OVARIAN-GENITAL CHARACTERISTICS AND EMBRYO RECOVERY RATE OF SUPEROVULATED NEW ZEALAND WHITE RABBIT DOES

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ABSTRACT

This study aimed to evaluate the effects of slaughter time (after 24 or 72 hours from matting) on ovarian and genital characteristics of superovulated female rabbits and quality and recovery rate of embryos. A total of 38 New Zealand White rabbit females were used in this study. All females were non-parous averaging 5 months of age and ranging between 2.7-2.9 kg LBW. All animals were superovulated by i.m. injection with 150 IU PMSG (Foligon), followed by 75 IU HCG (Pregnyl) at the time of natural mating (72 h after PMSG injection). All treated females were slaughtered either 24 (G1, n=18) or 72 (G2, n=20) hours after mating, where ovaries and reproductive tract of each female were separated to determine weight and length of the ovary, oviduct and uterus as well as number of follicles and corpora lutea (CL) on the ovarian surface. Embryos were collected from superovulated does in both groups Embryos were recovered from uterine horns by flushing with phosphate buffer solution with 10% fetal calf serum at room temperature (20-25 °C). Thereafter, the recovered embryos were counted and examined for different embryo stages using an inverted microscope fitted with a calibrated eye piece micrometer reflected on stereomicroscope. Results show that absolute ovarian weight increased (P<0.01) by 56% (0.26 vs. 0.41 g), ovarian weight relative to LBW increased (P<0.01) by 66.6% (0.09 vs. 0.15 g/kg) and ovarian length increased (P<0.05) by 18.5% (1.31 vs. 1.55 cm) in G2 compared with G1. Number of follicles/ovary did not differ between G1 and G2. However, number of CL/ovary increased (P<0.001) in G2 than in G1(6.4 vs. 8.2/ovary). There were insignificant differences in absolute weight, relative weight to LBW and length of both oviduct and uterus between G1 and G2. Number of embryos recovered per ovary was nearly similar in G1 and G2 (4.30 and 4.35/ovary). However, recovery rate was higher (P<0.01) in G1 than in G2 (66.7 vs. 53%). The highest distribution of embryos in G1 was recorded in 2-cell stage (zygote, 84.4%), followed by 5.2% in 4-cell stage, while 10.4% were in abnormal type vs. 84% in compacted morula stage, 3.4% in early morula stage, 2.9% in early blastocyst stage and 9.8 in abnormal type.

The current study may conclude that rabbit does should be superovulated by PMSG and HCG and slaughtered after 24 h of mating when embryos at early stage are needed for banking females of high genetic merit, and slaughtered after 72 h of mating if embryos at morula stage were needed for embryo transfer.

Keywords: Doe rabbits, superovulation, ovary, genitalia, embryo, recovery rate.

INTRODUCTION

Animal biotechnology used in conjunction with traditional animal breeding programmes can significantly improve the rate of genetic progress in domestic animals. But while improvements have been made, genetic diversity is being reduced as native animal populations become less isolated and breeding programmes become more globally oriented. Banking of desirable animals for unique genetic, production and disease resistance traits

Dorra, Tork M.I. et al.

will facilitate acquisition and characterization of potentially useful germplasm, ensure genetic variation through preservation of selected stocks, and facilitate use of useful germplasm in research and industry in the future. There are methods that adequately, but not optimally, preserve germplasm and embryos from genetically superior animals of most of livestock species (Dobrmsky, 1997).

Superovulation is considered to be an efficient economic method for producing additional embryos or oocytes from females of high genetic merit. The exogenous hormones as stimuli for ovulation induction in higher number of follicles (Superovulation) included both FSH and eCG (Schmidt *et al.*, 1992 and Joly *et al.*, 1996), In previous studies with rabbits, Besenfelder (1991) and Kauffman *et al.* (1998) reported a higher superovulatory response to eCG treatment than to FSH treatment. However, Rebollar *et al.* (2000) observed a higher superovulatory response to FSH treatment than to eCG treatment than to eCG treatment. Altough many authors have reported that eCG treatment improves the receptivity of rabbit does (Mirabito *et al.*, 1994, Theau-Clèment and Lebas, 1996 and Theau-Clèment *et al.*, 1998), others found a non-significant effect of eCG treatment on receptivity (Bourdillon *et al.*, 1992) and negative effects on conception rate, fertility and viability rate at birth (Canali *et al.*, 1991, Alabiso *et al.*, 1994 and Maertens and Luzi, 1995).

This study aimed to evaluate the effects of slaughter time (after 24 or 72 hours from matting) on ovarian and genital characteristics of superovulated does and on survival, quality and recovery rate of embryos.

