

ACEH CATTLE FOLLICLE DYNAMIC UNDER ENVIRONMENTAL HEAT STRESS

Teuku Armansyah¹, Tongku Nizwan Siregar², Dwinna Aliza³, Juli Melia^{2*}, Nellita Meutia⁴, Budianto Panjaitan⁵, Hafizuddin², Mulyadi Adam⁶, and Mahdi Abrar⁷

¹Laboratory of Pharmacology Faculty of Veterinary Medicine Syiah Kuala University, Banda Aceh, Indonesia

²Laboratory of Reproduction Faculty of Veterinary Medicine Syiah Kuala University, Banda Aceh, Indonesia

³Laboratory of Pathology Faculty of Veterinary Medicine Syiah Kuala University, Banda Aceh, Indonesia

⁴Indrapuri Superior Aceh Cattle Breeding Center, Aceh Besar, Indonesia

⁵Laboratory of Clinical Faculty of Veterinary Medicine Syiah Kuala University, Banda Aceh, Indonesia

⁶Laboratory of Physiology Faculty of Veterinary Medicine Syiah Kuala University, Banda Aceh, Indonesia

⁷Laboratory of Microbiology Faculty of Veterinary Medicine Syiah Kuala University, Banda Aceh, Indonesia

*Corresponding author: julimelia@unsyiah.ac.id

ABSTRACT

The objective of this research is to know Aceh cattle follicle dynamic under environmental heat stress condition. This research was conducted on December 2012 until August 2013. Animals used in this research were 20 Aceh cattle aged 5-8 years old, weighing of 150-250 kg, and had at least 2 regular cycles. All cattle used were clinically in good body condition score. The cattle were divided into two groups of 10, used in two different periods of times (December-January and July-August) to examine the effect of environmental heat to their follicle dynamic. Each group is separated into two different keeping management; one group was kept in pens while the others in the pasture, 5 cattle in each management group. Research timing was based on information acquired from Indrapuri Agency for Meteorology, Climatology, and Geophysics (BMKG) which predicted that extreme weather would last from July to August while December to January would be relatively normal in Aceh region. All cattle estrous cycle were synchronized by 5 mg/mL of PGF2 α intramuscularly (LutalyseTM, Pharmacia & Upjohn Company, Pfizer Inc.). Ultrasonography examination was performed to monitor ovary follicle's growth and dynamic during one cycle. Days during ovulation marked by standing heat was regarded as day 0 of estrous cycle. Follicular dynamic examination during estrous cycle on December-January and also on July-August, both penned and pastured cattle showed the follicle waves that was 3 follicle growth waves. The size of follicle growth on first wave (1st DF) on cattle kept in pen on July-August and December-January were relatively similar. The size of follicle growth on first wave for cattle kept in pasture on July-August and December-January were relatively similar. To conclude, Aceh cattle follicle dynamic is not change during environment heat stress condition and is not affected by different cattle-keeping management.

