the experiment

Yij= j-th observation on the i-th treatment

$\tau i = the\ i-th\ treatment\ effect$

$\in ij = random\ error\ present\ in\ the\ j-th\ observation\ in\ the\ i-th\ treatment$

# Results

The production efficiency of broiler breeder hen offered graded inclusion of dietary supplement of VE is presented in Table 3. Significantly higher ($p<0.05$) egg production (34.97 ± 0.89) was observed in hens fed 40 IU/kg which was noticeably greater ($p<0.05$) than obtained in other dietary group. The treatment supplemented with 100 IU/kg (13.07 ± 0.61) had the least ($p<0.05$) egg produced. The treatments supplemented with 60 IU/kg (22.97 ± 1.42) and those without VE supplementation, 0 IU/kg (22.23 ± 1.11) were of similar ($p<0.05$) production.

The highest number of egg set ($p<0.05$) was attained in group fed ration containing 40 IU/kg (34.27 ± 0.80) while those supplemented with 100 IU/kg (12.60 ± 0.53) was the lowest ($p<0.05$). The treatment offered 60 IU/kg in diet (22.50 ± 1.33) was similar ($p<0.05$) to those without VE supplementation, 0 IU/kg (21.60 ± 1.36). The egg transferred to the hatcher from the setter significantly varied ($p<0.05$) from the highest at 40 IU/kg (33.10 ± 0.92) to the lowest at 100 IU/kg (11.43 ± 0.25). The highest mean hatched chicks per treatment ($p<0.05$) was obtained at 40 IU/kg (32.10 ± 1.10) followed by 20 IU/kg (23.60 ± 0.73). The number of chicks hatched in treatment with 60 IU/kg (19.50 ± 1.30) supplemental VE was noticeably greater ($p<0.05$) than was obtained in treatment without any VE supplementation, 0 IU/kg (18.10 ± 0.92). The treatment supplemented with 100 IU/kg (9.9 ± 0.10) had the least hatched chicks.

The fertility obtained in treatment supplemented with 40 IU/kg (96.57 ± 0.80) was noticeably higher ($p<0.05$) comparable to other groups. The least fertility ($p<0.05$) was obtained in the chickens fed diets without supplementation, 0 IU/kg (90.01 ± 2.29) and those supplemented with 100 IU/kg (90.87 ± 2.40). However, those supplemented with 20 IU/kg (93.79 ± 0.59) was similar ($p<0.05$) to those on 60 IU/kg (93.91 ± 2.30) supplemental VE.

The hatchability of eggs from chickens on the treatment supplemented with 40 IU/kg (96.95 ± 1.50) was also significantly more ($p<0.05$) than in other treatments. However, the hatchability of those from other treatments was the same ($p>0.05$) except for the group offered ration containing 100 IU/kg (86.38 ± 1.90) which had the lowest hatchability ($P<0.05$).

## Relationships between egg hatchability and dietary supplement of vitamin E

The relationship between egg hatchability and supplemental vitamin E is presented in Figure 1. The hatchability of the eggs ranged from 86.38% to 96.95% but the optimum hatchability of 94.80% was obtained at 37 IU/kg inclusion of VE in the diet of broiler breeder hens. The relationship was significant ($p<0.05$), positive and quadratic. The relationship was represented by the equation below:

$$y = -0.0018x^2 + 0.1215x + 92.836 \quad (5)$$

$$(R^2 = 0.6251)$$

The equation above showed that 62% of the observed improvement in hatchability was adduced or attributed to dietary supplemental of $\alpha$-tocopherol.

The effect of varying inclusions levels of supplemental $\alpha$-tocopherol on relative organ weights of hatched chicks is shown in Table 4. The chicks fed diets without supplemental VE, (36.90 ± 1.57) had reduced ($p<0.05$) body weight compared with other groups while those with the higher chicks' weight ($p<0.05$) were those who fed on diets supplemented with 40 IU/kg (42.93 ± 1.30). All chicks in other treatment group had similar ($p>0.05$) body weights.

The heart weight of the chicks in the treatment supplemented with 40 IU/kg (0.83 ± 0.03) was significantly higher ($p<0.05$) than those without VE supplementation, while those on 0 IU/kg (0.67 ± 0.04) supplemental had the least ($p<0.05$) heart weight, followed by those supplemented with 20 IU/kg (0.72 ± 0.04) supplemental VE. The heart weight of the chicks on treatments supplemented with 80 IU/kg (0.77 ± 0.04) and 100 IU/kg (0.75 ± 0.04) was similar ($p>0.05$).

The liver of the chicks without VE supplementation, 0 IU/kg (2.72 ± 0.10) and those supplemented with 20 IU/kg (2.69 ± 0.1) were similar ($p>0.05$) but significantly lower ($p<0.05$) than in other treatment groups. Those supplemented with 40 IU/kg (3.26 ± 0.1), 60 IU/kg (3.12 ± 0.1), 80 IU/kg (3.18 ± 0.1) and 100 IU/kg (3.22 ± 0.1) showed no significant variation ($p>0.05$). The relative weights of yolk sac of the various treatment group on varying supplemental inclusion levels were similar ($p>0.05$). The values varied from 5.92 ± 0.2 in 60 IU/kg supplemental VE to 6.35 ± 0.3 in those without supplemental VE.

