



Study on Survival, Serum Immunoglobulin Levels and Some Blood Parameters of Holstein Calves Reared in Individual Calf Hutches

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ABSTRACT

Calf deaths occur most often from birth to two months of age. Sufficient amount of colostrum consumption, type of barn and hygiene are factors that positively affect the survival of calves. Calves that are fed with a sufficient amount of colostrum to obtain healthy and disease-resistant calves should be taken into the hutches prepared for a single calf. The aim of this study was to determine the effect of the birth season on calves living in individual calf hutches, livability, serum immunoglobulin level and some blood parameters. The material of the study consisted of blood samples of 88 female Holstein calves reared in individual fiberglass calf hutches. Birth season had no effect on calves blood serum total protein and glucose levels ($P>0.05$), while the age and seasonal factors had significant effect on the immunoglobulin levels and blood parameters ($P<0.05$). Blood serum IgG levels of calves in the first two months of age were significantly higher in autumn and winter than in spring and summer ($P<0.001$). Serum IgM levels were found to have higher mean scores in autumn than in other seasons ($P<0.001$). Serum IgA levels of calves in one-day age were found to be similar in spring and summer seasons, but the difference between all seasons at the other day periods was statistically significant ($P<0.001$). There was no difference in serum total protein levels between autumn and winter months for each measurement period, whereas it was found to be statistically significantly lower than the other seasons at first day of age ($P<0.01$). Serum albumin levels were found that the summer season was statistically lower than the other seasons ($P<0.001$). Serum globulin, urea, glucose levels were found to be statistically significant between seasons. As a result; In addition to the barn structure, it can be said that the seasonal effect is important in terms of providing adequate passive immunity between the births of calves to weaning age. It can be said calves in individual hutches reared in summer and spring months have negative effects on passive immunity transfer in Aydın province and surrounding area. In our study, it was found that calf hutches which are suitable for climatic conditions played an important role in preventing heat stress. It was concluded that the colostrum intake of calves will shed light on the studies investigating the interaction of blood parameters and environmental factors, which are indicative of passive transfer mechanism.

Keywords: Calf, Holstein, individual calf hutch, serum immunoglobulin

Bireysel Kulübelerde Barındırılan Holştayn Buzağlarının Yaşama Gücü, Serum İmmunglobulin Düzeyleri ve Bazı Kan Parametreleri Üzerine Bir Araştırma

ÖZET

Buzağı ölümlerinin en fazla görüldüğü doğum-iki aylık yaş arası dönemde; yeterli miktarda kolostrum tüketimi, barınak tipi ve hijyeni, buzağlarının yaşama gücünü olumlu yönde etkileyen faktörlerdir. Sağlıklı ve hastalıklara karşı dirençli buzağılar elde etmek için yeterli miktarda kolostrumla beslenmiş olan buzağuların, tek buzağı için hazırlanmış kulübelere alınması gerekmektedir. Çalışma, bireysel buzağı kulübelerinde barınan buzağuların yaşama gücü, serum immunglobulin düzeyi ve bazı kan parametreleri üzerine doğum mevsiminin ne ölçüde etkilediğinin araştırılması amacıyla yapılmıştır. Araştırmanın materyalini fibreglas bireysel buzağı kulübelerinde barındırılan 88 adet buzağıya ait kan örnekleri oluşturmuştur. Buzağuların serum total protein ve glikoz düzeyleri üzerine mevsimin bir etkisi bulunmamış ($P>0,05$), immunglobulin düzeyleri ve kan parametreleri üzerine ise buzağı yaşı ve mevsim faktörlerinin önemli düzeyde etkisi olduğu gözlenmiştir ($P<0,05$). Buzağuların ilk iki aylık yaşta kan serum İgG düzeyleri, sonbahar ve kış mevsimlerinde ilkbahar ve yaz mevsimlerinden istatistiksel olarak önemli derecede yüksek bulunmuştur ($P<0,001$). Serum İgM düzeylerinin her ölçüm dönemi için sonbahar mevsiminin diğer mevsimlere göre istatistiksel önemde daha yüksek ortalamalara sahip olduğu saptanmıştır ($P<0,001$). Buzağuların bir günlük yaşta Serum İgA düzeyi ortalamaları ilkbahar ve yaz mevsimlerinde benzer bulunurken, diğer yaş dönemlerinde tüm mevsimler arası fark istatistiksel olarak önemli bulunmuştur ($P<0,001$). Serum total protein düzeyleri arasında her ölçüm dönemi için sonbahar ve kış ayları ortalamaları bakımından farklılık bulunmazken bir günlük yaşta yaz ayı ortalamasının diğer mevsimlerden istatistiksel olarak önemli düzeyde düşük olduğu saptanmıştır ($P<0,01$). Serum albumin düzeylerinin, her bir ölçüm döneminde yaz mevsiminde diğer mevsimlere göre istatistiksel anlamda düşük olduğu saptanmıştır ($P<0,001$). Serum globulin, üre, glukoz düzeylerinin mevsimler arasında farklılığı istatistiksel olarak önemli bulunmuştur. Sonuç olarak; buzağuların süt kesim yaşına kadar olan dönem mortalitesinin azaltılmasında barınak yapısının yanı sıra mevsimsel etkinin de özellikle yeterli pasif bağışıklık sağlanması bakımından önemli olduğu söylenebilir. Çalışmanın gerçekleştirildiği Aydın ilinde özellikle yaz ve ilkbahar ayları bireysel kulübelerde barındırılan buzağularda sıcak stresine yol açarak pasif bağışıklık transferini olumsuz yönde etkilediği söylenebilir. Çalışmada bölge iklim koşullarına uygun buzağı barınakları yapılmasının sıcak stresini engellemede önemli rol oynadığı saptanmıştır. Araştırmanın, buzağularda kolostrum pasif transfer mekanizması göstergesi olan kan parametreleri ile çevresel faktörlerin etkileşimini inceleyen araştırmalara ışık tutacağı sonucuna varılmıştır.

