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Growth rate, feed intake, physiological responses and hormonal profile of Murrah buffaloes implanted melatonin during summer season

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ABSTRACT

In order to find out the effect of melatonin implantation on physiological responses, hormonal profile, feed intake and growth performance, 12 growing Murrah buffaloes were selected. These animals were further divided equally in two groups i.e. control and treatment (melatonin implantation @ 18 mg/50 kg body wt.). Melatonin was implanted subcutaneously in the thoracic region after every 45 days' interval in treatment group. Body weight and physiological parameters, viz. respiration rate (RR), rectal temperature (RT), pulse rate (PR) were recorded at fortnightly interval. Blood samples were also collected aseptically from both the group of animals at fortnightly intervals and plasma was separated for quantitation of hormones. The physiological responses were found to be significantly lower in treatment group than control group of growing buffaloes. The levels of growth and cortisol hormones were higher in treatment than control group. No significant variation was observed in the IGF-I levels. The melatonin implantation improved the growth rate, ADG and feed intake of growing buffaloes than control group. Based on the results of the present study, it is evident that melatonin implantation to growing buffaloes worked as an antioxidant and lowered the stress levels and enhanced growth rate during heat stress. Higher growth rate in treatment group of growing buffaloes will help in reducing the age of puberty and ultimately increase the total productive life.

Keywords: Buffalo, Growth rate, Hormonal profile, Melatonin, Physiological responses

The buffaloes are mainly reared for its milk, meat and drought purpose. It is an integral part of the Indian agricultural system and is known as Black Gold of India. Heat stress increases respiration rate and core body temperature, while reducing feed intake, milk yield and growth performance (West 2003). Exposure to extreme temperature elicits a series of changes in the animal's biological functions that include reduction in feed intake, efficiency and utilization, disturbances in metabolism of water, protein, energy and mineral balances, hormonal secretions, enzymatic reactions and blood metabolites. These changes result in impairment of growth, production and reproduction performances (Tao *et al*. 2011). Therefore, strategies should be taken to reduce the heat stress on dairy animals during summer.

Melatonin hormone is a chemical substance found in animals, plants, fungi and bacteria and it anticipates the daily onset of darkness (Hardeland 2005). Melatonin effectively protects against heat stress by a variety of mechanisms. In tropical continents, animals are exposed to heat stress all over the year; melatonin with its potential favourable effects can be useful as an anti-heat stress promoter to prevent the loss of production by dominating cardiovascular system and evaporative heat loss (Hyder *et al*. 2017). Melatonin has been shown to decrease thyroxine and cortisol levels under short term heat stress exposure.

It was revealed that melatonin implant significantly reduced oxidative stress, as it is a potent hormonal antioxidant and its metabolites showed powerful free radicals scavenging characteristics. Melatonin accrues higher concentration in mitochondria of the cells and manages the mitochondrial homeostasis. Due to higher production of free radicals, mitochondria wall is very susceptible to free radical's attack. Melatonin reduces oxidative stress (Sharma *et al*. 2013). Melatonin interacts with numerous cellular proteins such as signaling molecules, transporters, channels and enzymes (Liu *et al.* 2019, Hemati *et al.* 2020). Only scanty information is available regarding melatonin implantation and its effect on growth, feed intake, physiological responses and hormonal profile of Murrah buffaloes under tropical regions. In order to fulfill this gap, present research work was planned and carried out on buffalo heifers during summer season.

MATERIALS AND METHODS

Twelve healthy growing Murrah buffaloes (8 to 12 months) were selected from the Livestock Research Centre

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Table 1a. Physical and chemical composition of the basal diet fed to Murrah buffaloes

Ingredient	Parts (%)
Maize	28
Bajra	5
Ground nut cake	10
Soybean meal	15
Mustard oil cake	13
Wheat Bran	15
Rice Polish	11
Mineral Mixture	2
Salt	1

Table 1b. Proximate analysis of different fodders (nutritive value) of the basal diet

