A Trial to Ameliorate the Reproductive Performance of Native Egyptian Cows Suffering from Reduced Fertility

W.M. Ahmed, E.M. Hanafi and M.M. Zaabal

Department of Animal Reproduction and A.I. National Research Center, Giza, Egypt

Abstract: Problems of fertility hinder the productive and reproductive performances of farm animals. Moreover, it causes great economic losses, especially in small holder farms, whereas animals are exposed to a lot of stressful conditions. The current investigation was a trial to improve the reproductive performance of native Egyptian cows suffering from reduced fertility through administration of vitamins AD,E and selenium. A total number of 50 native Egyptian cows which calved since more than a year and did not show signs of estrous activity was used. These animals rose at small holder farms at Lower Egypt. Cows were divided into 2 comparable equal groups, the first group kept as the control group and the second group injected (i.m) with AD,E and selenium once weekly for 3 successive weeks. Blood samples were collected for plasma progesterone assay (ELISA). Estrous signs were checked and ovarian activity was monitored by both rectal palpation and ultrasonography. Results indicated that supplementation of native Egyptian cows suffering from reduced fertility with AD,E and selenium improved ovarian activity whereas, 80% of cows in the treated group Vs. 20% of cows in the control group showed estrous signs within 7-14 days post treatment. Also, the appeared estrous signs were more intense in the treated than the control groups. Graafian follicles and/or corpora lutea were detected in 88.00 vs, 28.00 % of the treated and control groups during the first month post treatment, respectively. The plasma progesterone level was undetectable in both groups before the start of treatment. The first raise in plasma progesterone level was detected in the treated group on day 7.44 ± 1.95 post treatments. After 2 months from treatment, 64 and 12% of cows in the treated and control groups were found pregnant, respectively. It was concluded that supplementation of low fertile Egyptian cows with AD,E and selenium improve the reproductive performance and consequently shortens the calving intervals and increases the income from breeding of livestock.

Key words: Cows • Ovarian activity • AD,E • Selenium • Estrus

INTRODUCTION

Ovarian inactivity hinders the productive and reproductive performances of farm animals and causes great economic losses, especially in small holder farms, whereas animals are exposed to a lot of stressful conditions.

Reproductive performance of farm animals depends on adequate balanced levels of vitamins and essential minerals due to their important roles in cellular metabolism, maintenance and growth [1]. Also, these nutrients have specific roles and requirements in reproductive tissues [2].

Beta carotene and vitamin E molecules constitute a major part of the antioxidant system, as they act as membranes protectant to maintain the integrity of phospholipids against oxidative damage and peroxidation [3]. Glutathione peroxidase is a selenium dependent enzyme that utilizes electrons from glutathione and other thiols to convert peroxides to water [4]. Production of free radicals could represent a source of infertility, because ovarian steroidogenic tissue [5], spermatozoa [6] and preimplantation embryos [7] are sensitive to free radicals damage. In some studies, administration of β-carotene [8] or vitamin E and selenium [9,10] improved fertility of
bovine, while in other studies, providing high amount of these antioxidants showed no beneficial effects on fertility[11].

The present study was a trial to improve ovarian activity and consequently the reproductive performance of native Egyptian cows suffering from reduced fertility using vitamins AD,E and selenium.

**MATERIALS AND METHODS**

The present experiment was carried out during regular field trips (2008 -2011) to small holder farms at areas of Lower Egypt which are deprived from ideal routine veterinary services.

**Experimental Cows:** A total number of 50 low fertile cows which calved at list since a year and showed no estrous signs was used in this study. Cows were fed on non sufficient amount of concentrated mixture, available crop residues beside rice straw and Egyptian clover (during December - May ad libitum).

**Experimental Design:** Fifty cows were divided into two equal comparable groups:

- The control group: kept without any treatment.
- The treated group: injected deeply intramuscular with the recommended dose (4 ml/week) of Aderon (Huron, Germany) for 3 successive weeks. This dose contains 400,000 IU vitamin A palmetate, 100,000 IU vitamin D₃ and 400 IU vitamin E acetate. Moreover, 10-ml sterile saline solution containing 40-mg selenium, as sodium selenite was simultaneously intramuscular injected [12].

