

# THE EFFECT OF GIVING BEAN SPROUTS (*PHASEOLUS AUREUS*) WITH SYNCHRONIZATION INTERNAL CONTROLLED DRUGS RELEASE (CIDR) AGAINST SYMPTOMS OESTRUS AND BEHAVIOR OESTRUS EWES (THIN TAILED SHEEP)

Hans Ririmasse, Ismaya & Zaenal Bachruddin

Postgraduate Programme, Faculty of Livestock, Department of Livestock Sciences, Gadjah Mada University, Yogyakarta, Indonesia

# ABSTRACT

This study aims to determine the oestrus symptoms and behaviour oestrus, thin tailed ewes that in synchronization using Internal Controlled Drugs Release (CIDR) and was treated with bean sprouts (*Phaseolus aureus*). Research material used 8 thin tailed ewes, each group consisting of 4 tail, group I (P<sub>0</sub>) having an average body weight of  $22.72 \pm 1.09$  kg and group II (P<sub>1</sub>)  $24.42 \pm 2.28$  kg by the age of 1.5 - 2 years. Then the two groups at synchronization using CIDR for 13 days by means of intravaginal. Group I (P<sub>0</sub>) is not given bean sprouts and group II (P<sub>1</sub>) given bean sprouts 250 g/tails/days. The results showed a 100% response oestrus after CIDR release. Early oestrus in group I (P<sub>0</sub>) and group II (P<sub>1</sub>) showed a mean  $33.50 \pm 1.73$  and  $29.00 \pm 3.16$  hours. While the end of oestrus showed a mean  $58 \pm 1.15$  and  $60 \pm 1.41$  hours. It was concluded that the synchronization of oestrus using CIDR showed a response of 100%. The early and end of each oestrus occurs in 32-36 and 26-33 hours, this response indicates a significant difference (P <0.05).

Keywords: Thin Tailed Sheep, Internal Controlled Drugs Release (CIDR), Oestrus, Bean Sprouts.

# 1. INTRODUCTION

Indonesian thin tailed sheeps is known as sheep native village and is one of germplasm that can potentially be developed for national meat supply. Thin tailed sheep have some advantages such as the ability gave birth to twins or more, the age of sexual maturity relatively quickly and do not know the breeding season so that they can give birth throughout the years (Sumoprastowo, 1987).

Reproduction is a major factor contributing to the efficiency of production is often not understood for sheep farmers, who generally still traditionally managed. So it can affect reproductive efficiency factors include physiological, genetic and environmental. Interpretation of the hormones estrogen and progesterone have different physiological stages



of the animal and is considered as one of the most important parameters for determining the fertility status (Zarkawi and Soukouti, 2001).

CIDR synchronization methods can cause oestrus symptoms together, within a short interval of time and can be predicted in a flock. Thus sinkronosasi CIDR aims to manipulate the reproductive process, so the cattle will be induced estrus, can be mated simultaneously and the results were normal fertility. CIDR synchronization method on the thin tailed sheep by administering the hormone progesterone will bring oestrus 24-36 hours after release (Rasad, 2004).

This synchronization method has a function to inhibit the secretion of LH, shortening the life of CL, shortening the onset of estrus and ovulation. With CIDR administration within a period of 12-13 days will cause regression of CL. In this case the progesterone will inhibit ovulation by pressing the LH secretion, so it will give effect to CL shrink. With this approach the concentration of progesterone in the blood increases. Furthermore, administration of exogenous progesterone will cause negative feedback on LH secretion immediately after CL regression. At the time of the revocation of the concentration of progesterone in the blood and the subsequent decline will occur follicle growth, the onset of oestrus and ovulation (Hafez, 2000; Rasad, 2004).

This understanding is also one of the factors that can control the start and end of the estrous cycle that can affect the use of natural feed ingredients containing hormones or phytohormones have been developed, one of which is a phytoestrogen found in bean sprouts (legume). Phytoestrogens are plant substrates which have estrogen-like activity (Glover and Assinder, 2006). Furthermore, according to Jefferson *et al.*, (2002) is the natural decomposition of phytoestrogens found in plants that have much in common with estradiol, a natural form of estrogen with the most potential.