Nutrient*	Compound feed	Maize green	Jowar green
DM	89.6	24.5	27.2
OM	91.2	90.4	93.5
CP	22.1	9.8	8.6
EE	4.3	2.3	1.6
NDF	24.3	63.1	61.3
ADF	14.2	30.4	32.4

*Nutrients in percentage.

of ICAR-National Dairy Research Institute, Karnal. These animals were divided into control and treatment (melatonin implantation @ 18 mg/50 kg body wt.) groups on the basis of body weight and body condition score (BCS). Blood samples were collected aseptically at fortnightly intervals and plasma was processed for quantitation of hormones. The rations consisted of dry fodder, green fodder and concentrate mixture. Fresh tap water was available for drinking round the clock to the animals. Both the groups of the animals were housed in a custom design animal shed throughout the study and dry matter content was estimated at fortnightly intervals throughout the experimental period and are given in Table 1a and 1b.

Micro climatic data, viz. dry bulb temperature (C_{db}) and wet bulb temperature (C_{wb}) were recorded at 7:30 AM and 2:30 PM with dry and wet bulb hygrometer (Zeal, UK) every day during experimental period. The temperature humidity index (THI) was calculated as method described by Mc Dowell 1972. The physiological parameters (rectal temperature, respiration rate and pulse rate) were recorded using standard procedure and blood samples were collected at fortnightly interval. Dry matter intake of individual animals was monitored by recording the feed offered and feed left in the manger. Body weight of growing buffaloes was recorded prior to actual experiment and subsequently recorded for 3 days using electronic weighing machine.

IGF-I, GH and cortisol was estimated in plasma samples using ELISA kit supplied by Bioassay Technology.

Experiment was conducted under the IAEC, constituted as per CPCSEA rules laid down by the Government of India (Reg. No. 1705/GO/ac/CPCSEA Dt. 3/7/2013). Statistical analysis of the obtained data was performed using software

Table 2. Fortnightly temperature humidity index (THI) during the experimental period

Fortnight	THI_{max}	THI_{min}	THI_{avg}
1	82.40±0.59	71.20±0.46	76.82±1.11
2	84.21±0.57	75.60±0.53	79.91±0.89
3	85.10±0.80	76.70±0.80	80.88±0.93
4	81.97±0.73	78.85±0.40	80.41±0.50
5	82.90±0.60	78.70±0.42	80.80±0.51
6	86.45±0.54	79.37±0.47	82.91±0.74
7	86.19±0.47	80.75±0.55	83.47±0.62
8	84.23±0.89	80.57±0.47	82.40±0.60
9	82.80±0.70	79.00±0.30	80.92±0.51
10	83.27±0.18	76.60±0.25	79.94±0.64

version (9.1) SAS Institute Inc., Cary, NC, USA Copyright© (2011). Two-way factor analysis of variance followed by post-hoc Tukey's studentised test. Evaluation of the correlation between all the factors in the respective set of animals was made using a correlation coefficient at the level of probability ($P<0.05$).

RESULTS AND DISCUSSION

Mean ± SE of THI during the experimental period have been depicted in Table 2. The 6^{th}, 7^{th} and 8^{th} fortnights showed most stressful period. A significant positive association of THI with RR and RT is in accordance with those of Kumar and Singh (2018) reported in Karan Fries and Sahiwal heifers. The impact of heat stress became more severe when relative humidity and sun exposure increased. There is positive correlation of THI with cortisol (Chaudhary *et al.* 2015). During heat stress, animals tries to reduce their metabolic heat production by reducing feed intake, which results in decreased growth rate and production. The average values of RT, RR and PR of growing Murrah buffaloes for control and treatment group have been presented in Table 3. Mean values of RT for control was significantly higher ($P<0.05$) than the treatment group. However, the highest values of RT of buffaloes were observed during the 5^{th} fortnight in control and treatment group. RT showed positive correlation with RR and PR and negative correlation with body weight, ADG and DMI. These results are in agreement with those of Indu *et al.* (2016) who reported significant increase in RT of Tharparkar and Karan Fries calves exposed to 44°C. RT increases only when physiological mechanisms of body are not able to counteract the unnecessary heat load (Siddiqui *et al.* 2011). During the present study, melatonin implantation reduced the RT significantly, compared to control group. Melatonin is linked with the control of body temperature and functionally related to metabolism and breathing (Dawson and van den Heuvel 1998). Melatonin administration lowers core body temperature by 2–3°C in mice (Charles *et al.* 1978). Reduction in core body temperature with melatonin administration has also been found in chicks (Barchas *et al.* 1967) and several mammalian species including humans (Strassman *et al.* 1991). The mean values for RR of treatment group was

