CONTENTS

LIST OF ABBREVIATIONS

AI	**Artificial Insemination**
ADLF	**Average diameter of largest follicles**
ADLUF	**Average diameter of largest unovulated follicles**
CL	**Corpus Luteum**
DF	**Dominant Follicle**
ECG	**Equine Chorionic Gonadotrophin**
E2	**Estrogen**
ET	**Embryo Transfer**
FD	**Follicular Diameter**
FN	**Follicular Number**
FSH	**Follicle Stimulating Hormone**
GnRH	**Gonadotrofin Releasing Hormone**
LD	**Largest Diameter**
HMG	**Human Menopausal Gonadotrophin**
LH	**Leutinizing Hormone**
MOET	**Multiple Ovulation and Embryo Transfer**

OPU	**Ovum Pick-Up**
P4	**Progesterone**
pFSH	**Porcine Follicle Stimulating Hormone**
PG	**Prostaglandin (PGF2α)**
PMSG	**Pregnant Mare Serum Gonadotrophin**
SO	**Superovulation**
SOR	**Superovulatory Response**
UOF	**Unovulated Follicle**

SUMMARY AND CONCLUSION

This study was conducted at Animal Production Department, Faculty of Agriculture, Mansoura University in co-operation with Animal Production Research Institute (APRI), Agricultural Research Center, Ministry of Agriculture. The experimental work was carried out at Animal Production Research Stations, Sakha and International Livestock Management Training Center, Sakha (ILMTC), belonging to APRI and located in the north eastern part of the Nile Delta, Kafer El Sheikh Governorate during the period from January to September 2009.

The aim of this study was to determine the effect of PMSG administration at mid-luteal stage of the oestrous cycle, before the selection of the dominant follicle, based on follicular diameter, on the superovulatory response obtained compared with PMSG adminstration on day 10 of oestrous cycle in relation to changes in ovarian characteristics and hormonal profile (P4 and E2) during superovulatory treatment period in Friesian cows.

A total of 20 Friesian cows (450-550 Kg LBW, 3.5-5.5 years old and 1-3 parities) were used as donor cows. Cows were divided into 4 groups (5 animals in each) according to time of PMSG injection. Before treatment, cows in all groups were injected with 2 ml $PGF_2\alpha$ (Estrumate) to start oestrous cycles. Cows in the 1^{st} group (Control, n=5) were i.m. injected with a single dose of

2500 IU PMSG on day 10 of the oestrous cycle. However, in the 2^{nd}, 3^{rd} and 4^{th} groups, cows were injected i.m. with a single dose of 2500 IU PMSG when diameter of the dominant follicle reach 5-7.5 mm (G2, n=5), >7.5-10 mm (G3, n=5) and >10 mm (G4, n=5). After 48 h of PMSG injection in each group, cows in all groups were injected with 2 ml Estrumate and artificially inseminated (AI) when excepted estrous. Ultrasonography device was used during treatment period to record the number of follicles and CL and diameter of the follicles for PMSG injection.

Flushing was conducted 7 days after AI to determine the ovulatory response**.** Collected embryos were evaluated morphologically and were classified into different grades (I, excellent; II, good; III, fair and IV, poor) on basis of their morphological symmetry. Measurements of embryos were recorded (Thickness (µm) of zona pellucida, Diameter (µm) of intrazonal and embryo with its coverings.

Blood samples were collected from all cows in each group. Serum was separated into labeled glass tubes and stored at -20^{o}C until assay of progesterone (P4) and estrogen (E2) hormones. Blood samples were collected on day of oestrus (Day 0), PMSG injection, $PGF_2\alpha$ injection, artificial insemination and flushing.

The obtained results could be summarized as the following:

I. Changes in ovarian characteristics and hormonal profile in experimental groups on treatment days:

Cows in all experimental groups exhibited oestrus activity within 48 h after PGF2α treatment

I.1. Day of oestrus:

1- Overall mean of follicular number (FN) was the lowest in G2 (4.2/cow), moderate in G4 (5.8/cow) and the highest in G1 and G3 (6.6 and 6.4/cow, respectively), but the differences were not significant. Overall mean of FN tended to be insignificantly higher on left than on right side (3.15 *vs.* 2.60/cow).

2- Overall mean of average follicular diameter (AFD) at different sizes was the lower in G1 and G4 (7.3 and 7.8 mm) than in G3 and G2 (8.7 and 9.9 mm), respectively, and was similar on right and left ovarian sides (8.8 and 8.1 mm, respectively). But, the differences were not significant.

3- Overall mean of average diameter of largest follicles (ADLF) ranged between 10.1 and 11.0 mm in different groups and was similar on right and left ovarian sides (10.6 and 10.5 mm, respectively).

