Jurnal Veteriner Juni 2010 Vol. 11 No. 2 : 87-91
ISSN : 1411 - 8327

Physiological Response of Bligon Buck to Transportation: Relation to Level of Thyroid Hormone

(RESPON FISIOLOGIS KAMBING BLIGON YANG MENGALAMI TRANSPORTASI: KETERKAITAN DENGAN KADAR HORMON TIROID)

Pudji Astuti [1], Sarmin [1], Asmarani Kusumawati [2], Claude Mona Airin, [1], Hera Maheshwari [3], Luthfiralda Sjahfirdi [4]

[1] Department of Physiology, [2] Department of Reproductive
Faculty of Veterinary Medicine, Gadjah Mada University
Jl. Fauna No.2 , Karang Malang, Yogyakarta.
Email: pastuti2001@yahoo.com, Phone: 08156851163
[3]Department of Physiology, Faculty of Veterinary Medicine,
Bogor Agricultural University, Dramaga, Bogor
[4] Department of Biology, Faculty of Mathematics and Natural Sciences,
University of Indonesia, Depok, Indonesia

ABSTRACT

Transportated animals may subject to a variety of physical stimuli including metabolism, crowding, noise, handling, isolation, agitation, and extreme temperature .The aim of this study was to determine the changes of serum T_4 and T_3 concentration, during animals transportation. Six adult Bligon buck with body weight ranging from 26-30 Kg were used in this study. Two weeks prior to the experiment, the animals were given anthelmintic Albendazole to eliminate egg worm. All animals were fed standard diet in their pen at 10% of their body weight per head daily and commercial concentrate also given everyday. Fresh water was provided *ad libitum.* All animals were transported around village for 16 hours starting from 18.00 pm until 10.00 am in open small truck (3 x 2 m); eye contact each others would be possible. Blood samples were withdrawn from jugular vein using vacutainer tubes containing heparin into 1.5 mL glass tubes, then centrifuged at 500 g for 15 minutes. Plasma was collected to be stored at –20° C. The blood were collected every 4 hours from 8 hours before transportation (at 10.00 am, 14.00 pm and 18.00 pm) until the time of arriving after transportation at 10.00 am. Plasma was harvested and stored at –20° C until T_3 and T_4 concentrations were measured using ELISA method (enzyme linked immunosorbent assay) product DRG, Germany. The result showed that transportation of Bligon bucks for 16 hours have an affect on level of T_4 only ($P<0.05$) and not for T_3 concentration ($P>0.05$) due to physical stimuli such as crowding, heat stress, noise, handling would be discarded so that the metabolic process was stable. During transportation, decreasing of T_4 levels indicated conversion of T_4 to T_3 to form active hormone.

Key words: Bligon bucks, transportation, Triiodothyronine (T_3), Thyroxine (T_4)

INTRODUCTION

Bligon goat is one of very popular livestock especially for meat consumption. Routine transportation of bucks between farm and markets or other cities for slaughtering has been running almost everyday. The length of time of transportation is vary starting from 2 until 16 hours. Transported animals may be exposed to a variety of physical and psychological stimuli including crowding, noise, handling, isolation and extreme temperatures (Al-Kindi et al. 2005). Transportation of animal is generally recognized as a stressful event. Although tremendous number of study on the effects of handling and transportation of cattle, pigs and poultry have been done, little work has been carried out to assess the effect of transportation of local goat on thyroid hormone concentration namely Triiodothyronin (T_3) and Thyroxine (T_4).

Thyroid hormones, T_3 and T_4, were reported to respond to variety of stressors in different fashions (Garriga et al. 2006 and Hangalapura et al. 2004). In cattle, the stress of transportation has been shown to cause lower carcass yields and dark cutters. Higher plasma levels of T_4 in

Suffolk ewes were shown to be positively related to larger body size and enhanced growth potential (Williams et al., 2004), whereas the decline in serum T_4 levels induced by feed restriction was greater in crossbreed ewes than in native Indian sheep (Naqvi and Rai, 1991). Stress of transportation induce an increase of activity of the hypothalamus-hypophysis-thyroid axis, together with peripheral tissue request (Fazio et al. 2005). T_3 stimulates mRNA transcription resulting in protein synthesis and anabolic effects. Other effects are in the form of increased temperature, behavioral, activity, weight loss, increased glucose turn-over, cholesterol catabolism, stimulation of growth and maturation (Altiner , 2006).

The aim of this study was to determine the change of thyroid hormone namely T_3 and T_4 in Bligon goat during long transportation (16 hours). These physiological parameters have been proposed as sensitive indices of physiological stress response of animals encountering long-term welfare problems such as handling and transportation.

