

## EFFECTIVENESS OF POST PARTICULAR PROLACTIN ADMINISTRATION ON FOLLICLE DEVELOPMENT THE COUNT IN MICE (*Mouse muscle*) FEMALE

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**Abstract:** This study aims to test the effectiveness of postpartum prolactin administration on follicle development in the count in mice (*Mouse muscle*). This type of research is a true experiment using a completely randomized design (RAL). The sample for this study was mice (*Mouse muscle*) female strain BALB/C, aged  $\pm 4$  months with a body weight between 25-30 gr in a healthy condition and not pregnant. The data obtained was analyzed using the One-way analysis of variance (F test) with a significance level of 0.05, followed by the Least Significant Difference test (BNT). The research results show that the sig. (p) = 0.001 <  $\alpha$  = 0.05, so it can be concluded that postpartum administration of prolactin has a significant effect on follicular development of the count in mice (*Mouse muscle*).

**Keywords:** *Prolactin, Postpartum, Follicles The count.*

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### INTRODUCTION

At this time, most postpartum mothers often experience breast milk production. The trigger factor is low levels of prolactin in the blood circulation. Prolactin is a type of glycoprotein hormone that has receptors in the mammary glands to stimulate the lactation process (the process of producing milk) [1-2]. Adequate levels of prolactin in the blood circulation influence increased secretion of breast milk. Increased levels of prolactin in the blood circulation provide negative feedback to the anterior pituitary gland, inhibiting the secretion of FSH (follicle-stimulating hormone) and LH (luteinizing hormone) [3]. [4] further stated that low levels of FSH and LH in the blood can indirectly cause obstacles to folliculogenesis. Folliculogenesis is the growth and development of follicles in the ovaries [4]. Folliculogenesis shows the stages of development, from primary, secondary, and tertiary follicles to becoming follicles that count. One of the hormonal disorders in one stage of folliculogenesis will cause follicles not to form so that ovulation will not occur [4]. Inhibition of follicle growth, the *count* accompanied by failure to ovulate, affects livestock fertility levels [5].

### RESEARCH METHODS

This study used a completely randomized design because the population units were assumed to have homogeneous characteristics [6]. The sample for this study was mice (*Mouse muscle*) BALB/C strain female,  $\pm 4$  months old with body weight between 25 – 30 grams, healthy, and not pregnant. A total of 32 mice were divided randomly into four groups, each group consisting of 8 mice. A total of 10 male mice were used as mice, aged  $\pm 4$  months with a body weight of 30 - 35 grams, in good health, and

having a high libido. Female and male mice were housed in one cage. After acclimatization, female and male mice were collected in one cage and monitored for mating. Female mice were mated with male macaques using the Harem Mating method [7] in 50 X 30 X 20 cm cages equipped with food and water containers. Food and water were provided ad libitum, and the laboratory room where the mice were kept during the research process was clean and dry, with good air circulation, a calm atmosphere, and stable room temperature. Female mice are left in the cage until parturition/giving birth. Mice mating was carried out by collecting four females and one male in each cage and leaving them for 24 hours. After it is believed that copulation has occurred, which is indicated by the presence of a vaginal plug, the males are separated. Treatment was given to the mother mice during the breastfeeding period. The treatment began to be given to the mother mice four days after breastfeeding her offspring and was given once a day for six consecutive days. Mice that had given birth were divided into four groups, each consisting of 8 mice, and given the following treatment: TO (control group) 0.5 ml NaCl 0.9%, T1, which was injected with prolactin at a dose of 5 IU per animal. T2 was injected with prolactin at a dose of 10 IU per head, and T3 was injected with prolactin at 20 IU per head intramuscularly. Treatment was given from the 4th to the 10th day after giving birth to stimulate the mother mice to produce maximum milk.

On the 11th day, the mother mice were sacrificed by dislocating the neck, then surgery was carried out by placing the mice on a section board, the abdominal skin was opened, and the left and right ovaries were taken. The ovaries were fixed with 10% formalin for further processing to make histology preparations. Counting the number of follicles, the

count Histology preparations were carried out in 4 observation areas: the front end, back end, and both sides. Each observation area requires five microscope views with a magnification of 400 times, assisted by a calculating tool, the Hand Counter. Observation and calculation of the number of follicles the count is based on the characteristics of the follicle and guided by the atlas. Data were analyzed using a one-way Analysis of Variance (F) test with a significance level of 0.05. If it turns out that there is effectiveness of prolactin on follicle development, the count in real terms, then proceed with the Least Significant Difference (BNT) test [8,9].

**RESULTS AND DISCUSSION**

Observation and counting of follicles The count in each group is presented in Table 1. Following Table 1.

Table 1. The average number of follicles *counted* in the ovaries of mice in the control group and the three treatment groups.

