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FACTORS AFFECTING COLOSTRUM QUALITY AND PASSIVE IMMUNITY FOR SUCKLING FRIESIAN CALVES UNDER CONDITIONS OF NILE DELTA,EGYPT

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ABSTRACT

One hundred and two Friesian cows and their offspring were used to identify factors associated with colostrum quality, risk factors associated with failure of passive transfer of immunity, and to monitor changes in serum IgG and total protein (TP) concentrations during the first week of age.The time from calving until the first milking, dam parity, the calving date, and calf gender were recorded.The volume, chemical composition of colostrum, and IgG levels were determined. Blood samples of each calf were collected daily until 7 days of age. Concentrations of IgG and TP in blood serum were measured. Significantly higher concentrations of TP and total solid and the percentage of cows that had IgG ≥50 g/L in colostrum were recorded in the winter season, fourth or greater lactation, and when cows were milked within two hours after calving.To assess the passive transfer of immunity for calves, both the IgG and TP concentrations were measured in serum at 24 h after colostrum feeding. The percentage of calves affected by the failure of passive transfer of immunity was 29.41%. Significantly higher concentrations of IgG and TP were recorded in the serum of Friesian calves that received ≥2 L from colostrum containing IgG ≥50 g/L at first feeding within 2h after birth, as well as calves born during the winter season or their dams of the second or more parity. The concentrations of serum IgG and TP in the calves showed no significant changes during the first week of age. They showed the highest values at 24 h after colostrum feeding (24.46 g/L and 6.09±1.44 g/dL, respectively).Thereafter,the values insignificantlydecreased gradually until the end of the first week of age. Concentrations of serum IgG and TP measured from day 2 to 7 were highly correlated with the value recorded 24 h after colostrum feeding (r ≥0.905 and r ≥0.765, respectively). Values of serum IgG and TP concentrations were highly correlated (r= 0.842;*P* <0.0001) during the first week of age. This indicates that determination of serum TP concentration up to day 7 can provide a reliable estimate of passive transfer as an alternative method for rapid and simple monitoring of passive transfer in calves.

Keywords:

Colostrum quality, IgG, total protein, passive immunity, calves

INTRODUCTION

Colostrum management is one of the most critical areas of calf care. Colostrum and subsequently milk provide a complete diet with all the essential nutrients for the neonate during the initial phase of its life. In ruminants, colostrum is the sole source of initial acquired immunity for the offspring **(Tóthová *et al.*, 2016).** Colostrum quality depends on several factors, including the volume produced, the time of collection, and the concentration of immunoglobulins **(Godden, 2008 and Atkinson *et al.*, 2017).** The concentration of IgG in colostrum has traditionally been considered the hallmark for evaluating colostrum quality, where IgG composes more than 85% of total Ig in colostrum. High-quality colostrum has an IgG concentration greater than 50 g/L **(Godden, 2008)**. In addition to immunoglobulins the protein fraction in bovine colostrum comprises bioactive components such as major milk proteins, hormones, growth factors and cytokines. Due to the presence of large amounts of these compounds, colostrum has a beneficial influence on newborn calves during the first days after calving. However, the concentrations of these proteins decrease over time after calving. Thus, the chemical and immunological qualities of colostrum decrease after the first milking. Passive transfer of immunity through colostrum is important for calf health. Colostrum intake is of vital importance for the protection of newborn calves against infectious agents during their first days of age. Failure of passive transfer of immunity (FPT) has been related to increased morbidity and mortality in calves, lower productivity, and increased risk of culling later in life **(Wilm *et al.*, 2018)**. For successful passive transfer of immunoglobulins the calf must ingest the colostrum soon after birth **(Weaver *et al.*, 2000).**

Passive transfer of immunity can be assessed by several methods. Radial immunodiffusion is considered the gold standard for directly assessing serum IgG concentrations.

The determination of serum TP is a useful and practical means of assessing immunoglobulin absorption in the neonatal calf **(Tóthová *et al.*, 2016)**. Serum TP concentration an indirect measure that is highly correlated with blood IgG concentration and is more practical for use on-farm **(Deelen *et al.*, 2014 and Wilm *et al.*, 2018)**. Also, maternal colostrum IgG has a half-life of approximately 10 days **(Hassig *et al.*, 2007 and Wilm *et al.*, 2018)**. Thus, some

researchers have suggested that IgG should be measured by 7 days of age **(Godden, 2008)**. Failure of passive transfer is defined as a serum IgG concentration below 10 g/L at 24 h after birth **(Quigley *et al.*, 2001 and Stanek *et al.*, 2019)**. Negative outcomes associated with FPT include: (1) increased preweaning morbidity and mortality, (2) increase in the duration of illness, contagiousness, and culling rates, and (3) reduced growth rate and milk production in the first lactation **(Atkinson *et al.*, 2017)**. Besides the time until first feeding after birth and the volume of colostrum ingested the immunoglobulin content of colostrum also plays an important role in the success (or failure) of transfer of passive immunity. A low colostralimmunoglobulin concentration can lead to FPT, and consequently increased risk of infectious diseases for the calf **(Reschke *et al.*, 2017)**.