The relationship between supplemental vitamin E and the body weights of hatched one day old chicks is illustrated in Figure 2. The relationship was cubic, positive and significant ($p<0.05$). The optimal weight was achieved with 24 IU/kg dietary supplement of VE. The relationship is summarised in equation 6 below:

$$y = 3E-05x^3 - 0.0062x^2 + 0.3162x + 36.78 \quad (6)$$

$$R^2 = 0.564$$

# Discussion

## Production efficiency of hens

Nutrients required for normal embryonic development are known to be derived from the maternal diets [17]. In the present study, dietary VE inclusion of up to 40 IU/kg resulted in improved egg production compared to other treatment groups. This was contrary to the report of Jiang YH, et al. that no significant variation in egg production of hens fed diets supplemented with 50, 100, 200, and 400 mg VE [18]. Conversely, Bollengier-Lee observed increased egg production when laying hen diets were supplemented with vitamin E during heat stress [19]. Similarly, Kirunda DF, et al. observed an increment in the hen day egg production of commercial layers supplemented with VE concentration of up to 60-65 mg/kg in feed [20]. The improved egg production observed in this study could be ascribed to VE capability in facilitating the release of vitellogenin and Very Low Density Lipoprotein (VLDLP) from liver to the blood streams [19]. The same trend was also observed for the egg set and egg transferred in this study. It was deduced that treatment with 40 IU/kg supplemental VE had significantly higher egg set and egg transferred compared with those on other treatments which may be connected to the reported lipo-peroxidative ability of VE in preventing tissue damage of the developing embryo [21]. This study did not find out some of the causes of embryonic mortality which might have resulted in decreased egg transferred to the hatcher for other treatment group, some of the reported vitamin E deficiency include abnormal vascular system and stunting of the embryo [22].

**Table 3.** Efficiency of production of broiler breeder hens fed varying levels of dietary supplement of vitamin E.

| Treatments (IU/kg) | Egg Produced | Egg Set | Transferred | Hatched | Fertility % | Hatchability % |
|---|---|---|---|---|---|---|
| 0 | 22.23+1.11$^c$ | 21.60+1.36$^c$ | 19.40+0.89$^d$ | 18.10+0.92$^d$ | 90.01+2.29$^c$ | 93.29+1.79$^b$ |
| 20 | 27.07+2.06$^b$ | 24.87+0.55$^b$ | 25.40+0.53$^b$ | 23.60+0.73$^b$ | 93.79+0.59$^b$ | 92.89+2.01$^b$ |
| 40 | 34.97+0.89$^a$ | 34.27+0.80$^a$ | 33.10+0.92$^a$ | 32.10+1.1$^a$ | 96.57+0.8$^a$ | 96.95+1.5$^a$ |
| 60 | 22.97+1.42$^c$ | 22.50+1.33$^c$ | 21.13+1.25$^c$ | 19.50+1.3$^c$ | 93.91+2.3$^b$ | 93.16+0.9$^b$ |
| 80 | 17.40+0.57$^d$ | 17.00+0.51$^d$ | 15.70+0.48$^e$ | 14.33+0.5$^e$ | 92.39+1.9$^c$ | 91.24+1.7$^b$ |
| 100 | 13.07+0.61$^e$ | 12.60+0.53$^e$ | 11.43+0.25$^f$ | 9.90+0.1$^f$ | 90.87+2.4$^c$ | 86.38+1.9$^c$ |
| SEM | 0.95 | 0.88 | 0.54 | 0.79 | 0.81 | 0.58 |

$^{abcd}$Means with different superscripts along the same column are significantly different ($P<0.05$), SEM: Standard Error of Mean