The successful implementation of reproductive technologies are also influenced by aspects of reproduction depends on the characteristics of the production of hormonal changes. Is this problem can be solved by the application of reproductive biotechnology in the form of weft or oestrus synchronization with CIDR method. Given this information, this experiment was designed to test the effect of bean sprouts and synchronization using CIDR for oestrus response, oestrus symptoms and oestrus behaviour on a thin tailed sheep. Thus the analysis that is required is to determine the length of estrus cycle and estimated time of ovulation.

# 2. MATERIALS AND METHODS

## 2.1 Animals and management

In this study, eight ewes selected at random using a random sampling method have age  $\pm 1.5$  - 2 years, ever given birth and had a healthy condition. Research consists of two groups each of four tails. The average weight for Group I (P<sub>0</sub>) 22.72  $\pm 1.09$  kg and group II (P<sub>1</sub>) 24.42  $\pm 2.28$  kg. Group I (P<sub>0</sub>) is not given bean sprouts and group II (P<sub>1</sub>) be given bean sprouts, each ewes 250 g/tail/day.

## 2.2 Reproductive and management

CIDR installation in thin tailed ewes for oestrus synchronization in this study conducted over 13 days. CIDR began inserted into the applicator, the applicator then smeared with jelly with a view to facilitating easily entry through to vulva until in vagina. Part of the vulva cleaned using alcohol with raised tail ewes that will be installed CIDR. Then the applicator tip with gently inserted into the vagina. Then driven by pressing hadel applicator. After CIDR occupies the anterior part of the vagina, the applicator is pulled out so it will look rope that directly related to the CIDR, will be seen hanging in part of the vulva.

CIDR release performed on day 14, CIDR in the vagina removable or withdrawn slowly by pulling a rope that depends in vulva that looks from the outside and connected directly to the CIDR. Mating between ewes and rams do naturally. Mating performed at days 16 and 17. After the release of CIDR and oestrus symptoms and oestrus behavior in 24-36 hours. Then mating is done in the morning, noon, and night. Observations in the form of symptoms and behavioral changes that occur during oestrus symptoms include; changes in the condition of the vulva, include; red vulva, vulva visible swelling around the red and warm to the touch. While the observation of behaviour change include; silent heat



and ewes agitated when brought near a stud. Observation of the onset of oestrus symptoms and oestrus behaviour for 24-36 hours after CIDR release for 14-16 days.

Activity	Date/Days to												
Installation CIDR	1	2	3	4	5	6	7	8	9	10	11	12	13
The release of CIDR + Observation oestrus	14	15											
Mated	16	17											

#### Table 1. Stages Field Research

#### **Description :**

Installation CIDR
The release of CIDR
Observation oestrus
Mated

## 2.3 Data analysis

The variables were observed in this study include; the early of oestrus and end of oestrus, symptoms oestrus and oestrus behaviour which is conducted after treatment CIDR release, divided by the number of sheep multiplied by one hundred percent, in group I ( $P_0$ ) are not given bean sprouts and group II ( $P_1$ ) be given bean sprouts, oestrus response; the early of oestrus and end of oestrus in Group I ( $P_0$ ) and II ( $P_1$ ). Oestrus symptoms and oestrus behavior tested with Chi-Quadrat. Early emergence of oestrus and late oestrus were analyzed using SPSS version 20.0 for Windows (IBM, USA, 2012).

# 3. **RESULTS AND DISCUSSION**

#### 3.1 Oestrusf symptoms and oestrus behaviour

From the observation of oestrus symptoms and oestrus behaviour on 8, thin tailed sheep were synchronized using CIDR, oestrus symptoms and oestrus behaviour showed 100% (Table 1). Results the percentage oestrus obtained from of group I ( $P_0$ ) without bean sprouts and group II ( $P_1$ ) which be given bean sprouts and synced using CIDR by means of intravaginal for 13 days effective. Rizal, (2003) reported that the implants CIDR-G during the 13 days in the vagina effectively, in an attempt to synchronize estrus in Garut sheep showed an average of 100% of the estrous response. Results of several studies have reported that the implants CIDR in vagina for 7, 12, and 14 days in Holstein Friesian cows (Vargas et al., 1994); 6, 9, and 12 days in Suffolk sheep (Fukui et al., 1994); 12 days in St. Croix sheep (Feradis, 1999) and 7 and 14 days in sheep Etawah (Ngangi, 2002) can result in estrus percentage of 88-100%.