Group	Number of Mice Mothers	Number of Follicles The <i>count</i> ( X ± Sd)
T0 (Control)	8	5.500 ± 2.138 <sup>a</sup>
T1 (Treatment 1)	8	3.125 ± 1.642 <sup>b</sup>
T2 (Treatment 2)	8	2.500 ± 1.069 <sup>b</sup>
T3 (Treatment 3)	8	2.125 ± 0.991 <sup>b</sup>

Note: Different letters in the same column indicate significant differences (P < 0.05).

Table 2. Results of One-Way Variance Analysis of the Number of Follicles The *count*

Diversity Source	DB	JK	KT	F <sub>count</sub>	P
Treatment	3	55.12	18.38	7.83	0.001
Error	28	65.75	2.35		
Total	31	120.8			

Table 3. BNT Test Results for Number of Follicles The *count*

	T1	T2	T3	T0
T1	-	0.625	1.000	2.375*)
T2	-	-	0.375	3.000*)
T3	-	-	-	3.375*)

Note: The superscript sign indicates significantly different (P<0.05).

The BNT test in Table 3 shows a significant difference P <a (0.05) between the number of

follicles the count in group T0 with groups T1 and T2 and T3. Meanwhile, between groups T1 and T2 and group T1 and group T3, group T2 and group T3 were not significantly different.

Many factors can influence the growth and development of follicles in the ovaries during the estrus cycle, such as animal species, reproductive phase, environmental conditions, age of the mother, and genetic factors [10,11]. In the growth and development of follicles from primordial follicles to follicles, the count and ovulation are influenced by the gonadotropin hormone produced by the anterior pituitary gland. The gonadotropin hormones consist of Follicle Stimulating Hormone (FSH) and Luteinising Hormone (LH). The main function of FSH is to stimulate the growth and development of follicles in the ovaries in the primordial phase so that the follicles become mature (the count) but do not cause ovulation [12]. FSH causes follicles to become more sensitive to LH [13]. Luteinizing hormones cause ovulation by causing the breakdown of the follicle wall and the count and release of egg cells [5]. According to [14], LH influences the corpus luteum formation from the remaining follicles, the count that had ruptured due to ovulation. Normally, dopamine is produced by the hypothalamus, which has an important role in inhibiting prolactin secretion [15].

The results of the one-way analysis of variance test in Table 2 show that there is a significant difference P (0.001) <a (0.05) from the four treatments, so it can be concluded that giving prolactin to postpartum mice is effective in reducing the number of follicles in the count. [16,17] said that high levels of prolactin in blood serum will increase the synthesis and secretion of milk by the mammary glands. Furthermore, [18] high milk secretion affects the hypothalamus, inhibiting the secretion of gonadotropin-releasing hormones (GnRH). Low GnRH levels result in suppressed FSH and LH secretion from the anterior pituitary. Low levels of FSH and LH in blood serum inhibit folliculogenesis, follicle maturation, and follicle formation [17]. Under normal conditions, the hormone prolactin can function properly in the body. However, suppose prolactin levels are too low (lack of prolactin) or too high (excess prolactin). In that case, a hormonal imbalance can harm the growth and development of follicles in the ovaries [19]. According to [2,13,20], prolactin has the synonym LH (Luteotropic Hormone) or mammotropin or lactogenic hormone.

The action of the prolactin hormone is directly on the target tissue and does not regulate the function of other endocrine glands. The anterior pituitary gland produces the hormone prolactin, namely by Lactotroph cells from acidophil cells [19]. Prolactin

is a single-chain protein hormone; female birds have target cells or receptors in the ovaries [11]. At high enough levels in the blood, it inhibits the growth and development of follicles in the ovaries [13]. According to [14], high prolactin levels or hyperprolactinemia when the chicken enters the brooding and caring phase of its young causes regression of the ovaries so that no growth and development of follicles occurs, which results in the chicken not producing eggs. It follows the statement [7,21] which states that when a chicken enters the brooding phase, physiologically, it will induce the hypothalamus to release prolactin-releasing hormone (PRH), which will stimulate the anterior pituitary to produce the hormone prolactin. The prolactin hormone that has been produced will flow through the bloodstream to its target organ, namely the follicles of the ovaries. High prolactin presence will increase the follicle cells' response in the ovaries, thereby causing the formed follicles to regress. The regression of these follicles will ultimately result in an egg not being formed. In more depth, [19] said that the hormone prolactin targets organs directly on the ovaries and initiates ovarian regression so that no growth and development of follicles occurs.

Furthermore, the hormone prolactin can cause anti-gonadal effects [12,20,22]. This effect causes inhibition of follicular growth and development. The anti-gonadal effect continues to affect egg production. Meanwhile, according to [19], prolactin is an endogenous hormone that has the effect of causing ovarian regression. Ovarian regression is a condition where follicle growth occurs in the ovaries, and they will be lysed and absorbed by the body, so there is no follicle growth. It means that ovulation and egg formation will not occur. Animals that cannot produce eggs indicate that their ovaries are not growing and developing follicles [4].

