



## Effect of Extreme Seasons on Some Blood Biochemical Parameters in Beetal and Toggenberg Goats of Jammu Region

Sumeet Kour\*, Jonali Devi, Kawardeep Kour, Dibyendu Chakraborty, Aafreen Wahid Ganai, Rizwana Zargar and Pallavi Khajuria

*Division of Veterinary Physiology and Biochemistry, FVSc and AH, SKUAST-J, R.S. Pura, J&K, INDIA*

*\*Corresponding author: S Kour; Email: ssonya558@gmail.com*

**Received:** 23 Feb., 2017

**Revised:** 17 July, 2017

**Accepted:** 22 July, 2017

### ABSTRACT

The influence of extreme environmental conditions on biochemical parameters were studied in 36 healthy adult female (2-4 years of age) Beetal (n=18) and Toggenberg (n=18) goats in Jammu (J&K, India). Blood glucose, total cholesterol, total protein, albumin and blood urea nitrogen (BUN) levels were studied during moderate (autumn), extreme hot (summer) and extreme cold (winter) ambiances. Ambient temperature ( $^{\circ}\text{C}$ ) and relative humidity (%) were recorded and temperature-humidity index (THI) was calculated. For the season and breed interaction, significantly higher ( $P<0.01$ ) total protein and cholesterol levels were found in Toggenberg goats in all the seasons. Significantly higher ( $P<0.01$ ) levels of total protein, albumin and BUN levels were found during summer as compared to autumn and winter seasons. The concentrations of glucose and cholesterol were significantly higher ( $P<0.01$ ) in winter season. Among the two breeds, all the values were significantly higher ( $P<0.01$ ) in Toggenberg. Biochemical parameters change in different seasons and THI values were found higher during summer season and is considered as danger status.

**Keywords:** Biochemical parameters, goat, extreme season

Environmental temperature of the Jammu region ranges between  $2^{\circ}\text{C}$  in winter to  $48^{\circ}\text{C}$  at summer season and this wide range of environmental temperature may alter the physiological status of the animal which leads to stress condition. There is great variation in biochemical parameters as observed between breeds of goats and goats reside at different climatic conditions (Azab and Abdel-Maksoud 1999; Tambuwal *et al.*, 2002). Among the breeds of goats of Jammu and Kashmir, Beetal is one of most important one owing to its contribution to the state economy in terms of milk and meat production. Whereas, Toggenberg is an exotic breed and is amongst the best dairy goats found in this area. Despite their importance in the economy of the state, no well established data on the effect of various seasons on the basic physiological parameters were available. Again, it is known that different animal breeds show different degrees of heat tolerance because of varied genetic make-up, which is a function of long time interaction between gene and exposed environmental

condition. Therefore, the present investigation was undertaken to study the effect of extreme season (heat and cold) on metabolic profile of Beetal (local breed) and Toggenberg (exotic) breeds of goat.

### MATERIALS AND METHODS

Blood samples were collected from the goat flock maintained at Government Dairy Goat Farm, Jammu and the collected blood samples were analyzed in the division of Division of Veterinary Physiology & Biochemistry, Faculty of Veterinary Science and Animal Husbandry, SKUAST-J, Jammu.

### Animals and experimental design

A total of 36 healthy, female Beetal and Toggenberg goats of 2-4 years of age were selected for the study. The animals were divided into two groups, Beetal (n=18) and

Toggenberg (n=18) breeds of goat. Blood samples were collected from each animal in three seasons that is summer (June-July), autumn (October-November) and winter (January-February) at 10 days interval.

### Climate parameters

Ambient temperature (°C) was recorded two times a day (morning and evening) at 10 days interval using dry and wet bulb thermometer. The psychrometric tables were used to calculate relative humidity in percentage using dry and wet bulb recording. A temperature-humidity index (THI) is a single value representing the combined effects of air temperature and humidity. The mean THI was calculated as per U.S. Weather Bureau (Wiersma and Armstrong, 1989) using the following equation:

$$THI = 0.72 (\text{Dry bulb temp. } ^\circ\text{C} + \text{Wet bulb temp. } ^\circ\text{C}) + 40.6$$

Mean climatological values of ambient temperature (°C), relative humidity (%) and THI for the experimental periods are shown in Table 3. A THI of 74 or less is considered normal, 75-78 is alert status, 79-83 is danger status and a THI equal to or above 84 is an emergency (Hernandez *et al.*, 2010).