**RESULTS**

Ovarian activity was followed up through observation of estrous signs, rectal palpation as well as ultrasonography. Cows’ owner were advised to observe estrous signs twice a day, at morning and at evening at least. Rectal palpations were carried out weekly for determining the ovarian changes and the presence of physiological structures (Graafian follicles - corpora lutea). Ultrasonography was carried out using a machine from PiaMedical Falcs e’Saote (Netherlands) with an endorectal linear array of 8.6 M hertz.

Blood samples were weekly collected from all cows by jugular vein puncture before and after treatment until the observation of heat signs. Plasma samples were separated (X1500g at 4° C for 15 minutes) from heparinized tubes and kept at -20° C until assaying of progesterone by ELISA kits from DIMA (Germany). The kit had a sensitivity of 2.0 pg/ml with inter- and intra- run precision coefficient of variations of 2.9 and 4.85, respectively.

Cows showed estrus have been mated with proven fertile bull at the appropriate time (12 hr5 after the appearance of signs).

**Statistical Analysis:** Data were statistically elaborated with Analysis of Variance [13] using Mc Graw Hill software [14].

Table 1 shows the effect of administration of vitamins AD,E and selenium on the ovarian activity of native Egyptian cows suffering from reduced fertility. 80% of cows in the treated group showed estrous signs within 7-14 days post treatment versus 20 % only in the control group (P< 0.01). Also, the appeared signs were more

<table>
<thead>
<tr>
<th>Reproductive parameter</th>
<th>Control group (N=25)</th>
<th>Treated group (N=25)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Appearance of estrous signs during 3 months post treatment (%)</td>
<td>20.00</td>
<td>80.00</td>
</tr>
<tr>
<td>1st observed heat ( days post treatment)</td>
<td>14 - 28</td>
<td>7 - 14</td>
</tr>
<tr>
<td>Intensity of signs(1-5 scores;1 weak ; 5 strong)</td>
<td>1.33 ±0-78</td>
<td>3.46 ± 1.33**</td>
</tr>
<tr>
<td>Presence of ovarian physiological structure (%)</td>
<td>28.00</td>
<td>88.00**</td>
</tr>
<tr>
<td>1st elevation of progesterone (day post treatment)</td>
<td>24.00 ± 6.22</td>
<td>7.44 ± 1.95**</td>
</tr>
<tr>
<td>Conception rate (%)</td>
<td>12.00</td>
<td>64.00**</td>
</tr>
</tbody>
</table>

** P<0.01

Table 2: Effect of administration of vitamin AD,E and selenium on plasma progesterone value (ng/ml) in low fertile native Egyptian cows (Mean ± SE).

<table>
<thead>
<tr>
<th>Reproductive phase</th>
<th>Control group</th>
<th>Treated group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before treatment</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>At appearance of heat signs</td>
<td>ND</td>
<td>0.22 ± 0.03</td>
</tr>
<tr>
<td>A week after heat</td>
<td>ND</td>
<td>2.12 ± 0.44**</td>
</tr>
<tr>
<td>Early pregnancy ( 30-35 day)</td>
<td>2.12 ± 0.23</td>
<td>2.16 ± 0.22</td>
</tr>
</tbody>
</table>

** P<0.01 ND= non detectable
intense (P< 0.01) in the treated (3.46 ± 1.33) than the control (1.33 ±0.78 ) groups when measured on 1-5 score system (1 weak - 5 very strong).

Ultrasonography revealed the presence of Graafian follicles and/or corpora lutea (P<0.01) in 88.00 vs, 28.00 % of the treated and control groups during the first month post treatment, respectively.

The level of plasma progesterone was undetectable in both groups before the start of treatment. The first raise was detected in the treated group on day 7.44 ± 1.95 post treatments, while the level was still undetectable in most of cows in the control group and it rose on day 24.00±6.22 in few of control cows. Progesterone level was 0.22 ± 0.03 ng/ml on the day of heat and 2.12 ± 0.44 ng.ml 7 days later in the treated group and remained undetectable in the control group.

