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EFFECT OF DIFFERENT DIETARY LEVELS OF INORGANIC ZINC OXIDE ON OVARIAN ACTIVITIES, REPRODUCTIVE PERFORMANCE OF EGYPTIAN BALADI EWES AND GROWTH OF THEIR LAMBS

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Summary

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The experiment was conducted on 48 Baladi ewes divided into four groups of 12 ewes each. Group 1 was kept as a control and was fed the basal diet consisting of roughage and concentrate mixture. Group 2 (ZnO 50 ppm) was fed the basal diet supplemented with 50 ppm zinc oxide. Group 3 (ZnO 100 ppm) and group 4 (ZnO 150 ppm) received the same basal diet supplemented with 100 and 150 ppm zinc oxide, respectively. Fertility traits of ewes and weight of lambs were recorded. Laparoscopic examination was carried out 3 weeks after the beginning of the experiment. Ovarian follicles were counted and classified according to their diameter into small (<2 mm), medium (2–4 mm) and large follicles (>4 mm). The numbers of corpora lutea on both ovaries as well as the ovulation rates were recorded. Data indicated that the supplementation of different levels of inorganic zinc oxide significantly increased the incidence of oestrus, pregnancy, lambing rates and resulted in shorter onset of oestrus as compared to the control group. A similar trend was observed in the mean body weight and daily gain of lambs born to supplemented ewes. A significantly ($P<0.05$) higher population of large follicles (2.5±0.5 and 3.0±0.6 follicles) were found in ewes supplemented with 100 and 150 ppm ZnO than in control and 50 ppm ZnO groups (1.0±0.2 and 1.0±0.3 follicles, respectively). The ovulation rate was significantly ($P<0.05$) higher in ewes supplemented with 150 and 100 ppm ZnO than those in 50 ppm ZnO and controls. In conclusion, the supplementation of 100 to150 ppm zinc oxide to the basal diet of Baladi ewes significantly improved the reproductive performance by decreasing the numbers of days to oestrus, increasing the incidence of oestrus, pregnancy, lambing rates and enhancing the body weight gain of their lambs. The number of large follicles and ovulation rates were also increased.

Key words: Baladi ewes, ovarian activity, reproductive performance, zinc oxide

INTRODUCTION

Trace elements may function as cofactors of enzymes, or stabilizers of secondary molecular structure. Their function is involved in cellular metabolism. There has been special interest in effects of dietary trace element deficiencies on physiological function and particularly on reproduction. Severe dietary deficiencies of copper, selenium and zinc are commonly seen in ruminants (Hidiroglou, 1979; Minson, 1990). Zinc is a trace element essential for every living being (Underwood & Suttle, 1999), and poor growth is a prominent characteristic of Zn deficiency in animals and plants (Dijkuizen *et al.,* 2001). Zinc is known to affect growth, reproduction and immune system of animals by influencing the enzyme activity and gene expression of proteins (Chester, 1997) or by its effect on mitogenic hormones, signal transduction, gene transcription and RNA synthesis (MacDonald, 2000). Lamb production before weaning can be influenced by ewe nutrition during critical life cycle stages of ewes (Rattray, 1987).

Many investigators have focused their research on the effect of organic form of zinc on the reproductive performance of goats (Hayat *et al.,* 2010), cows (Campbell & Miller, 1998) and buffaloes (Abou-Zeina *et al.,* 2009). Greater bioavailability and more positive effects on milk quality and udder health have been reported for organic Zn when compared to inorganic forms (Spears, 1989). Recent studies showed that Zn supplemented through the organic sources had a higher retention and tissue concentrations (Lardy *et al.,* 1992; Cao *et al.*, 2000).

Little is known about the effect of inorganic zinc sources on reproductive performance and ovarian activity of sheep. Thus the main objective of the current study was to evulate the effects of different levels of dietary zinc oxide on ovarian activity, the reproductive perfomance of Baladi ewes and the growth of their lambs.

MATERIALS AND METHODS

The study was conducted during the summer season at the experimental sheep farm in the Sharkia Governorate, Egypt located in the north part of Nile Delta (latitude 30'01" N; longitude 31'21" E).