### Experimental procedure and analyses of different parameters

About 10 ml of blood was collected from each animal by venipuncture aseptically. 2 ml of blood was taken in sodium flouride vials for estimation of blood glucose. The rest of blood sample was allowed to clot and serum was separated and collected in storage vials. The samples were stored at -20°C and other biochemical parameters were analyzed. Blood glucose, total protein, albumin, blood urea nitrogen and cholesterol was analyzed as per GOD-POD end point colorimetric method (Basak, 2007), direct

Biuret method (Doumasa *et al.*, 1971), Bromocresol green method (Doumasa *et al.*, 1971), GLDH-Urease method (Kaltwassar and Schlegal, 1966) and CHOD-PAP method (Roeschlau *et al.*, 1974), respectively using analytical kits.

For all the observed data in the present experiment, the standard statistical procedures recommended by Snedecor and Cochran (2004) were followed. The data were presented by as mean and standard error and significant differences in mean values were statistically analyzed by Two Way ANOVA. Analysis of variance (ANOVA) was done with the help of Duncan's Multiple Range Test by Statistical Software Sigma Stat.

## RESULTS AND DISCUSSION

### Climatic parameters

The environmental temperature, relative humidity and THI levels on the experimental periods are presented in Table 1. From the data we observed that average temperature (°C) values during summer were higher than the critical temperature of 24-27°C for most species. In the study, THI was higher during summer and is considered as danger status.

### Blood Glucose

Table 2 represented the glucose level (mg/dl) between different seasons within Beetal and Toggenberg breeds. Significantly higher (P<0.01) glucose concentrations were recorded in winter as compared to summer and autumn when comparing the values between different seasons in both the breeds. When comparing the glucose level in between breeds within seasons (Table 2), significantly higher (P<0.01) concentration was found only in summer season in Toggenberg goats as compared to Beetal. The reason might be due to more feed intake by the exotic

**Table 1:** Mean ambient temperature (°C), relative humidity (%) and temperature-humidity index (THI) during different seasons

Period of study	Average temperature (°C)	Relative humidity (%)	THI
Summer	37.75	46.48	80.17
Autumn	21.21	60.58	67.49
Winter	11.43	79.50	53.16

**Table 2:** Biochemical parameters (Mean  $\pm$  S.E.) of Beetal and Toggenberg goats in different seasons

Parameters	Beetal			Toggenberg			Seasons		
	Summer (n=36)	Autumn (n=36)	Winter (n=36)	Summer (n=36)	Autumn (n=36)	Winter (n=36)	Summer (n=72)	Autumn (n=72)	Winter (n=72)
Glucose (mg/dl)	55.28 <sup>ap</sup> $\pm 0.45$	56.22 <sup>a</sup> $\pm 0.45$	60.56 <sup>b</sup> $\pm 0.45$	56.50 <sup>aq</sup> $\pm 0.45$	56.86 <sup>a</sup> $\pm 0.45$	60.97 <sup>b</sup> $\pm 0.45$	55.89 <sup>x</sup> $\pm 0.32$	56.54 <sup>x</sup> $\pm 0.32$	60.76 <sup>y</sup> $\pm 0.32$
Cholesterol (mg/dl)	81.69 <sup>ap</sup> $\pm 0.41$	83.11 <sup>bp</sup> $\pm 0.41$	86.19 <sup>cp</sup> $\pm 0.41$	83.64 <sup>aq</sup> $\pm 0.41$	84.64 <sup>aq</sup> $\pm 0.41$	87.83 <sup>bq</sup> $\pm 0.41$	82.67 <sup>x</sup> $\pm 0.29$	83.88 <sup>y</sup> $\pm 0.29$	87.01 <sup>z</sup> $\pm 0.29$
Total Protein (g/dl)	6.64 <sup>bp</sup> $\pm 0.04$	6.56 <sup>bp</sup> $\pm 0.04$	6.27 <sup>ap</sup> $\pm 0.04$	7.01 <sup>cq</sup> $\pm 0.04$	6.86 <sup>bq</sup> $\pm 0.04$	6.50 <sup>aq</sup> $\pm 0.04$	6.82 <sup>z</sup> $\pm 0.03$	6.71 <sup>y</sup> $\pm 0.03$	6.39 <sup>x</sup> $\pm 0.03$
Albumin (g/dl)	2.70 <sup>b</sup> $\pm 0.15$	2.40 <sup>b</sup> $\pm 0.15$	1.73 <sup>a</sup> $\pm 0.15$	3.03 <sup>b</sup> $\pm 0.18$	2.50 <sup>b</sup> $\pm 0.15$	1.83 <sup>a</sup> $\pm 0.16$	2.87 <sup>z</sup> $\pm 0.12$	2.54 <sup>y</sup> $\pm 0.11$	1.78 <sup>x</sup> $\pm 0.11$
BUN (mg/dl)	22.50 <sup>b</sup> $\pm 1.759$	19.50 <sup>b</sup> $\pm 1.759$	14.00 <sup>a</sup> $\pm 1.759$	27.25 <sup>b</sup> $\pm 1.759$	22.50 <sup>b</sup> $\pm 1.759$	15.50 <sup>a</sup> $\pm 1.759$	24.88 <sup>z</sup> $\pm 1.24$	21.00 <sup>y</sup> $\pm 1.24$	14.75 <sup>x</sup> $\pm 1.24$