After 2 months from treatment, a significant (P<0.01) higher percent of treated (64.00) cows were found pregnant versus a percent of 12.00 of the control group with a level of plasma progesterone averaged 2.16 ± 0.22 and 2.12 ± 0.23 ng.ml in the treated and control group, respectively.

**DISCUSSION**

Nutrition and management mainly determined the reproductive performance of livestock. Adequate nutrition could encourage farm animals to express their biological genetic potentials, alleviate the negative effect of a harsh physical environment and managemental techniques particularly during the postpartum period [5,15].

Vitamins are essential micronutrients for proper function of various vital organs including genital organs and endocrine glands due to their obvious roles in cellular metabolism, maintenance and growth [16].

In the present study, administration of vitamins AD,E and selenium in native Egyptian cows suffering from reduced fertility improved the ovarian activity and consequently the reproductive traits as monitored by the conception rate as compared with the control group.

The present favorable effect of supplementation on the reproductive performance was previously reported in cattle [17] and buffalo [18]. In this respect, it has been reported that administration of vitamin E and selenium could improve uterine health through enhancing neutrophils functions, promotes removal of microorganisms and supports uterine tissue [19]. Also selenium may stimulate uterine contraction and motility that enhance uterine involution [20] and consequently ovarian rebound.

The first elevation of progesterone level as well as the first observed estrous signs were recorded in treated group earlier than control one. Moreover, 88% of treated cows Vs.28 % of control cows exhibited estrous signs within the first month of the study period and 64 Vs.12% of cows conceived after less number of services within the first 2 months post treatment these finding were previously recorded and attributed to the fast uterine involution and clearance from the postpartum contaminants as well as ovarian rebound responding to the gonadotropins[8] that may be enhanced under the influence of vitamin D supplementation [21]. In the same time, it has been reported that vitamin E and selenium may increase fertilization rate in cattle and sheep as it is traced to increase the rate of sperm transport as well as uterine motility and embryo survival [20].

The significant positive effect of supplementation on progesterone values in the treated group could be explained according to Mistry *et al.* [22] who reported that vitamin A changes into retinolic acid in corpora lutea and stimulate luteal cells to utilize low-density lipoproteins for progesterone synthesis. High levels of progesterone during the mid luteal phase enhance implantation and conception and consequently maintains pregnancy [23].

The mechanism by which vitamins AD,E and selenium administration could improve reproductive traits in livestock was previously investigated. Retinol (the active metabolites of vitamin A), is secreted from the liver, bound to retinol binding protein [24], converted to retinolic acid in target cells and acts by a mechanism similar to steroid hormones-receptors complex in altering gene expression in the nucleus and induces cell division and differentiation, RNA and protein synthesis and steroid synthesis[15]. Also, it was reported that vitamin D is responsible for regulation of intra-cellular calcium and calcium binding protein in reproductive organs and pituitary[15]. Recently immunohistochemical analysis revealed that Vitamin D receptors mRNA are expressed in denderetic cells, macrophages and luminal and glandular epithelial cells of the endometrium, granulosa and cumulus oophorus cells of the ovary and fallopian epithelial cells, particulary during estrous phase suggesting its immunological role during this period [21].

On the other hand, Vitamin E and selenium affect reproductive tissue through their antioxidant roles as well as involvement in prostaglandin synthesis [22]. Vitamin E has been implicated in the control of phospholipase A2 activity which is responsible for cleaving arachidonic acid from membrane phospholipids for synthesis of prostaglandins [24]. Selenium preferentially accumulates...
in ovary, pituitary, placenta and adrenal glands suggesting specific requirements for selenium in these tissues for proper function [25].

In conclusion, administration of vitamins AD, E and selenium for native Egyptian cows suffering from reduced fertility improved their reproductive performance as indicated by resumption of ovarian activity and conception rate. It could be recommended to supply livestock with such elements to get favorable income from animal husbandry.

REFERENCES