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# RESEARCH ARTICLE

# EFFECT OF FOOD RESTRICTION ON PLASMA FSH AND TIMING OF THE NEW FOLLICULAR WAVE EMERGENCE IN CATTLE

## \*Mohamed Ali

Department of Animal Production, Qassim University, Al-Qassim, Saudi Arabia

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#### **ABSTRACT**

Friesian cows were used to determine the effect of undernutritionon concentrations of the follicle stimulating hormone (FSH) in serum and emergence of new follicles after estradiol cypionate (ECP). Cows of G1 and a control group were fed on concentrated feed and dry clover while G2 was fed on hay only (restricted diet) for a period of 60 days before initiation of the treatment with ECP. After 60 days, cows of G1 and G2 were given 1 mg of ECP. On Day 0 (day to receive ECP), blood serum from all groups was collected at 12 hour interval for 3 days, and concentrations of FSH were quantified in blood serum. A well-defined increase and synchronization in FSH concentration was observed in G1.FSH concentrations were greater for G1 (0.86 mIU/ml) than for other groups. Emergence of a new wave of follicular development was observed 48 hours after beginning the study in G1 and contrasted with delayed emergence of a new follicular wave in the control group (48±24 hours). However, long-term feed deprivation decreased FSH concentrations in serum and absence emergence of new follicular waves.

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## INTRODUCTION

Reproduction efficiency is a major factor affecting the production and economic efficiency of dairy and beef cow herds. The ideal calving interval of one year can only be achieved if the interval between parturition and successful service is less than 85 days (Stagg et al., 1995). This ideal calving interval can only be attained with accurate detection of oestrus and timing of insemination relative to ovulation (Stagg et al., 1995). Inadequate nutrition impairs reproductive function in most mammals. The adverse effects of underfeeding on the reproductive performance of cattle may be due to hormonal disturbance at the level of the ovary, anterior pituitary gland and/or hypothalamus (Schillo, 1992). Henricks et al. (1986) found that undernutrition in postpartum beef heifers resulted in a decrease in the serum concentration of oestradiol and delayed follicular growth. Suppression of both frequency and amplitude of pulses of LH was observed (Day et al., 1986) in heifers which underwent a restriction in dietary energy intake when compared with heifers fed an adequate energy diet. Later, Kurtz et al. (1990) observed that restriction of dietary energy inhibited the prepubertal increase in LH pulse frequency both in intact and ovariectomized heifers. Collectively, it can be concluded that undernutrition can disturb pulsatile LH secretion and this may disturb follicular development to the preovulatory stage. Murphy et al. (1991) demonstrated that low dietary energy intake reduces the maximum diameter of the dominant

\*Corresponding author: Mohamed Ali

Department of Animal Production, Qassim University, Al-Qassim, Saudi Arabia.

follicle and shortens its persistence on the ovary. Also, McDougall et~al.~(1995) observed that the first dominant follicle in Friesian heifers that calved with a low body condition score took longer to reach its maximum size compared to heifers that calved at a higher body condition score  $(14.0 \pm 1.3 \text{ versus} 10.6 \pm 0.7 \text{ d})$ . Specific research on the impact of nutrition on the response of follicular development and FSH secretion after estradiol injection in cattle is limited. Therefore, the aims of this study were, 1) to determine the effect of long term under nutrition on concentration of FSH and 2) to examine the time of emergence of a new follicular wave after estradiol injection.

## **MATERIALS AND METHODS**

# Animals and treatment

This study was conducted at the Research Center of the Agriculture and Veterinary College, Qassim University, Saudi Arabia. A total of 10 adult cows were used for this study. These animals were selected to be cycling and non-pregnant for the research. The selection was dependent on rectal palpation (when the ovary contain active corpus luteum or follicles), and herd records. The age of the animals was more than 3 years and free from any diseases. Fresh water was given ad libitum. Ten cows were subdivided to three groups (4 animals in Control, 3 animals in G1, and 3 animals in G2 groups). The animals in G1 and the control group were supplemented with a commercial concentrated food and dry clover. While G2 group was only supplemented with hay for 60 days before initiation of oestradiol cypionate treatment. All

cows in G1 and G2 groups received 1 mg of oestradiol cypionate (ECP) by intramuscular injection.

The ovaries of all cows were examined by an ultrasound scanner with a 6 MHz transrectal probe (piemedical, Netherlands) in order to determine the size of follicle. Before insertion of the probe, fecal material was removed from the rectum, and subsequently the location of ovary was determined. The diameters of ovarian structures were monitored by transrectal ultrasonography on a twice daily basis from Day 1 (ECP treatment) until the emergence of a new wave of follicular development was confirmed retrospectively. The day of new follicular wave emergence was defined as the day on which the largest follicle of a growing cohort was 4–5 mm in diameter (Burke, *et al.*, 2003).

#### **Blood samples**

The cows were restrained in a cattle-crush and blood samples were collected by coccygeal vein into 5 ml vacutainers containing sodium heparin. Following collection, the labeled blood samples were immediately placed in ice (3°C) and centrifuged for 15 min at 1340 x g. The plasma was transferred into a 2 ml polypropylene tube and stored frozen at -20 °C until assay. Blood samples for hormone assay were collected twice daily beginning with ECP treatment and continued for 3 days. In each sample, the concentration of follicle stimulating hormone (FSH) was evaluated by ELISA. (Easilisa Co.) according to the manufacturer's instructions.

