

## **INDUCTION OF OVULATION IN RABBITS USING DIFFERENT METHODS**

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### **ABSTRACT**

A total number of fifty six Baladi Black (BB) rabbit does were used in this study to compare different methods for induction of ovulation using Gn-RH, vasectomized bucks; mechanical stimulation and natural mating. The animals were randomly allotted into four equal groups. Rabbit does in the 1<sup>st</sup> group were naturally mated (controlled group). Does in the 2<sup>nd</sup> group were intramuscularly injected with 0.2 ml/doe of Gn-RH analogue. The vaginas of does in the 3<sup>rd</sup> group were mechanically stimulated using a thermometer. In the 4<sup>th</sup> group, ovulation was induced by a mating with using a vasectomized buck. All rabbit does in the 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> groups were artificially inseminated with fresh diluted semen using a Tris based extender. The dose of insemination was  $50 \times 10^6$  motile spermatozoa for each insemination. Abdominal palpation was practiced 10 days after AI to detect pregnancy. Number of pregnant does, conception rate %, gestation length, litter size, litter weight at birth, 21 and 28 days and pre-weaning mortality percentage were recorded.

Gn-RH increased conception rate ( $P<0.05$ ) as compared with the other groups. Litter size at 21 and 28 days of age were significantly increased when using the mechanical vaginal stimulation or a vasectomized buck compared to those either naturally mated or injected with Gn-RH. However, insignificant differences between groups were recorded in pregnancy length and litter size at birth. Rabbit does mechanically stimulated showed the best results ( $P<0.01$ ) for litter weight at birth, 21 and 28 days and kits weight at birth and to 21 days and litter weight gain from birth to 21 or 28 days and from 21 to 28 days of age. Also, mated does showed the lowest values for kits weight and litter weight gain ( $P<0.01$ ) at different ages.

*However, kits weight at 28 days of age was not significantly affected by treatments. Does stimulated by a vasectomized buck recorded the lowest values ( $P < 0.01$ ) for pre-weaning mortality percentage from birth to 21 days and from birth to 28 days as compared with the other groups, however, the differences were not significant in pre-weaning mortality percentage from 21 to 28 days. In conclusion, an intramuscular injection of 0.2 ml Gn-RH (Receptal®)/doe at insemination was as effective as natural mating on conception rate.*

**Keywords:** Rabbit, ovulation induction, vasectomized buck, vaginal mechanical stimulation, insemination

## INTRODUCTION

Artificial insemination has become an effective and essential mean for controlling breeding programs not only in experimental scale but also in commercial rabbitries (Lavara *et al.*, 2000, Nizza *et al.*, 2000, Riad, 2003 and Seleem, 2005). However, in rabbits, ovulation is a neuroendocrine reflex that is physiologically induced by mating (Sabrah *et al.*, 2001). The mechanisms whereby rabbit does become spontaneous ovulators are still unclear, but are likely associated to factors interfering with the control of the gonadal axis involving the hypothalamic centers responsible for Gn-RH release (Theau-Clement, 2008). In rabbits, the use of artificial insemination (AI) requires an ovulation inducer which stimulates the hypothalamus to release gonadotropin-releasing hormone that causes the LH peak able to initiates the ovulatory process (Ramirez and Beyer, 1988). Ovulation in rabbit does occurs 10- 13 hours after copulation or after any other mechanical, electrical or hormonal stimuli (Hafez, 1987). Some investigators successfully used vasectomized bucks to stimulate the ovulation in inseminating rabbit does. Using vasectomized rabbit buck as ovulation inducer at the time of artificial insemination resulted the best response for induction of rabbit doe ovulation compared to hCG or Gn-RH, and has no side effects on hormonal balance of doe (Khalifa *et. al.*, 2002). Gonadotropin releasing hormone (Gn-RH) appears to have solved the problem of induction of artificial ovulation in rabbits (Seleem, 2003).

The aim of the present study was to compare several ovulation methods such as a using Gn-RH injection, vasectomized bucks and a vaginal mechanical stimulation with natural mating, in order to develop an artificial insemination technique in Egyptian rabbit farms.

## MATERIALS AND METHODS

This study was carried out at Rabbits Research Unit, Department of Animal and Poultry Production, Faculty of Technology & Development, Zagazig University, Zagazig, Egypt, during September 2009 and lasted 5 months.

A total number of fifty six nulliparous Baladi Black (BB) rabbit does ( $31520 \pm 178$ g live body weight and 6-8 months of age) were randomly allotted into four groups (N=14/group). In first group, rabbit does were naturally mated (control). Does in the 2<sup>nd</sup> group were intramuscularly injected with 0.2 ml/doe of Gn-RH analogue (Receptal®, Intervet International B.V. Boxmeer-Holland). In the 3<sup>rd</sup> group, vagina of each doe was mechanically stimulated with a thermometer. Does of the 4<sup>th</sup> group, were mated with a vasectomized rabbit buck. Does of groups 2, 3 and 4 were artificially inseminated with fresh diluted semen (Tris extender), using 1 ml containing  $50 \times 10^6$  motile spermatozoa/doe. Pregnancy was detected by abdominal palpation 10 days after mating or AI.