Therefore, the objectives of this study were as follows: (1) to evaluate colostrum quality in terms of its IgG content and nutrients in Friesian cows and to identify risk factors associated with poor colostrum quality, (2) to monitor changes in serum IgG and TP concentrations from birth until 7 days of age to describe the relationship between them, and to determine when reliable estimates of passive transfer of immunity could be acquired and (3) to identify risk factors associated with failure of passive transfer of immunity in calves.

MATERIAL AND METHODS

This study was conducted at El-Qarada Experimental Station, belonged to an Animal Production Research Institute, located in the Nile Delta (Kafr El-Sheikh governorate). A total of 102 Friesian cows (30 primiparous and 72 multiparous) and their calves were used in this study. Cows were housed in open sheds and fed traditional summer ration consisted of concentrate feed mixture (CFM), berseem (alfalfa) hay, rice straw, and corn silage and traditional winter ration consisted of concentrate feed mixture, fresh berseem, and rice straw. Cows were fed to cover the recommended requirements according to Animal Production Research Institute Recommendation **(APRI, 1997)** for dairy Friesian cows. Animals were fed in groups that were assigned according to life body weight, milk yield, and reproductive status. Water was available for animals all over the day round. The average dry period for multiparous cows was 90 days (range: 70 to 120 days).

None of the calves suckled from their dam before they were separated after birth. Calves were weighed and housed in individual rice-straw bedded pens. The dams were milked and the

time from calving until the first milking was recorded, also dam parity and the calving date were recorded. The volume of the colostrum (liter) was estimated, and the IgG concentrations were measured using a colostrometer - hydrometer readings (Kruuse Colostrum Densimeter, Langeskov, Denmark). The calves were fed colostrum through a nipple bottle (colostrum produced from their mother). The time from birth until the first feeding and colostrum volume were recorded.

After that all calves were fed individually on milk at a rate of 10% of body weight given in two meals for six weeks. The milk allowances were reduced gradually until weaning at about 15 weeks of age. Calf starter and hay (high quality) were available in front of calves from the beginning of the third week of age. While fresh and clean drinking water was available in front of them from the third day of age.

Colostrum and blood sampling and analysis:

Colostrum samples of each dam were taken from the first milking used for the first feeding of the calf and stored frozen at -20°C until analyzed. Before analysis, all samples were thawed at room temperature and then were diluted by adding distilled water to reduce sample viscosity and prevent technical difficulties encountered with highly viscous samples. Colostrum composition of fat, total protein, lactose, and total solids were determined by using Milko-Scan (Model 133B, N. Foss Electric PHYSIOLOGICAL Denmark).

Blood samples of each calf were collected daily from the jugular vein into untreated evacuated tubes. The first sample was taken before colostrum feeding, the second sample was taken at 24 h following colostrum feeding, and subsequent samples were taken each day until 7 days of age. All blood samples were centrifuged at 3000 r.p.m. for 10 minutes to separate blood serum which was kept frozen at -20°C until analyzed in the laboratory. Determination of levels of IgG was done by Bovine IgG ELISA kits according to the procedure outlined by the manufacturer (Alpha Diagnostic International, Texas, USA). Concentrations of TP (g/dL) in blood serum were measured according to the procedures described by the manufacturer (BIODIAGNOSTIC, Dokki, Giza, Egypt). Determination methods were colorimetric using spectrophotometer.

Statistical analysis:

The data were statistically analyzed using the General Linear Model (GLM) procedure, **SAS (2002)**. The significant differences among means were tested using Duncan's Multiple

Range Test **(Duncan, 1955)**. Probability values ≤5% were considered significant. Descriptive statistics of cows that had IgG more or less than 50 g/L in colostrum were calculated. Three statistical analysis were performed, the first to evaluate factors affecting colostrum quality and categorical variables included colostrum yield (three levels: ≤3, >3-6, and >6 kg), calving season (four levels: winter, spring, summer, and autumn), lactation number (four levels: first, second, third, and fourth or greater parity), time to first milking postpartum (three levels:<2, 2-6, and >6 h). The second analysis was to evaluate factors affecting serum IgG and TP concentrations and categorical variables included colostrum quality (two levels: IgG ≥50 and IgG<50 g/L), time of first feeding (three levels: <2, 2-6, and >6 h), colostrum volume at first feeding (two levels: ≥2 and <2 L), birth season (four levels: winter, spring, summer, and autumn), Dam parity (three levels: first, second, and third or greater parity), and calf gender (two levels: male and female). In the third analysis, the Pearson correlation was used to measure (1) the correlation between serum IgG concentration measured at 24 h after colostrum feeding and each of day 2 to day 7, (2) the correlation between serum TP concentration measured at 24 h after colostrum feeding and each of day 2 to day 7, and (3) the correlation between serum IgG and TP concentrations measured during the first week of age.

RESULTS AND DISCUSSION

1. Factors affecting colostrum quality:

Table 1 illustrates the overall means for compositions of colostrum, the percentages of cows that had IgG more or less than 50 g/L in colostrum, and the factors affecting its quality.