#### RESULTS

Concentrations of FSH did not differ among treatments at hour 12.Awell-defined increase and synchronization in FSH concentration was observed in the G1 (feeding plus ECP group). In the other groups (non-feeding plus ECP group or control group) were not evident (Figure 1). However, 3 cows of the control group (75%) were observed with good concentrations of FSH but great variability of time was evident (28±16 hours) (Table 1). Therefore, the FSH level was not consistently observed (Figure 1).

Emergence of a new wave of follicular development was observed 48 hours after beginning the study in 2 cows of the G1(feeding plus 1ECP)and contrasted with delayed emergence of a new follicular wave in the control group (48±24 hours) (Table 1). The effect of ECP with good feeding in synchronization with the time to emergence of a new wave of follicles was evident and followed a similar effect on the timing of the FSH peak (Table 1). However, in the G2 (nonfeeding plus ECP group) showed no high concentration of FSH and emergence of new follicle compared to G1 or control group. It is demonstrated that the peak of FSH after ECP treatment was suppressed and growth of new dominant follicle was restricted. Therefore, there is no result in G2shown on the table (1). The interval between peak FSH and new follicular wave emergence (12 h) was not different among groups.

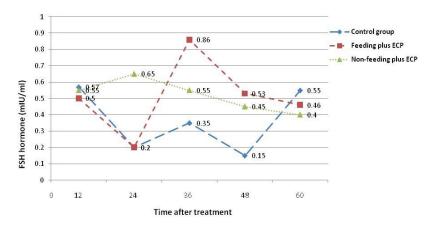


Figure 1. Mean concentrations of FSH hormone in circulation of cows receiving 0ECP (control group), 1ECP with feeding and 1ECP without feeding of concentrated food mg ECP i.m./cow, Day 0 is day of ECP injection

Table 1. Intervals from treatment initiation to time of maximal concentration of FSH (FSH peak) and emergence of a new follicular wave in cows receiving 0ECP (control group), 1ECP with feeding and 1ECP without feeding of concentrated food mg ECP i.m./cow

	Groups (%)		
	1ECP with feeding	1ECP without feeding	0ECP (control
	(G1)	(G2)	group)
Animals (n)	3	3	4
Mean time to peak of FSH (hrs.)	36±0.0 (66.6%)	nr	28±16 (75.0%)
Range(hrs.)	36		12-60
Mean time from peak of FSH to follicle emergence (hrs.)	12±0.0 (66.6%)	nr	12±0.0 (50.0%)
Mean time from beginning study to follicle emergence			
(hrs.)	$48 \pm 0.0$	nr	48±24
Range(hrs.)	48		24-72

nr: no result.

## **DISCUSSION**

The main finding of this experiment is that a long feeding restriction, that is, for a period of 60 days, resulted in failure of the development of a new follicular wave. Our results suggest that failure of the emergence of new follicular waves was always associated with the absence of an FSH surge especially in the group with restricted feeding (hay only). However, time of emergence of the new follicle wave and the rise in FSH was delayed by ECP. Mean time from ECP injection to peak FSH was from 28 to 36 hours in groups with high maintenance feeding. Thus, nutritional deprivation may have a greater effect on FSH secretion and ovaries which produce proteins, peptides and steroids that are inhibitory and stimulatory to FSH secretion. Therefore, all cows feeding on hay only showed inactive ovaries and no development of new follicular waves for a period 5 days after ECP injection. Feed restriction can affect FSH in several ways. Looper et al. (1996) reported an increase in FSH concentrations after short-term feed restriction and a decrease in FSH after long-term feed deprivation in ovariectomized cows. However, Gutierrez et al. (1997) found no effects of feed deprivation on FSH concentrations in Hereford X Friesian heifers. Cows show reduced fertility and loss of body condition as indicated by lower BCS when the nutrient requirements for maintenance and lactation exceed intake (Santos et al., 2004). The nutrition of an animal is involved in regulating GnRH secretion which has an effect on the LH pulse frequency and FSH (Bolafioset al., 1998 and Diskin et al., 2003). It is clearly evident the effect of feed restriction in other animal like ewes (Kiyma et al., 2004), sow (Kauffold et al., 2007) and chickens (Bruggeman et al., 1999).

In our study, all cows in G2 (restricted food plus ECP treatment) showed that the follicle size (< 4 mm) did not change throughout the study. Effects of nutrition on follicular waves have also been explored (Comin, et al., 2002 and Driancourt et al., 2001). Both studies demonstrated that poor nutrition in cattle was associated with a reduced size of the dominant follicle of all waves, and by a reduced persistence of the dominant follicle of the first wave. In addition, low dietary intake tended to increase the proportion of the cycles with 3 follicular waves. In summary, this experiment confirmed that long restriction, that is, for a period 60 days, suppresses the growth rate and maximum diameter of dominant follicles. We suggest that suppressed growth of follicles and decreased FSH concentration is due to the absence of an LH/FSH surge. Therefore, dairy cow management, such as feeding, must be taken in consideration for milk production, body condition score and on certain aspects of reproductive functions in cattle.

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