Anahtar Sözcükler: Bireysel buzağı kulübesi, buzağı, Holştayn, serum immunglobulin.

Introduction

Weaning age of the calves is most important life period in dairy cattle farms from birth until 2 months of age. Calf deaths are seen most common in weaning period. Higher birth weight; colostrum consumption, shelter type and hygiene are the factors that affect the survival of calves positively in this period. In order to obtain healthy and disease-resistant calves, sufficient colostrum feeding should be obtained in individual calf hutches. In modern enterprises, calves are generally breeding in hutches and then transferred to paddocks. It has been reported in some researches about intensive dairy cattle breeding that breeding of calves in individual hutches during weaning period significantly reduce problems of health, growth and maintenance-feeding (Cummins and Brunner, 1991; Gudin, 1991; Quigley et al., 1995; Shukanov, 1992; Virtala et al., 1999; Zhekov et al., 1997). It has been reported that calf hutches made from wood, polyester, water plywood, corrugated cement board, galvanized sheet, cheap and easy to find different materials are most suitable system for calf breeding (Tümer, 1995; Virtala et al., 1999). Macaulay et al. (1995) reported that polythene individual calf hutches deployed inside the barn have the hottest micro-climate; this is followed by wood and polymer hutches outside the barn. Serum immunoglobulin measurement is a practical and important application in newborn calves. Various factors interact with the amount of passive immunoglobulin to detect the presence of the disease. These factors include farm management, environment, hygiene, presence of infection, virulence of infectious organisms and antibody specificity. The risk of disease is high because immunoglobulin passive transfer is not fully realized in neonatal calves. Similarly, newborns which have received adequate passive transfer can easily become ill if housed in unhygienic shelters (Weaver et al., 2000). In a study about some blood parameters of calves housed in hutches using various building materials, the mean total protein, albumin and globulin values of calves in polyethylene hutches were found lower than calves housed in hutches by polymer, wood and metal building materials (McKnight et al., 1999). In some studies on the survival of calves, mortality was observed

in summer lower than in winter (Ghose et al., 1994; Mishra and Taneja, 1991; Streit and Ernst, 1992). Some studies on calf shelter types, was researched about environmental, yield and behavioral differences between individual hutches and barn conditions (Hanekamp et al., 1994; Virtala et al., 1999). It has not been observed in any scientific study about this issue in Turkey. It is important to determine the effect of fiberglass individual calf huts, which are becoming more common in intensive dairy cattle breeding, on calf survival, serum immunoglobulin level, and some blood parameters and shelter microclimate.

The aim of this study was to determine the effect of the birth season on calves living in individual calf hutches, livability, serum immunoglobulin G (IgG) which is an important indicator of full immunity against diseases, immunoglobulin A (IgA) and immunoglobulin M (IgM) levels and some blood parameters (total protein, albumin, globulin, urea, glucose, ionized calcium, sodium and potassium levels).

This study; The survival of Holstein calves who spent the weaning period in fiberglass calf hutch, some blood parameters (total protein, albumin, globulin, urea, glucose, ionized calcium, sodium and potassium levels) and serum immunoglobulin G which is an important indicator of full immunity against diseases (IgG), immunoglobulin A (IgA) and immunoglobulin M (IgM) levels on the effect of birth season were investigated.