MATERIAL AND METHODS:

Animals: Six adult bligon bucks ranging in body weight 26-30 Kg were used in this study. Two weeks prior to the experiment, the animals had been given anthelmintic Albendazole to eliminate egg worm. All animals were fed standard diet in their pen at 10% of their body weight per head daily and commercial concentrate also given everyday. Fresh water was provided *ad libitum.*

Treatment: All animals were transported around village for 16 hours starting from 18.00 pm until 10.00 am in open small truck (3 x 2 m); eye contact among animals would be possible. Blood samples were withdrawn from jugular vein using vacutainer tubes containing heparin was transferred into 1.5 mL glass, then centrifuged at 500 g for 15 minutes. Plasma was collected and stored at –20° C until T_3 and T_4 concentrations were measured using commercial ELISA (enzyme linked immunosorbent assay) kit product DRG, Germany. The blood was collected every 4 hours from – 8 hours before transportation (at 10.00 am, 14.00 pm and 18.00 pm) until the time of arriving after transportation at 10.00 am.

ELISA for plasma hormone concentration: The Triiodothyronine and Thyroxine kit is a solid phase enzyme–linked immunosorbent assay based on the principle of competitive binding. The microtitter wells are coated with monoclonal antibody directed towards an antigenic site on the T_3 and T_4 molecules. Endogenous hormone of sample competes with a T_3 and T_4-horseradish peroxidase conjugate for binding to the coated antibody. After incubation, the unbound conjugate was washed off. The amount of bound peroxidase conjugate is inversely proportional to the concentration of hormone in sample. After addition of the substrate and stop solution, the intensity of color would be inversely to concentration of hormone. Concentrations of T_3 and T_4 were determined in duplicate samples by EIA.

Statistical analysis: The differences in level of T_3 and T_4 were contrasted using ANOVA and continued by Tukey HSD test.

RESULTS AND DISCUSSIONS

Plasma T_3 and T_4 levels of bucks are given in Table 1 and Figure 1. Concentration of T_3 tended to lower in 4 hours after transportation, but no significant differences were found before and after transportation ($P>0.05$). Before loading, level of T_4 was normal then got to increase at night and early in the next morning until the time of arriving; there was significant differences were found before and after transportation ($P<0.05$).

Table 1. Average plasma levels of T_3 and T_4 before and after transportation

Indices	Period of transportation (hours)						
	-8	-4	0	4	8	12	16
T_3(nmol/dL)	1.0 ± 0.1	1.7± 0.7	1.0± 0.3	0.8±0.1	1.0 ± 0.1	2.5±0.3	1.0 ± 0.1
T_4(nmol/dL)	27.2±8.07	28.2±9.9[a)]	33.0±2.9[b)]	33.5±6.9	30.2±10.2	27.9±14.0[a)]	37.1±7.6

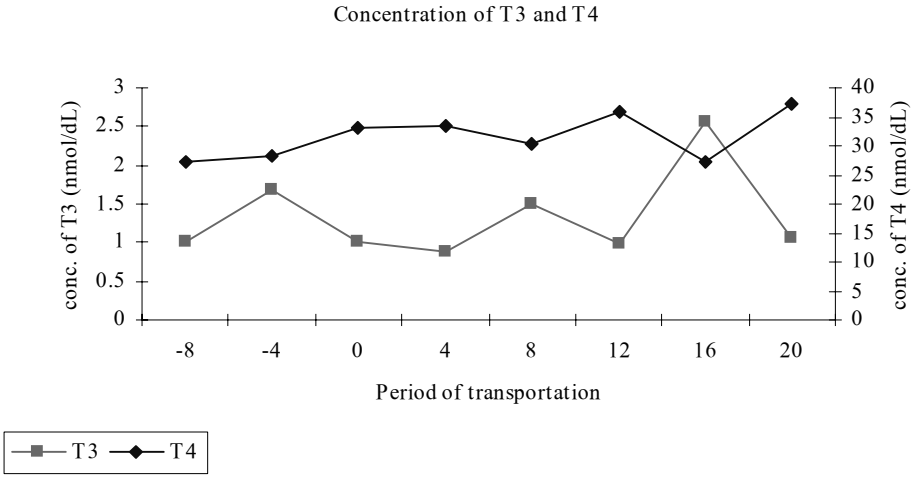

Figure1. Changes of plasma T3 and T4 activity in Bligon bucks during transportation. Values are express as average ± SD. Minus symbol (-) represent before loading and 0 just loading.