Shoon group	Shoop oodo	Oestrus Symptoms			
Sheep group	Sheep code -	Oestrus	No Oestrus		
	P <sub>0-</sub> 14	+	-		
Group I (P <sub>0</sub> )	P <sub>0-</sub> 13	+	-		
CIDR without Sprouts	P <sub>0-</sub> 12	+	-		
	P <sub>0-</sub> 06	+	-		
Total	4	4	0		
Estrus Percentage P <sub>0</sub> (%)	100	100	0		
	P <sub>1</sub> .09	+	-		
Group II (P <sub>1</sub> )	P <sub>1</sub> .08	+	-		
CIDR + Sprouts	P <sub>1</sub> .07	+	-		
	P <sub>1</sub> .02	+	-		
Total	4	4	0		
Oestrus Percentage P <sub>1</sub> (%)	100	100	0		

**Table 2.** Percentage of sheep oestrus after the installation of Internal Controlled Drugs Release (CIDR)











Figure 2. Symptoms appear estrus after CIDR revoked in group II (P<sub>1</sub>). A. Red vulva and swelling, B. Vulva swelling, C. Red vulva and swelling, D. Red vulva and swelling

The symptoms of oestrus is shown in Figure 1 and 2 is a manifestation of the increased levels of the hormone estrogen in the blood that is synthesized and secreted by the follicle after release CIDR. Frandson *et al.*, (2003), estradiol stimulates the thickening of the walls of the vagina, resulting in increased vascularization of the external genitalia, causing swelling and redness as well as an increase in vaginal secretions.

These images correspond to the physical description of genitalia in Figures 1 and 2. This incident seems to have a close relationship with the corpus luteum (CL) that developed during the luteal phase and increased production of progesterone. Toelihere, (2003) lysis of the CL will be followed by the secretion of gonadotropin hormone that causes estrus and ovulation. Gonadotropin hormone is a protein hormone secreted by the pituitary gland and including luteinizing hormone (LH) and follicle stimulating hormone (FSH).

LH and FSH regulate estrous cycle is the cyclical patterns that affect ovarian activity and facilitate the transition from female animals between the reproductive period of non-acceptance and acceptance as well as allow mating and subsequent pregnancy. Alila and Hansel, (1984) reported that CL comes from ovulatory follicle cells and LH is the primary hormone luteotropic (LTH) in female animals, is responsible for stimulating the luteinizing theca and granulosa cells of pre-ovulatory follicles into luteal cells.

This opinion is reinforced by Stronge *et al*, (2005); Starbuck *et al*, (2001); McNeill *et al*, (2006); Parr *et al*., (2012) which states that CL function is to produce enough progesterone concentrations during the luteal phase of the estrous cycle to maintain the gestation and during gestation, reduce the secretion of gonadotropins and prevent the occurrence of estrus behavior.

Roche, (1996) stated that the estrous cycle is regulated by the hypothalamic hormone gonadotropin releasing hormone (GnRH), anterior pituitary follicle stimulating hormone (FSH) and luteinizing hormone (LH), ovaries (progesterone, estradiol and inhibins) and uterus (prostaglandin F2 $\alpha$ , PGF2 $\alpha$ ). This hormone works through a system of positive and negative feedback to regulate the estrus cycle. The onset of estrus after CIDR release caused by ewes entering estrus cycle. Installation which contains the hormone progesterone CIDR will produce a negative feedback



effect on the secretion of gonadotropin hormone (GnRH), which FSH and LH. Hansel, (1993) that this synchronization technique will exert negative feedback on LH hormone that inhibits the secretion of endocrine events and cause preovulasi follicle maturation and ovulation.

Inhibition of gonadotropin secretion during installation CIDR will continue, until the accumulation of gonadotropin hormone in the anterior pituitary. At the time of release CIDR, a decline in the concentration of progesterone in the blood so that negative feedback is lost. This resulted gonadotropin hormone is secreted in large amounts that can stimulate folliculogenesis process (the process of follicular development in the ovaries) and formed follicles mature. The mature follicle will synthesize the hormones estrogen and secreted into the blood circulation then ewes oestrus and excreted in the form of the appearance of symptoms of oestrus.