Table 3. Effect of melatonin implantation on physiological responses of growing Murrah buffaloes during summer season

Fortnight	Rectal temperature (°C)		Respiration rate (breaths/min)		Pulse rate (beats/min)	
	Control	Treatment	Control	Treatment	Control	Treatment
1	37.60^{AX}±0.03	37.53^{AY}±0.02	33.17^{AX}±1.56	31.17^{AY}±1.42	66.00^{AY}±2.53	68.00^{AX}±1.15
2	38.45^{BX}±0.15	38.40^{BY}±0.03	33.17^{A}±0.60	32.50^{AB}±0.67	68.50^{AB}±1.82	67.33^{A}±2.11
3	38.58^{CX}±0.02	38.47^{BY}±0.03	36.00^{BCX}±0.86	34.83^{CY}±1.01	70.33^{BX}±1.74	68.67^{AY}±2.29
4	38.73^{DX}±0.04	38.62^{CY}±0.03	36.33^{BCX}±1.15	33.00^{ABCY}±0.37	74.33^{CX}±1.82	72.67^{BCDY}±1.34
5	38.78^{DX}±0.03	38.70^{CY}±0.03	34.33^{AB}±0.99	34.17^{BC}±0.65	73.67^{CX}±1.41	72.00^{BCY}±2.25
6	39.48^{EX}±0.16	38.70^{CY}±0.03	36.83^{CX}±1.42	32.67^{ABY}±0.67	74.67^{CDX}±1.33	72.67^{BCDY}±1.34
7	38.43^{BX}±0.02	38.33^{BY}±0.02	37.50^{CX}±1.50	34.33^{BCY}±0.76	77.67^{DX}±0.95	74.33^{CDEY}±0.17
8	39.72^{FX}±0.03	38.67^{CY}±0.03	40.33^{DX}±1.41	33.33^{BCY}±1.26	81.00^{EX}±1.84	76.67^{EY}±1.34
9	38.23^{GX}±0.03	38.10^{DY}±0.04	35.83^{BX}±1.35	32.83^{ABCY}±0.95	80.67^{DEX}±0.67	75.33^{DEY}±1.69
10	38.18^{GX}±0.02	37.92^{EY}±0.05	36.33^{BCX}±1.12	33.83^{BCY}±1.01	77.84^{DEX}±1.33	70.67^{BY}±1.76
Mean± S.E.	38.62^{X}±0.08	38.34^{Y}±0.05	35.98^{X}±0.44	33.27^{Y}±0.30	74.47^{X}±0.78	71.83^{Y}±0.63

Values with different superscripts A–G within a column and X, Y within a row differ significantly (P<0.05); Values of mean ± SE are observations on six animals.

Table 4. Effect of melatonin implantation on body weight, average daily gain and dry matter intake of growing Murrah buffaloes during summer season