Animals and experimental design

Forty-eight Baladi ewes, 2 to 3 years old, weighting 40 kg on the average were used in the present study. All ewes were healthy and clinically free of external and internal parasites. Animals were housed in semi open yards. All animals were individually fed a daily basal diet of 1.5 kg dry matter intake consisting of roughage and concentrate mixture. The concentrate mixture was comprised of yellow corn (30%), wheat bran (29%), cotton seed meal (25%), soybean meal (6%), rice bran (4%), molasses (3%), limestone (2%) and common salts (1%). Diet was formulated to meet the nutrient requirements of NRC (1985) for sheep. All ewes were fed Egyptian clover (*Trifolium alexandrium*), green maize (Darawa) and wheat straw as a source of roughage. They were randomly allotted into four groups of 12 ewes each. Group 1 was kept as a control and was fed the basal diet. Group 2 was fed the basal diet supplemented with 50 ppm zinc oxide (50 ppm ZnO). Groups 3 (100 ppm ZnO) and 4 (150 ppm ZnO) received the same basal diet supplemented with 100 and 150 ppm zinc oxide, respectively. The supplementation period was 7 months.

Reproductive performance

All ewes were synchronized for oestrus using intravaginal sponges impregnated with 60 mg medroxy progesterone acetate (MAP, Veramix, Upjohn, Egypt) for 12 days three weeks after the treatment began, to allow adaptation to ration. Oestrus was detected every day by visual observation between 9–12 AM and 2–5 PM using a vasectomized ram. Ewes were considered to be in oestrus when standing to be mounted. Ewes in oestrus were naturally inseminated by a fertile ram. Fertility traits including oestrus response, the time interval to onset of oestrus, pregnancy; lambing rates and fecundity were recorded.

Lamb performance

Ewes lambed indoors and born lambs were numbered and weighed at day of birth and at 4 months of age (weaning) to determine body weight gain.

Laparoscopic examination

Follicular populations, size and ovulation rates were determined by laparoscopies (7 mm in diameter) after infiltration with 2% lidocaine at the puncture site as previously reported by Kelly & Allison (1976). Laparoscopies were performed in four ewes of each group 3 weeks after the start of supplementation. Preovulatory examination was studied for three successive days after sponge removal to record the follicular activity.

The luteal stage was assessed 7 days after sponge removal and then at weekly intervals to determine the ovulation rate. Ovarian follicles were counted and classified according to their diameter into small (<2 mm), medium (2–4 mm) and large follicles (>4 mm). The number of corpora lutea on both ovaries were recorded.

Statistical analysis

Data were expressed as mean ± SEM. The data were analyzed statistically by ANOVA and Duncan's test to detect differences among means using SPSS® Statistical Software (SPSS ®11.01 for Windows, 2001). Reproductive performance percentages were analyzed by the chi-square test.

RESULTS

The oestrus response, onset of oestrus, pregnancy and lambing rates for control and supplemented groups are presented in Table 1. The incidence of oestrus was significantly (P<0.05) higher in ewes supplemented with 100 and 150 ppm ZnO (100% in both groups) than in control ones (75.0%). Ewes supplemented with 150 ppm ZnO showed oestrus symptoms earlier (16.6±0.30 days, P<0.05) than the control (20.3±0.42 days) and 50 ppm ZnO groups (18.4±0.37 days). No significant difference existed between groups supplemented with 150 ppm and 100 ppm ZnO with regard to onset of oestrus. In-

Table 1. Effect of zinc supplementation on the reproductive performance of ewes

Reproductive performance	Control	50 ppm ZnO	100 ppm ZnO	150 ppm ZnO
Oestrus response, number (%)	9 (75.0) ^{b}	11 (91.7) ^{ab}	12 (100.0) ^{a}	12 (100.0) ^{a}
Onset of oestrus (days)	20.3±0.42 ^{b}	18.4± 0.37 ^{b}	17.7±0.34 ^{ab}	16.6±0.30 ^{a}
Pregnancy rate, number (%)	8 (88.80) ^{a}	11(100.0) ^{a}	12 (100.0) ^{a}	12 (100.0) ^{a}
Lambing rate, number (%)	7 (87.50) ^{a}	10 (90.90) ^{a}	11 (91.7) ^{a}	12 (100.0) ^{a}
Fecundity	142.8	150.0	154.5	158.3

Values in the same row with different superscripts (a, b) are different (P<0.05).