4- Overall P4 concentration was <0.5 ng/ml, ranging between 0.14 and 0.32 ng/ml in all groups and reflecting incidence of oestrus and ovulation in cows of all groups. Overall E2 concentration tended to be higher in G1 than in groups (G2, G3 and G4), but the differences among groups were not significant.

I.2. Day of PMSG treatment (pre-PMSG injection):

1- Overall mean of FN ranged between 7.6/cow in G3 and 4.8/cow in G4, while number of follicles was higher in G1 (6.8/cow) than in G2 (6.0/cow) with insignificant differences and was insignificantly higher on right than left ovarian sides (3.25 *vs.* 3.05/cow).

2- Overall mean of AFD was higher ($P<0.05$) in G4 (9.6 mm) than in G2 and G3 (5.6 and 5.4 mm), but did not differ significantly from that in G1 (7.5 mm). Overall mean of AFD was insignificantly higher on right than left ovarian sides (7.4 *vs.* 6.6 mm).

3- Overall mean of ADLF was higher ($P<0.05$) in G4 (11.9 mm) than in G2 and G3 (6.6 and 7.3 mm, respectively), but did not differ significantly from that in G1 (9.8 mm). Overall mean of ADLF was insignificantly higher on right than left ovarian sides (9.1 *vs.* 8.7 mm).

4- Overall P4 concentration was almost above 1.0 ng/ml, being lower in G2 (1.38 ng/ml) than in G1, G3 and G4 (4.72, 3.93 and 3.74 ng/ml), respectively, reflecting presence of complete CL in all cows. However, E2 concentration was higher ($P<0.05$) in G1 (3.50 pg/ml) than in other treated groups, being 1.48, 1.38 and 1.10 pg/ml in G2, G3 and G4, respectively.

I.3. Day of PGF2α treatment (pre-PG and post-PMSG injections):

1- Overall mean of FN ranged between 9.4/cow in G1 and 6.0/cow in G4, while number of follicles tended to be higher in G1 and G3 (8.8 and 9.4/cow) than in G2 and G4 (6.2 and 6.0/cow), respectively, with insignificant group

differences. Overall mean of FN was insignificantly higher on right than left ovarian sides (4.0 *vs.* 3.6/cow).

2- Overall mean of AFD ranged between 7.2 and 9.0 mm, however, the differences were not significant. Overall mean of AFD was insignificantly higher on right than left ovarian sides (8.4 *vs.* 7.4 mm).

3- Overall mean of ADLF ranged between 10.4 and 12.4 mm, however, the differences were not significant. Overall mean of ADLF was insignificantly higher on right than left ovarian sides (11.4 *vs.* 10.2 mm).

4- Overall P4 concentration was lower ($P<0.05$) in G2 (1.87 ng/ml) than in G1, G3 and G4 (6.81, 7.83 and 5.26 ng/ml, respectively). Overall E2 concentration was not affected significantly by treatment, ranging between 1.02 and 2.75 pg/ml) in all experimental groups.

I.4. Day of artificial insemination (AI):

1- Overall mean of FN was the highest (13.0/cow, $P<0.05$) in G3, moderate in G1 (10.0/cow) and the lowest in G2 and G4 (6.2 and 8.6/cow). Overall mean of FN was insignificantly higher on right than left ovarian sides (5.40 *vs.* 4.25/cow).

2- Overall mean of AFD ranged between 8.0 and 9.4 mm, however, the differences were not significant and were nearly similar on the right and left ovarian sides (8.6 *vs.* 8.7 mm).

3- Overall mean of ADLF ranged between 11.7 and 13.4 mm, however, the differences were not significant and were nearly similar on the right and left ovarian sides (12.3 *vs.* 12.1 mm).

4- Overall P4 concentration was almost less than 1.0 ng/ml in all groups, ranging between 0.33 and 0.40 ng/ml. However, E2 concentration was the highest in G1 (5.53 pg/ml, $P<0.05$), followed by G3 and G4 (3.70 and 3.48 pg/ml, respectively), and the lowest in G2 (1.62 pg/ml, $P<0.05$).

I.5. Day of flushing:

1- Overall number of unovulated follicles (UOF) was insignificantly lower (1.4/cow) in G1, moderate in G2 and G3 (2.6 and 2.4/cow) and the highest in G4 (3.6/cow) and was insignificantly higher on right than left ovarian sides (1.55 *vs.* 0.95/cow).

2- Overall mean of average diameter of unovulatory follicles at different sizes was the highest ($P<0.05$) in G4 (13.1 mm), moderate in G2 and G3 (9.8 and 7.7 mm) and the lowest in G1 (3.7 mm) and was insignificantly higher on the right than left ovarian sides (10.0 *vs.* 7.0 mm).