Fortnight	Body weight (kg)		Average daily gain (g)		Dry matter intake (kg/100 kg bwt)	
	Control	Treatment	Control	Treatment	Control	Treatment
I	112.80^{A}±9.32	113.17^{A}±6.86	–	–	2.63^{A}±0.24	2.61^{A}±0.15
II	120.03^{AB}±9.06	123.10^{AB}±6.59	520.00^{AX}±27.00	506.67^{ABCY}±17.04	2.80^{AY}±0.22	3.02^{BX}±0.17
III	127.02^{ABC}±8.90	131.98^{BC}±6.47	482.23^{BY}±21.34	525.56^{AX}±10.53	3.00^{AY}±0.23	3.23^{BX}±0.17
IV	133.70^{BCD}±8.81	139.88^{C}±6.62	465.56^{BCY}±16.00	515.56^{ACX}±9.22	3.17^{ABY}±0.23	3.40^{BCX}±0.18
V	140.18^{CDY}±8.77	148.17^{CDX}±6.36	445.56^{CDEY}±11.85	507.78^{ABCX}±8.33	3.22^{BY}±0.20	3.50^{CDX}±0.17
VI	146.52^{DEFY}±8.76	157.07^{DEX}±6.88	432.23^{DEY}±10.53	537.78^{BX}±12.49	3.37^{BY}±0.20	3.68^{DX}±0.17
VII	152.77^{EFGY}±8.74	164.74^{EFX}±6.79	422.23^{DEY}±4.77	522.23^{AX}±11.12	3.51^{BCY}±0.20	3.87^{DX}±0.17
VIII	158.95^{FGHY}±8.73	172.22^{FGX}±6.55	408.89^{EY}±2.23	520.00^{AX}±14.50	3.30^{BY}±0.20	3.60^{DX}±0.18
IX	165.08^{GHY}±8.75	180.60^{GX}±6.90	412.22^{EFY}±4.01	492.22^{CX}±14.34	3.80^{CDY}±0.20	4.20^{EFX}±0.18
X	172.34^{HY}±8.61	188.74^{GX}±7.51	466.67^{BCY}±11.55	556.67^{DX}±10.00	3.96^{DY}±0.20	4.39^{FX}±0.18
Mean± S.E.	142.94^{Y}±3.54	151.97^{X}±3.67	450.62^{Y}±6.48	520.49^{X}±4.48	3.28^{Y}±0.08	3.55^{X}±0.08

Values with different superscripts A–H within a column and X, Y within a row differ significantly (P≤0.05); Values of mean ± SE are observations on six animals.

lower (P<0.05) than the control group of growing buffaloes. The highest value for control and treatment group was observed on 8th and 3rd fortnight, respectively. RR showed positive correlation with RT, PR, body weight, and negative correlation with DMI. RR is the most susceptible index to the environmental conditions than any other physiological responses and is the first indicator to animal's stress (Omran *et al*. 2011). An increase was observed in RR with the increase in temperature which may be due to more demand of oxygen by the tissues (Haque *et al*. 2012). The results are in agreement for higher values of RR in control groups of heifer compared to antioxidant supplemented group (Kumar and Singh 2018). Physiological responses showed significant positive correlation among them and negative correlation with DMI, growth rate and ADG (Table 5). The mean values of PR showed significant (P<0.05) difference during fortnights and groups. Positive correlation was observed among PR and RT, RR and negative correlation with B wt and DMI. When exposed to hot ambient temperature, dairy cows were found to show increased HR, respiratory frequency and RT maintain homeotherm (Avendano-Reyes *et al*. 2011). The results are in agreement as reported 70.2, 68.8 and 69.3 HR (beats/min) of Surti buffaloes during hot dry, hot humid and comfort zone respectively (Chaudhary *et al*. 2015). To maintain normal body temperature and prevent hyperthermia, the physiological adjustments are crucial, which leads to increased RT, RR and PR in dairy heifers during summer season (Kumar and Singh 2018).

The mean values of IGF-I for control animals were lower than the treatment group (Fig. 1A). IGF-I showed positive correlation with GH and cortisol. No significant variation between fortnights, groups and within the groups were found during the present study which might be because the growing animals were only one-year-old and were under heat stress, therefore the concentration of IGF-I remained unaffected between the groups. During stress, there is uncoupling of the somatotropic axis and hence level of

