

Effect of Fenprostalene 14 Days after Fertirelin Treatment on Intervals from Treatment to Conception in Cows with Follicular Cysts Diagnosed by Milk Progesterone Test

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ABSTRACT. Of 170 cows with ovarian cysts which were detected by palpation of the ovaries per rectum, 88 cows (51.8%) were diagnosed as having follicular cysts on the basis of milk progesterone concentrations. Eighty-three of the 88 cows were used for treatment trials. A group of 56 cows injected subcutaneously with fenprostalene, prostaglandin F₂α analog, 14 days after treatment with fertirelin, an analog of gonadotropin-releasing hormone, compared with another 27 cows treated only with fertirelin (controls), showed a higher pregnancy rate within 100 days after the initial treatment (66.1 vs 48.1%) and shorter intervals from the treatment to conception (30 ± 21 vs 43 ± 27 days). It was concluded that the administration of fenprostalene following fertirelin treatment is effective in shortening the interval from treatment to conception in cows with follicular cysts.—**KEY WORDS:** cattle, follicular cyst, gonadotropin-releasing hormone, prostaglandin F₂α.

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The ovarian follicular cyst is one of the most common reproductive disorders in dairy cattle. Cows with follicular cysts show either continuous or irregular estrus or become anestrus and then infertile. Since high milk producing cows are prone to develop follicular cysts, prevention by means of genetic selection or by feed management is virtually impossible. Correct diagnosis and adequate treatment with hormones such as gonadotropin-releasing hormone (GnRH) or its analog would be a beneficial solution [2, 8].

It is often difficult to diagnose correctly ovarian follicular cysts by palpation per rectum under field conditions [7] as the cysts may include luteal cysts, cystic corpus luteum or normal Graafian follicles co-existing with follicular cysts. When treated with GnRH, these cysts of the ovaries different from follicular cysts may show the different responses to the treatment [7]. This may cause equivocal and variable results of field experiments on the hormone therapy such as a combined administration of GnRH and prostaglandin (PG) F₂α [3, 6, 11]. It has not been concluded yet whether the administration of PGF₂α 9 to 14 days after GnRH treatment is effective in shortening the recovery period of follicular cysts.

In this field experiment, the ovarian cysts detected by palpation per rectum were classified into follicular cysts, luteal cysts, cystic corpus luteum, and normal Graafian follicles co-existing with follicular cysts based on milk progesterone concentrations and only cows with follicular cysts were used for a treatment trial. The study aimed to clarify the effect of fenprostalene, a PGF₂α analog of fenprostalene, luteolytic agent having a long biological half life, given 14 days after the treatment with a GnRH analog (fertirelin acetate), which acts on the anterior pituitary to release luteinizing hormone and follicular

stimulating hormone to cause luteinization of follicular cysts, on shortening the recovery period.

MATERIALS AND METHODS

One hundred and seventy Holstein Friesian cows diagnosed as having follicular cysts by palpation per rectum received a milk progesterone test. Milk samples were first collected the day after the palpation per rectum and sent by mail to the laboratory of the authors' department. Progesterone concentrations in milk were measured by a commercial enzyme immunoassay kit (Ovucheck^R, Cambridge Life Science, U.K.) [12]. Cows showing a milk progesterone concentration of 10 ng/ml or higher were considered to have luteal cysts, cystic corpora lutea or corpus luteum co-existing with follicular cysts and therefore discarded from the treatment trial. Second milk samples were collected from cows showing a milk progesterone concentration less than 10.0 ng/ml 7 days after the first sampling. Cows in which a milk progesterone concentration increased up to 10 ng/ml or higher at the second test were considered to have corpora lutea and were therefore excluded from the experiment. Only cows showing milk progesterone concentrations less than 10 ng/ml at the two tests were diagnosed as having follicular cysts and were used for the treatment trial.

Cows with follicular cysts were divided into two groups. The first group of cows (average age; 5.2 ± 2.7 years) was initially injected intramuscularly with 100 μg fertirelin acetate, followed by a subcutaneous injection of 1.0 mg fenprostalene. The second group of cows (average age; 5.0 ± 2.0 years) was treated only with fertirelin. Response of ovaries after the treatment was also monitored by the milk progesterone test.