MATERIALS AND METHODS

The present study was carried out at IVF Laboratory, International Livestock Management Training Center (ILMTC), Sakha, Kafer El-Sheikh Governorate, belonging to the Animal Production Research Institute, Ministry of Agriculture in cooperation with Poultry Production Department, Faculty of Agriculture, Mansoura University, during the period from September 2005 to June 2006.

Animals:

A total of 38 New Zealand White rabbit females was used in this study. All females were non-parous, averaging 5 months of age and between 2.7-2.9 kg LBW. All females were housed in individual galvanized wire batteries arranged in raw, located in conventional building ventilated by electric fans. Females were fed *ad-libitum* on a commercial pelleted complete feed diet, containing 2.3% fat, 16.5% crude protein, 13% crude fiber, 17% ash and 2740 Kcal digestible energy (DE)/kg on dry mater basis.

Superovulation protocol:

Superovulation was carried out by intramuscular injection of each female with 150 IU PMSG (Foligon) in legs, followed by 75 IU HCG (Pregnyl) in ear vein at the time of natural mating (72 h after PMSG injection). Embryos were collected by slaughtering all treated females either 24 or 72 hours after mating, where ovaries and reproductive tract of each female including oviduct, horns and uterus were separated for morphological measurements. Weight (g) and length (cm) of the ovary, oviduct and uterus as well as

number of follicles and corpora lutea on the ovarian surface were recorded after slaughtering.

Embryo recovery:

Embryos were collected from superovulated does slaughtered after 24 or 72 hours of mating and HCG injection. Embryos were recovered either after 24 hours from 18 does or after 72 hours from 20 does.

Embryos were recovered from uterine horns by flushing with phosphate buffer solution (PBSC) with 10% fetal calf serum (FCS) at room temperature (20-25 °C). Thereafter, the recovered embryos were counted and examined for different embryo developmental stages using an inverted microscope fitted with a calibrated eye piece micrometer reflected on stereomicroscope.

Statistical analysis:

The statistical analysis was performed using a software package (SAS, 2000). Frequency distribution of different embryo stages recovered from does in G1 and G2 were analyzed using chi square analysis. The obtained data for Ovarian characteristics of superovulated does in G1 and G2 were subjected to t test.

RESULTS AND DISCUSSION

Superovulation protocol of rabbit does:

Ovarian characteristics of superovulated does:

Ovarian characteristics of superovulated NZW does slaughter after 24 or 72 hours of mating are presented in Table (1). The present results revealed that averages of absolute weight, weight relative to LBW and length of the ovary were significantly (P<0.01, P<0.01 and P<0.05, respectively) higher in does slaughtered after 72 than 24 hours of mating, according to protocol of superovulation of these does.

Absolute ovarian weight of does slaughtered after 72 significantly (P<0.01) increased by about 56% as compared to that in does slaughtered after 24 hours of mating. The corresponding increases in relative weight and length of the ovary in does slaughtered after 72 than 24 hours were 66.6 and 18.5%, respectively (Table 1).

It is of interest to note that the pronounced increase in average absolute weight of the ovaries in does slaughtered after 72 hours of mating was indicated in term of significant (P<0.01) increase in ovarian weight relative to LBW as compared to does slaughtered after 24 hours of mating.

The present ovarian weight of superovulated does in both groups is heavier than the average ovarian weight of multiparous NZW does, being 0.24 g as reported by Hafez and Rajakoski (1964).

Item	G1	G2	T-Value	P-Value
Live body weight (kg)	2.74±0.065	2.80±0.076	0.62	0.540
Ovarian weight (g)	0.26±0.028	0.41±0.038	3.12	0.004
Ovarian weight (g/kg LW)	0.09±0.010	0.15±0.010	2.99	0.005
Ovarian length (cm)	1.31±0.035	1.55±0.084	2.65	0.013
Number of follicles/ovary	4.00±0.325	4.50±0.245	1.36	0.183
Number of CL/ovary	6.40±0.397	8.20±0.327	3.57	0.001

Table (1): Ovarian characteristics of superovulated does in G1 and G2.

This was in accordance with the results of Gorabner *et al.* (1987), who reported that using stimuli to induce ovarian activity was found to increase ovarian weight of doe rabbits. Least square means for pair ovarian weight in non-parous NZW injected with 6-methoxybenzoxazlinone at three levels were 0.44, 0.34, 0.59 and 0.42 g as compared to 0.35 g in the control does, respectively.