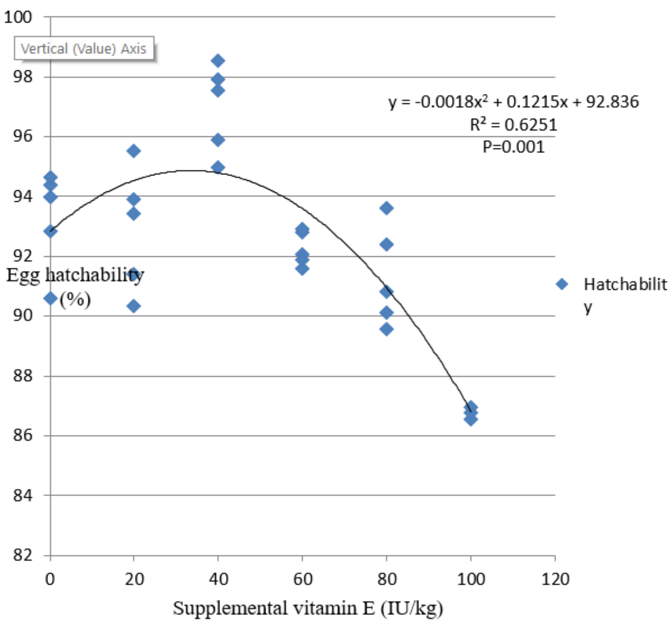


**Figure 1.** Relationship between egg hatchability and supplemental vitamin E.

**Table 4.** Effect of varying levels of α-tocopherol on relative organ weights of hatched chicks.

| Treatment (IU/kg) | Weight (g) | Heart | Liver | Yolk Sac |
|---|---|---|---|---|
| 0 | 36.90 ± 1.57$^{c}$ | 0.67 ± 0.04$^{d}$ | 2.72 ± 0.1$^{b}$ | 6.35 ± 0.3 |
| 20 | 40.23 ± 1.3$^{b}$ | 0.72 ± 0.04$^{c}$ | 2.69 ± 0.1$^{b}$ | 6.30 ± 0.5 |
| 40 | 42.93 ± 1.3$^{a}$ | 0.83 ± 0.03$^{a}$ | 3.26 ± 0.1$^{a}$ | 6.17 ± 0.4 |
| 60 | 39.07 ± 1.2$^{b}$ | 0.78 ± 0.05$^{ab}$ | 3.12 ± 0.1$^{a}$ | 5.92 ± 0.2 |
| 80 | 40.00 ± 0.6$^{b}$ | 0.77 ± 0.04$^{bc}$ | 3.18 ± 0.1$^{a}$ | 6.21 ± 0.1 |
| 100 | 39.27 ± 0.6$^{b}$ | 0.75 ± 0.04$^{bc}$ | 3.22 ± 0.2$^{a}$ | 6.34 ± 0.1 |
| SEM | 0.38 | 0.01 | 0.05 | 0.06 |

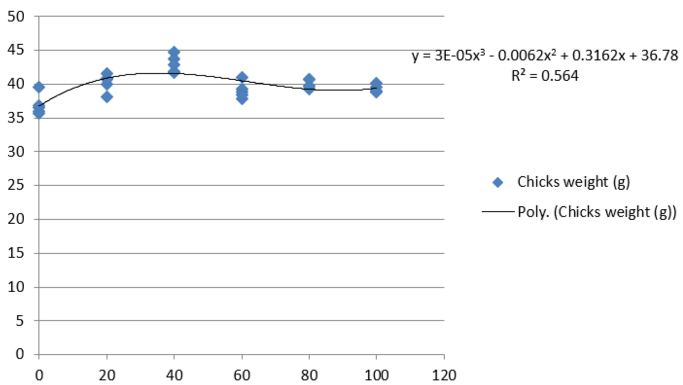


**Figure 2.** Relationship between dietary vitamin E supplement and one day old chicks' weight.

The report on hatchability in this study aligned with the findings of Romanoff AL and Romanoff AJ on feed supplementation with graded levels of VE kg$^{-1}$ for Ross breeder [23]. They reported that those on 50 mg/kg VE produced the greatest number of fertile eggs and chicks at week 40 of chickens' age. However, Hossain SM, et al. observed no significant difference on hatchability of Taiwan native chickens when VE was supplemented [24]. Leeson S and Summers JD made a recommendation of 20-25 mg of VE/kg for inclusion in breeders' broiler feed [25]. The report above showed that newer broiler breeders strain requires additional VE supplementation. Therefore, optimum inclusion of 37 IU kg$^{-1}$ supplemental VE in the diets of Arbor Acres plus broiler breeder was recommended for improved hatchability in this study.

### Relative weights of chicks organ

The varied dietary supplementation of α-tocopherol for Arbor Acres breeder hens significantly influenced the relative weights of organ of hatched chicks. Higher body, heart and liver weights were obtained from chicks of hens on 40 IU/kg supplemental α-tocopherol. The higher body weight observed in the chicks may be due to the low lipoperoxidative damage to the yolk during the process of incubation. The α-tocopherol being a fat soluble vitamin is deposited mainly in the yolk compartment of the egg for embryonic development. This observation conformed with the work of Surai PF, et al. that dietary VE derived from the ration of the hen are deposited in the egg and subsequently to embryonic tissue before being utilised by the chicks [26]. Selim S, et al. observed a higher body weight in ducklings produced from eggs inoculated in-ovo with VE [27]. Although, authors observed a declined relative heart and liver weights of the ducklings. According to Kling LJ and Soares JH, VE is important for hatchability [28]. The VE requirement of breeder hens is usually assessed based on egg production and hatchability alongside other requirements (such as physiological requirements) in consideration, this assertion may be contrary to an earlier report by Tengerdy RP, et al. [29]. However, there was no specific VE requirement for broiler breeders as the required amount of 10 mg kg$^{-1}$ in the diet had been documented for the layer breeders on a daily ration consumption of 100 g/hen/day [30].

## Conclusion

Supplemental VE for the Arbor Acres plus breeder broiler hens improved fertility and eggs hatchability in Ibadan, Nigeria. Optimum hatchability of 94.80% was obtained at 37 IU/kg inclusion of dietary tocopherol. The optimum chicks' weight of 41grams was achieved with 24 IU/kg dietary inclusion level of supplemental VE for the breeding broiler chickens

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## Conflicts of Interest

The authors declare no conflict of interest.

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