Materials and Methods

The research protocol of the study was approved by the Ethic Committee of Adnan Menderes University (Approval Number: 2009/057). The study was conducted in a private dairy cattle farm in Aydin, Turkey. The material of the study consisted of 88 female Holstein calves born between 1 March 2010 and 28 February 2011. The calves were born at the calving pens and were transferred to the fiberglass calf hutches within 24 hours, regardless of gender. Calves were fed with colostrum and ad libitum fresh water with intensive and coarse feed supplements. All of the calf hutches are 110 cm×162×137 cm width and height. The calves were fed twice daily, at 9.00 am and 5.00 pm, with a total of 4 liters of preheated milk per day, 2 liters per meal. The milk was given daily with soft rubber mouthed, plas-

Table 1. Interactions of some blood mean parameters with immunoglobulins (F)

| Parameter | In-group (Time) | Between Groups (Seasons) | Factor x Seasons |
|------------------|-----------------|--------------------------|------------------|
| Immunoglobulin G | 310.609*** | 25.209*** | 4.334** |
| Immunoglobulin M | 1770.919*** | 8.545*** | 204.933*** |
| Immunoglobulin A | 756.792*** | 10.680*** | 493.934*** |
| Total Protein | 4.985* | 1.322 ^{NS} | 3.413* |
| Albumin | 332.124*** | 6.578*** | 49.450*** |
| Globulin | 356.395*** | 5.658*** | 43.055*** |
| Urea | 588,303*** | 4,824*** | 21,219*** |
| Glucose | 568,584*** | 1,136 ^{NS} | 132,837*** |
| Calcium | 3696,734*** | 20.297*** | 63.965*** |
| Potassium | 30.068*** | 2.632** | 5.386** |
| Sodium | 77.070*** | 2.257* | 15.037*** |

*: P<0.05; **: P<0.01; ***: P<0.001; NS: Non-significant

Table 2. Immunoglobulin levels of calves in different seasons in the first two months after birth (g/L, n= spring: 21, summer: 25, autumn: 20, winter: 18; Mean±Standard error)

| | Seasons | Days | | | | F |
|------------------|---------|-------------------------|-------------------------|-------------------------|-------------------------|-----------------------|
| | | 1 th day | 4 th day | 30 th day | 60 th day | |
| Immunoglobulin G | Spring | 13.1±1.4 ^{bd} | 24.8±2.2 ^{cc} | 43.4±2.9 ^{bb} | 59.0±5.0 ^{ba} | 40.15 ^{***} |
| | Summer | 13.8±1.3 ^{bd} | 35.2±2.0 ^{bc} | 44.5±2.7 ^{bb} | 62.8±4.6 ^{ba} | 41.28 ^{***} |
| | Autumn | 26.2±1.5 ^{ad} | 52.4±2.2 ^{ac} | 62.0±3.0 ^{ab} | 93.7±5.1 ^{aa} | 55.43 ^{***} |
| | Winter | 21.6±1.5 ^{ad} | 47.0±2.3 ^{ac} | 53.6±3.2 ^{abb} | 91.0±5.4 ^{aa} | 51.12 ^{***} |
| | F | 18.32 ^{***} | 29.97 ^{***} | 8.47 ^{***} | 12.73 ^{***} | |
| ImmunoglobulinM | Spring | 0.19±0.06 ^{cc} | 0.27±0.09 ^{cb} | 0.90±0.01 ^{ba} | 0.82±0.01 ^{ca} | 552.68 ^{***} |
| | Summer | 0.27±0.05 ^{bd} | 0.37±0.09 ^{bc} | 0.96±0.02 ^{ba} | 0.79±0.01 ^{cb} | 515.88 ^{***} |
| | Autumn | 0.50±0.06 ^{ab} | 0.52±0.01 ^{ab} | 1.13±0.27 ^{aa} | 1.08±0.01 ^{aa} | 444.90 ^{***} |
| | Winter | 0.25±0.06 ^{bc} | 0.30±0.07 ^{cb} | 0.94±0.02 ^{ba} | 0.98±0.01 ^{ba} | 574.39 ^{***} |
| | F | 487.23 ^{***} | 116.92 ^{***} | 15.13 ^{***} | 65.18 ^{***} | |
| Immunoglobulin A | Spring | 0.13±0.01 ^{cd} | 2.16±0.6 ^{dc} | 3.02±0.92 ^{db} | 3.99±0.17 ^{da} | 69.66 ^{***} |
| | Summer | 1.57±0.09 ^{cd} | 3.52±0.68 ^{cc} | 4.59±0.85 ^{cb} | 5.98±0.15 ^{ca} | 216.27 ^{***} |
| | Autumn | 3.16±0.10 ^{bd} | 4.54±0.69 ^{bc} | 6.05±0.95 ^{bb} | 6.65±0.17 ^{ba} | 153.66 ^{***} |
| | Winter | 3.86±0.11 ^{ad} | 5.04±0.71 ^{ac} | 6.55±0.10 ^{ab} | 7.95±0.18 ^{aa} | 154.63 ^{***} |
| | F | 133.29 ^{***} | 348.16 ^{***} | 279.05 ^{***} | 84.27 ^{***} | |

