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Cortisol and Estradiol Profile in Cross-bred Ettawa Does: The Effects of Body Condition Scoring (BCS).

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Abstract

Body Condition Scoring (BCS) is an estimation of the muscle and fat development of an animal. Thin ewes that are fighting to maintain their own body weight and low concentration of cortisol are not able to ovulate as ewes in a more desirable condition due to lack of oestradiol concentration. The aims of this research are to monitor the cortisol and oestradiol profile in Cross-bred ettawa does and to determine effect of BCS on the cortisol and oestradiol profile. Eight does were used in this research. These animals were devided equally into 2 groups based on Body Condition Scoring (BCS), namely BCS 2, which body weight range between 25-30 kgs as group I (n=4) and BCS 3 which consists of ettawa with body weight range between 33-40 kg as group II (n=4). All animals were synchronized using implant of CIDR and PGF2alpha. Blood from jugular vein were collected every 3 and 6 hours as soon as oestrus until 72 hours. Serum contained cortisol and oestradiol then assayed using ELISA method. Cortisol and oestradiol concentrations were compared between groups by T test. The results showed that average concentration of cortisol is 47.17 ± 42.19 ng/mL for BCS 2 and 112.40±74.41ng/mL for BCS 3 ($P<0.05$), whereas concentration of oestradiol is 72.25±30.62 pg/mL for BCS 2 and 145.72±100.18 pg/mL for BCS 3 ($P<0.05$). Either cortisol or oestradiol have very synchronized wave except 2 of animals from BCS 2 (50%), which has tendency to suppress each other. It was concluded that profile of cortisol and oestradiol hormone have a very similar pattern, and BCS can affect hormone profile.

Keywords : cortisol, estradiol, body condition score, ettawa

Introduction

Body Condition Scoring (BCS) is an estimate of the muscle and fat development of an animal. This scoring is very important especially for 3 conditions such as breeding, late gestation and lambing. Thin ewes, that are fighting to maintain their own body weight are not able to ovulate as ewes in a more desirable condition due to lack of oestradiol concentration. Cortisol is one of cortex adrenal hormones which is responsible for gluconeogenesis (Litwack and Schmidt, 2002). More modest changes in nutrient intake can influence the plasma concentration of a range of counter regulatory metabolic hormones, including those secreted from the adrenal and thyroid glands, the pituitary, as well as leptin release from adipose tissue (Bispham *et al.* 2003). Brook and Marshall (1996) reported that cortisol also stimulates the appetite which is necessary to compensate for lack of body weight. The profile of cortisol, both in ideal or poor BCS of ewes, is unclear. The cortisol level in poor BCS can disturb or furthermore suppress oestradiol as an important reproductive hormone. Breen *et al.* (2005) reported that plasma cortisol interfere the follicular phase by suppressing the

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development of high frequency LH pulses, which compromises the preovulatory estradiol rise and LH and FSH surges. Moreover, the suppression of LH pulse frequency provides indirect evidence that cortisol acts centrally to suppress pulsatile GnRH secretion in follicular-phase cow (Gentry, 2007) and ewes (Breen *et al*. 2004). The Elevations of glucocorticoids suppress pulsatile LH secretion in sheep, but the neuroendocrine sites and mechanisms of this disruption remain unclear (Tilbrook *et al.* 2000). In the previous theory, elevations in glucocorticoids inhibit reproductive neuroendocrine activity in a variety of species, ranging from rodents to primates and domestic animals (Tilbrook *et al.* 2000; Tilbrook *et al*. 2002).

This work was conducted to determine profile or pattern of cortisol and oestradiol in Crossbreds ettawa does and also to determine effect BSC on concentration of cortisol and estradiol

Materials and methods.

Eight adult (1,5-2 years old) goat with a body weight of 25-40 kg were used in this study. Animals were maintained in the Farm Unit of Peranakan Etawah does in Balai Pembibitan Ternak dan Hijauan Makanan Ternak (BPT-HMT), Desa Toyomerto, Singosari, Malang. They were fed a standart of ransum which contains 5 kg/day/animal of King grass (rumput gajah), 500-800 g/day/animal concentrate, and *ad libitum* of water. Prior to blood withdrawn, animals were assessed for parasites.