3- Overall mean of average diameter of largest unovulated follicles was higher ($P<0.05$) in G4 and G2 (15.2 and 11.2 mm) than in G3 and G1 (8.8 and 4.2 mm). While, it was higher on the right than left side (11.4 *vs.* 8.2 mm).

4- Overall number of CLs markedly increase to the maximal number, being higher ($P<0.05$) in G1 and G3 (8.0 and 7.4/cow) than in G2 and G4 (1.0

and 3.0/cow) and was insignificantly higher on right than left ovarian sides (2.55 *vs.* 2.30/cow).

5- Overall P4 concentration was the highest (P<0.05) in G1 and G3 (13.51 and 18.29 ng/ml), moderate in G4 (5.88 ng/ml) and the lowest (less than 1.0 ng/ml) in G2. However, E2 concentration was insignificantly higher in G4 (3.86 pg/ml) than in G1, G2 and G3 (1.52, 1.84 and 1.08 pg/ml), respectively.

II. Changes in ovarian characteristics and hormonal profile during treatment period in each experimental group:

1- Average FN in all groups showed marked increase, reaching the maximal number on day of AI, and then reduced to the minimal numbers on day of flushing. These changes were significant (P<0.05) in G1 and G3 and insignificant in G2 and G4 during treatment period.

2- On day of AI, FN was higher in cows of G1 and G3 than in G2 and G4.

3- Average FD showed changes (P<0.05) only in G2, being the highest on day of flushing and the lowest on day of PMSG. However, changes in follicular diameter in G1, G3 and G4 were not significant during treatment period.

4- Average LD increased to the maximal values on day of AI in G1 and G3 and on day of flushing in G2 and G4. However, the changes were significant (P<0.0) only in G1 and G2.

5- The present results showed insignificant differences in DF of the 1st and 2nd waves in all experimental groups.

6- Reaching the FN, AFD and ALD of follicles in G1 and G3 to the maximal values on day of AI was in parallel with increasing E2 concentration to the highest level and P4 to the lowest level on day of AI.

III. Evaluation of embryo production:

1- Day of PMSG treatment averaged 7.2, 9.6 and 8.2 for cows in G2, G3 and G4, respectively.

2- All cows in G1 and G3 (100%) produced CL *vs.* 60 and 80% of cows in G2 and G4, respectively.

3- Response rate of embryo production was only detected in G1 and G3, being 60 and 80%, respectively. No embryos were recovered from cows in G2 and G4.

4- Average number of CLs/cow was higher ($P<0.05$) in G1 and G3 (8.0 and 7.4) than in G2 and G4 (1.0 and 3.0), respectively.

5- Cows in G1 and G3 showed the highest ($P<0.05$) total response (CLs and UOF), being 9.4 and 9.8/cow, respectively.

6- Averages number of total and transferable embryos were higher ($P<0.05$) in G3 by about 42 and 44% than in G1.

7- Recovery rate of total and transferable embryos were higher in G3 than in G1 (73.0 and 70.0% *vs.* 47.5 and 45.0%, respectively).

8- Yield of embryos at morula and blastocyst stages was higher in G3 than in G1. Cows in G3 showed the highest distribution of embryos at morula stage (37.0%) versus the highest distribution of compact morula in G1 (42.1%).

9- Cows in G3 increased yield of excellent embryos/cow (2.2 *vs.* 1.0) and good embryos/cow (3.0 *vs.* 1.8) as compared to those in G1. Excellent and good embryos represented 40.7 and 55.6% in G3, being higher than 26.3 and 47.4% in G1. The differences in production of embryos at different stages or grades were not significant.

10- Zona pellucida was thicker ($P<0.05$) in embryos of G1 (21.7, 21.3 and 20 μm) than in G3 (18.8, 17.2 and 18.2 μm) at morula, compact morula and in transferable embryos, respectively. However, it had close values in embryos at blastocyst stage.

11- Intrazonal diameter reduced ($P<0.05$) in embryos of G1 at all stages and transferable embryos as compared to G3. However, embryos at blastocyst stage and transferable embryos showed significantly ($P<0.05$) higher total diameter in G3 than in G1.

The present study may conclude that the potentiality of PMSG injection timing to induce hight superovulatory response in cattle. Under the experimental conditions of present study, appropriate time for superovulation in Friesian cows with 2500 IU of PMSG was when diameter of the dominant follicle reached a rang from >7.5-10 mm (day from 8-10 of synchronized oestrus) to reflect the highest ovulatory response, the best number of transferable embryos of excellent grade.