The average number of calving and interval between the last calving and treatment in fertirelin-fenprostalene-treated and fertirelin-treated groups were 3.4 ± 2.3 and 147 ± 63 days, and 3.3 ± 1.7 and 138 ± 77 days, respectively.

RESULTS

Of 170 cows with follicular cysts which were detected by palpation per rectum, 88 cows (51.8%) were diagnosed as having follicular cysts on the basis of the results of the milk progesterone test (Table 1). Of the 88 cows with follicular cysts, 83 cows were forwarded to the treatment trial; 56 cows were treated with a combination of fertirelin and fenprostalene, 27 cows with fertirelin only, and the remaining 5 cows were culled without any treatment.

Thirty-seven (66.1%) of 56 cows treated with fenpros-

Table 1. The diagnosis for differentiating follicular cysts, follicular cysts with a Graafian follicle and luteal cysts or cystic corpora lutea in cows on the basis of milk progesterone concentrations

Milk progesterone (ng/ml)		Diagnosis	Number of cows	
day 0 ^{a)}	day 7		cows	Percentage
<10	<10	Follicular cysts	88	51.8
<10	10<	Follicular cysts with Graafian follicle	38	22.4
10<		Luteal cysts or cystic corpora lutea	44	25.9
Total			170	100

a) The day when cows were diagnosed as having follicular cysts on the basis of palpation per rectum.

Table 2. Reproductive performance in cows with follicular cysts after the treatment with a combination of fertirelin and fenprostalene or fertirelin only

	Treatment ^{a)}	
	Fertirelin and fenprostalene	Fertirelin
No. of cows treated	56	27
No. of cows coming into estrus within 6 days after fenprostalene (%)	37 (66.1)	0
No. of cows inseminated within 100 days after initial treatment (%)	49 (87.5)	20 (74.1)
Days from initial treatment to insemination (mean \pm SD)	22.7 \pm 11.7	26.2 \pm 17.4
No. of cows conceiving within 100 days after initial treatment (%)	28 (56.0)	10 (37.0)
No. of cows retreated	17	15
Total No. of cows conceiving within 100 days after initial treatment (%)	37 (66.1)	13 (48.1)
Days from initial treatment to conception (mean \pm SD)	30.4 \pm 20.7	43.1 \pm 27.2

a) Fertirelin: 100 μ g by an intramuscular injection, fenprostalene: 1.0 mg by a subcutaneous injection 14 days after fertirelin injection.

Table 3. Effect of fertirelin and fenprostalene on follicular cysts in cows shown on the basis of milk progesterone concentrations

Milk progesterone concentration (ng/ml)				Effect		No. of cows
day 0 ^{a)}	day 7 ^{b)}	day 21 ^{c)}	day 24 ^{d)}	Fertirelin	Fenprostalene	
<10	<10	10<	<10	Positive	Positive	37
<10	<10	10<	10<	Positive	Negative	2
<10	<10	<10	<10	Negative	N.K. ^{e)}	12
<10	<10	<10	10<	Negative	N.K.	2
Total						53 ^{f)}

- a) The day when cows were diagnosed as having follicular cysts by palpation per rectum.
 b) Day of treatment with fertirelin.
 c) Day of treatment with fenprostalene.
 d) Three days after fenprostalene.
 e) Effect of fenprostalene was not known, since fenprostalene was given to cows having no luteinized cysts.
 f) Milk sample was not collected on day 24 from 3 cows, which were not included.

talene 14 days after fertirelin exhibited estrus within 6 days after the fenprostalene injection (Table 2). Cows with follicular cysts treated with fenprostalene after fertirelin showed a higher pregnancy rate within 100 days after the initial treatment than the cows treated with fertirelin only (Table 2). The average interval from the initial treatment to conception in the fertirelin-fenprostalene-treated cows were 13 days shorter than that in fertirelin-treated cows.

Effect of fenprostalene given 14 days after fertirelin was also shown by the response of milk progesterone in 53 of the 56 cows treated (Table 3). Of 39 cows having luteinized cysts following the fertirelin treatment, 37 cows (95%) responded to fenprostalene with a significant decrease in milk progesterone concentrations 3 days after the injection.