The study of group I ( $P_0$ ) and II ( $P_1$ ) after the release of CIDR showed symptoms of oestrus 100%, this indicates that the hormone progesterone contained in CIDR effective to perform the function of negative feedback, preventing the secretion of hormones gonadotropin are synthesized by the anterior pituitary. Toelihere, (1993) stated that the increase in the secretion of the hormone estrogen in the blood resulting in estrus female animals characterized by vulva symptoms such as red, swollen and silent when ridden stud. The same was reported by Fukui *et al.*, (1994) using implants CIDR-G combined with 600 IU PMSG on the day before CIDR-G removed and Hastono *et al.*, (1997) using 40 mg implant fluorogstones acetate (FGA) in vagina. Falkenburg *et al.*, (1971) reported that the percentage rate of 100% oestrus occurs using 375 mg of progesterone in the form of crystalline silicon rubber and methods implanted subcutaneously in combination with the hormone estradiol. Oestrus expression after CIDR release occurs 100%, this shows the effectiveness of the CIDR granted for 13 days resulted in a sensitivity of ovarian activity and ensure the expression of oestrus at an acceptable level.

#### 3.2 Oestrus response

In this study, related to the provision of additional treatments bean sprouts, then the thin tailed ewes were divided into two groups with different bean sprouts administration. Two groups  $(P_0-P_1)$  sheep are all mounted CIDR, but bean sprouts are only given to group II  $(P_1)$ . Additional treatments differences were eventually able to influence the timing of oestrus early and end of oestrus, as shown in Tables 3 and 4.

Group P <sub>0</sub> (CIDR -	+ Without Sprouts)	Group P <sub>1</sub> (CIDR + Sprouts)			
Numbers Ewes	Early of Oestrus	Numbers Domba	Early of Oestrus		
	(Hours)		(Hours)		
P <sub>0</sub> -14	32	P <sub>1</sub> -09	30		
P <sub>0</sub> -13	36	P <sub>1</sub> -08	33		
P <sub>0</sub> -12	33	P <sub>1</sub> -07	26		
P <sub>0</sub> -06	33	P <sub>1</sub> -02	27		
Total	134	Total	116		
Average (±SD) 33,50±1,73		Average (±SD)	29,00±3,16		

**Table 3.** Average early of oestrus (hours) the group of sheep  $P_0$  and  $P_1$  after the release CIDR



Group P <sub>0</sub> (CIDR	+ Without Sprouts)	Group $P_1$ (CIDR + Sprouts)			
Numbers Ewes	End of Oestrus (Hours)	Numbers Domba	End of Oestrus (Hours)		
P <sub>0</sub> -14	59	P <sub>1</sub> -09	59		
P <sub>0</sub> -13	57	P <sub>1</sub> -08	60		
P <sub>0</sub> -12	57	P <sub>1</sub> -07	59		
P <sub>0</sub> -06	59	P <sub>1</sub> -02	62		
Total	232	Total	240		
Average (±SD)	58±1,15	Average (±SD)	60±1,41		

Table 4. Average end of oestrus (Hours) the group of sheep  $P_0$  and  $P_1$  after the release CIDR

Based on tables 3 and 4, group I (P<sub>0</sub>) indicates that the initial emergence of oestrus occurs 32-36 hours after release CIDR to the average ( $\pm$ SD) of 33.50  $\pm$  1.73 hours and the group II (P<sub>1</sub>) early emergence of estrus 26-33 hours after release CIDR to the average of 29.00  $\pm$  3.16 hours. While end of oestrus on group I (P<sub>0</sub>) occur between 57-59 hours during oestrus with a mean of 58  $\pm$  1.15 hours and the end of oestrus on group II (P<sub>1</sub>) 59-62 hours with an average of 60  $\pm$  1.41 hours.

Distribution of oestrus symptoms after release CIDR in Table 1 and illustrated in Figures 2 and 3 show that did not show significant differences. While at the early and end of oestrus, group I ( $P_0$ ) and II ( $P_1$ ) respectively occur within 32-36 hours and 26-33 hours, this response indicates that the response to the early and end of oestrus showed a significant difference (P<0.05). Time interval, the early and end of the onset of oestrus on group I ( $P_0$ ) and II ( $P_1$ ) showed significant differences (P<0.05) after the lifting of CIDR group II ( $P_1$ ) time interval estrus sooner or faster and longer than group I ( $P_0$ ).