## CONCLUSION

From the results of this study, it can be concluded that postpartum administration of prolactin has a significant effect on the development of de Graafian follicles in mice (*Mus musculus*).

## REFERENCES

- [1] HENDARTI, G. A., Safitri, E., & Hariadi, M. (2008). Anti Prolaktin Sebagai Penghambat Pengeraman Pada Ayam Kampung Bertelur.
- [2] Safitri, E. (2005). *Karakterisasi dan Produksi Antibodi Poliklonal Anti Prolaktin (Abpo-Prol) Sebagai Penghambat Proses Moulting* (Doctoral dissertation, UNIVERSITAS AIRLANGGA).
- [3] Rejeki, P. S., Putri, E. A. C., & Prasetya, R. E. (2019). Ovariectomi pada tikus dan mencit.
- [4] Ningtyas, N. S. I. I. (2017). Pengaruh Pemberian Minyak Buah Merah (*Pandanus conoideus* Lam.) Terhadap Histopatologi Folikel De Graaf Pada Mencit (*Mus musculus*) Model Infertil. *Jurnal Sangkareang Mataram*, 3(3), 36-38.
- [5] Ramadhani, S. A., Supriatna, I., Karja, N. W. K., & Winarto, A. (2017). Pengendalian folikulogenesis ovarium dengan pemberian ekstrak biji kapas. *Jurnal Sain Veteriner*, 35(1), 71-80.
- [6] Zainuddin, M. (2020). *Metodologi Penelitian Kefarmasian Edisi 2*. Airlangga University Press.
- [7] Hafez, E. S. E., & Hafez, B. (Eds.). (2013). *Reproduction in farm animals*. John Wiley & Sons.
- [8] Steel, R. G. D., Torrie, J. H., & Sumantri, B. (1991). Prinsip dan prosedur statistika: suatu pendekatan biometrik. (*No Title*).
- [9] Abdurahman, M., Somantri, A., & Muhidin, S. A. (2011). *Dasar-Dasar Metode Statistik Untuk Penelitian*.
- [10] Hardjopranjoto, S. (1995). Ilmu kemajiran pada ternak.
- [11] Ramesh, R., Kuenzel, W. J., & Proudman, J. A. (2001). Increased proliferative activity and programmed cellular death in the turkey hen pituitary gland following interruption of incubation behavior. *Biology of reproduction*, 64(2), 611-618.
- [12] Bédécarrats, G., Guémené, D., Morvan, C., Kühnlein, U., & Zadworny, D. (1999). Quantification of prolactin messenger ribonucleic acid, pituitary content and plasma levels of prolactin, and detection of immunoreactive isoforms of prolactin in pituitaries from turkey embryos during ontogeny. *Biology of reproduction*, 61(3), 757-763.
- [13] Hardjopranjoto, S. (1995). Ilmu kemajiran pada ternak.
- [14] Anwar, H., & Safitri, E. (2005). Anti-prolaktin sebagai penghambat proses moulting. *BERKALA PENELITIAN HAYATI JOURNAL OF BIOLOGICAL RESEARCHES*, 11(1), 25-29.
- [15] Smith, J. R. (1995). Produksi Serum Hiperimun. *Teknologi ELISA dalam Diagnosis dan Penelitian*. Gadjah Mada University Press. Yogyakarta, 15-32.
- [16] Wodzicka-Tomaszewska, M., Utama, I. K., Putu, I. G., & Chaniago, T. D. (1991). *Reproduksi tingkah laku dan produksi ternak di Indonesia*. PT Gramedia Pustaka.

- [17] Yamamoto, I., Wakita, M., & Tanaka, M. (2003). Tissue distribution of prolactin receptor mRNA during late-stage embryogenesis of the chick. *Poultry science*, 82(1), 155-157.
- [18] Plant, T. M., & Zeleznik, A. J. (Eds.). (2014). *Knobil and Neill's physiology of reproduction*. Academic Press.
- [19] Jabbour, H. N., & Kelly, P. A. (1997). Prolactin receptor subtypes: a possible mode of tissue-specific regulation of prolactin function. *Reviews of reproduction*, 2(1), 14-18.
- [20] Freeman, M. E., Kanyicska, B., Lerant, A., & Nagy, G. (2000). Prolactin: structure, function, and regulation of secretion. *Physiological reviews*.
- [21] Marhiyanto, B. (2000). Sukses beternak ayam arab. *Indonesia: Difa publisher*.
- [22] HENDARTI, G. A., Safitri, E., & Hariadi, M. (2008). Anti Prolaktin Sebagai Penghambat Pengeraman Pada Ayam Kampung Bertelur.
- [23] Proudman, J. A., & Wentworth, B. C. (1996). Pulsatile secretion of prolactin in laying and incubating turkey hens. *Domestic Animal Endocrinology*, 13(3), 277-282.
- [24] Safitri, E., Yuniarti, W. M., & Hariadi, M. (2007). Peningkatan produksi telur pada ayam dengan pemberian a PRL.