Triiodothyronine

Triiodothyronine (T3) is one of thyroid hormone which derivates from the amino acid tyrosine. T3 stimulates mRNA transcription, resulting in protein synthesis and anabolic effects. Also, it stimulates Na+, K+-ATPase at the cell membrane, thus increasing the oxygen consumption. Small amounts of the active hormone T3 come from the thyroid, but in adult sheep at least 50% of serum T3 and 97% of serum rT3 derive from monodeiodination of T4 in peripheral tissues (Fisher et al., 1972; Chopra et al., 1975)

Plasma total T3 concentrations significantly correlated with energy and nutrition balance. The overall effects are to increase the basal metabolic rate, to make more glucose available to cells, to stimulate protein synthesis, to increase lipid metabolism and to stimulate cardiac and neural functions (Capen and Martin, 1989 *cit* Todini, 2007). In the current experiment, concentration of T3 was not significantly different between before and after transportation (P>0.05). There are many possibilities for such a result: 1) even the animals were transported for 16 hours, they still had positive energy balanced so that their basal metabolism rate was still stable. Riis and Madsen, (1985); Todini (2007) reported that circulating thyroid can be considered as indicators of metabolic and nutritional status of the animals. 2) Period of 16 hours of transportation was not long enough to make severe stress. Thus, the body could compensate for such unsevere stress by regulating their normal metabolism status so that concentration of T3 was stable. Sarmin et al. . 2009 (*in press*) reported that in Bligon buck which was transported for 16 hours , level of cortisol increased 4 hours post-transportation then decreased until 16 hours due to animals adaptation. Even statistically there were no significant differences observed, cortisol became higher as soon as the subjects arrived at home**.** Ingram et al. (2002); Toscano and Friend, (2001) reported that orientation during transport had no effect on stress levels, but transport is stressful and orientation affects balance during transport (3) Different from previous studies, in this experiment, the small truck was opened and loaded with 6 animals only so physical stimuli such as crowding would be discarded. Heat stress, noise would also be discharded because the transportation was held from evening to the next morning. Todini *et al*. (2007) reported that many factors transportation animals may be exposed to a variety of physical stimuli including crowding and high stocking density (Cockram et al. 2004), noise, handling, isolation, agitation, metabolic and extreme temperature. It has been demonstrated that excessively high stocking density during transportation leads to increased injury and stress (Tarrant, 1990; Cockram et al., 1996). The initial period of transportation is the most stressful time for animals (Broom et al., 1996). Driver behavior and road quality are also factors affecting sheep (Cockram et al., 2004) and cattle (Fazio *et al*. 2005) during transportation.

Thyroxine

T4 can be deiodinated to form the biologically active hormone T3 by a 5'-deiodinase enzyme and to the inactive reverse T3 (rT3) (Leonard and Koehrle 2000; Hernandez and Germain, 2003).

The thyroxine level, which is major metabolic hormone, revealed any significant changes in treatment group before and after transportation ($P<0.05$) (Figure 1). Table 1 showed that increased of T_3 would be matched by decreased of T_4 levels. Four hours after the course of transportation, , concentration of T_4 was stable, whereas levels of T_3 decreased. In the same time, cortisol which can be considered as indicator of stress increased (Sarmin, 2009 *in press*). The finding of increased T_3 and decreased T_4 levels suggest that its may peripheral T_4 to T_3 conversion to form active hormone was decreased since cortisol was dominated this situation. Cortisol and T_3 are very important in increasing the metabolic process. Because of stress, metabolism and energy demand became higher, cortisol dominated the situation so that conversion of T_4 to T_3 was decreased. Christiansen *et al* (2007) illustrated the similar phenomenon in human, increased in T_3 and decreased in T_4 levels suggest that lack of cortisol may stimulate peripheral conversion of T_4 to T_3 to form active hormone.

CONCLUSION

Transportation of Bligon bucks for 16 hours affected level of the T_4 only and not for T_3 concentration due to physical stimuli such as crowding, heat stress, noise, would be discarded. During transportation, decreasing of T_4 levels indicated conversion of T_4 to T_3 to form active hormone. For accurate assessment of an animal reaction, combination of behavioral and physiological aspect must be measured for determination of discomfort experiencing by the animals.

ACKNOWLEDGMENTS

This work was granted by the Comp. Grant 2008 of the Faculty of Veterinary Medicine, University of Gadjah Mada, Yogyakarta. Special thanks go to our students for kindly helping in the field.

