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EFFECT OF DIFFERENT ANTI PMSG ADMINISTRATION TIME ON MICE EMBRYO AND UNWEANED PRODUCTION

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ABSTRACT

The research was aimed to find the best time for giving Anti-PMSG to produce embryos and unweaned in mice. The research used twenty-eight mice for producing embryo and twenty-eight mice for produce unweaned, divided into four groups. Control group was superovulated with 5 I.U. PMSG and 5 I.U hCG subcutaneously. The groups 1, 2, 3 were superovulated by 5 I.U. PMSG/SC, then, after twice, were treated with 5 LU. hCG/SC and each group treated with 1 : 20 dilution of anti-PMSG 1 hour before, during and 1 hours after LH administration.

The result showed that optimal time for giving anti-PMSG was determined at 1 hour before the administration and there was an increase in the number of mice embryos and unweaned ($p < 0.01$).

Keywords: anti-PMSG, 1 hour before, during, and after LH administration, mice embryo, unweaned mice

INTRODUCTION

Lower or higher birth rate from a mother generally depends on reproductive problems. A study in cattle reproduction is needed to increase cattle production. Some of combined hormones routinely used form embryo production through superovulation program are Pregnant Mare Serum Gonadotropin (PMSG) and Human Chorionic Gonadotropin (hCG).

Although PMSG is highly potential in stimulating ovarian function, the result of embryonal harvesting remains unsatisfactory. This is because of high cyalic acid content in PMSG molecules, rendering the half-life of PMSG becomes longer, i.e., 118 - 123 hours (Putro, 1993).

To overcome the negative effect of PMSG, agent that can limit the action time or half-life of PMSG should be given. The agent will produce anti-PMSG. However, the proper time of anti-PMSG administration should be determined, whether it is one hour before, at the same time, or one hour after LH administration in mice superovulation program using PMSG and hCG.

MATERIALS AND METHODS

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Effect of anti-PMSG polyclonal antibody administration on the production of mice embryo and the number of unweaned

Twenty-eight female and male Balb/e strain mice aged 2 months (to find embryo production) and 28 female and male Balb/e strain mice, also aged 2 months (to find the number of unweaned) were randomly grouped into four treatments, each of which received seven repetitions:

PO (control) : mice were injected with 5 I.U PMSG subcutaneously (SC) without anti-PMSG polyclonal antibody. Two days later, these mice received 5 I.U hCG injection and mated with male mice.

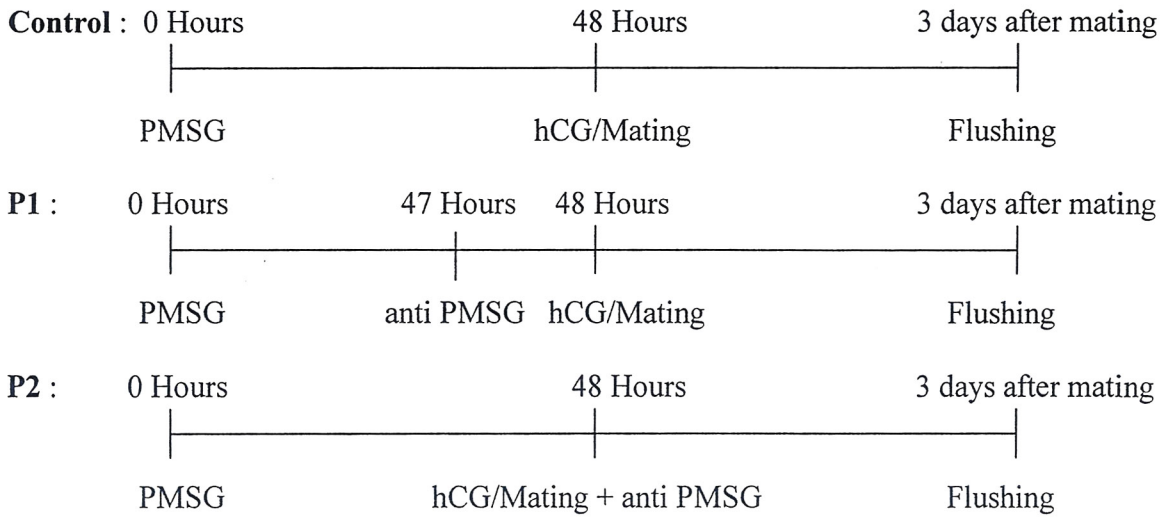
P1 : mice were injected with 5 I.U PMSG SC, and two days later were injected with 5 I.U hCG. One hour prior to LH administration, 0.1 ml anti-PMSG polyclonal antibody was given from 20 dilution SC, and mated with male mice.

P2 : mice were injected with 5 I.U PMSG SC, and two days later were injected with 5 I.U hCG, and, along with LH administration, these mice were injected with 0.1 ml anti-PMSG polyclonal antibody from 20 dilution SC, and mated with male mice.

P3 : mice were injected with 5 I.U PMSG SC, and two days later were injected with 5 I.U hCG. One hour after LH administration, 0.1 ml anti-PMSG polyclonal antibody was given from 20 dilution SC, and mated with male mice.

Observation to mice embryo production was carried out three days after mating using operation. Observation to the number of unweaned mice were done at day 21 after mating (at the time of delivery).

Schedule of embryonal harvesting in mice



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P3 : 0 Hours — 48 Hours — 49 Hours — 3 days after mating

PMSG — hCG/Mating — anti PMSG — Flushing

PMSG Dose : 5 I.U.
Anti PMSG dose : 0,1 ml from 20 dilution
HCG dose : 5 I.U.