*** P<0.001; a, b, c, d: The difference between the means carrying different letters in the same column is statistically significant; A, B, C, D: The difference between the means carrying different letters in the same line is statistically significant (P<0.05)

tic body calf feeding bottles attached to the hangers in front of the hutches. In order to determine blood serum immunoglobulin levels and some blood parameters at various age periods of calves, 5 ml of blood from vena jugularis of each calf in the first 24 hours after birth (1st day), on the 4th, 30th and 60th days of age was collected. Total protein, albumin, globulin, urea and glucose levels were determined by spectrophotometric commercial test kits from serum samples to be obtained from blood Aydin Adnan Menderes University, Faculty of Veterinary Medicine, Department of Biochemistry laboratories in Aydin. IgG, IgA and IgM levels were determined by using commercial Elisa Kits (Cusabio, China) using microtiter plates and an ELISA reader (Anthos 2010, Anthos Labtec Instruments, Salzburg, Austria). Levels of ionized calcium, sodium and potassium, total protein, albumin, globulin, urea and glucose were determined by Microlab 2000 (Merck) using commercially available kits (Biomedical Systems, Barcelona, Spain) from serum samples to be obtained from blood Aydin Adnan Menderes University, Faculty of Veterinary Medicine, Department of Biochemistry laboratories. The analyses were carried out according to the manufacturer's recommendations.

Since the number of milking cows in the enterprise was sufficient, blood samples were taken from 22 calves in each season and the data of the calves dying during the study were evaluated as missing data in statistical calculations.

Data are presented with mean and standard error values. The difference between the 1st, 4th, 30th and 60th day measurements, within and between the seasons was determined by the General Linear Model, in terms of blood parameters, IgG, IgA and IgM. For repeated measures, multivariate analysis was performed. A p-value of less than 0.05 was considered to show a statistically significant result.

Results

Five of 88 female calves in the birth season groups have died at different stages of the study from the beginning. One of these calves died in spring and one died in autumn months and three of them died in winter, and the mortality values for spring, summer, autumn and winter were 4.5%, 0%, 4.5% and 13.6%,

respectively. The data obtained from these calves as long as they lived were evaluated.

General differences of all parameters within the group (time) and between groups (season) are given in Table 1. There was no effect of season on serum total protein and glucose levels of calves (P>0.05), but it was observed that time and season factors had significant effect on immunoglobulin levels and blood parameters (P<0.05). Calf serum immunoglobulin levels were measured starting with spring in every season starting with birth in the 4th, 30th and 60th days after birth. The highest IgG levels on day 1 and day 60 were 26.2 g/L and 93.7 g/L, respectively. In the same periods, the highest values were found to be 0.50 g/L and 1.08 g/L for IgM, 3.86 g/L and 7.95 g/L for IgA, respectively. It was seen that IgG and IgM levels reach the highest values in autumn and IgA levels reach the highest value in winter months (Table 2). In terms of serum IgG levels, the difference between the seasons was statistically significant for each measurement day (P<0.001). Serum IgG levels determined in autumn and winter were significantly higher than spring and summer values on all four measurement days (P<0.001). Serum IgG levels showed a linear increase between 1-60 days in all four seasons and the difference between the measurement periods was statistically significant (P<0.001). When the serum IgM levels were examined, it was found that the autumn season had statistically higher means than the other seasons for each measurement period (P<0.001). Serum IgM levels increased significantly between 1-30th days in spring, summer and autumn (P<0.001), this level decreased on day 60, but this decrease was only statistically significant in summer compared to the previous period (P<0.001). In winter, there was a statistically significant increase between all measurement periods, and the 30th and 60th day means were significantly higher than the 1st and 4th day measurements (P<0.001). The mean serum IgA levels were found to be similar in the spring and summer means on the first day and the difference between all seasons was statistically significant (P<0.001). It was observed that the means showed a linear increase from spring to winter. When the seasons were examined separately, it was found that serum

Table 3. Total protein, albumin and globulin values of calves in blood in different seasons in the first two months after birth (g/dL, n= Spring: 21, summer: 25, autumn: 20, winter: 18; Mean±Standard error)