The experimental rabbits were housed individually in wire cages (60 x 555 x 40 cm) provided with galvanized feeders and nipple drinkers. The animals were kept under the same environmental and managerial conditions. Rabbits were fed isonitrogenous and isoenergetic diets (17% crude protein and 2700 Kcal ME/kg DM). The diet was formulated to meet the nutrient requirements of adult rabbits according NRC (1977). The feeding and water was provided *ad libitum*.

Number of mated or inseminated does, number of pregnant does, conception rate (%), gestation length and litter size, litter weight and pre-weaning mortality percentage were recorded. Data were statistically studied by analysis of variance (ANOVA) according to Snedecor and Chochran (1982) using SPSS system (1998). The differences between means were tested by using Duncan's New Multiple Range test, (Duncan, 1955). Fertility (conception rate measured by abdominal palpation) was analyzed using the contingency tables according to Everitt (1977). Pre-weaning mortality percentages were subjected to arc-sin transformation before being analyzed in order to approximate normal scale distribution. Means were transformed to the original scale before being illustrated.

## RESULTS AND DISCUTION

The effects of different methods of induction of ovulation on reproductive and productive traits are presented in Tables 1, 2 and 3.





**Table 3: Effect of type of mating on pre-weaning mortality percentage.**

Classification	Pre-weaning mortality percentages		
	Birth-21	Birth-28 days	21-28 days
Natural mating	35.92 <sup>a</sup>	38.73 <sup>a</sup>	4.38
0.2 mL Receptal + AI	43.97 <sup>a</sup>	47.72 <sup>a</sup>	5.83
Mechanical stimulation + AI	35.00 <sup>a</sup>	35.00 <sup>a</sup>	00.00
Vasectomized buck + AI	14.84 <sup>b</sup>	18.61 <sup>b</sup>	4.44
Significance	**	**	NS

Means with different superscripts in the same column differ significantly ( $P < 0.05$ ).

\*\* =  $P < 0.01$  and NS = Not significant

Fertility was higher when ovulation was induced by a Gn-RH injection ( $P < 0.05$ ) compared to others methods. Neither the pregnancy length nor the litter size at birth were influenced by the method to induce ovulation. In a contrast, litter size at 21 and 28 days of age were increased ( $P < 0.01$ ) when ovulation was induced by a mechanical stimulation or a vasectomized buck, compared to natural mating or a Gn-RH injection. Similar results were reported by Quintela *et al.* (2004), Ondruska *et al.* (2008) and Kalaba and Abdel-Khalek (2011) who found that Gn-RH introduced in the seminal dose increased kindling rate as compared to the control group. Some investigators used vasectomized bucks to stimulate the ovulation in artificially inseminated rabbits. Using a vasectomized buck as an ovulation inducer at the time of artificial insemination produced the best response for ovulation induction in rabbits doe compared to hCG or Gn-RH, and has no side effects on hormonal balance in does (Khalifa *et al.*, 2000). The last authors emphasized that, using vasectomized buck to induce ovulation is like natural copulation especially with the presence of the male around the female, insertion and friction of penis in the vagina, also the pressure from mounting and contact with the perineal and pudenda regions, may stimulate release of more fertilizable ova than the use of other ovulation inducers.

Rabbits does mechanically stimulated showed the best results ( $P < 0.01$ ) for litter weight at birth, at 21 and 28 days and for the average of kits weight at 21 days and for litter weight gain from birth to 21 days or to 28 days and from 21 to 28 days of age. Also, the doe mated naturally showed the lowest values for the kit weight at 21 days ( $P < 0.01$ ) and litter weight gain between birth and 28 days ( $P < 0.05$ ). However, the kits weight at 28 days of age was not affected by treatments. The group of does having been mated by a vasectomized buck recorded the lowest values ( $P < 0.01$ ) for pre-weaning mortality percentage from birth to 21 days and from birth to 28 days compared to the others groups. The last week before weaning, the mortality was not influenced by the treatments. Generally, using Gn-RH in low fertile and/or

heat stressed rabbit does improved conception and kindling rates and litter size and weight at birth (El-Gaafary, 1994; El-Sherbiny, 1994; Daader *et. al.*, 2000; Seleem, 2003 & Seleem and Riad, 2005).

Conclusively, it could concluded that the induction of ovulation in rabbit does by 0.2 ml Gn-RH Receptal/doe can be successfully used to improve fertility. However, intra-vaginal mechanical stimulation or using vasectomized buck could be recommended to avoid problems associated with repeated injection of Gn-RH.

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