These animals were devided equally into 2 groups namely BCS 2, body weight 25-30 kg as group I (4 animals) and BCS 3 body weight 33-40 kg as group II (4 animals). Determination of BCS was chosen based on the previous studies (Sphar, 2005) which assessed from the vertical bone protrusion (spinous process) and the horizontal protrusion (transverse process) of the loin . Ewe are scored from 1 (Emaciated) to 5 (Obese) based on the level of muscling and fat deposition around the loin. Healthy, well nourished ewe should not be over fat or very thin and free of a large parasites. To make similar reproductive status, animals were synchronized using implant of CIDR and PGF2alpha then blood collection from jugular vein were done every 3 and 6 hours as soon as oestrus until 72 hours. The blood was drawn with 3-mL syringes and transferred into 5-mL glass tubes containing 1 drops of heparin. Plasma was harvested and stored at –20° C until cortisol, and oestradiol concentrations were measured. using ELISA method (DRG, Germany).

Results and Discussion

Our result showed, average of cortisol concentration in BCS 2 is 47.17 ± 42.19 ng/mL whereas in BCS 3 is higher than group I 145.72±100.18 ng/mL ($P<0.05$) (Table 1). For oestradiol, average of this hormone in BCS 2 is 72.25±30.62 pg/mL whereas in group II (BCS 3) is 145.72±100.18 pg/mL ($P<0.05$) (Table 2). It would be predicted this is caused by lacking of metabolism both carbohydrate or lipid and glyconeogenesis according to their thin body. Guyton& Hall (1996) reported that cortisol is one of steroid hormone which give responsibility of carbohydrate, lipid, and protein metabolism also glyconeogenesis. In human, obese people have higher cortisol levels than lean people (Spudich, 2007).

Table 1. Concentration of Cortisol (ng/ml) in group I (BCS 2) and group II (BCS 3)

	Animal 1 Cort (ng/mL)	Animal 2 Cort (ng/mL)	Animal 3 Cort (ng/mL)	Animal 4 Cort (ng/mL)	Average Cort (ng/mL)
BCS 2	63.15± 82.29	40.85±16.15	27.51±27.80	57.29±38.80	47.16±42.19[a]
BCS 3	115.98±42.28	109.08±135.23	160.49±83.60	64.06±36.56	112.40±74.41[b]

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

Table 2. Concentration of oestradiol (pg/mL) in group I (BCS 2) and group II (BCS 3)

	Animal 1 Oestradiol (pg/mL)	Animal 2 Oestradiol (pg/mL)	Animal 3 Oestradiol (pg/mL)	Animal 4 Oestradiol (pg/mL)	Average Oestradiol (pg/mL)
BCS 2	95.25± 60.01	70.96±26.01	26.38±4.20	96.40±32.25	72.25±30.62 [a]
BCS 3	187.45±82.4	28.14±30.03	290.08±221.11	77.21±67.13	145.72±100.18 [b]

Note: different superscript indicates significantly dif ferent (P<0.05).

Based on the hormone profile, cortisol is fluctuated (Figure 1 and 2) by hours to hours.

Cortisol exhibited significant fluctuation over time and may, like testosterone be subject to pulsatile regulation by the episodic release of ACTH from the anterior pituitary (Liota&Krieger, 1990). Beside releasing of ACTH, this fluctuation has occurred due to feed back mechanism by releasing of CRF (Corticotropin Releasing Factor) from hypothalamus (Guyton& Hall, 1996). If it compared to profile of oestradiol for each animal, it seems very synchronized profile (figure 1 and 2), both of them have a similar tendency unless BCS 2 (1) and BCS 2 (3). In this case (BCS 2, animal number 1 and 3), their profile seem to be contraindication; when concentration of cortisol is getting decrease the oestradiol become increase and their opposite. There are 2 reasons which have predicted: first, the contradictive profile maybe due to competition of receptor sites between cortisol and progesterone. Brown and Pentland (2005) demonstrated that long term low level stress and/or poor nutrition fatigue the adrenals which causes a cortisol deficiency. This stress hormone competes with progesterone for receptor sites, leading to a condition of estrogen dominance and less active progesterone. This state is similar as our result especially in group I (animals no. 1 and 3). Furthermore, progesterone is used to produce three different types of estrogen, testosterone, cortisol, and aldosterone. Second, as we know