| | Seasons | Days | | | | |
|----------------------|---------|--------------------------|--------------------------|--------------------------|-------------------------|----------------------|
| | | 1 th day | 4 th day | 30 th day | 60 th day | F |
| Total protein | Spring | 5.17±0.14 ^{acD} | 5.50±0.13 ^{bcC} | 5.75±0.13 ^{bbB} | 6.14±1.55 ^A | 25.40 ^{***} |
| | Summer | 4.96±0.13 ^{bcD} | 5.12±0.12 ^{bc} | 5.37±0.12 ^{bb} | 5.70±1.42 ^A | 18.61 ^{***} |
| | Autumn | 5.59±0.15 ^{aD} | 6.16±0.13 ^{ac} | 6.60±0.13 ^{ab} | 7.05±1.59 ^A | 71.75 ^{***} |
| | Winter | 5.60±0.16 ^{aD} | 5.98±0.14 ^{acc} | 6.31±0.14 ^{ab} | 10.34±1.67 ^A | 34.50 ^{***} |
| | F | 4.61 ^{**} | 13.24 ^{***} | 18.45 ^{***} | 1.69 ^{NS} | |
| Albumin | Spring | 2.75±0.07 ^{aD} | 2.97±0.07 ^{bc} | 3.14±0.06 ^{bb} | 3.36±0.05 ^{ba} | 19.46 ^{***} |
| | Summer | 2.10±0.06 ^{bd} | 2.42±0.06 ^{dc} | 2.68±0.05 ^{cb} | 3.02±0.05 ^{ca} | 54.94 ^{***} |
| | Autumn | 2.66±0.07 ^{aD} | 3.19±0.07 ^{abc} | 3.52±0.06 ^{ab} | 3.76±0.06 ^{aA} | 62.77 ^{***} |
| | Winter | 2.77±0.08 ^{aD} | 3.33±0.07 ^{ac} | 3.63±0.06 ^{ab} | 3.86±0.06 ^{aA} | 55.05 ^{***} |
| | F | 19.94 ^{***} | 34.43 ^{***} | 53.17 ^{***} | 45.20 ^{***} | |
| Globulin | Spring | 2.47±0.05 ^{aD} | 2.84±0.06 ^{bcC} | 3.01±0.06 ^{bb} | 3.24±0.06 ^{ba} | 36.89 ^{***} |
| | Summer | 2.10±0.05 ^{bc} | 2.49±0.05 ^{db} | 2.62±0.05 ^{cb} | 2.89±0.05 ^{ca} | 48.79 ^{***} |
| | Autumn | 2.54±0.06 ^{aD} | 2.98±0.06 ^{cc} | 3.33±0.06 ^{ab} | 3.58±0.06 ^{aA} | 62.51 ^{***} |
| | Winter | 2.59±0.06 ^{aD} | 3.15±0.07 ^{ac} | 3.56±0.06 ^{ab} | 3.77±0.06 ^{aA} | 74.94 ^{***} |
| | F | 15.60 ^{***} | 19.55 ^{***} | 43.16 ^{***} | 40.49 ^{***} | |

P<0.01; * P<0.001; NS: Non-significant; a, b, c, d: The difference between the means carrying different letters in the same column is statistically significant; A, B, C, D: The difference between the means carrying different letters in the same line is statistically significant (P<0.05).

IgA levels increased between 1-60 days of age and this increase was statistically different (P<0.001).

The means of total protein, albumin and globulin obtained from blood sera obtained from calves are presented in Table 3. Total protein mean was determined as the first day of measurement in summer (4.96 g/dL) and the highest winter season in day 60 (10.34 g/dL).

The lowest and highest values for albumin were determined as the first day measurement of summer (2.10 g/dL) and 60 days of winter (3.86 g/dL) respectively, while the lowest and highest values for globulin were summer first day measurement (2.10 g/dL) and winter season 60th day measurement (3.77 g/dL). It was found that the mean of summer at 1 day age was significantly lower than the other seasons (P<0.01). Serum total protein 4th day means were not different between winter and autumn and spring, whereas autumn and winter were significantly higher than summer mean and autumn mean (P<0.001). Serum total protein levels in the autumn and winter were significantly higher in the 60th day measurements than in the spring and summer (P<0.001). Serum total protein level increased as the measurement days progressed in each season and a statistically significant difference was found between the measurement days (P<0.001). When the change in serum albumin level is considered, it was found that the summer season was statistically lower than the other seasons at each measurement period (P<0.001). The first day of measurement in summer serum albumin mean is lower than other seasons; the mean taken from since the fourth day were significantly higher in autumn and winter than in spring and summer. Serum albumin level was found to increase linearly with increasing age and this increase was statistically significant in all seasons (P<0.001). Serum globulin levels were also found to have statistically significant differences between the seasons at each measurement day and between days in each season (P<0.001). It was determined that the summer season had a significantly lower value on the first day compared to the other days, and that the spring and summer months had lower values compared to the autumn and winter months in the 4-60 day period. The mean serum glucose lowest level was 54.40 mg/dL on the

60th day of autumn, and the highest level was 118.11 mg/dL on the first day of winter. The difference between seasons of serum urea levels for each measurement day was statistically significant (P<0.001).

In terms of serum urea means on the first, fourth and 30th days, the summer and winter months were found to be significantly higher than the spring and autumn months, whereas the 60th day spring mean was significantly lower than the other seasons. When the serum glucose level was taken into consideration, it was determined that the differences between the measurement days and the seasons were statistically significant (P<0.001). In the 1st, 30th and 60th day measurements, it was found that the spring and winter means were significantly higher than the autumn and summer means, and the calves born during the summer also had significantly higher serum glucose concentrations than the calves born during the autumn season (P<0.001). On the 4th day, the highest serum glucose concentration was observed in the winter. Spring, autumn and summer months' levels followed winter months respectively. It was also found that serum glucose concentration showed a linear decrease with time progressing in all four seasons (Table 4). Calcium, potassium and sodium ratios obtained from blood serum obtained from calves are presented in Table 5. In all seasons, there were statistically significant differences in serum calcium concentrations of calves between all age stages and a linear decrease was observed with increasing age (P<0.001). While calf serum potassium level is maximum 4.55 mEq/L, it occurs in winter in 1 day age period; the lowest level was 3.68 mEq/L with summer 4-day age period. In the first day of summer, serum potassium levels were significantly lower than the other seasons (P<0.05), whereas in the fourth day, summer and autumn months had significantly lower values than spring and winter (P<0.01). The mean of serum potassium levels the fourth day in summer, autumn and winter was significantly lower than the other days (P<0.001, P<0.01). Serum sodium levels were statistically significant between the seasons and between 1-60 days of age blood measurement periods (P<0.001). Sodium levels in calf blood serum taken at 1 and 30 days of age were significantly higher in autumn and winter compared to