Key words: Aceh cattle, estrous synchronization, follicular dynamic, heat stress

ABSTRAK

Tujuan penelitian ini adalah mengetahui dinamika perkembangan folikel sapi aceh di bawah kondisi stres panas lingkungan. Penelitian ini dilaksanakan dari bulan Desember 2012 sampai dengan Agustus 2013. Materi yang digunakan dalam penelitian ini adalah 20 ekor sapi aceh, umur 5-8 tahun, mempunyai berat 150-250 kg, dan mempunyai minimal dua siklus regular. Sapi yang digunakan secara klinis sehat dan mempunyai skor kondisi tubuh dengan kriteria baik. Seluruh sapi dikelompokkan dalam dua kelompok, masing-masing berjumlah 10 ekor yang digunakan pada dua periode yang berbeda (Desember-Januari dan Juli-Agustus) untuk menguji efek panas lingkungan terhadap dinamika folikel sapi aceh. Masing-masing kelompok dibagi atas dua manajemen pemeliharaan yang berbeda, yakni sapi yang dipelihara dalam kandang dan sapi yang dipelihara di padang penggembalaan masing-masing terdiri atas lima ekor sapi. Pemilihan waktu kegiatan berdasarkan informasi yang diperoleh dari Badan Meteorologi Klimatologi dan Geofisika (BMKG) Indrapuri, yang memperkirakan pada bulan Juli dan Agustus terdapat suhu yang ekstrim sedangkan pada bulan Desember dan Januari suhu relatif normal di Provinsi Aceh. Seluruh sapi disinkronisasi berahi menggunakan PGF2 α sebanyak 5 mg/ml secara intra muskular (LutalyseTM, Pharmacia & Upjohn Company, Pfizer Inc.). Pemeriksaan ultrasonography dilakukan untuk memonitor pertumbuhan dan dinamika folikel ovarium dan dilakukan selama satu siklus. Hari pada saat ovulasi yang ditandai dengan standing heat dianggap sebagai hari ke-0 siklus estrus. Pengamatan terhadap dinamika folikuler selama siklus estrus baik pada Desember-Januari maupun Juli-Agustus, dikandangkan dan tidak dikandangkan menunjukkan jumlah gelombang folikel yang sama yakni sebanyak tiga gelombang pertumbuhan folikel. Perkembangan folikel terbesar pada gelombang pertama (1stDF) pada sapi yang dipelihara di kandang baik pada Juli-Agustus dan Desember-Januari relatif sama. Perkembangan folikel terbesar pada gelombang pertama (1stDF) pada sapi yang dipelihara tanpa kandang baik pada Desember-Januari dan Juli-Agustus relatif sama. Dari hasil penelitian dapat disimpulkan bahwa dinamika folikel sapi aceh tidak berubah pada kondisi stres panas lingkungan dan tidak dipengaruhi oleh manajemen perkandangan yang berbeda.

Kata kunci: sapi aceh, sinkronisasi berahi, dinamika folikel, stres panas

INTRODUCTION

Heat stress caused infertility and become the main source of loss in cow management (Silanikove 2000; Rensis and Scaramuzzi, 2003). It is predicted that the impact of heat stress will be more significant in the future along with the rise of average world temperature on land and sea due to global warming effect (Lendrum and Woodruff, 2006). Earth global warming might directly decrease livestock reproduction efficiency. This effect would increase body temperature which

could disturb follicle growth in the ovary and reduce steroid concentration from its normal condition. Cattle under heat stress suffered decreasing duration and intensity of estrous as the result of lower level of gonadotropin and steroid (Wolfenson *et al.*, 1995; Wijayagunawardane, 2009). The same condition was also found on goats (Ozawa *et al.*, 2005).

The disturbance in reproduction affected by heat stress is called follicle development. Cattle in heat stress would reduce their feed intake that caused lowering frequency of luteinizing hormone (LH) pulse and

lengthening the follicular wave. Lengthened follicular wave resulted in dominant follicle formed to be smaller than normal (Badinga *et al.*, 1993; Sartori and Wilbank, 2002). Follicle is responsible for estrogen hormone production. Smaller follicle has lower estrogen production which will result in lower estrogen activity (Jordan, 2003; West, 2004). Wolfenson *et al.* (2000) added that even though there is an attempt to cool temperature down had been done, cattle fertility decline would still persist.

Aceh cattle, a cross between local cattle (*Bos sondaicus*) with Zebu crossbred cattle from India (*Bos indicus*), is among one of local cattle biodiversity in Indonesia (Martoyo, 2003). Although its growth is not as fast as crossbred cattle, local beef cattle show maximum productivity and economic efficiency in limited condition. Local cattle have the advantages in terms of feed consumption efficiency, adaptation ability to Indonesia's environment (hot, humid, low quality feed, ectoparasite, and endoparasite) and its slaughter weight is suitable with the demand of local market which made it more suitable and economic to be cultivated by local people's pattern and farming condition (Romjali *et al.*, 2007).

In order to increase Aceh cattle productivity, exploration on the potency of Aceh cattle needs to be conducted. There have been many researches on Aceh cattle recently. Reproduction technology implementation by PGF2 α and progesterone has been practiced (Siregar *et al.*, 2015). Embryo transfer utilizing follicle stimulating hormone (FSH) based on the presence of dominant follicle (Siregar *et al.*, 2012) and using hypophysis extract (Arum *et al.*, 2012) has also been done. Furthermore in the last few years, vaginal epithelial cell proportion throughout estrous cycle has also been reported (Siregar *et al.*, 2016a). However, Aceh cattle reproduction under environmental heat stress has never been reported. Therefore, this research aimed to know Aceh cattle follicular dynamic under environmental heat stress.