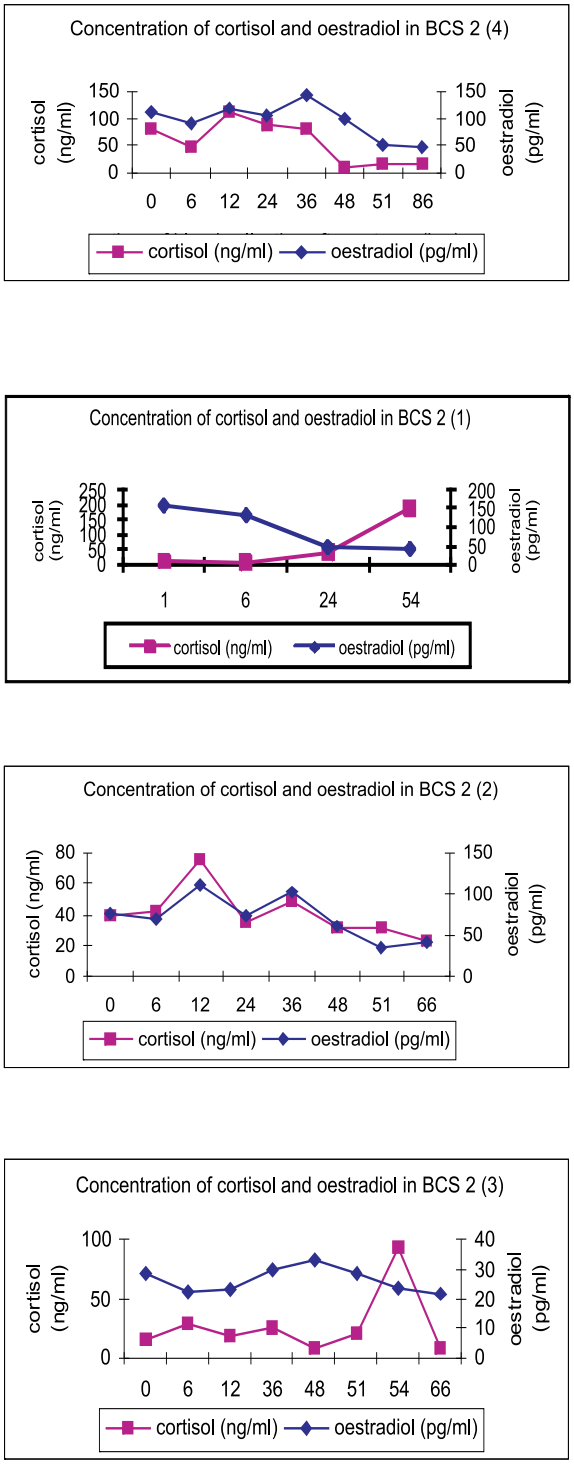

Figure 1. Profile of cortisol and oestradiol in animals which categorized BCS 2 (group 1)