Table 2. Effect of zinc supplementation on the production performance of lambs up to weaning age (mean±SEM)

Production performance	Control	50 ppm ZnO	100 ppm ZnO	150 ppm ZnO
No. of lambs born	10	15	17	19
Weight (kg) at birth	3.10 ±0.31	3.30±0.37	3.60±0.29	4.60±0.5
Weight (kg) at weaning	16.2 ±0.52^{c}	19.9 ±0.97^{b}	21.6±1.1^{b}	24.8±1.5^{d}
Body weight gain (kg)	13.10 ±0.31^{d}	16.6 ±0.73^{c}	18.0 ±0.99^{b}	20.2 ±1.5^{a}
Litter size at birth	1.43±0.13	1.50±0.18	1.55 ±0.20	1.58 ±0.16
Litter size at weaning	1.14±0.15	1.40 ±0.18	1.55 ±0.20	1.58 ±0.16

Values in the same row with different superscripts (a, b, c, d) are different (P<0.05).

Table 3. Effect of zinc supplementation on ovarian activity of ewes (mean±SEM; n=4)

	Control	50 ppm ZnO	100 ppm ZnO	150 ppm ZnO
Number of follicles				
small (<2 mm)	4.0 ±1.0	4.5 ±1.2	3.8±0.9	4.5 ±1.3
medium (2–4 mm)	3.0 ±0.5	3.5 ±0.6	3.0 ±0.4	3.6 ±0.7
large (>4 mm)	1.0 ±0.2 ^{b}	1.0 ±0.3 ^{b}	2.5±0.5 ^{a}	3.0 ±0.6 ^{a}
Ovulated ewes, number (%)	2 (50.0)^{c}	2 (50.0)^{c}	3 (75.0) ^{b}	4 (100.0)^{a}
No. of corpora lutea	3	4	8	10
Ovulation rate	1.0 ±0.3 ^{b}	1.3 ±0.4 ^{b}	2.0 ±0.5 ^{a}	2.5 ±0.7 ^{a}

Values in the same row with different superscripts (a, b, c) are different (P<0.05).

clusion of 50, 100, and 150 ppm ZnO increased pregnancy rates compared to controls (100.0, 100.0, 100.0 vs. 88.8%, respectively) but without statistical significance. Similar tendency was observed in lambing rates. The highest lambing rate was in ewes supplemented with 150 ppm ZnO (100%) and the lowest in controls (87.5%). The differences between supplemented and control groups were not significant in relation to the fecundity.

The data presented in Table 2 show that litter size at lambing as well as at weaning was not significantly affected by Zn supplementation. The average body weight of lambs at birth was not significantly different among groups. At weaning however, the body weight of lambs was significantly (P<0.05) improved in the group supplemented with 150 ppm ZnO (24.8±1.5 kg) as compared to controls and groups that received 50 and 100 ppm ZnO (16.2±0.52, 19.9±0.97, 21.6±1.1 kg, respectively). A similar trend was observed in the average body weight gain of lambs born to ewes supplemented with 150 ppm ZnO at weaning age (4 months). It was significantly higher (20.2±1.5 kg; P<0.05) vs control, 50 and 100 ppm ZnO groups (13.10±0.31, 16.6±0.73 and 18.0±0.99 kg, respectively).

The results from the laparoscopic examination of ovarian activity of Baladi ewes are illustrated in Table 3. No significant difference was found among different groups regarding the population of small and medium follicles. However, a significant effect was observed in the large follicle population, where groups given 100 and 150 ppm ZnO had the highest population with 2.5±0.5 and 3.0±0.6 follicles respectively (P<0.05). The lowest population of follicles found in this category was for control and 50 ppm ZnO groups with 1.0±0.2 and 1.0±0.3 follicles. In terms of the percentage of ewes that ovulated and that had a functional corpus luteum, the group supplemented with 150 ppm ZnO group had significantly higher values (100%, $P<0.05$) than control, 50 ppm and 100 ppm ZnO groups (50.0, 50.0, 75.0%, respectively). Regarding the effect of Zn supplementation on ovulation rate, the groups that received 150 and 100 ppm ZnO had significantly ($P<0.05$) higher ovulation rates, with an average of 2.5±0.7 and 2.0±0.5 than those in control and 50 ppm ZnO groups (1.0±0.3; 1.3±0.4, respectively).