MATERIALS AND METHODS

The research was conducted in Indrapuri Superior Aceh Cattle Breeding Center (BPT-HMT) from December 2012 until August 2013. Animal used in this research were 20 Aceh cattle aged 5-8 years, weighing 150-250 kg, with at least 2 regular cycles. All cattle used were owned by Indrapuri Superior Aceh Cattle Breeding Center, Aceh Besar. The cattle used were clinically and have good body condition score. Cattle were fed both concentrate and forage. Cattle were sorted into two research groups, 10 cattle in each group, used in two different periods (December-January and July-August) to evaluate environmental heat's effect on Aceh cattle reproduction ability. Each group was divided into two cattle-keeping managements, cattle kept in pens and cattle kept in pastures with 5 cattle in each treatment. Research time allocation was based on information obtained from Indrapuri Agency for Meteorology, Climatology, and

Geophysics (BMKG), which predicted that there would be extreme weather for July and August while being relatively normal on December and January around Aceh region.

Estrous Synchronization

All cattle were estrous synchronized using PGF2 α 25 mg/mL intramuscularly (LutalyseTM, Pharmacia & Upjohn Company, Pfizer Inc.). Cattle used were not in gestation, but had corpus luteal in their ovaries. The cattle were fed with forage twice a day and concentrate once a day with ad libitum drinking water.

Body Temperature Measurement

Body temperature was measured by digital thermometer (Electronic Digital Clinical Thermometer MT-B132F) with the measured temperature ranged between 32° C to 43° C with 0.1° C of accuracy. Temperature was measured by inserting proof thermometer 2-3 cm into rectal for 2-3 minutes on 14.00 (GMT+7). Measurement took place every day on 14.00 (GMT+7).

Ultrasonography

Transrectal ultrasonography was performed by ultrasound scanner equipped by 5MHz transducer (Ultrascan 90 Alliance, Quebec, Canada). Ultrasonography examination on the cattle was performed to monitor the growth and dynamic of ovary follicle. The examination was performed for one cycle. Ovulation time was marked by standing heat and regarded as day 0 of estrous cycle.

Data Analysis

Data of follicle growth and dynamic were analyzed descriptively.

RESULTS AND DISCUSSION

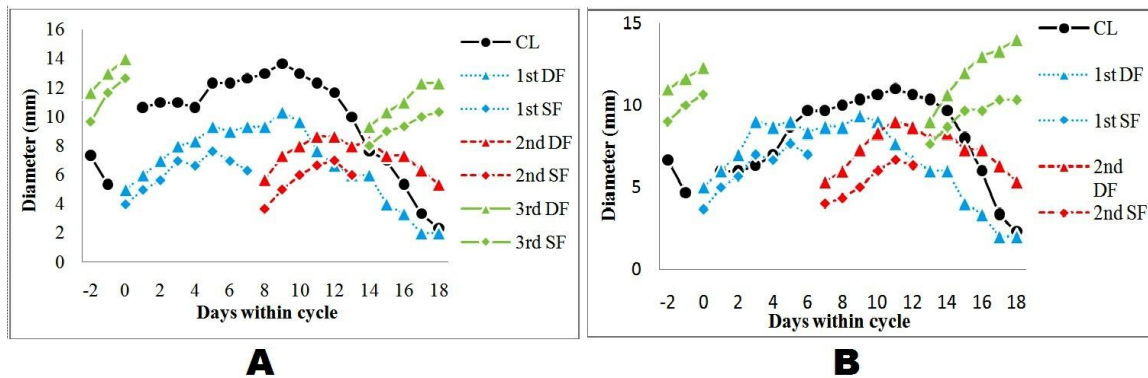
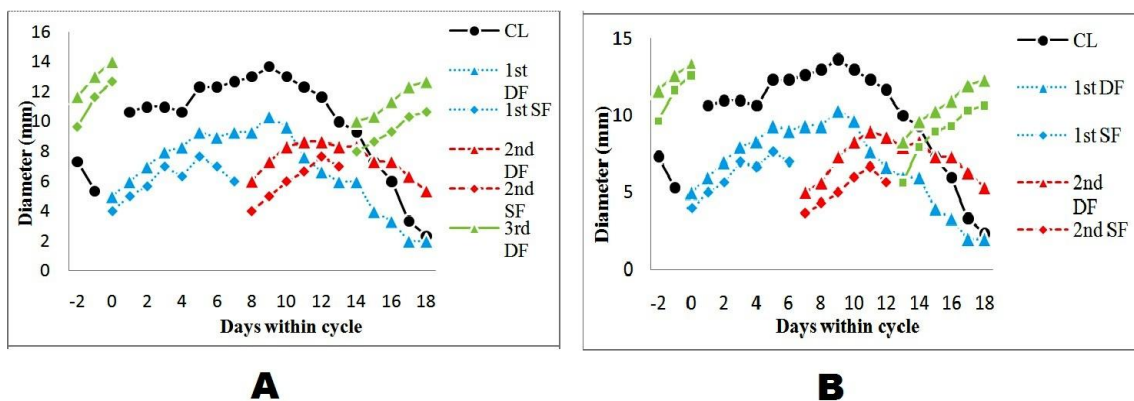
The average temperature and humidity on July-August and December-January are 26.5 \pm 1.52° C and 85 \pm 3.20% and 30.5 \pm 1.32° C and 88 \pm 4.68%, respectively. Converted into temperature humidity index (THI), July-August was categorized as 3 (mild stress) while December-January categorized as 4 (stress tendency). Examination on follicle dynamics during stress cycle on December-January and July-August for both penned and pasture cattle showed the same result of three follicle waves. The characteristics of follicular dynamic during estrous cycle on two different cattle-keeping managements were presented on Table 1. Follicular wave of Aceh cattle estrous cycle in two hot environments and two different cattle-keeping managements were presented on Figure 1 and Figure 2.