In nearly agreement with the obtained results, Fukunari *et al.* (1990) found that the ovarian weight of mature Japanese White rabbit aged 4 months slaughtered 72 h after treatment with PMSG (50 IU) was significantly higher in treated than untreated does. Also, Gosalves *et al.* (1994) recorded that females treated with PMSG had a heavier ovary weight than those injected with the saline solution, being 0.60 and 0.18 g, respectively). On the other hand, El–Gaafary *et al.* (1994) found non-significant differences between ovarian weight for NZW rabbit does injected with HCG (50 IU) and untreated rabbits, being 0.83 vs. 0.67 g.

The significant increase in ovarian weight of does slaughtered after 72 h of mating was mainly attributed to significant increases in ovarian length (P<0.05) and in number of CL/ovary (P<0.001, Table 1). In this respect, Gosalves *et al.* (1987) found that California multiparous does had heavier ovaries than non-parous ones (0.37 vs. 0.29 g) and ovaries weight increased with increasing number of litters as a result of increasing number of CL on ovaries.

On the other hand, number of follicles/ovary did not differ significantly between does slaughtered after 72 or 24 hours of mating, but may tend to be higher than that in untreated ones. This trend may indicate similar effect of superovulation protocol on development of not ovulated follicles having shorter diameter and small cavity (Table 1).

Fukunari *et al.* 1990) found that total number of follicles was significantly higher in immature Japanese White rabbits treated with 50 IU PMSG than untreated does. The same trend was obtained by Gosalves *et al.* (1994), who mentioned that does treated with PMSG had a greater number of antral follicles than those injected with saline solution followed by 20 mg Fertagyl (LHRH), 2 days later.

Also, Bonhoff and Adams (1985) found an increasing degree and number of follicular development when rabbit does treated with HCG or LHRH. In supporting the previous results, El-Gaafary *et al.* (1994) found that the number of mature follicles on ovaries of NZW rabbits increased to 16 follicles/ovary when doe rabbits were injected with 50 IU of HCG.

Generally, number of follicles was affected by type (Peinado *et al.*, 1995) and doses of hormones (Mehaisen, 2005). Marai *et al.* (1994) found that injection of rabbits with 50 IU of HCG gave the highest number of follicle/doe than 100 IU dose. The improvement in ovarian activity of does injected with 50 IU may be attributed to the effectiveness of that level which exerts mostly LH and slightly FSH- like effects and gets in circulation quickly before the release of the endogenous LH. While the higher level (100 IU HCG) may inhibit the release of LH and FSH hormones through the negative feed back mechanism.

J. Agric. Sci. Mansoura Univ., 32 (8), August, 2007

The observed significantly increase in number of CL/ovary in does slaughtered after 72 than 24 hours of mating was indicated by Peinado *et al.* (1995), who found that average number of CL was higher (13.0) in does slaughtered after 72 than 14 h from insemination (8.0). NZW does were treated with 25 IU of PMSG followed at 48 h either with 50 IU of hCG. All does were previously artificially inseminated to avoid endogenous LH surge. Half of the animals were killed at 72 h after the hormone administration, and the remaining half were killed at 14 days.

The recorded number of CL in this study (6.4/ovary or 8.2/ovary) was nearly similar to 17.6/doe treated with 72 IU of PMSG and 36 IU of HCG (in a single intramuscular injection, Gravance, 1994). However, it was higher than that reported by Gosalves *et al.* (1994), who estimated 11.0/doe for rabbit female aged 17 weeks and injected with 100 IU PMSG, followed by 20 mg LHRH (Fertagyl) 2 days later. On the other hand, Lee *et al.* (1991) observed higher number of ovulation points averaging 19.2/female rabbit superovulated with PMSG.

Generally, number of CL/doe was affected mainly by dose of hormonal treatment, being 7.4 to 10.3 in different rabbit breeds treated with 0.2 or 0.4 ml GnRh (El-keraby *et al.*, 1991) and 19.2, 15.5 and 12.2 for 200 IU of eCG, 50 IU eCG and cotrol, respectively (García-Ximénez and Vicente, 1990). Also, using different types of hormonal administeration to induce ovulation (superovulation) for the R and V line rabbits, number of ovulation sites was 15.3 and 15.9 (Mehaisen, 2005); 14.3 and 13.8 (Viudes-de-Castro *et al.*, 1995) and 13.5 and 13.2 (Vicente *et al.*, 2003) for R and V lines, respectively. These differences may be due to the effect of the superovulation treatment that was used.

Genital characteristics of superovulated does:

Means and analysis of variance of genital characteristics of superovulated NZW does slaughtered after 72 or 24 hours of mating are presented in Table (2). Data shown in Table (2) cleared insignificant differences in averages of absolute and relative weight and length of both oviduct and uterus between does slaughtered after 72 or 24 hours of mating.