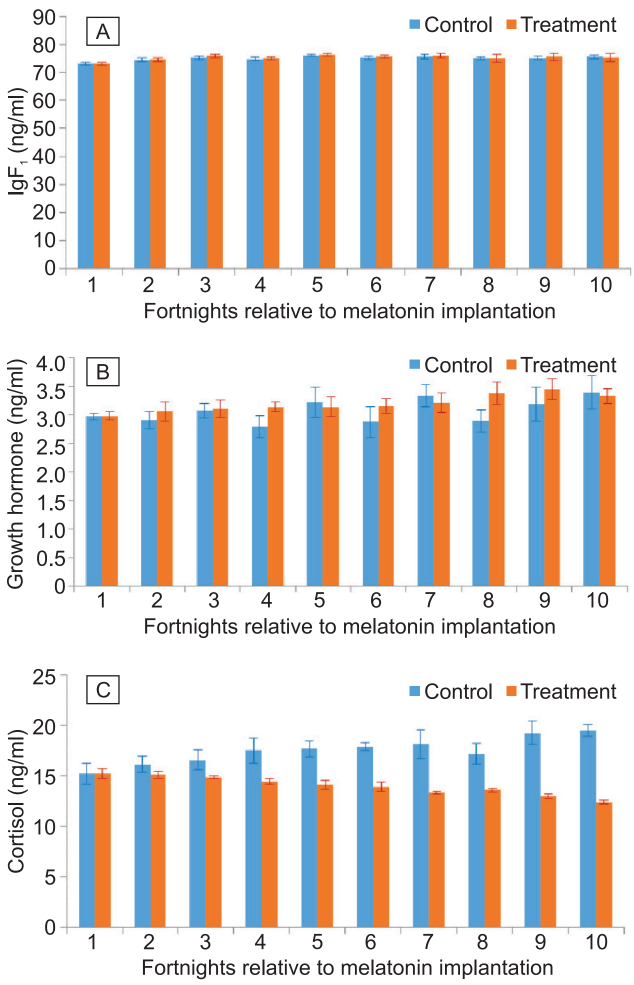

Fig.1. Effect of melatonin implantation on IGF1, growth and cortisol hormone levels of growing Murrah buffaloes during summer season.

IGF-I reduced. Similar trend of higher mean values of IGF-I was also observed in astaxanthin (antioxidant) supplemented group of Sahiwal and Karan Fries heifers during summer (Kumar and Singh 2018). The mean values of GH were lower in control than the treatment group of buffaloes (Fig. 1B). GH was positively correlated with IGF-1 while negatively correlated with cortisol. During hot humid season, the values of GH in control was significant (P<0.05) lower than the treatment group. GH improved the cognitive capacity and melatonin should be attributed to its antioxidant, anxiolytic and neuro-protective properties (Barcelo *et al.* 2016). During chronic heat stress GH concentration decreased, whereas, depicted static concentration of GH during acute heat stress, due to GH act as a calorigenic hormone (Mitra *et al.* 1972). Administration of melatonin at the beginning of dark period restores the melatonin secretion in the pineal gland and the serum levels of GH and IGF-I in a rat model (Rudnitskaya *et al.* 2015). Cortisol is used as an indicator of stress and the results obtained during the present study have been presented in Fig. 1C. Average values of cortisol hormone were higher (P<0.05) in control than the treatment group. Analysis of variance showed significant (P<0.05) difference between fortnights and groups. Cortisol showed negative correlation with GH. Acute stressor activates the hypothalamus-pituitary-adrenal axis and results in rise of cortisol concentration in stressed farm animals (Dantzer and Hormede 1983); Increased levels of cortisol in acute heat stress in Holstein steers (Habeeb *et al.* 1996). The physiological significance of higher plasma cortisol is to mobilize amino acids by protein catabolism for gluconeogenesis (Bhan *et al.* 2013). Sharma *et al.* (2013) also reported higher (P<0.05) cortisol levels in control compared to melatonin treated group during various exposure temperatures which reveals its protective role against heat alleviation and therefore suppressed serum cortisol level.

Table 5. Correlation among various physiological and nutritional parameters of growing Murrah buffaloes

	T_{rec}	RR	PR	DMI	B Wt	ADG
T_{rec}	1					
RR	0.398**	1				
PR	0.336**	0.381**	1			
DMI	–0.001	–0.093	–0.189*	1		
B Wt	–0.078	–0.172	–0.273**	0.960**	1	
ADG	–0.445**	–0.101	–0.236**	0.420**	0.389**	1

*, Correlation is significant at the 0.05 level; **, Correlation is significant at the 0.01 level.