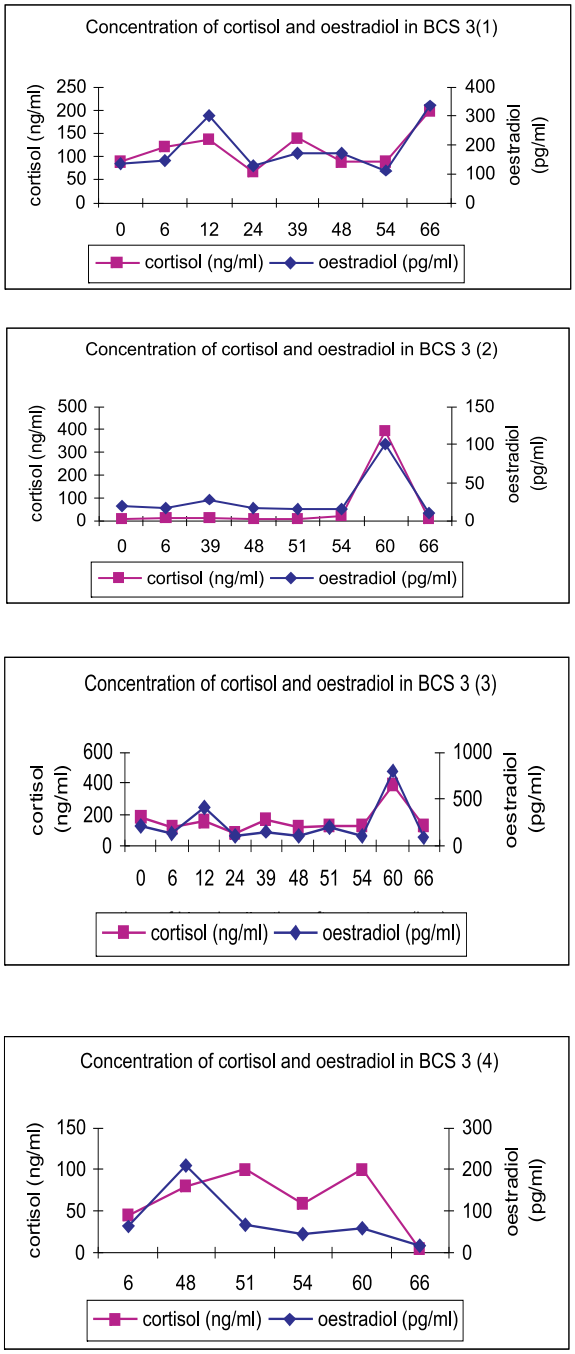

Figure 2. Profile of cortisol and oestradiol in animals which categorized BCS 3 (group II).

that oestradiol is steroid hormone which synthezied from cholesterol, so if metabolism of lipid is only a little as in BCS 2, not surprising that BCS 2 has very low concentration both cortisol and oestradiol. Brown and Pentland (2005) reported, theca internal cells continually release estrogen, which caused the pituitary gland and therefore the release of follicle stimulating hormone; too little fat cells in the body (being more than 15% underweight) causes estrogen levels to drop and difficult for the body to reach a state where ovulation can occur. In addition, a reduction in maternal cortisol during a prolonged period of nutrient restriction is likely to be due to a decrease in maternal cortisol secretion. A reduction in maternal cortisol, together with lower plasma leptin, IGF-I, and T_4, may act to reduce maternal carbohydrate oxidation and promote lipolysis, as indicated by the rise in maternal NEFA, (Bispham *et al.* 2003). In nutritionally restricted ruminants decreased serum concentration of luteinizing hormone (LH) and decreased LH pulse frequency characterize nutritional anestrus (Richards *et al.* 1989), thus negatively impacting follicular growth. In a review of reproduction, Randel (1990) speculated that the nutritional mechanism controlling ovarian activity may have its effect on the hypothalamus, pituitary or ovary directly. Because ovarian function is controlled by the hypothalamic release of GnRH causing a release of gonadotropins from the pituitary, the hypothalamic-pituitary axis is where control of the nutritional mechanism is most likely located. cortisol has been found to reduce GnRH receptor message and protein expression in sheep, although this effect requires the presence of estradiol (Adams *et al.* 1999). Furthermore Breen *et al.* (2004) reported that cortisol inhibits reproductive neuroendocrine function at the pituitary level. Specifically, an acute, stress-like elevation in cortisol inhibits pituitary responsiveness to

physiologically relevant pulses of GnRH. The important thing that cortisol does not acutely inhibit GnRH pulsatility in the absence of gonadal steroids.

Stress-induced inhibition of tonic gonadotropin secretion below levels required for follicular maturation would be expected to inhibit ovulation. An additional mechanism whereby acute stressors could interfere with ovulation is through inhibition of the preovulatory gonadotropin surge (Lujan *et al*. 2002). However, in nonpregnant adult sheep, 4 d of food withdrawal has no effect on plasma cortisol, and in late gestation, a 50% reduction in maternal food intake only causes a transient rise in maternal plasma cortisol and has no effect on cortisol in the fetus (Edwards *et al*. 2001). Whether other metabolic hormones, particularly leptin, may be consistently affected by under nutrition during pregnancy also remains unclear. The reason why maternal nutrient restriction has a markedly different effect on cortisol between early to mid gestation compared with late gestation (Edwards *et al*. 2001) may be due to the much higher fetal glucose demands in late gestation (Molina *et al*. 1991).

Fortunately, even concentration of cortisol is lower than group II, they still have good respond of oestrus (Table 3) but none animal to conduct ovulation

Table 3. Respond of oestrus after oestrus synchronization using CIDR and PGF2alfa

Variable	Group I (BCS 2)	Group II (BCS 3)
Number of animal	4	4
Respond of oestrus (%)	100	100
Ovulation (%)	0	100

In BCS 3 group, its appear that hormone profile in all of animals are very synchronized each other, and peak of oestradiol as a sign of oestrus and very important for praovulatory also very clear. Different from BCS 2, onset of ovulation in all of animals in the BCS 3 are very clear too. This result is supported by Lujan *et al*. (2002), they demonstrated that in monkey, sheep and other rodents, the rise of oestradiol is an essential stimulus for the preovulatory that induces the LH surge.