Item	G1	G2	T-Value	P-value
Number of treated does	18	20	-	-
Oviduct weight (g)	0.504±0.04	0.586±0.05	1.21	0.237
Oviduct weight (g/kg LW)	0.18±0.020	0.21±0.021	0.98	0.334
Oviduct length (cm)	9.531±0.42	9.753±0.44	0.36	0.722
Uterine horn weight (g)	3.449±0.17	3.877±0.39	0.98	0.336
Uterine horn (g/kg LW)	1.26±0.040	1.38±0.056	1.21	0.237
Uterine horn length (cm)	8.128±0.26	7.884±0.39	0.52	0.608

Table (2): Genital characteristics of superovulated does in G1 and G2.

This finding was expected where the effect of superovulation protocol is mainly effective on the ovaries as target organs and secondarily affects tunica mucosa of oviduct and myometrium layer of the uterine horns. Such effects were represented in a tendency of higher absolute weight and relative weight of oviduct and uterus in does slaughtered after 24 hours than 72 hours of mating. However, length of oviduct slightly increased and uterine horn

length decreased in does slaughtered after 72 hours as compared the those slaughtered after 24 hours of mating (Table 2).

Recovery rate of embryos:

Characteristics of embryos collected from oviduct and fallopian tube are presented in Table (3). The present data revealed that number of embryos recovered per ovary was nearly similar in superovulated does slaughtered after 24 or 72 hours of mating (4.35 and 4.30 embryos/ovary). However, recovery rate calculated on the basis of number of CL (Table 2) was significantly (P< 0.01) higher for does slaughtered after 24 h (67.2%) than those slaughtered after 72 h (53%).

Table (3): Number of embryos and embryo recovery rate (9	%) of does in
G1 and G2.	-

Item	G1	G2	T- Value	X ²	P- Value
Number of treated does	18	20	-	-	-
Total number of CL/doe	6.4±0.39	8.2±0.32	3.57	-	0.001
Number of embryos/ovary	4.30	4.35	1.36	-	0.183
Recovery rate (%)**	66.7	53.0	-	10.37	0.001

* Collected from oviduct and fallopian tube.

** Calculated as (number of collected embryos/number of CL) x 100

The observed difference in recovery rate of embryos was mainly related to significantly (P<0.01) greater number of CL in does slaughtered after 72 than 24 hours of mating, in spite of the nearly similarity in number of collected embryos per ovary between both groups of does.

In comparing the present recovery rate with other studies, the response to superovulation treatment depends mainly on the type of hormone and the method of administration. In this respect, El-keraby *et al.* (1991) found that lower recovery rates of embryos collected from does treated with 0.2 or 0.4 ml GnRh ranging between 8.9-36.7% in treated groups. However, higher embryo recovery in rabbits was reported when PMSG was injected intramuscular (Besenfelder *et al.*, 2000) or when FSH was used for superovulation (Joly, 1997).

Also, Mehaisen (2005) evaluated the effect of different doses of eCG administered subcutaneously (0, 50 and 200 IU) and the hormonal induction of ovulation (GnRH or hCG) on recovery rate in R and V line-rabbit does. Administration of 200 IU of eCG significantly decreased recovery rate (28.8 vs. 47.7 and 48.7) as compared to 50 IU and 0 IU eCG, respectively.

Moreover, the response to superovulation treatment differs between rabbit breeds (Bolet *et al.*, 2000). Using different types of hormonal administeration to induce ovulation (superovulation), recovery rate was 43.2 and 40.3% (Mehaisen (2005), 63 and 82% (Viudes-de-Castro *et al.*, 1995) and 77 and 74% (Vicente *et al.* (2003) for R and V line rabbits, respectively.

The obtained number of recovered embryos/ovary in this study ranged between 4.30-4.35, being 8.6-8.7 per doe (Table 3). However, Al-Hasani *et al.* (1984) recorded that the average number of oocytes recovered from ovarian follicles by puncture was 11.7 oocytes/ rabbit during different times after p-LH injection.

The difference in recovery rate of embryos was affected by type of hormonal treatment. Bourdage and Halbert (1988) observed that the use of 50 IU hCG provoked an alteration in oviductal motility causing an accelerated transit of the embryos. This acceleration could be responsible for the higher number of embryos found in the uteri of does treated with hCG.

Concerning the differences in frequency distribution of embryos collected from both doe groups at different stages (Table 4), it is of interest to note that all embryos were in 2-4 cell stage for does slaughtered after 24 hours of mating, while all embryos were in morula and blastocyst stages in those slaughtered after 72 hours of mating. Such phenomenon was expected, whereas the time elapsed from ovulation and fertilization was 24 hours, which allowed to embryos to develop within the fallopian tube up to 8-cell stage. However, in does slaughtered after 72 hours of mating, embryos developed over 8-cell stage to be in compacted morula and early blastocyst stages.