The mean values of body weight, ADG and DMI of control and treatment group of animals have been presented in Table 4. The mean values of body weight for treatment group of growing buffaloes were higher (P<0.05) than the control group. The body weight gain was significantly (P<0.05) different among the groups and fortnights. Body weight gain was positively correlated with DMI and ADG and negatively correlated with $RT_,$ RR and PR. Ronchi *et al.* (2001) also reported lower body weight gains in control animals compared to treatment group due to lower DMI. Reduction in feed intake and body weight gain was reported in heat exposed animals by Singh *et al.* (2014). During the present study the ADG was higher (P<0.05) in treatment than the control group of growing buffaloes. ADG was positively correlated with DMI and body weight and negatively correlated with $RT_,$ RR and PR. ADG declined with decrease in DMI during heat stress (Ronchi *et al.* 2001). DMI was positively correlated with body weight and ADG while it was negatively correlated with RT, RR and PR. The results of the study corroborate with Ronchi *et al.* 2001 who reported a decrease in DMI due to heat stress exposure. Ahmad and Tariq (2010) also reported decline in DMI of buffaloes with an increase in ambient temperature.

Heat stress severely affected the feed intake, average daily gain, physiological responses and hormonal levels in control group whereas subcutaneous injection of melatonin in treatment group protected the animals from the adverse effects of heat stress. Melatonin implantation reduced the plasma levels of cortisol significantly in treatment group of heifers. Due to heat stress, ameliorative action of melatonin helped in conservation of energy which resulted in an increase in growth performance, which could be otherwise used in dissipation of heat stress. Hence, it is very pertinent that melatonin implantation is able to protect growing buffaloes against heat stress and is boon to increase the growth rate and lowering the age of puberty.

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REFERENCES

Ahmad S and Tariq M. 2010. Heat stress management in water buffaloes: A review. *Review Vetenaria* **21**(1): 297–310.

Avendano-Reyes L, Alvarez-Valenzuela F D, Correa-Calderon A, Saucedo-Quintero J S, Robinson P H and Fadel J G. 2011. Effect of cooling Holstein cows during the dry period on postpartum performance under heat stress conditions. *Livestock Science* **281**: 2535–47.

Barcelo P, Nicolau C, Gamundí A, Fiol M A, Tresguerres J A F, Akaarir M and Rial R V. 2016. Comparing the behavioral effects of exogenous growth hormone and melatonin in young and old wistar rats. *Oxidative Medicine and Cellular Longevity* **2016***:* 17.

Barchas J, Da Costa F and Spector S. 1967. Acute pharmacology of melatonin. *Nature* **214**: 919–20.

Bhan C, Singh S V, Hooda O K, Upadhyay R C and Beenam. 2013. Influence of temperature viability on physiological, hematological and biochemical profiles of growing and adult Karan Fries cattle. *Indian Journal Animal Sciences* **83**(10): 1090–96.

Charles L, Ralph B T, William A G and David W O. 1978. The pineal complex and thermoregulation. *Biological Reviews* **54**: 41–72.

Chaudhary S S, Singh V K, Upadhyay R C, Puri G, Odedara A B and Patel P A. 2015. Evaluation of physiological and biochemical responses in different seasons in Surti buffaloes. *Veterinary World* **8**(7): 727–31.

Dantzer R and Hormede P. 1983. Stress in farm animals: A need for revaluation. *Journal of Animal Science* **57**(6):1594–1600.

Dawson D and Van den heuvel J. 1998. Integrating the actions of melatonin on human physiology. *Annals of medicine* **30**: 95–102.

Habeeb A A M, Maria I F M, Kamal T H, Phillips C and Piggins D. 1996. Heat stress in farm animals and environment. *Journal of the Japanese Society of Grasslands* **41**(4): 280–86.

Haque N, Ludri A, Hossain S A and Ashutosh M. 2012. Alteration of metabolic profiles in young and adult Murrah buffaloes exposed to acute heat stress. *International Journal of Applied Animal Science* **1**(1): 23–29.