Figures with parenthesis indicates number of observations

\*a, b, c describe significant differences between seasons within breed

\* p,q describe significant differences between breeds within season

\*x, y, z describe significant differences between seasons

breeds (Gupta *et al.*, 1992; Singh and Singh and Upadhaya, 1997) as compared to the local breed.

When considering variations between three different seasons irrespective of breed (Table 2), glucose concentration was found significantly higher ( $P < 0.01$ ) in winter as compared to other two seasons. Cold stress induces gluconeogenesis through glucocorticoid secretion with maintenance of blood glucose in stressed animals (Weber *et al.*, 1965). During heat stress feed consumption decreases, which comparatively lowers the blood glucose level (Kataria *et al.*, 2002) or it might be due to increased glucose oxidation (Collier *et al.*, 2008) during summer stress. Decreased gluconeogenesis and glycogenolysis were observed in cows during heat stress (Itoh *et al.*, 1998). The finding was similar to the observations of Kataria *et al.* (1993) in Marwari goats and Rasooli *et al.* (2004) in heifers. On the other hand, Sejian and Srivastava (2009) reported that glucose level increased significantly ( $P < 0.05$ ) after thermal exposure. Shrikhande *et al.* (2008) also reported that in summer season the average blood glucose level was higher whereas, lowest level was recorded during rainy season in cattle.

### Cholesterol

Table 2 represented the cholesterol level (mg/dl) between different seasons within Beetal and Toggenberg breeds. In Beetal, significantly higher ( $P < 0.01$ ) values were recorded in winter followed by autumn than that of summer season. In Toggenberg breed, significantly higher ( $P < 0.01$ ) values were found in winter season than those of the other two. When comparing the cholesterol level in between Beetal and Toggenberg within different seasons (Table 2), significantly higher ( $P < 0.01$ ) concentration was found in Toggenberg goat in all seasons.

When considering variations between three different seasons irrespective of breed (Table 2), cholesterol level was found significantly higher ( $P < 0.01$ ) in winter followed by autumn season. Decrease cholesterol concentration with high ambient temperature might be due to the decrease in acetate concentration, which is the primary precursor for the synthesis of cholesterol (Darcan, 2000; O'cak *et al.*, 2009). Plasma cholesterol concentrations would also be reduced during stress (Peinado *et al.*, 1993). Marai *et al.* (1995) and Habeeb *et al.* (1996) reported that total cholesterol decreased significantly with prolonged

exposure to high environmental temperature. Such phenomenon may be due to the increase in utilization of fatty acids for energy production as a consequence of the decrease in glucose level.

### Total Protein

Table 2 represented the protein level (g/dl) between different seasons within two breeds of goat. Significant variation at 1% level was observed in between all the seasons in Toggenberg goats, significantly lower values found in winter followed by autumn and summer; whereas in Beetal, the values were significantly lower in winter as compared to other two seasons. In Beetal goats, no significant difference was observed in between autumn and summer seasons. When comparing the protein level in between Beetal and Toggenberg within seasons (Table 2), significantly higher ( $P<0.01$ ) total protein concentrations were found in Toggenberg than those of Beetal goats in all seasons.