Follicle growth characteristics obtained in this research are almost the same with the result reported by Siregar *et al.* (2016b) who found three follicular waves on Aceh cattle. This condition also supported by Adams (1999) who stated that >95% of cattle during their estrus cycles will have two or three follicular waves. In addition, usually cattle with two follicular waves have

Table 1. Characteristics of follicle growth in Aceh cattle on two hot environments and two different cattle-keeping managements

Characteristics	Cattle in pens		Cattle in pasture	
	July-August	December-January	July-August	December-January
Length of estrus cycle (days)	17.67±0.58	18.00±1.00	18.00±1.00	18.33±1.15
Number of follicular wave	3	3	3	3
Post-synchronization ovulated follicle diameter (Ov. DF) (mm)	13.00±1.73	11.67±0.58	14.00±3.00	12.67±1.55
Ovulating follicle diameter (3 rd DF) (mm)	12.67±1.15	12.33±0.58	12.33±0.28	14.00±2.00
Largest CL diameter (mm)	H9 13.67±2.08	H9 13.67±2.08	H9 13.67±2.08	H11 11.00±1.00

DF= Dominant follicle; Max= Maximum; SF= Subordinate follicle

**Figure 1.** Aceh cattle follicular dynamics during dry season (A= Kept in pens; B= Kept in pastures)**Figure 2.** Aceh cattle follicular dynamics during rainy season (A= Kept in pens; B= Kept in pastures)

19-20 days of estrous cycle length whereas cattle with 3 follicular waves have 22-23 days. The previous statement did not match the result on Aceh cattle. The average estrous cycle length in Aceh cattle is 18 days with 3 follicular waves. The number of follicular waves detected was the same with the result reported by Perez *et al.* (2003) in FH cattle, Amrozi *et al.* (2004) in Japanese black cattle, and Melia (2010) in PO cattle.

Normal estrous cycle consists of two or three follicular waves, which comprised of emergence, growth, deviation, dominant, atresia, or ovulation period. Each wave involves follicle growth which ranging from big, average, to small. Deviation period consists of the elimination or stalling of second largest follicle (subordinate follicle = SF) and the largest follicle turns dominant and becomes larger. This deviation period is a selection mechanism that controls

several ovulating follicle into a single ovulating follicle (Ginther *et al.*, 2003).

Figure 1 and 2 show that the largest follicle in the first wave (1st DF) in normal season is relatively similar. The growth of follicle has begun since day 0 which is during estrous (5.0 mm vs 4.0 mm) and reached its peak on day 5 (9.33 mm vs 9.33 mm) and started to turn atresia since day 6 (9.0 mm vs 9.33 mm) until the end of cycle. The second largest follicle (1st SF) increased from day 0 (7.67 mm vs 7.67 mm) then turned atresia until day 7 (6.33 mm vs 6.0 mm). On second wave, the largest follicle (2nd DF) increased from day 8 (5.67 mm vs 6.0 mm) and reached its peak on day 12 (8.75 mm ±0.50 mm) then turned atresia since day 13 (8.00±0.82 mm) until the end of the cycle. Second largest follicle (2nd SF) increased from day 8 (4.75±0.96 mm) and reached its peak on day 12 (8.67