In the control group of 27 cows treated with fertirelin only, 11 cows (40.7%) had luteinized cysts 14 days after the treatment. These 11 cows showed a lower pregnancy rate (36.4% vs 66.7%) and a significantly longer interval from the initial treatment to conception than the 39 cows with luteinized cysts treated with fenprostalene (62 ± 23 vs 23 ± 13 days) ($P < 0.05$).

DISCUSSION

The accuracy of rectal palpation for the diagnosis of follicular cysts assessed by the milk progesterone test was 51.8% in the present study. Cows having luteal cysts, cystic corpora lutea or cystic follicles co-existing with normal Graafian follicles or corpus luteum might have erroneously been interpreted as cases with follicular cysts on the basis of palpation per rectum. Similar findings have already been reported by the present authors [7].

In their anatomical investigation on cystic ovaries in cattle, Al-Dahash *et al.* [1] found corpora lutea associated with follicular cysts in 31% of the cases. In this study 26% of cows with follicular cysts clinically diagnosed had luteal structure as assessed by milk progesterone concentrations. Likewise, 22% of cows with follicular cysts had co-existing normal Graafian follicles, all of which ovulated and became corpora lutea as evidenced by an increase in milk progesterone concentration. Sawamukai *et al.* [9, 10] already reported that most of cows with follicular cysts showing estrus at a regular interval had co-existing Graafian follicles and that they showed a normal conception rate when inseminated without giving any treatment. Under field conditions, it is sometime difficult to differentiate correctly follicular cysts with co-existing Graafian follicles from multiple follicular cysts on the basis on palpation per rectum. Repetition of milk progesterone tests may be a solution to this problem. It should be emphasized that cows with follicular cysts diagnosed by palpation per rectum be tested for milk progesterone concentrations. Cows with a high milk progesterone concentration don't need to be treated or can be treated with $\text{PGF}_2\alpha$, while cows with a low milk progesterone concentration should be re-tested for milk progesterone 7

to 14 days later. Only cows with low progesterone concentrations at the first as well as the second tests should be given treatment.

Inducing estrus in cows with luteinized cysts following the treatment with GnRH or hCG may be beneficial to shortening of the interval from treatment to conception in cows with follicular cysts [8]. Kesler *et al.* [3] reported that an administration of $\text{PGF}_2\alpha$ 9 days after the treatment with GnRH in cows with follicular cysts resulted in shorter intervals from treatment to estrus as well as to conception and in a higher conception rate compared with controls treated with GnRH only. Later, Nakao *et al.* [4-6] also reported similar beneficial effect of the GnRH- $\text{PGF}_2\alpha$ combination treatment on follicular cysts. Stolla *et al.* [11], however, could not find any beneficial effect of $\text{PGF}_2\alpha$ given 14 days after GnRH. Accuracy of rectal palpation to judge luteinization of follicular cysts prior to $\text{PGF}_2\alpha$ injection and efficiency of heat detection are considered to be major factors influencing results of the GnRH- $\text{PGF}_2\alpha$ combination treatment of follicular cysts. Results of the present experiment clearly showed that an administration of a $\text{PGF}_2\alpha$ or its analog 14 days after GnRH was effective in reducing the recovery period of follicular cysts. The administration of fenprostalene caused regression of luteal tissue of ovarian cysts formed after fertirelin treatment, followed by development of new Graafian follicles and expression of normal estrous signs. The beneficial effect of fenprostalene as shown in this study may attribute to the early detection of estrus after the treatment and an artificial insemination at an appropriate time resulting in a high conception rate. In economical view of the treatment, shorter period from treatment to estrus, higher pregnancy rate and shorter intervals from treatment to conception following the GnRH- $\text{PGF}_2\alpha$ therapy may overcome the cost of $\text{PGF}_2\alpha$ administration 14 days after GnRH injection.

In conclusion, the milk progesterone test for accurate diagnosis of follicular cysts and an administration of a $\text{PGF}_2\alpha$ analog following treatment with GnRH are recommended as effective methods for the treatment of follicular cysts in cows.

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