Hamra *et al.*, (1986), Welch *et al.*, (1984) and Carlson *et al.*, (1989) reported that the results of the estrous cycle is the time constant estrus begins within 24 hours and estrus were observed for 30 hours after release CIDR. Robertson, (1980) and MacNatty *et al.*, (1988) reported the highest frequency of estrus approximately 36 hours after administration of the PMSG and ovulation approximately 27.4 hours after the onset of estrus is consistent. Best estrus observed in groups 1, 2 and 3 with 70, 90 and 80%, respectively with no show significant differences (P>0.05) between the three groups.

There are significant differences in the duration of the period of estrus between groups I ( $P_0$ ) are not given bean sprouts with group II ( $P_1$ ) which is given bean sprouts. This possibility is suspected by the influence of phytoestrogens contained in bean sprouts so as to accelerate the occurrence of oestrus. The influence of phytoestrogens on female genital tract resulted in a change of onset of oestrus, puberty progress and increase the length of the estrous cycle (Delclos *et al.*, 2009). In the opinion of Wilcox *et al.*, (1990) found that supplementation with phytoestrogens menu led to increased maturation of vaginal cytology.

Gunnar, (2006) reported that the phytoestrogens affect the hypothalamic-pituitary center and organize feedback from the regulatory hormones in sheep. Similarly with the opinion Mathieson and Kitts, (1980) which states that the effect of estrogenic hormone levels found in the reproductive organs caused by changes in hormone levels in animals and affect estrogenic hormones in the reproductive organs.

Phytoestrogens affect hormone levels in sheep. This is due to the biological response of phytoestrogens in animals depend on factors species, age, sex, dose, mode of administration and metabolism (Hernawati, 2009). While the opinion of Burroughs *et al.*, (1985) has been repeatedly reported that further research in several countries presented research conducted ooforektomi (expenditure ovaries) is done bilaterally, proving that phytoestrogens remain in the neonatal period and has a lasting effect the hypothalamic hypophyseal axis in the ovary.

The results obtained showed that the thin tailed sheeps are given bean sprouts can cause different estrus. The onset of estrus can be caused by a given dose, pattern or observation methods and factors the condition of livestock as



well as the effect of feed given. High levels of the hormone estrogen may increase the proliferation (repetition) ovarian granulosa cells and prevent or non-occurrence of egg follicles, so that the egg is diovulasikan more (Papaji, 2009).

Kelcey, (2009) reported phytoestrogens are estrogenic compounds found in plants, has a chemical structure similar to mammalian estrogen (estradiol-17 $\beta$ ). With this similarity, feed containing phytoestrogens once ingested can enter the circulate and are associated with estrogen receptors in the body, mimicking the effect on the body that occur naturally estradiol-17 $\beta$ . Estrogen occurs naturally in males and females that play a role in sexual reproductions and sexual behavior.

In the opinion of Glover and Assinder, (2006) stated that phytoestrogens plant substrates which have estrogenlike activity and has similarities with estradiol (Jefferson *et al.*, 2002) and has the effect of a safe (Achdiat, 2003). Reinforced by the opinions of Jefferson *et al.*, (2002) that the decomposition of phytoestrogens are naturally found in plants that have much in common with estradiol, which is a natural form of estrogen with the most potential. The use of phytoestrogens have the effect of better security than synthetic estrogen or hormones replacement such as hormone replacement therapy (HRT) (Achdiat 2003 cited in Hernawati, 2008).

# 4. CONCLUSIONS

The observations of the study include: symptoms of oestrus, oestrus behavior; respons oestrus: early of oesrus and end of oestrus. Symptoms of oestrus and behavior oestrus indicates red vulva, swollen, warm vulva, silent heat, agitated and aggressive look when brought closer to the ram. After releasing CIDR, early oestrus on Group I ( $P_0$ ) 32-36 hours with a mean ( $\pm$  SD) 33.50  $\pm$  1.73 hours and the end of oestrus 57-59 hours with an average of 58  $\pm$  1.15 hours. While early of oestrus the group II ( $P_1$ ) 26-33 hours with a mean ( $\pm$  SD) 29.00  $\pm$  3.16 hours and the end of oestrus 59-62 hours with an average of 60  $\pm$  1.41 hours. So the conclusion from the results of this study indicate that the synchronization of oestrus using Internal Control Drugs Release (CIDR) by means intravaginal on the thin tailed ewes with a percentage of 100% effectively and showed symptoms of oestrus with oestrus behavior. Early emergence of the fastest and longest oestrus and late oestrus was found in group II ( $P_1$ ), which is given sprouts as a natural feed, the early and end of oestrus respectively 29.00  $\pm$  3.16 and 60  $\pm$  1.41 hours.

