# Response of Serum LH and Milk Progesterone to Two GnRH Agonists Fertirelin and Buserelin in Cows with Ovarian Follicular Cysts

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# Introduction

Milk progesterone assays have been used in the diagnosis of follicular cysts in dairy cows, and a combination with rectal palpation is suitable for an accurate diagnosis of follicular cysts.<sup>1</sup>

Fertirelin acetate and Buserelin acetate, analogs of GnRH, have been used for the treatment of follicular cysts. To this day, however, there is no report comparing the two GnRH agonists including their effects on LH response in cows with follicular cysts.

The objectives of this study were to examine the accuracy of rectal palpation for the diagnosis of follicular cysts based on milk progesterone tests and to compare the effectiveness of the GnRH analogs on LH response, luteinization and subsequent reproductive performance in cows with "authentic" follicular cysts.

## **Materials and Methods**

One hundred and seventy-two Holstein-Friesian cows in 50 days or later postpaturm were tentatively diagnosed as having ovarian follicular cysts upon palpation per rectum conducted at the first examination.

Milk samples were collected at the first examination. The progesterone levels of skim milk were determined by progesterone enzyme immunoassay kit.<sup>2</sup> Milk samples were again collected seven days after the first examination from 117 out of 172 cows having less than 1.0 ng/ml skim milk progesterone level. Then, the third milk samples were collected seven days after the second milk sampling from 94 cows having low skim milk progesterone level. Finally, 71 cows were diagnosed as having "authentic" follicular cysts confirmed by skim milk progesterone level showing less than 1.0 ng/ml at three consecutive times or for 15 days. The cows with follicular cysts (n=71) were assigned alternately to one of the two study groups in the order in which they were diagnosed as having follicular cysts. The fertirelin-treated group received an intramuscular injection of 200  $\mu$ g fertirelin, while the buserelin-treated group received 20  $\mu$ g buserelin i.m., 14 days after the first examination. Milk samples were collected seven days after the first treatment to determine progester-one concentration.

Blood samples were collected twice, namely, immediately before GnRH injection and 2 hours after injection (for fertirelin-treated group) or 2.5 hours after injection (for buserelin-treated group) from the coccygeal vessels to determine serum LH concentrations by radioimmunoassay.

#### Results

Of the 172 cows, 71 cows (41.3%) were confirmed to have follicular cysts by progesterone levels in defatted milk less than 1.0 ng/ml at weekly three consecutive tests. Of the 71 cows, 38 were treated with fertirelin and 33 with buserelin. The other 101 cows were excluded from the experiment, since they showed a milk progesterone level 1.0 ng/ml or higher either on the first examination, seven or 14 days later. Of the 101 cows, 55 cows (32.0%) were diagnosed as having luteal cysts or cystic corpus luteum, 37 cows (21.5%) as having Graafian follicles which luteinized seven or 14 days after the first examination. In the other nine cows missing milk samples, the diagnosis of follicular cysts was not made.

Response of serum LH after treatment was examined in 57 out of 71 cows with follicular cysts. All the 57 cows responded to the treatment with an increase of serum LH. The serum LH level in 30 cows before and 2 hours after an administration of 200  $\mu$ g of fertirelin

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and that in 27 cows before and 2.5 hours after an administration of 20  $\mu$ g of buserelin are summarized in Table 1. There was no significant difference in the LH response between the two GnRH treated groups.

Luteinization was defined in terms of an increase of progesterone in skim milk up to 1.0 ng/ml or higher. Percentages of the cases luteinized did not differ significantly between two treated-groups (Table 1). There was no significant difference in skim milk progesterone concentrations seven days after the treatment between the two groups.

The perentages of cows that conceived within 180 days after the treatment and the average days from treatment to conception in the fertirelin- and buserelintreated groups were 70.8% and  $71\pm41$  days, and 61.9% and  $63\pm53$  days, respectively. There was no significant difference in reproductive performance between the two groups.

Table 1. Serum concentrations of LH (mean  $\pm$  SD) in cows with follicular cysts before and after treatment with fertirelin or buserelin and percentage of the cases luteinized after treatment.

Treatment	Number of cows	Serum LH after treatment (ng/ml)		Percentage of
		0	2 or 2.5 hrs*	cases luteinized
Fertirelin 200 µg i.m.	30	$1.2\pm0.6$	$22.5\pm10.7$	76.7% (23/30)
Buserelin 20 µg i.m.	27	$1.1\pm0.4$	$20.6 \pm 11.4$	66.7% (18/27)

\* Two hours after treatment with fertirelin or 2.5 hours after treatment with buserelin.

### Discussion

The correspondence between the diagnosis of follicular cysts by palpation per rectum and that by milk progesterone test diminished as the weekly milk progesterone test was repeated two or three times. It indicates that two or three times milk progesterone test weekly is a useful method to diagnose follicular cysts accurately.

There was no significant difference in LH levels in cows with follicular cysts after an injection of  $200 \,\mu$ gfertirelin and  $20 \,\mu$ g-buserelin. In all cases, the LH levels increased after GnRH injection more than four times of the basal LH level. Therefore, most of the cases were considered to have normal pituitary function. However, there was a large variation in LH response among individual cows. Causes of the variation were not well understood.

Percentages of cows showing luteinization of folli-

cular cysts after GnRH treatment in this study were similar to those in previous studies.<sup>3,4,5</sup> No significant differences between treatment groups was shown for skim milk progesterone concentrations.

It is concluded that measuring progesterone levels in milk before treatment appears to be a reliable method to differentiate the follicular cysts from other ovarian states and that 200  $\mu$ g of fertirelin and 20  $\mu$ g of buserelin have an equal therapeutic effect in the treatment of ovarian follicular cysts in dairy cows.

#### Summary

Of 172 Holstein-Friesian cows which were diagnosed as having follicular cysts by rectal palpation, 71 cows (41.3%) were confirmed to have follicular cysts by the weekly milk progesterone tests. The effect of fertirelin and buserelin on cystic ovaries in secreting progesterone and on the subsequent reproductive performance were studied in 68 of the 71 cows. Response of serum LH after treatment was examined in 57 of the 68 cows. The serum LH level in 30 cows before and 2 hours after an administration of 200 µg fertirelin was  $1.2 \pm 0.6$  and  $22.5 \pm 10.7$  ng/ml (mean  $\pm$  SD). The serum LH in 27 cows treated with 20  $\mu g$  buserelin were 1.1  $\pm$ 0.4 and  $20.6 \pm 11.4$  ng/ml before and 2.5 hours after the treatment. Seventy-five percent (27/36) of cows treated with fertirelin and 72% (23/32) of cows treated with buserelin showed an increase of progesterone level up to 1.0 ng/ml or higher. There was no significant difference between the two groups in terms of subsequent reproductive performance.

#### Acknowledgements

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