Result presented in table (4) show that, in does slaughtered after 24 hours of mating, the highest distribution of embryo was recorded in 2-cell stage (zygote, 84.4%), which considered as very early embryonic stage immediately post-fertilization. Frequency distribution of embryos in 4-cell stage was the lowest, being 5.20%, while 10.4% of embryos were abnormal. However in does slaughtered after 72 hours of mating, embryos in compacted morula stage showed the highest distribution (84.0%), while those in early morula stage were more frequent than those in early blastocyst stage (3.4 vs.2.9%).

Some authors attributed the differences in frequency distribution of embryonic stages to protocol of superovulation. In this line, Peinado *et al.* (1995) treated NZW does receiving 25 IU of PMSG followed at 48 h either with 50 IU of r-hLH or hCG. All does were artificially inseminated and killed at 72 h after the hormone administration. The results indicated that embryonic development was more homogeneous in the animals receiving r-hLH (8 to >or = 16 cells) compared to those receiving hCG (2 to > or = 16 cells). The median of embryos still in oviducts at 72 h was significantly higher in the hCG than in the r-hLH group (group (6 vs. 0, P= 0.41).

Embryo stage	G1		G2	
	N	%	N	%
2-cell embryo	130	84.40	-	0.0
4-cell embryo	8	5.20	-	0.0
8-cell embryo	-	.00	-	0.0
Abnormal embryo	16	10.40	-	0.0
Total	154	100	-	0.0
Compacted morula	-	0.0	146	84.0
Early morula	-	0.0	6	3.4
Early blastocyst	-	0.0	5	2.9
Abnormal embryo	-	0.0	17	9.8
Total	-	0.0	174	100

 Table (4): Frequency distribution of different embryo stages recovered from does in G1 and G2.

On the other hand, abnormal embryos distributed 9.8%, which was slightly lower than that observed in does slaughtered after 24 hours of mating (10.4%). This difference between both doe groups in distribution of abnormal embryos may be related to the higher measurements (weight and length) of genitalia in does slaughtered after 72 than 24 hours of mating, which may facilitate the embryo recovery. Quality of embryo was affected by type of hormonal treatment.

Peinado *et al.* (1995) treated NZW does with 25 IU of PMSG and either with 50 IU of r-hLH or hCG at 48 h later. All does were artificially inseminated and killed at 72 h after the hormone administration. The results indicated that the percentage of good-quality, intermediate-quality and degenerated embryos was 71.4, 25 and 0% in the r-hLH vs. 33.3, 33.3 and 33.3% in hCG group, respectively. However, when Vicente and Garcia-Ximenez, 1991) recovered embryos 64-66 h post-coitus by flushing the oviducts and uteri, they found that normal embryos were obtained from all hCG-treated donors (G2) and from 69% of untreated does (G1). About 21% of embryos were recovered from the uterus of G2 in contrast to 6% from G1.

On the other hand, Mehaisen (2005) observed no significant differences in embryo recovery between hCG and GnRH treatments. Vicente and Garcia-Ximenez (1991) found higher proportion of morphologically abnormal embryos in the uterus (37%) in comparison with the proportion in the oviduct (5.5%), which could be attributed to the effects of an inadequate uterine environment (Beier, 1976).

The current study may conclude that rabbit does should be superovulated by PMSG and HCG and slaughtered after 24 h of mating when embryos at early stage are needed for banking females of high genetic merit, and slaughtered after 72 h of mating if embryos at morula stage were needed for embryo transfer.

REFERENCES

- Alabiso, M.; A. Bonanno; M.L. Alicata and B. Portolano (1994). Trattammento "differenziato" con PMSG su coniglie inseminate artificialmente, Coniglicoltura .,31: 25–30.
- Al-Hasani, S.; J. Hahn ; K. Dankowski and V. Schneider (1984). In vitro fertilization and embryo transfer of rabbit oocytes collected at different time after P-LH injection. European J. Obstetrics, Gynecology and Reproductive- Biology. 17: 417-423.
- Beier, H.M. (1976). Uteroglobin and related biochemical changes in the reproductive tract during early pregnancy in the rabbit .J. Reprod. Fertil. Suppl., 25:53-69.
- Besenfelder, .U (1991) .Untersuchungen zum Einfluβ von β-carotin auf Fertilitätsparameter in Embryo transfer programmen beim Kaninchen. Thesis Munich. Germany.
- Besenfelder, U.; C. Haas and G. Brem (2000). Reproduction technology and gene transfer in rabbits, Proceedings of 7th World Rabbit Congress, A Valencia, Spain., 37–59.