# 5. ACKNOWLEDGEMENTS

Thank you profusely to BPPS DIKTI Scholarship year of 2012/2013 for funding this study.

## 6. **REFERENCES**

- [1] Alila, H W., and Hansel, W. (1984). Origin of different cell types in the bovine corpus luteum as characterized by specific monoclonal antibodies. Biol. Reprod. 31, 1015-1025.
- [2] Burroughs, C.D., H.A. Bern, and E.L.R. Stokstad. 1985. Prolonged vaginal cornification and other changes in mice treated neonatally with coumestrol, a plant estrogen. J Toxicol Environ Health. 15: 51-61.
- [3] Carlson, K.M., H.A.Pohl., J.M. Marcek., R.K. Muser, dan J.E. Wheaton. 1989. Evaluation of progesterone controlled internal drug release dis-pensers for synchronization of estrus in sheep. Anim. Reprod. Sci. 18: 205-218.
- [4] Delclos, K.B., C.C. Weis., T.J. Bucci., G. Olson., P. Mellick., N. Sadovova, J.R. Latendresse, B. Thorn, and R.R. Newbold. 2009. Overlapping but distinct effects of genistein and ethinyl estradiol (EE2) in female Sprague-Dawley rats in multigenerational reproductive and chronic toxicity studies. Reproductive Toxicology. 27: 117-132.



- [5] Falkenburg, J.A., C.V. Hulet, and C.C. Kaltenbach. 1971. *Effect of hormone combinations on estrus, ovulation and fertility in ewes.* J. Anim. Sci. 32: 1206-1211.
- [6] Feradis, 1999. Penggunaan Antioksidan dalam Pengencer Semen Beku dan Metode Sinkronisasi Estrus pada Program Inseminasi Buatan Domba *St. Croix*. [Disertasi]. Program Pascasarjana, Institut Pertanian, Bogor.
- [7] Frandson R.D., W.L. Wilke, and A.D. Fails. 2003. *Anatomy and Physiology of Farm Animal*. 7h edition. Philadelphia: Lippincott Williams and Wilkins. Pp. 395-404.
- [8] Fukui, Y., K. Tabuchi, A. Yamada, N. Hayashi, and K. Tanaka. 1994. Effect of insertion periods of controlled internal drug release (CIDR-G) on conception rate by fixed-time intrauterine insemination with frozen semen in seasonally anestrous ewes. J. Reprod. Dev. 40: 221-226.
- [9] Glover, A., and S.J. Assinder. 2006. Acute exposure of adult male rats to dietary phytoestrogen reduces fecundity and alters epididymal steroid hormon receptor expression. Jour. Endoc. 189: 565-573.
- [10] Gunnar, S. E. 2006. *Effects of phytoestrogen and mycoestrogens in domestic animals*. In: Endocrine Disrupters. Oslo: The Norwegian Academy of Science and Letters.
- [11] Hafez, E. S. E. 2000. *Reproduction in Farm Animals*. 7th ed. Lippincott Williams and Wilkins. A Wolters Kluwer Company. Philadelphia.
- [12] Hamra, A., Y. Massri., J. Marcek, and J. Wheaton. 1986. Animal reproduction science. 11: 187.
- [13] Hansel, W., and Convey, E.M. 1993. Physiology of estrus cycle. J. Anim Sci. 57: 104-412.
- [14] Hastono, I. Inounu, dan N. Hidayati. 1997. Penyerentaran Birahi pada Domba Betina St. Croix. Makalah Seminar nasional Peternakan dan Veteriner, Ciawi, Bogor.
- [15] Hernawati. 2008. Perbaikan Kinerja Reproduksi Akibat Pemberian Isoflavon dari Tanaman Kedelai. [Skripsi]. Fakultas Matematika dan Ilmu Pengetahuan Alam, Universitas Pendidikan Indonesia. Bandung.
- [16] Jefferson, W.N., B.E. Padilla, G., Clark, and R.R. Newbold. 2002. Assessing estrogenic activity of phytochemicals using transcriptional activation and immature mouse uterotrophic responses. J. Chromatogaphy. 777(1-2): 179-189.
- [17] Kelcey, S. 2009. Animal Nutrition, Technical Services Manager Information Sheet GMO's Ranch-Way Feeds.
- [18] Macnatty K.P., N.L. Hudson., M. Balland, and S. Forbes. 1988. Treatment of seasonally anestrous Romney Marsh ewes with continuous infusions of low doses of GnRH. Effects on estrus, ovulation and plasma progesterone concentrations. Theriogennology. 30: 953-960.
- [19] Ngangi, L.R. 2002. Efektivitas Lama Pemberian Implan Progesteron Intravaginal dan Waktu Inseminasi terhadap Penampilan Reproduksi Kambing Peranakan Etawah. Tesis. Program Pascasarjana. Institut Pertanian Bogor. Bogor.
- [20] Papaji, 2009. Sistem Reproduksi dan Lamanya Perjalanan Sebutir Telur. <u>http://papaji.forumotion.com/t504-sistim-reproduksi-dan-lamanya-perjalanan-sebutir-telur.</u>