When considering variations between three different seasons irrespective of breed (Table 2), total protein concentration in winter was significantly lower ( $P<0.01$ ) than those of other two seasons. El-Nouty *et al.* (1989) found that the rise in ambient temperature during summer was associated with a significant increase in albumin and the slight rise in globulin concentrations in goat. El-Nouty *et al.* (1980) reported that plasma total protein content decreased by 11.9% in buffaloes and increased by 4.4% in Baladi cattle when exposed to direct solar radiations. Similar observation was also recorded in other species (Rasooli *et al.* 2004; Shrikhande *et al.* 2008). In the other studies (Hooda and Naqvi, 1990; Abdelatif *et al.*, 2009), it was found that season had no significant effect on the concentration of total protein in goat and sheep.

Between three different seasons, irrespective of breeds (Table 3), significantly higher ( $P<0.01$ ) total protein concentration was recorded in summer than autumn followed by winter season.

### Albumin

Table 2 represented the albumin level (g/dl) between different seasons within two breeds of goat. Significantly lower ( $P<0.01$ ) value found in winter as compared to other two seasons in both the breeds. When comparing the

albumin level in between Beetal and Toggenberg within seasons (Table 2), no significant variation in albumin concentrations were observed between Beetal and Toggenberg goats.

Considering variations between three different seasons irrespective of breed (Table 2), albumin level was found significantly higher ( $P<0.01$ ) in summer followed by autumn when compared to winter season. Higher albumin level in summer might be due to the loss of extracellular fluid due to heat exposure. Rasooli *et al.* (2004) also reported that the increase in plasma albumin concentrations in summer compared to winter in non-pregnant Holstein heifers. Higher concentration of albumin during summer season was also recorded by Ganaie *et al.* (2013) in cows and buffalo calves, in which they described that albumin is the major extracellular source of thiols, which are scavengers of free radicals allowing albumin to function as an antioxidant (Rasooli *et al.*, 2004). Similar finding was also reported by Koubkova *et al.* (2002) in bovines. On the contrary, other workers reported heat stress induced reduction in plasma albumin concentrations in other breeds of goat (Al-Eissa *et al.*, 2012) and sheep (Abdoun *et al.*, 2012). Shrikhande *et al.* (2008) reported higher serum albumin level during winter and summer as compared to rainy season in cattle.

### Blood Urea Nitrogen (BUN)

Table 2 represented the blood urea nitrogen level (mg/dl) between different seasons within two breeds of goat. Significantly lower ( $P<0.01$ ) value found in winter as compared to other two seasons in both the breeds. When comparing the BUN level in between Beetal and Toggenberg within seasons (Table 2), no significant variation in BUN concentration was observed between Beetal and Toggenberg goats.

Considering variations between three different seasons irrespective of breed (Table 2), BUN level was found significantly higher ( $P<0.01$ ) in summer followed by autumn when compared to winter season. The high concentration of blood urea in summer months probably as a result of loss of extra-cellular fluid due to heat exposure (Rasooli *et al.* 2004). Again, increased level of BUN during heat exposure might be due to increased utilization of amino acids through mobilization of protein from muscle (Sreedhar *et al.*, 2013). This finding corroborate

the studies in dairy cows being carried out by different workers (Rowlands *et al.*, 1979). Shrikhande *et al.* (2008) found that BUN level did not differ significantly during summer and winter, but was significantly low during rainy season.

## CONCLUSION

The influence of environmental stress (heat and cold stress) on biochemical parameters were studied in Beetal and Toggenberg goats in Jammu region in different seasons. The biochemical parameters were changed with environmental stress. THI was higher during summer and was considered as danger status. Among the two breeds, all the values were found to be significantly higher ( $P < 0.01$ ) in Toggenberg than those of Beetal goats. Being the original habitats, the local breeds of animals are highly adapted to the climatic condition and can withstand the extreme hot and cold climate of Jammu region.

## ACKNOWLEDGEMENTS

The authors acknowledge the support and facilities provided by the Government Dairy Goat Farm, Rajbagh, Kathua, Jammu (Jammu and Kashmir).