- Bolet, G., J.M. Brun, M. Monnerot, F. Abeni, C. Arnal, J. Arnold, D. Bell, G. Bergoglio, U. Besenfelder, S. Bosze, S. Boucher, N. Chanteloup, M.C. Ducourouble, M. Durand-Tardif, P.J. Esteves, N. Ferrand, A. Gautier, C. Haas, G. Hewitt, N. Jehl, T. Joly, P.F. Koehl, T. Laube, S. Lechevestrier, M. Lopez, G. Masoero, J.J. Menigoz, R. Piccinin, G. Queney, G. Saleil, A. Surridge, Van Der Loow, J.S. Vicente, M.P. Viudes de Castro, J.S. Virag and J.M. Zimmermann (2000). Evaluation and conservation of European rabbit (Oryctolagus cuniculus) genetic resources. First results and inferences, Proceedings of 7th World Rabbit Congress, vol. A Valencia, Spain., 281–315.
- Bonhoff, A.J and C.E. Adams (1985). Relationship of hormonally induced developmental changes in preovulatory follicles of the rabbits . Laboratory Animals. 19:27-31.
- Bourdage, R. J. and S. A .Halbert (1988). Distribution of embryos and 500µm microspheres in the rabbit oviduct: Control for acute motion analysis during transport. Biol. Reprod., 38: 282-291.
- Bourdillon, A.; F. Chmitelin, D. Jarrin, V. Parez and H. Rouillere (1992). Effects of a PMSG treatment on breeding results of artificially inseminated rabbits, Proceedings of 5th World Rabbit Congress., 530– 537.
- Canali, C.; C. Boiti ; D. Zampani ; C. Castellini and M. Battaglini (1991). Correlazione tra fertilitá e titolo anticorpale anti-PMSG di coniglie trattate ripetutamente con gonadotropine nel corso della loro carriera riproduttiva, Atti IX Congresso Nazionale ASPA., 671–678.
- Dobrmsky, J.R (1997). Cryopreservation of pig embryos .J. Reprod. And Fertil. Supplem., 52:301-312.
- El-Gaafary, M.N; I. F.M.marai; K. A.O. Yamani ; F.R. Abd-allah ; R.S. Amin and H.M.E. El- Kelawy (1994). Growth performance and reproductive efficiency of weaned NZL rabbits as affected by HCG injection. Proceeding of the first international conference held in Cairo , Egypt, 6-8 Septemper, 8:321-331.
- El-keraby, F.; Tork M.Dorra; A.I. Abd El-Ghani and S.A. Fawzy (1991). Effect of GnRh administration rate and embryo recovery in some rabbits breeds. J.Agric. Sci. Mansoura Univ.16(8);1749-1757.
- Fukunari, A.; T.Maeda ; T. Terada and Y. Tsutsumi (1990). The recovery of follicular oocytes from the rabbit ovary during prepubertal age and in vitro maturation of their oocytes. Japanese J. Anim. Reprod., 36:1-8.
- García-Ximénez, F. and J.S. Vicente (1990). Effect of PMSG treatment to mating interval on the superovuatory response of primiparous rabbits, J. Appl. Rabbit : 13 :71–73.
- Gorabner, M.A.; P.R.; Cheeke; N.M.; patton and R.J.; Nachman (1987). The effect of 6-methoxybenzoxazolinone (6-MBOA) on ovarian and uterine weight in nonporous rabbit.J.Appl. Rabbit Res., 10:62-64.
- Gosalves, L.E.; J.M.R .Alvarino and P.T.M . Diaz (1994). Influence of age ,stimulation by PMSG or flushing on the ovarian response LHRHa in young rabbit females. World- Rabbit- Science. 2:41-45.