Table 4. Blood urea and glucose values of calves in different seasons in the first two months after birth (mg/dL, n= Spring: 21, Summer: 25, Autumn: 20, Winter: 18; Mean±Standard error)

| | Seasons | Days | | | | F |
|---------|---------|---------------------------|---------------------------|---------------------------|--------------------------|----------------------|
| | | 1 th day | 4 th day | 30 th day | 60 th day | |
| Urea | Spring | 13.76±0.45 ^{bd} | 15.38±0.53 ^{bc} | 19.42±0.73 ^{db} | 24.23±0.81 ^{ba} | 55.44 ^{***} |
| | Summer | 17.00±0.41 ^{ad} | 19.68±0.49 ^{ac} | 24.60±0.67 ^{acB} | 29.80±0.75 ^{aA} | 90.51 ^{***} |
| | Autumn | 14.20±0.46 ^{bcD} | 16.80±0.55 ^{bc} | 23.35±0.75 ^{bcB} | 28.10±0.84 ^{aA} | 91.27 ^{***} |
| | Winter | 15.61±0.49 ^{acD} | 20.88±0.57 ^{aC} | 26.61±0.79 ^{aB} | 30.61±0.88 ^{aA} | 81.62 ^{***} |
| | F | 11.33 ^{***} | 21.56 ^{***} | 16.23 ^{***} | 11.78 ^{***} | |
| Glucose | Spring | 112.38±1.45 ^{aA} | 101.38±1.42 ^{bb} | 83.90±1.51 ^{aC} | 73.90±1.77 ^{ad} | 65.90 ^{***} |
| | Summer | 105.08±1.33 ^{ba} | 95.52±1.30 ^{cb} | 76.24±1.39 ^{bc} | 66.92±1.62 ^{bd} | 75.75 ^{***} |
| | Autumn | 96.90±1.49 ^{ca} | 84.90±1.46 ^{db} | 65.90±1.55 ^{cC} | 54.40±1.81 ^{cd} | 75.43 ^{***} |
| | Winter | 118.11±1.57 ^{aA} | 109.61±1.54 ^{aB} | 86.00±1.64 ^{aC} | 74.16±1.91 ^{ad} | 71.04 ^{***} |
| | F | 36.85 ^{***} | 48.61 ^{***} | 33.59 ^{***} | 25.80 ^{***} | |

*** P<0.001; a, b, c, d: The difference between the means carrying different letters in the same column is statistically significant; A, B, C, D: The difference between the means carrying different letters in the same line is statistically significant (P<0.05)

spring and summer (P<0.001). Fourth day autumn serum sodium levels were significantly higher than the spring and summer seasons (P<0.001), but there was no significant difference between autumn and winter seasons. Blood samples taken at the age of 60 days; again, it was found that serum sodium levels were significantly higher (P<0.001) in the autumn season than in the spring and summer seasons. Serum sodium levels in the 60-day age period were significantly lower than the one and four-day age period (P<0.001); there was no statistically significant difference between the ages of 30-60 days in summer and winter seasons.

Discussion

In our study, 30th and 60th days IgG values were found higher than the values obtained by Broucek et al. (1990). While Serum IgG concentrations of newborn calves were found to be lower than those determined by Streit and Ernst (1992); the values at the age of 30 days were similar and higher than those at the age of 60 days. It can be said that colostrum immunoglobulin intake of calves in our study is better and the immune systems of calves are stronger with advancing time. Yuceer, (2008), reported the development of lower passive immunity in calves born in autumn. The lowest colostrum IgG concentration was found in cows giving birth in summer months. Similarly, serum IgG levels of calves born in summer were lower. These data were found to be consistent with the results of our study. Earley and Fallon (1999) reported that the serum IgG levels of calves born in spring were higher than those of calves born in autumn, and that the serum serum IgG levels of one month old calves reached the highest level in autumn compared to 22.6 mg/ml. In our study, it was shown that blood serum IgG levels obtained in autumn and winter months reached significantly higher levels than those obtained in spring and summer months. It can be said that calf blood serum IgG levels may change in different climatic conditions.