## REFERENCES

- Abdelatif, A.M., Ibrahim, M.Y. and Hassan, Y.Y. 2009. Seasonal variation in erythrocytic and leukocytic indices and serum proteins of female Nubian goats. *Middle-East J. Sci. Res.*, **4**(3): 168-174.
- Abdoun, K.A., Samara, E.M., Okab, A.B. and Al-Haidry, A.I. 2012. A comparative study on seasonal variation in body temperature and composition of camels and sheep. *J. Anim. Vet. Adv.*, **11**(6): 769-773.
- Al-Eissa, M.S., Alkahtani, S., Al-Farraj, S.A., Saud, A.A., Al-Dahmash, B. and Al-Yahya, H. 2012. Seasonal variation effects on the composition of blood in Nubian ibex (*Capra nubiana*) in Saudi Arabia. *Afr. J. Biotechnol.*, **11**(5): 1283-1286.
- Azab, M.E. and Abdel-Maksoud, H.A. 1999. Changes in some haematological and biochemical parameters during pre partum and post partum periods in female Baladi goats. *Small Rumin. Res.*, **34**: 77-85.
- Basak, A. 2007. Development of a inexpensive plasma glucose estimation by two-point kinetic method based on glucose oxidase-peroxidase enzymes. *Indian J. Clin. Biochem.*, **22**:156-160.
- Chand, D. and Georgie, G.C. 1989. Influence of season and genetic group on the blood plasma cholesterol in neonate calves. *Indian J. Anim. Sci.*, **59**: 149-153.
- Christi, W.W. 1981. Lipid metabolism in ruminant animals. *Pergamon press New York.*, 20-150.
- Collier, R.J., Collier, J.L., Rhoads, R.P., and Baumgard, L.H. 2008. Genes involved in the bovine heat stress response a invited review. *J. Dairy Sci.*, **91**: 445-454.
- Darcan, N. 2000. A study on adaptation mechanism of crossbred goat types in Cukurova sub-tropical climate conditions. M.Sc. Thesis, Cukurova University Adana, Turkey.
- Doumasa, B.T, Arendis, R.L. and Pinto, P.C. 1971. *Standard Method of Clinical Chemistry.* Academic Press, Chicago. pp. 175-189.
- Ganaie, A.H., Shanker, G., Bumla, N.A., Ghasura, R.S., Mir, N.A., Wani, S.A. and Dudhatra, G.B. 2013. Biochemical and physiological changes during thermal stress in bovines. *J. Veterinar. Sci. Technol.*, **4**: 126.
- Gupta, A., Rao, M.V.N. and Upadhyay, R.C. 1992. Effect of different speeds and ingredients up on blood constituents in crossbred males. *Indian J. Dairy Sci.*, **52**: 121-124.
- Habeeb, A.A., El-Masry, K.A., Aboul-Naga, A.I. and Kamal, T.H. 1996. The effect of hot summer and level of milk yield on blood biochemistry and circulating thyroid and progesterone hormones in Friesian cows. *Arab J. Nucl. Sci. Appl.*, **29**: 161-173.
- Hernandez, A., Dominguez, B., Cervantes, P., Munoz-Melgarejo, S., Salazar-Lizan, S. and Tejeda- Martinez, A. 2011. Temperature-humidity index (THI) 1917-2008 and future scenarios of livestock comfort in Veracruz, Mexico. *Atmosfera.*, **24**(1): 89-102.
- Hooda, O.K. and Naqvi, S.M.K. 1990. Changes in some blood constituents in different breeds of sheep exposed to elevated temperature and feed restriction. *Indian Vet. J.*, **62**: 1121-1125.
- Itoh, F., Obara, Y., Rose, M.T. and Fuse, H. 1998. Heat influences on plasma insulin and glucagon in response to secretagogues in non-lactating dairy cows. *Domest. Anim. Endocrine.*, **15**: 499-510.
- Kaltwassar, H. and Schlegal, H.G. 1966. NADH- dependent coupled enzyme assay for urease and other ammonia producing systems. *Anal. Biochem.*, **16**: 132.
- Kataria, A.K., Kataria, N., Bhatia, J.S. and Ghosal, A.K. 1993. Blood metabolic profile of Marwari goats in relation to seasons. *Ind. Vet. J.*, **70**: 761-762.
- Kataria, N., Kataria, A.K., Agarwal, V.K., Garg, S.L. and Sahani, M.S. 2002. Effect of long term dehydration on serum constituents in extreme climatic conditions in camel. *Indian J. Physiol. Pharmacol.*, **46**: 218-222.