- Gosalves, L.E.; J.U.; Rodriguez.; P. Diaz and S. Gomez (1987). Ovarian and body weight changes in rabbit before and after parturition. J. Appl. Rabbit Res., 10:126-129.
- Gravance, C.G. (1994). Effect of single combined dose of pregnant mares serum gonadotrophin and human chorionic gonadotrophin on ovulation rate in young rabbits. Anim. Reprod. Sci., 34:299-305
- Hafez, E.S.E and E.Rajakosaki (1964). Growth and survival of blastocysts in the domestic rabbit. II. Quantitive effect of oxygenous progesterone following ovariectomy. J. Reprod. Fertil., 1:241.
- Joly, T. (1997). Etablissement dune cryobanque de semence ou dembryons pour la conservation ex situ de la diversité génétique chez les mammifères domestiques: Lexemple du lapin (Oryctolagus cuniculus). Thèse, Institut National des Sciences Appliquées de Lyon, France.
- Joly, T; J.C. Vicente; M.F. Theau-Cle` ment ;F. Garcı´ a-Xime´ nez ; U.Besenfelder and j.P. Renard (1996).Cryopreservation of genetic resources in rabbit species: practical application.In: Lebas F(ed.), 6th World Rabbit Congress, INRA Toulouse Research Center, France, Vol. 2, pp. 293–298.
- Kauffman, R.D.; P.M. Schmidt, W.F. Rall and J.M. Hoeg (1998). Superovulation of rabbits with FSH alters in vivo development of vitrified morulae. Theriogenology., 50: 1081–1092.
- Lee, T.S; K.T. Lim and J.S. Na (1991). Superovulation treatment and short term storage of embryos in domestic rabbit . Korean J. Anim. Sci., 33: 18-24.
- Maertens, L. and F. Luzi (1995). Note concerning the effect of PMSG stimulation the mortality rate at birth and the distribution of litter size in artificially inseminated does, World Rabbit Sci., 3: 57–61.
- Marai, I.F.M.; K.A.O.Yamani.; M.N.El-Gaafary and H.M.E.; El- Kelawy (1994). Effects of human chorionic gonadotrophin on reproductive performance in female New Zealand white rabbits. Proceeding of the first international conference held in Cairo, Egypt, 6-8 Septemper, 8:355-365.
- Mehaisen, G.M.K ; J.S. Vicentea ; R. Lavaraa and M.P. Viudes-de- astrob (2005). Effect of eCG dose and ovulation induction treatments on embryo recovery and in vitro development post-vitrification in two selected lines of rabbit does. Animal Reproduction, Science ., 90: 175-84.
- Mirabito, L. Galliot and C. Souchet (1994). Programme lumineux ou PMSG pour améliorer la réceptivité des lapines, 6es Journées de la Mirabito Recherche Cunicole en France. 169–178.
- Peinado, J.A.; I. Molina; M. Pla; J.A. Tresguerres and A. Romeu (1995). Recombinant-human luteinizing hormone (r-hLH) as ovulatory stimulus in superovulated does. J Assist Reprod Genet., 1: 61-64.
- Rebollar, P.G.; E. Ubilla, P.L. Lorenzo, M. Sánchez-Dávil, J. Sánchez, L. Tucker and J.M.R. Alvaiño (2000). Ovulation and embryo implantation rate in synchronized artificial inseminated multiparous lactating does, Proceedings of 7th World Rabbit congress,. A Valencia, Spain., 239– 244.

SAS,(2000). SAS Institute Inc. SAS User's Guide, Statistics. Cary, NC.

- Schmidt, P.M.; V.M. Hollifield; X. Lin and D.E. Wildt (1992). Induced ovulation and adequate embryo recoveries in New Zealand white rabbits treated with a low PMSG/HCG dose or single, daily injections of FSH-P. Theriogenology., 37: 293.
- Theau-Clèment et al., M. Theau-Clèment, F. Lebas, B. Poujardieu and P. Mercier (1998). Journées de la Recherche Cunicole en France, Effet de différentes doses de PMSG sur linduction de la réceptivité sexuelle et la productivité des lapines conduites en insémination artificielle, pp. 221–224.
- Theau-Clèment, M. and F. Lebas (1996). Effect of a systematic PMSG treatment 48 h before artificial insemination on the productive performance of rabbit does, World Rabbit Sci., 4: 47–56.
- Vicente, J.S. and F. Garcia-Ximenez (1991). Effects of hCG, treatment on morula recovery in the rabbit and their survival after synchronous transfer. Anim. Reprod. Sci., 24:347-353.
- Vicente, J.S.; M.P. Viudes-de-Castro; M.L. García and M. Baselga (2003). Effect of rabbit line on a program of cryopreserved embryos by vitrification, Reprod. Nutr. Dev., 43: 137–143.
- Viudes-de-Castro, M.P.; F. García-Ximénez and J.S. Vicente (1995). Embryo recovery from eliminating does of three selected rabbit strains for an embryo bank, Investigación Agraria: Producción y Sanidad Anim., 10: 145–152.