Murphy et al. (2005) reported newborn calf blood serum IgG level as 27.1 mg/ml. This result was found to be similar to the values of winter and autumn seasons found in our study, but it was found to be higher than IgG values obtained from calves born in spring and summer months. It can be said that this difference is caused by working with calves of different breeds in both studies. In addition, especially in summer and spring months, it can be said that the climatic conditions of our region have high environmental temperature values which are prone to causing hot stress, and this affects both colostrum quality

and passive immunity transfer negatively. In a study conducted by Genç (2015) in Erzurum province, blood serum IgG levels of Brown-Swiss and Holstein calves were 12.6 mg/ml, 9.8 mg/ml, 11.97 mg/ml and 12.81 mg/ml in the spring, summer, autumn and winter months, respectively. In both studies, it was observed that calf serum IgG levels were higher in autumn and winter months.

In our study it was found that the serum IgM levels were the lowest in the spring months, followed by the summer and winter months, and the highest levels of serum IgM were reached in the autumn months. Earley and Fallon (1999) reported that the highest serum IgM levels of calves at the one month of age were reached in autumn with a rate of 1.37 mg/ml. These results, which are consistent with our study, suggest that calves gain better passive immunity during the cold season. When the total immunoglobulin level is examined, it is seen that higher levels of immunoglobulin are measured in autumn compared to spring months. This result is consistent with the means obtained from our study. Blood serum IgA levels of newborn calves were found to be similar to the values determined by Streit and Ernst (1992) for spring and summer, but higher than those of autumn and winter. It can be said that the climatic conditions of our study are close to the appropriate humidity and temperature values for cattle in autumn and winter months and increase the colostrum immunoglobulin level and calf blood serum immunoglobulin level. Earley and Fallon (1999) found that calf serum IgA levels in spring were higher than those of autumn months. According to the results of our study, IgA levels were found to be the lowest in autumn in parallel to IgG and IGM levels, and the highest means for winter months were obtained. When some blood parameters obtained as a result of our study were examined in terms of seasonal changes; Calf blood serum total protein levels were significant in newborn, four day and one month old calves. While serum total protein levels of these age periods decreased to the lowest levels in summer months, they reached the highest levels in autumn and winter months. The total protein values of two-month-old calves were highest in winter and lowest in summer. McKnight et al. (1999) reported serum total protein levels in 42-day-old calves showed differences between seasons. The lowest total protein values in summer were followed by autumn and winter, respectively, and the highest total protein levels were reached in spring. It can be said that the ration quality varies in different seasons, in different regions and this affects the total protein level of colostrum positively or negatively. One and two month

Table 5. Calcium, potassium and sodium levels of calves in blood in different seasons, in the first two months after birth (n= Spring: 21, Summer: 25, Autumn: 20, Winter: 18; Mean±Standard error)

| | Seasons | Days | | | | F |
|----------------------|---------|---------------------------|----------------------------|----------------------------|----------------------------|-----------------------|
| | | 1 th day | 4 th day | 30 th day | 60 th day | |
| Calcium (mg/dL) | Spring | 9.82±0.17 ^{CA} | 9.10±0.15 ^{CB} | 5.21±0.06 ^{bCC} | 4.24±0.03 ^{BD} | 353.58 ^{***} |
| | Summer | 9.00±0.15 ^{dA} | 8.60±0.13 ^{BB} | 4.85±0.06 ^{dC} | 4.00±0.03 ^{CD} | 352.67 ^{***} |
| | Autumn | 11.46±0.17 ^{aA} | 10.95±0.15 ^{aB} | 5.64±0.07 ^{aC} | 4.61±0.04 ^{aD} | 532.94 ^{***} |
| | Winter | 10.51±0.18 ^{bA} | 10.01±0.16 ^{bB} | 5.42±0.07 ^{aCC} | 4.60±0.04 ^{aD} | 352.55 ^{***} |
| | F | 39.80 ^{***} | 48.03 ^{***} | 26.13 ^{***} | 59.73 ^{***} | |
| Potassium (mEq/L) | Spring | 4.41±0.10 ^{ac} | 4.18±0.09 ^a | 4.39±0.07 ^D | 4.27±0.07 | 1.66 ^{NS} |
| | Summer | 4.13±0.09 ^{bcA} | 3.68±0.08 ^{BB} | 4.41±0.06 ^A | 4.33±0.07 ^A | 16.73 ^{***} |
| | Autumn | 4.33±0.11 ^{acA} | 3.79±0.09 ^{bcB} | 4.58±0.07 ^A | 4.50±0.07 ^A | 16.04 ^{***} |
| | Winter | 4.55±0.11 ^{aA} | 4.05±0.10 ^{acB} | 4.46±0.08 ^A | 4.47±0.08 ^A | 6.00 ^{**} |
| | F | 2.70 [*] | 6.55 ^{**} | 1.36 ^{NS} | 2.03 ^{NS} | |
| Sodium (mEq/L) | Spring | 136.42±0.69 ^{bA} | 135.00±0.66 ^{bcB} | 133.04±0.66 ^{bc} | 131.42±0.68 ^{BD} | 23.38 ^{***} |
| | Summer | 135.60±0.63 ^{bA} | 134.52±0.61 ^{bcB} | 133.76±0.61 ^{bcC} | 132.84±0.62 ^{bcC} | 10.82 ^{***} |
| | Autumn | 141.05±0.71 ^{aA} | 139.05±0.68 ^{aB} | 138.10±0.68 ^{aB} | 136.10±0.69 ^{aC} | 30.68 ^{***} |
| | Winter | 139.33±0.74 ^{aA} | 136.83±0.72 ^{acB} | 137.22±0.72 ^{aBC} | 135.27±0.73 ^{acC} | 33.30 ^{***} |
| | F | 13.66 ^{***} | 9.67 ^{***} | 13.85 ^{***} | 9.83 ^{***} | |