- [21] Parr, M.H., M.P. Mullen., M.A. Crowe., J.F. Roche., P. Lonergan., A.C.O. Evans. 2012. The repeatability of embryo survival, and the relationship between plasma progesterone in the early luteal phase and embryo survival in dairy heifers. J. Dairy Sci. 95 :2390–2396
- [22] Rasad, S.D. 2004. Teknologi Reproduksi Ternak. Penerentakan Birahi. Lab. Reproduksi Ternak Fakakultas Peternakan. UNPAD.
- [23] Rizal, M. 2003. Pengaruh Implantasi Progesteron Intravaginal Terhadap Timbulnya Estrus Pada domba Garut Betina. J. Indon. Trop. Agric. 30 (3).
- [24] Robertson, H.A. 1980. Diagnosis of pregnancy in ewe at mid-gestation. Animal production Science. Pp. 369-76.
- [25] Roche, J.F. (1996). Control and regulation of folliculogenesis a symposium in perspective. Reviews of Reproduction 1: 19-27.
- [26] Starbuck, G.R., A.O. Darwash., G.E. Mann, and G.E. Lamming. 2001. The detection and treatment of post insemination progesteroneinsufficieny in dairy cows. In: Diskin MG, editor. Fertility in the High Producing Dairy Cow BSAS Occasional Pulication. 2:447-50.
- [27] Stronge, A.J.H., J.M. Sreenan., M.G. Diskin, J.F. Mee., D.A. Kenny, and D.G. Morris. 2005. *Post insemination milk progesterone concentration and embryo survival in dairy cows*. Theriogenology. 64(5): 12-24.
- [28] Sumoprastowo. 1987. Beternak Domba Pedaging dan Wol. Jakarta : PT Bhatara Karya Aksara.
- [29] Toelihere, M. R. 1993. Inseminasi Buatan pada Ternak. Angkasa, Bandung.
- [30] Toelihere, M.R. 2003. Increasing the success rate and adoption of artificial insemination for genetic improvement of Bali cattle. ACIAR Proceeding strategies to improve Bali cattle in eastern Indonesian. Pp. 110.
- [31] Vargas, R.B., Y. Fukui., A. Miryamoto, and Y. Terawaki. 1994. *Estrus synchronization using CIDR in heifer*. J. Reprod. Dev. 40: 59-64.
- [32] Welch, R.A.S., W.D. Andrewes., D.R. Barnes., K. Bremner, and T. G. Harvay. 1984 *The 6th International Congress of Animal Reproduction and Artificial Insemination*. Urbana-chamapaign. Pp. 354.
- [33] Wilcox, G., M.L. Wahlqvist, dan H.G. Burger, et al. 1990. Oestrogenic effects of plant foods in postmenopausal women. BMJ. 301: 905–6.
- [34] Zarkawi, M. and A. Soukouti, 2001. Serum progesterone levels using radioimmunoassay during oestrous cycle of indiginous Damascus does. New Zealand J. Agri. Res. 44: 165-169.