DISCUSSION

The results of the present study revealed that inorganic zinc oxide supplementation was beneficial for the reproductive performance of native Baladi ewes as indicated by decreased mean interval to oestrus and increased oestrus response. In this respect, Campbell & Miller (1998) reported that cows and heifers supplemented with zinc had fewer days to first oestrus than non-supplemented. Furthermore, Socha & Johnson (1998) stated that dairy cows fed supplemental organic minerals reduced the mean interval to 1st service and open days. The pregnancy and lambing rates recorded in the present study were greatly improved in the groups supplemented with zinc as compared to controls. The pregnancy rates were 100% in all supplemented ewes and 88.8% in controls. The lambing rate was 100% only in ewes supplemented with 150 ppm Zn. Ahola *et al.* (2004) found that cows receiving trace minerals (Zn, Cu and Mn) had higher pregnancy rates than non-supplemented. Inadequate zinc levels have been associated with decreased fertility, abnormal oestrus, abortion and altered myometrial contractility with prolonged labour (Duffy *et al.,* 1977; Maas, 1987). Zn has a critical role in the repair of damaged uterine tissue and maintenance of uterine lining following parturition, speeding return to the normal reproductive function and oestrus (Green *et al.,* 1998). Improvement of reproductive performance in Zn-supplemented dairy cows was also reported by Manspeaker *et al.* (1987). This may be due to increased plasma β-carotene level that correlates directly to the improved conception rates and embryonic development (Hayat *et al.,* 2010).

The profitability of many animal production units is dependent upon optimum gain and efficient feed conversion of livestock. The present study revealed a significantly higher body weight at weaning and faster weight gain of lambs born from dams supplemented with 150 ppm as compared to those of control one. The body weight of lambs at birth was not affected by Zn supplementation. These results go in parallel with those reported in goats (Hayat *et al.,* 2010). Hatfield *et al.* (1992; 1995) reported that Zn supplementation increased feedlot lamb performance and had a positive effect on lamb weaning. Furthermore, Abdelrahaman *et al.* (2003) reported a higher average daily gain and feed conversion efficiency of Zn supplemented lambs as compared to controls. Hermansen *et al.,* (1995) observed that feeding a high concentration of Zn only tended to increase zinc concentration in the whole milk. This high level of zinc in the milk consumed by lambs enhances their growth at weaning (Grag *et al.,* 2008) and explains the increased body weight of lambs from supplemented dams at weaning and the faster weight gain from birth to weaning. However, we did not investigate that in the present study. As shown by Engle *et al.* (1997), one of the first indicators of marginal zinc deficiency is depression in gain and conversion that are often present prior to any change in blood or liver levels.

Providing adequate levels of bioavailable trace minerals can affect growth performance. Zinc is essential for epithelial tissue integrity, cell division, repair and uptake transport mechanism and utilization of vitamins A and E. In the present study, groups supplemented with 100 and 150 ppm zinc oxide had a higher population of large follicles as compared to the other groups. This was reflected in significantly ($P<0.05$) higher ovulation rates in groups supplemented with 100 and 150 ppm ZnO. The highest percentage of ovulated ewes was achieved with supplementation with 150 ppm ZnO (100%).

Follicles population consisted mainly of small follicles, followed by medium follicles, which were similar in all studied groups. This shows that all groups studied had the same potential of ovarian activity in the reproductive stage. Research results in beef cattle indicate that first-calf heifers had confirmed pregnancies 10 days earlier (Swenson *et al.,* 1998), 17 to 35% improvement in artificial insemination (AI) conception rates (Spears & Kegley, 1991; Ansotegui *et al.,* 1994) and an increase in number of ova ovulated per heifer (6.3 versus 2.8) when females fed complexed trace minerals were compared to those fed iso-sulfate forms. Manspeaker *et al.* (1987) found that supplementation of dairy heifers with Cu, Zn, Mn, Fe and Mg (chelated form) exhibited higher number of mature follicles 30–80 days post-partum compared to non-supplemented (35 vs. 20%, respectively). Swenson *et al.,* (1998) supplemented Cu, Zn, Co and Mn as either inorganic sulfate or in a complexed form to first-calf heifers and showed that even though the percentage of significant structures (follicles greater than 12 mm and/or corpora lutea as determined by rectal palpation) and cows exhibiting oestrus by day 45 was lower when complexed minerals were supplemented, the percentage of cows bred by AI was improved.

In conclusion, the supplementation of 100 to150 ppm zinc oxide to the basal diet of Baladi ewes significantly improved the reproductive performance by decreasing the number of days to oestrus, increasing the incidence of oestrus, pregnancy, lambing rates and enhancing the body weight gain of their lambs as well as increasing the number of large follicles and ovulation rates.

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