Hardeland R. 2005. Antioxidative protection by melatonin: multiplicity of mechanisms from radical detoxification to radical avoidance, *Endocrine* **27**(2): 119–30.

Hemati K, Pourhanifeh M H, Dehdashtian E, Fatemi I, Mehrzadi S, Reiter R J and Hosseinzadeh A. 2020. Melatonin and morphine: potential beneficial effects of co use. *Fundamental and Clinical Pharmacology*. doi.org/10.1111/fcp.12566.

Hyder I, Sejian V, Bhatt R and Gaughan J B. 2017. Biological role of melatonin during summer season related heat stress in livestock. *Biological Rhythm Research* **48**(2): 297–314.

Indu B, Hooda O K and Upadhyay R C. 2016. Effect of thermal stress on physiological, hormonal and haematological parameters in Tharparkar and Karan Fries calves. *Indian Journal of Dairy Science* **69**(4): 467–72.

Kumar S and Singh S V. 2018. Influence of astaxanthin supplementation on attainment of puberty and lipid peroxidation in Sahiwal and Karan Fries (Holstein× Tharparkar) heifers during summer season. *Biological Rhythm Research* **51**(1): 15–28.

Liu L, Labani N, Cecon E, and Jockers R. 2019. Melatonin target proteins: Too many or not enough? *Frontiers in Endocrinology* (Lausanne) **10**: 791.

Mc Dowell R E. 1972. *Improvement of Livestock Production in Warm Climates,* pp. xi + 711. W.H. Freeman and Company, San Francisco, 2nd Edition CA.

Mitra R, Christison G I and Johnson H D. 1972. Effect of prolonged thermal exposure on growth hormone secretion in cattle. *Journal of Animal Science* **34**: 776–79.

Omran F I, Ashour G, Youssef M M and Shafie M M. 2011. Responses of hematology, blood metabolites, mineral ions and hormonal profile to heat stress for Egyptian buffalo-calves. *Egyptian Journal of Agriculture Research* **89**(3): 1129–40.

Ronchi B, Stradaioli G, Verini Supplizi A, Bernabucci U, Lacetera N, Accorsi P A, Nardone A and Seren E. 2001. Influence of heat stress and feed restriction on plasma progesterone, estradiol-17b, LH, FSH, prolactin and cortisol in Holstein heifers. *Livestock Production Science* **68**: 231–41.

Rudnitskaya E A, Maksimova K Y, Muraleva N A, Logvinov S V, Yanshole L V, Kolosova N G and Stefanov N A. 2015. Beneficial effects of melatonin in a rat model of sporadic Alzheimer's disease. *Biogerontology* **16**(3): 303–16.

Sharma S, Ramesh K, Hyder I, Uniyal S, Yadav V P, Panda R P, Maurya V P, Singh G, Kumar P, Mitra A and Sarkar M. 2013. Effect of melatonin administration on thyroid hormones, cortisol and expression profile of heat shock proteins in goats (*Capra hircus*) exposed to heat stress. *Small Ruminant Research* **112**: 216–23.

Siddiqui M H, Al-Whaibi M H and Basalah M O. 2011. Interactive effect of calcium and gibberellin on nickel tolerance in relation to antioxidant systems in *Triticum aestivum*. *Protoplasma* **248**: 503–11.

Singh S V, Hooda O K, Narwade B, Beenam and Upadhyay R C. 2014. Effect of cooling system on feed and water intake, body weight gains and physiological responses of Murrah buffaloes during summer conditions. *Indian Journal of Dairy Science* **67**(5): 426–31.

Strassman R J, Qualls C R, Lisansky E J and Peak G T. 1991. Elevated rectal temperature produced by all night bright light is reversed by melatonin infusion in man. *Journal of Applied Physiology* **71**: 2178–82.

Tao S, Bubolz J W, do Amaral B C, Thompson I M, Hayen M J, Johnson S E and Dahl G E. 2011. Effect of heat stress during the dry period on mammary gland development. *Journal of Dairy Science* **94**: 5976–86.

West J W. 2003. Effects of heat-stress on production in dairy cattle. *Journal of Dairy Science* **86**(6): 2131–44.