خصائص المبيض والقناة التناسلية ومعدل الحصول على الأجنة من أرانب النيوزلندي الأبيض المحدث لها تعدد التبويض ترك محمد إبراهيم دره* ، مغازى السيد علي المغازى* ، حاتم عبد السلام جاد** ، محمود حسن ربيع* و هائى نبيل فهيم* * قسم أنتاج الدواجن- كلية الزراعة – جامعة المنصورة ** معهد بحوث الإنتاج الحيواني – مركز البحوث الزراعية

تهدف الدراسة لمعرفه مدي تأثير وقت جمع البويضات بعد التلقيح عن طريق ذبح إنات الأرانب بعد ٢٤ و ٢٢ ساعة من التلقيح على خصائص المبيض و القياسات التناسلية بعد أجراء تعدد التبويض ومدى تأثيرها علي جوده وحيوية الاجنه الناتجة منها استخدم في هذه الدراسة ٣٨ من إنات النيوزلندي الأبيض لم يسبق لها الولادة ومتوسط أعمارها ٥ شهور وأوزانها ٢,٧- ٢,٩ كجم /وزن حي.

أجريت عمليه تعدد التبويض عن طريق حقن هرمون الفوليجون في العضل بمعدل ١٥٠ وحده دوليه متبوعة بالحقن بالبريجنيل بمعدل ٧٥ وحده دوليه في نفس وقت التلقيح الطبيعي بعد ٢٢ ساعة من الحقن بالفوليجون . جميع الأمهات المعاملة ذبحت بعد ٢٤ ساعة (المجموعة الأولي وعددها ١٨ أم وبعد ٢٢ ساعة (المجموعة الثانية وعددها ٢٠ أم)بعد التلقيح . وقد تم استئصال الجهاز التناسلي وذلك لتقدير وزن و طول المبيض وكذلك قناة المبيض و الرحم مع عد الأجسام الصفراء و الحويصلات المبيضيه . الأجنة تم جمعها عن طريق الغسيل بواسطة محلول منظم الفوسفات+ ١٠ % سيرم اجنه الأبقار تم عد وفحص الأجنة المجمعة في مراحل تطور ها المختلفة بواسطة الميكروسكوب الضوئي . و يمكن تلخيص النتائج فيما يلي :-

Dorra, Tork M.I. et al.

- ١- حدثت زيادة معنوية (عند مستوى ١٠,٠١) في الوزن المطلق للمبيض بنسبه ٥٦% (١٠,٠٠-١,٤١ جرام) وكذلك في الوزن النسبي منسوبا للوزن الحي بنسبه ٦٦,٦% (١٩,٠-٥,٠٩ جرام/كجم).
- ٢- حدثت زيادة معنوية (عند مستوى ٩٠,٠٠) في طول المبيض بنسبه ١٨,٥% (١,٣١ مقابل
 ٢- منابل من المجموعة الثانية مقارنه بالأولي.
- ٣- لم يكن هناك اختلاف بين المجموعتين (١ و ٢) بالنسبة لعدد الحويصلات/المبيض ولكن زاد عدد الأجسام الصفراء/المبيض معنويا (عند مستوي ١٠,٠٠١) في المجموعة الثانية مقارنه بالأولى (٦,٤ مقابل ٨,٢/مبيض)
- ٤- لم يكن هناك فروق معنوية في الوزن المطلق والنسبي لطول قناة المبيض والرحم بين المجموعتين (١ و ٢). ذ
- ٥- كان عدد الأجنة المتحصل عليها لكل مبيض متشابه في كلا المجموعتين (٤,٣٠ مقابل ٤،٥٣/ معابل ٤، ٣٠/ مبيض) بينما معدل الحصول على الأجنة من المجموعة الأولي أكبر معنويا (عند مستوي (٠,٠١ مقابل ٥٣ %)
- ٦- كان توزيع الأجنة خلال مراحل انقسامها كما يلي: مرحلة ٢ خليه (٤,٤,٨%)، مرحله ٤ خلايا (٥,٢%) بينما نجد أن نسبه الأجنة المشوهة (١٠,٤%)وذلك في المجموعة الأولى وذلك مقابل ٨٤% في مرحله الجسم التوتى المدمج (موريولا) و ٣,٤% في مرحله الجسم التوتى المبكر و٢,٩% في مرحله الطور الجرثومي المبكر (البلاستوسيست) و نسبه الأجنة المشوهة تصل إلى ٩,٨% في المجموعة الثانية .

توضح هذه الدراسة مدى أهميه الاستفادة من تعدد التبويض لإناث الأرانب بذبحها بعد ٢٤ ساعة من التلقيح و الحصول علي الأجنة لاستخدامها علي نطاق واسع في مجال الهندسة الوراثية وكذلك ذبح الأمهات بعد ٢٢ ساعة من التلقيح في مرحله الجسم التوتى للاستفادة منها في مجال نقل الأجنة.