- Koubkova, M., Knizkova, I., Kunc, P., Hartlova, H., Flusser, J. and Dolezal, O. 2002. Influence of high environmental temperatures and evaporative cooling on some physiological, haematological and biochemical parameters in high yielding dairy cows. *Czech J. Anim. Sci.*, **47**: 309-318.
- Kweon, O.K., Ono, H., Osasa, K., Onda, M., Oboshi, K., Uchisugi, H., Kurosawa, S., Yamashina, H. and Kanagawa, H. 1986. Factors affecting serum total cholesterol level of lactating Holstein cows. *Jpn. J. Vet. Sci.*, **48**: 481-486.
- Marai, I.F.M., Habeeb, A.A.M., Daader, A.H. and Yousef, H.M. 1995. Effects of Egyptian sub-tropical conditions and the heat stress alleviation techniques of water spray and diaphoretics on the growth and physiological functions of Friesian calves. *J. Arid Environ.*, **30**: 219-225.
- Nath, R. 2006. Seasonal variation of blood biochemical parameters in crossbred calves. *Indian Vet. J.*, **83**: 800 – 801.
- O'cak, S., Darcan, N., Cankaya, S. and Inal, T.C. 2009. Physiological and biochemical responses in German Fawn kids subjected to cooling treatments under Mediterranean climate conditions. *Turk. J. Vet. Anim. Sci.*, **33**(6): 455-461.
- Peinado, V.I., Fernandez-arias, A., Viscor, G. and Palo-Meque, J. 1993. Haematology of Spanish ibex (*Capra pyrenaica hispanica*) restrained by physical or chemical means. *Vet. Rec.* **132**: 580-583.
- Prakash, P. and Rathore, V.S. 1991. Seasonal variation in blood serum profiles of Triiodothyronins and thyroxine in goat. *Indian J. Anim. Sci.*, **61**: 1311-1312.
- Rasooli, A., Nouri, M., Khadjeh, G.H., Rasekh, A. 2004. The influences of seasonal variation on thyroid activity and some biochemical parameters of cattle. *Iranian J. Vet. Res.*, **5**: 1383-2004.
- Roeschlau, P., Bernt, E. and Gruber, W.A. 1974. Enzymatic determination of total cholesterol in serum. *Clin. Biochem.*, **12**: 226-228.
- Rowlands, G.J., Little, W., Stark, A.J. and Manston, R. 1979. The blood composition of dairy herds and its relationship with season and lactation. *Brit. Vet. J.*, **135**: 64-74.
- Sejian, V. and Srivastava, R.S. 2009. Effects of melatonin on adrenal cortical functions of Indian Goats under thermal stress. *Vet. Med. Int.*, **2010**: 1-6.
- Shrikhande, G.B., Rode, A.M., Pradhan, M.S. and Satpute, A.K. 2008. Seasonal effect on the composition of blood in cattle. *Vet. World.*, **1**: 341-342.
- Singh, S.V. and Upadhyay, R.C. 1997. Physiochemical responses of the buffaloes during cart loads under hot season. *Indian J. Dairy Sci.*, **50**: 341-342.
- Snedecor, G.W. and Cochran, W.G. 2004. Statistical methods. 8<sup>th</sup> edn., Oxford and IBH Pub. Co., Kolkata.
- Sreedhar, S., Rao, K.S., Suresh, J., Moorthy, P.R.S. and Reddy, V.P. 2013. Changes in haematocrit and in some serum biochemical profile of Sahiwal and Jersey X Sahiwal cows in tropical environments. *Veterinarskiarhiv.*, **83**: 171-187.
- Tambuwal, F.M., Agale, B.M. and Bangana, A. 2002. Haematological and biochemical values of apparently healthy Red Sokoto goats. *Indian J. Anim. Sci.*, **57**: 1200-1204.
- Weber, G., Singhal, R.L. and Srivastava, S.K. 1965. Effect of nutrition on hormonal regulation of liver enzymes. *Can. J. Biochem. Cell b.*, **43**: 1549-1553.
- Wiersma, F. and Armstrong, D.V. 1989. Microclimatic modification to improve milk production in hot arid climate. *Dairy Sci. Abstract.*, **52**(6): 3848.