REFFERENCES

Al-Kindi, Kadim LT, Mahmoud LY, Mahgoub O, Plude J, Al-Maani, Bakheit CS. 2005. Physiological Response of Two Ages Groups of Omani male Goats to Short Road Transportation in Relation to Circulating Levels of Gonadotropins, Cortisol, Thyroid Hormones, Sex Steroids and Plasma Chemistry. *J Anim Vet Adv* 4(8): 737-741

Altiner A. 2006. Study of Serum of Growth Hormone, 3,5,3'- triiodothyronin, Thyroxine, Total Protein and Fatty Acids Levels During Parturition and Early Lactation in Ewes. *Bull Vet Inst Pulawy* 50, 85-87, 2006

Broom DM. 2003. Transport stress in cattle and sheep with details of physiological, ethological and other indicators. *German Vet J* 3: 83-90

Cockram MS. 2004. A review of behavioral and physiological responses of sheep to stressor to identify potential behavioral signs of distress. Anim. Welfare. 13:283-291

Cockram, MS, Baxter EM, Smith LA, Bell S, Howard CM, Prescott RJ, and. Mitchell MA. 2004. Effect of driver behaviour, driving events and road type on the stability and resting behaviour of sheep in transit. *Anim Sci* 79:165-176.

Chopra IJ, Sack J, Fisher DA 1975. 3,30,50-Triiodothyronine (Reverse T3) and 3,3,50-Triiodothyronine (T3) in fetal and adult sheep: studies on metabolic clearance rates, production rates, serum binding, and thyroidal content relative o thyroxine. *Endocrinology* 97, 1080–1088.

Christiansen JJ, Christian B, Djurhuus, Claus H. Gravholt, Per Iversen, Christiansen JS, Schmitz O, Jørgen W, Lunde JO, Jørgensen Møller N. 2007. Effects of Cortisol on Carbohydrate, Lipid, and Protein Metabolism: Studies of Acute Cortisol Withdrawal in Adrenocortical Failure. *J of Clin. Endocrinol Metabl* 92, No. 9 3553-3559

Fazio E, Medica P, Alberghina P, Cavaleri S, Ferlazzo. 2005. Effects of Long Distance Road Transport on Thyroid and Adrenal Function and Hematocrit Values in Limousin Cattle: Influence of Body Weight Decrease. *Vet Research Com* 29 (8): 713-719

Fisher DA, Chopra IJ, Dussault JH 1972. Extrathyroidal conversion ofthyroxine to triiodothyronine in sheep. *Endocrinology* 91, 1141–1144.

Garriga C, Hunter RR, Amat C,. Planas JM,. Mitchell MA, Moreto M.2006. Heat stress increases apical glucose transport in the chicken jejunum, *Am J Physiol Regul Integr Comp Physiol* **290** R195–R201.

Hangalapura BN, Nieuwland MG, Buyse J, Kemp B, Parmentier HK, 2004. Effect of duration of cold stress on plasma adrenal and thyroid hormone levels and immune responses in chicken lines divergently selected for antibody responses, *Poult Sci* **83** 1644–1649

Hernandez A, St Germain DL 2003 Thyroid hormone deiodinases: physiology and clinical disorders. *Curr Opin Pediatr* 15:416–420

Ingram JR, Cook CJ, Harris PJ. 2002. The effect of transport on core and peripheral body temperatures and heart rate of sheep. *Anim Welfare* 11:103-112

Leonard JL, Koehrle J. 2000 Intracellular pathways of iodothyronine metabolism. In: Braverman LE, Utiger RD, eds. The thyroid. Philadelphia: Lippincott Williams & Wilkins Pp.136–173

Naqvi SMK, Rai AK 1991. Influence of dietary energy-level on sheep for mutton during winter. 2. Effect on cardiorespiratory responses, rectal temperature, some blood metabolites, enzymes and thyroidal hormones. *Indian J Anim Sci* 61, 1126–1131.

Riis PM, Madsen A. 1985. Thyroxine Concentration and Secretion rates in relation to pregnancy, lactation and energy balance in goats. *J of Endocrinol*.: 107: 421-427

Tarrant PV. 1990. Transportation of cattle by road. *Appl Anim Behav Sci* 28: 153-170

Todini L. 2007. Thyroid hormones in small ruminants: effects of endogenous, environmental and nutritional factors. *Animal* : 1 (7), 997–1008.

Toscano MJ, Friend TH. 2001. A note on the effects of forward and rear-facing orientation on the movement of horses during transport. Appl. Anim. Behav. Sci. 73: 281-287

Williams CC, Calmes KJ, Fernandez JM, Stanley CC, Lovejoy JC, Bateman HG, Gentry LR, Gantt DT, Harding GD 2004. Glucose metabolism and insulin sensitivity in Gulf Coast native and Suffolk ewes during late gestation and early lactation. *Small Ruminant Res* 54, 167–171.