*: P<0.05; **: P<0.01; *** P<0.001; NS: Non-significant ; a, b, c, d: The difference between the means carrying different letters in the same column is statistically significant; A, B, C, D: The difference between the means carrying different letters in the same line is statistically significant (P<0.05).

of age Holstein calves serum total protein, glucose and albumin averages were found consistent with Khan et al. (2007^b). Within the scope of our study, it can be said that the total protein values of calves born during the winter are higher than the age of two months because the calves are given more concentrated feed in order to meet the energy requirement due to environmental temperature change. Khan et al. (2007^a) found blood serum total protein, glucose and albumin levels of one and two months old Holstein calves, 6.35-6.45 g/dL and 6.40-6.90 g/dL; 93.44-96.62 g/dL and 70.20-84.13 g/dL; 4,11-4,23 g/dL and 4,13-4,45 g/dL respectively. These results are consistent with the total protein, glucose and albumin mean of the one-month-old calves obtained from our study and higher than the average of the two-month-old calves. It can be said that the fact that the winter mean obtained in our study was higher than the mean of serum total albumin and globulin levels were due to the application of a more qualified nutrition program in those months and the positive effect of micro-conditioning conditions on calf's survival and feed efficiency. Swan et al. (2007) determined the maternal colostrum and commercial colostrum substitutes feed serum calves fed the total protein levels of 5.5 g/dL and 4.6 g/dL respectively. In a study designed by Smith and Foster (2007) on Holstein calves, colostrum-fed calf blood serum total protein levels were reported to be 5.4 g/dL, colostrum replacement feed was reported to be 4.4-4.7 g/dL. Godden et al. (2009) reported maternal colostrum-fed newborn Holstein calves and one and two doses of colostrum substitutes fed with total protein levels of 5.7 g/dL, 4.9 g/dL and 5.5 g/dL respectively. The results of these studies are close to the total protein levels of calf blood serum obtained from our study. Johnson et al. (2007) found serum total protein levels of calves fed with raw and heated colostrum at the age of one day as 5.9 g/dL and 6.34 g/dL, respectively. These levels are higher than the levels obtained as a result of our study. In a study by Pauletti et al. (2003), the effect of IgG levels on total protein level was investigated; Total blood serum protein levels of 1 day old and 1 month of age calves with 20 mg/mL and below IgG levels were found as 6.25 mg/mL, and 7.29 mg/mL; while calves with 30 mg/mL and upper IgG levels were as 8.99

mg/mL and 7.92 mg/mL respectively. These results are higher than the mean serum total protein values obtained on the first day of our study, but are similar to the first and second month values. According to the 42th day data, serum globulin showed a statistically significant difference between the seasons. These results are different from the results of our study. As a result of this research, the highest levels were reached in winter, followed by autumn, spring and summer months. It can be said that this difference is caused by the heat stress that occurs in the summer months and that the studies were conducted in different environments. In the same study, the mean serum urea reached the highest level with 4.4 mmol/L in summer and the lowest level with 2.6 mmol/L in spring. The lowest serum urea levels in the spring months are consistent with the data obtained from our study. Serum glucose and calcium levels were highest in winter and autumn, while the lowest values were achieved in autumn and summer. In terms of both biochemical parameters, the seasonal differences obtained from our study were similar.

As a result; healthy development of newborn calves and reduction of mortality in the period up to weaning; it can be said that the calf house structure and seasonal effect are important, especially in terms of providing adequate passive immunity. Moreover, it can be said that calves receiving adequate and qualified colostrum affect calves' blood serum immunoglobulin levels positively. It can be said that the calves which are housed in individual hutches during the summer and spring months in Aydin province where the study was carried out cause heat stress and affect passive immunity transfer negatively. Our study concluded that the construction of calf houses suitable for the climatic conditions of the region plays an important role in preventing heat stress. It is concluded that our study could be guide to new studies investigating the interaction of blood parameters that are indicative of colostrum passive transfer mechanism in calves and environmental factors.

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Conflict of interest

The authors declare that they have no competing interests.

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