mm vs 8.67 mm) then turned atresia, with deviation day on day 13 (6.0 mm vs 7.0 mm). On the third cycle, there was a very rapid growth from largest follicle (3rd DF) and could clearly be observed since day 14 (9.3 mm vs 10.00 mm) until it became dominant follicle ready for ovulation on day 18 (12.33 mm vs 12.67 mm). Second largest follicle (3rd SF) increased from day 14 (8.0 mm vs 8.0 mm) and reached its peak on day 18 (11.00±1.4 mm) with deviation day also on day 18 (10.67 mm).

The growth of largest follicle on first wave (1st DF) in cattle kept in pasture on both December-January and July-August are relatively similar. Follicle growth had started since day 0 which was estrous (5.0 mm vs 4.0 mm) and reached its peak on day 5 (9.0 mm vs 9.33 mm) then turned atresia since day 6 (8.33 vs 9.00 mm) until the end of cycle. The second largest follicle (1st SF) increased from day 0 (3.67 vs 4.0 mm) and reached its peak on day 5 (7.67 vs 7.67 mm) then turned atresia until day 7 (6.0 vs 6.0 mm). On second wave, the largest follicle (2nd DF) increased from day 7 (5.33 vs 5.00 mm) then reached its peak on day 11 (9.0 vs 9.0 mm) and turned atresia on day 12 (6.33 vs 5.67 mm) until the end of cycle. Second largest follicle (2nd SF) increased from day 8 (4.00 vs 3.67 mm) then reached its peak on day 11 (6.67 vs 6.67 mm) then turned atresia, with deviation day on day 12 (6.33 vs 6.67 mm). On third wave, a very rapid growth was observed from largest follicle (3rd DF) and clearly observed since day 13 (9.00 vs 8.33 mm) until it turned into ready to ovulate dominant follicle on day 18 (14.00 vs 12.33 mm). Second largest follicle (3rd SF) increased from day 13 (7.67 vs 5.67 mm) and reached peak on day 18 (mm) with deviation day also on day 18 (10.33 vs 10.67 mm).

During the wave, the process of follicle growing and turning atresia was slower than in other wave. Probably it was caused by the delay of progesterone hormone on gonadotropin hormone secretion (Fortune, 1993). The small follicle atresia on second wave was caused by corpus luteal and dominant follicle. It was proven by how the taking of dominant follicle from the ovary would be followed by the growth of the next small follicles (Hariadi, 2005). It was also in line with Adams *et al.* (1994) who stated that the presence of dominant follicle will hinder the emergence of the next follicular wave. Ovulation during the wave happens because of corpus luteal regression that causes progesterone level to fall, and further result in the disappearance of negative feedback on gonadotropin. Senger (2003) stated similarly that ovulation can't happen when progesterone level is dominant.

Several reports on follicular dynamics during estrous cycle research, such as Gaur and Purohit (2007) reported that Ratih cattle (*Bos indicus*) has two and three waves within its estrous cycle. The report showed that as many as 78.57% of cattle have two follicular waves while the rest have three follicular waves (21.42%). Putro *et al.* (2008) reported that FH crossbred cattle have two follicular waves in their estrous cycle.

According to Perez *et al.* (2003), cattle with three follicular waves have average 1st DF (anovulatory) diameter larger than other follicles, but smaller than those in cattle having two follicular waves. This finding is different with the result obtained from this research, with Aceh cattle exhibiting 3 follicular waves having average 1st DF diameter smaller than average 3rd DF diameter (ovulatory). The result in this Aceh cattle is the same with the one found on PO cattle that has been reported by Melia (2010).

Even though in this research two follicular waves were found, but as a comparison, more than three follicular waves has been found by many cattle researchers in Europe (Savio *et al.*, 1990; Sirois and Fortune, 1988) and in *Bos indicus* cattle (Viana *et al.*, 2000; Gaur and Parohit, 2007). In this research no four follicular waves or more found like in gir cattle (Viana *et al.*, 2000), which is a part of a small proportion of cattle showing four or more follicular waves during estrous cycle. In conclusion, Aceh cattle follicular dynamics does not change under environmental heat stress and is not affected by different cattle-keeping management.

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