It would be concluded that in general, profile of cortisol and oestradiol seem to be have a similar pattern. Also, BCS is very important component because it can affect the hormone profile.

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Refferences

Adams TE, Sakurai H, Adams BM. 1999. Effect of stress-like concentrations of cortisol on estradiol-dependent expression of gonadotropin-releasing hormone receptor in orchidectomized sheep. *Biol Reprod.*, 60:164–168

Bispham J, Gopalakrishnan GS, Dandrea S, Wilson V,Budge H, Keisler DH, Broughton F, Pipkin, Stephenson T and Symonds ME. 2003. Maternal Endocrine Adaptation throughout Pregnancy to Nutritional Manipulation: Consequences for Maternal Plasma Leptin and Cortisol and the Programming of Fetal Adipose Tissue Development. *Endocrinol.*,(144):(8):3575-3585

Breen KM, and Karsch FJ. 2004. Does Cortisol Inhibit Pulsatile Luteinizing Hormone Secretion at the Hypothalamic or Pituitary Level? *Endocrinol.*, (145): (2): 692-698

Breen KM, Heather J. Billings, Elizabeth R. Wagenmaker, Emily W. Wessinger and Fred J. Karsch, 2005. Endocrine Basis for Disruptive Effects of Cortisol

on Preovulatory Events, *Endocrinol.*, (146): (4): 2107-2115

Brook CGD and Marshall NJ. 1996. *Essential Endocrinology*, 3rd edition. Blackwell Science Ltd. United Kingdom.

Brown L and Pentland S. 2005. *Hormone: Keeping the Balance*.ealth Information Organization Health Information.http//:www.spence@acubalance.ca

Edwards LJ, McMillen IC. 2001 Maternal undernutrition increases arterial blood pressure in the sheep fetus during late gestation. *J Physiol.,* 533:561–570

Gentry GT Jr . 2007. Effect of Age, Body Condition, Pregnancy and Lactation on Circulating Leptin Concentrations in Beef cattle. *Dissertation.* Faculty of the Louisiana State University and Agricultural and Mechanical College in The Interdepartmental Program in Animal and Dairy Sciences.

Guyton MD, Hall JE. 1996. Textbook of Medical Physiology.9th ed.. WB. Saunders Company, Philadelphia, Pennsylvania.

Liotta AS, Krieger DT. 1990. ACTH and Related Peptide. In; Baulieu EE, Kelley PA, editors. Horones: from molecules to disease. New York: Champman and Hall

Litwack, G and Schmidt TJ. 2002. Biochemistry of Hormones II: Steroid Hormones,*in*: Textbook of Biochemistry and Clinical Correlations 5th ed.John Wiley and Sons.Pp 959-988

Lujan M.E, MacTavish, Alicja A. Krzemien, Michael W. Bradstock and Dean A. Van Vugt . 2002. Estrogen-induced gonadotropin surge in rhesus monkeys is not inhibited by cortisol synthesis inhibition or hypoglycemia. *Endocrinol.*,(19):(2):169-176

Molina RD, Meschia G, Battaglia FC, Hay WW. 1991 Gestational maturation of placental transfer of glucose. *Am J Physiol.,* 261:697–704

Randel RD. 1990. Nutrition and postpartum rebreeding in cattle. *J. Anim. Sci. 68:853-62*

Richards, MW, Wettemann RP, and Schoenemann HM 1989. Nutritional anestrus in beef cows: Body weight change, body condition, luteinizing hormone in serum and ovarian activity. *J. Anim. Sci.* 67:1520-6.

Sphar LI. 2005. Body Condition Scoring in meat Goats. *Penn State College of Agricultural Science*., 23:11-15

Spudich T. 2007. *Cortisol and Weight*. Project Aware. North Carolina, USA. Aware-editor@project-aware.org

Tilbrook AJ, Turner AI, Clark IJ. 2002 Stress and reproduction: central mechanisms and sex differences in non-rodent species. *Stress.,* 5:83–100

Tilbrook AJ, Turner AI, Clark IJ. 2000 Effects of stress on reproduction in non-rodent mammals: the role of glucocorticoids and sex differences. *Rev Reprod.,* 5:105–113