Schedule of unweaned counting

Control : 0 Hours — 48 Hours — 21 days after mating

PMSG — hCG/Mating — Flushing

P1 : 0 Hours — 47 Hours — 48 Hours — 21 days after mating

PMSG — anti PMSG — hCG/Mating — Flushing

P2 : 0 Hours — 48 Hours — 21 days after mating

PMSG — hCG/Mating + anti PMSG — Flushing

P3 : 0 Hours — 48 Hours — 49 Hours — 21 days after mating

PMSG — hCG/Mating — anti PMSG — Flushing

PMSG Dose : 5 I.U.
Anti PMSG dose : 0,1 ml from 20 dilution
HCG dose : 5 I.U.

Data Analysis

This study was analysed by Complete Randomized Design, and obtained data on embryo and unweaned were analyzed using Anova and, if the result showed difference, the result was subjected to Least Significant Difference test (Steel and Torrie, 1995).

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RESULTS AND DISCUSSION

Effect of anti-PMSG polyclonal antibody administration on the production of mice embryo

Mice embryo examination using operation was carried out at day 3 after mating, characterized with the presence of vaginal plug. Flushing was done both uterine cornua directed from cornua to fallopian tube. The data can be seen in Table 1.

Table 1. Mean of mice embryo in control and treatment groups injected with anti-PMSG polyclonal antibody obtained three days after mating.

Treatment	N	Range	Average
PO	7	7-13	8,57± 2,07[d]
P1	7	18-29	22,71± 3,45[a]
P2	7	16-22	18,71± 2,06[b]
P3	7	10-18	14,43± 2,64[c]

Numbers with different superscript in the same column are significantly different ($p < 0.01$)
Notes: PO : Control
P1 : anti-PMSG injection 1 hour before LH
P2 : anti-PMSG injection along with LH
P3 : anti-PMSG injection 1 hour after LH

It is statistically apparent that control and treatment groups showed highly significant difference ($p < 0.01$) in the number of mice embryo. This indicates that anti-PMSG polyclonal antibody with 20 dilution injected in different time could affect the number of embryo obtained.

Keller and Tepker (1990) stated that the use of PMSG for superovulation could produce immature ova and abnormal embryo that led to early embryonal death. This problem can be overcome by anti-PMSG polyclonal antibody administration. If being given concomitantly with hCG administration, anti-PMSG polyclonal antibody administration could increase the number of mice embryo superovulated with PMSG.

According to Katagiri et al (1991), the administration of anti-PMSG antiserum for mice in appropriate time and dose may increase superovulatory response. Nakajima et al (1992) remarked that the use of anti-PMSG antiserum in bovine given 12 hours after the onset of estrous could improve obtained embryonal quality, as it could prevent highly estrogenic uterine condition. PMSG antiserum administration 5 hours post-estrous leads to the shortening of bovine estrous period. The length of estrous could be reached for 25.8 hours, while in control it was 51.3 hours. Produced ova were 17.9 and 12.5 transferable embryos, while in control the numbers were 5.9 and 2.9 (Kummer et al., 1981).

The administration of anti-PMSG antiserum pre-LH surge in cow may increase ovulatory rate to 20.3 ± 2.6, while the administration at the time of LH surge the rate was 6.3 ± 2.3 (Vos et al., 1994). According to Kuran et al. (1996) the administration of PMSG antibody can neutralize PMSG effect in vivo and increase transferable embryo count if it is given in pre-ovulatory LH. PMSG neutralization after pre-ovulatory LH may depress the effect of PMSG at the end of follicular maturation and increase the ovulatory rate (Dielman and Bevers, 1987).

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Gonzales et al (1994) studied bovines by injecting PMSG antiserum given 48 - 60 hours after PGF2a injection and obtained higher number of corpus luteum from group injected with PMSG antiserum after 60 hours as compared from that injected after 48 hours. Total ova, fertile ova, embryo, and transferable embryo were higher in both groups injected with PMSG antiserum compared to control.

Effect of anti-PMSG polyclonal antibody administration on the production of unweaned mice

The examination and counting of unweaned mice was carried out after delivery Table 2.

Table 2. Mean of unweaned mice in control and treatment groups injected with anti-PMSG polyclonal antibody obtained after delivery

Treatment	N	Range	Average
PO	7	6-10	$7,42\pm 1,40^{c}$
P1	7	10-16	$12,00\pm 2,00^{a}$
P2	7	7-12	$9,43\pm 1,62^{b}$
P3	7	6-10	$7,86\pm 1,46^{bc}$

Numbers with different superscript in the same column are significantly different ($p < 0.01$)
Notes: PO : Control
P1 : anti-PMSG injection 1 hour before LH
P2 : anti-PMSG injection along with LH
P3 : anti-PMSG injection 1 hour after LH

It is statistically apparent that control and treatment groups showed highly significant difference ($p < 0.01$) in the number of unweaned mice. This indicates that anti-PMSG polyclonal antibody with 20 dilution injected in different time could affect the number of unweaned mice.

It confirms that the administration of anti-PMSG polyclonal antibody can increase the number of unweaned mice. Anti-PMSG polyclonal antibody administration is able to cease PMSG action, resulting in the production of mature follicles and the number of ovulated oocytes is increased. Abnormalities in hormonal balance, fertilization and embryonal development can be eliminated by the provision of anti-PMSG polyclonal antibody. After being mated, higher number of embryo and unweaned mice can be obtained.

CONCLUSION

The administration of anti-PMSG polyclonal antibody 1 hour before LH administration in superovulatory program using PMSG and LH can produce highest number of embryo and unweaned mice compared to that obtained by administration along with or 1 hour after LH administration.

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