The overall mean value of fat content was 5.50% ± 0.98% which is lower as compared with that cited by **Kehoe *et al.*, 2007** (6.7%±4.2%), greater than that reported by **Zarei *et al.*, 2017** (4.6%±3.4%) and similar to the **Morrill *et al.*, 2012** (5.3%±0.5) in Holstein cows. Energy from fat in colostrum is critical for thermogenesis and body temperature regulation **(Godden, 2008)**. Low fat content in colostrum affects calf viability, as newborn calves are born with limited fat (380 to 600 g) and calves less than 10 days of age exhibit high rates of metabolism **(Morrill *et al.*, 2012)**.

The overall mean value of protein content was 13.68% ± 2.74% which is lower as compared with that cited by both **Kehoe *et al.*,2007**(14.9%±3.3%) and **Zarei *et al.*,2017** (18.5%),greater than that reported by **Morrill *et al.*, 2012** (12.5%±0.7) in Holstein cows. Protein is the more

important component as a source of amino acids and is the third component of colostrum that can be an energy source, also colostrum contains more total protein than milk (14.0 vs. 3.2%) and a greater percentage of IgG 6.0 vs. 0.09%; **(Morrill *et al.*, 2012)**.

The overall mean value of lactose content was 2.48% ± 0.49% which is lower as compared with that cited by both **Kehoe *et al.*, 2007** (2.5%±0.7%) and **Morrill *et al.*, 2012** (3.0%±0.1%) and, greater than that reported by **Zarei *et al.*, 2017** (2.0%±0.9%) in Holstein cows. Lactose is the primary carbohydrate in colostrum and milk, this proportion of colostrum nutrients is physiologically necessary for the neonate calf and reports confirm that lactose continues to increase until 30 days postpartum **(Morrill *et al.*, 2012 and Zarei *et al.*, 2017)**.

The overall mean value of total solid content was 22.86% ± 3.12% which was consistent with the values reported by **Morrill *et al.*, 2012** (22.2%±0.9%) but is lower than that found by **Zarei *et al.*, 2017** (27.6%±5.8%) in Holstein cows. **Godden (2008)** reported that much of this increase in colostrum solids content is attributed to a more than fourfold increase in protein content of colostrum versus milk, this being because of significant increases in Ig and casein content, also the crude fat content of first milking Holstein colostrum (6.7%) is significantly higher than for milk (3.6%).

Colostrum samples with IgG concentration less than 50 g/L were considered as being of insufficient quality **(Godden, 2008 and Reschke *et al.*, 2017)**.Based on the data obtained from this study, only 22.55% of the samples collected had IgG content <50 g/L. This percentage is lower as compared with that cited by **(Lorenz *et al.*, 2011 and Morrill *et al.*, 2012)** who found that the percentage of cows those had IgG ≥50 g/L in colostrum was 32% and 29.4%, respectively. In contrast, this percentage is greater than that reported by **(Reschke *et al.*, 2017)** (15.5%) in Swiss dairy herds. Many factors can affect IgG concentration of colostrum and may have been explaining the variation in sample means across these studies. Volume of colostrum produced, parity, dry period length, vaccination, and other factors have been reported to affect IgG content **(Weaver *et al.*, 2000)**. Current industry recommendations include discarding colostrum with less than 50 g of IgG/L. In this study, 22.55% of colostrum had IgG concentrations less than the recommended IgG level, thus potentially putting almost 23% of calves at risk of failure of passive transfer.

Effect of colostrum volume:

The overall mean for the volume of first milking colostrum was 5.49 ±1.80 kg (range: 2.00 to 8.00 kg), volume of colostrum was found to affect neither colostrum components, nor its quality (P >0.05). This result agrees with that reported by **Zarei *et al.* (2017**) who indicated that no correlation could be established between the volume of first milking colostrum and colostrum quality or its components.Also, **Godden(2008)** reported that there is no predictable relationship between colostrum IgG concentration and colostrum volume produced at first milking. **Silva-del-Río *et al.* (2017)** stated that cows producing less than 8.5 kg of colostrum at first milking were more likely to produce high-quality (IgG ≥50 g/L) colostrum than cows producing higher quantities of first milking colostrum (≥8.5 kg). In our study, cows produced below this threshold and are thus more likely to meet quality standards based on the concentration of colostrum from IgG.

Effect of calving season:

The results indicate that calving season has a significant effect (P <0.05) on concentrations of total solid and total protein and the percentage of cows that had IgG ≥50 g/L in colostrum (Table 1). Significantly higher concentrations of total solid and total protein and the percentage of cows that had IgG ≥50 g/L in colostrum were recorded in the winter (24.44%, 15.22%, and 84.21%, respectively). Whilst the lowest concentrations of total solid and total protein and the percentage of cows that had IgG ≥50 g/L in colostrum were recorded in the summer (20.39%, 11.05%, and 62.50%, respectively). These results agree with these of **Godden (2008)** who reported that exposure to high ambient temperatures during late pregnancy is associated with poorer colostrum composition, including a lower mean concentration of colostral IgG, and lower mean percentages of total protein, casein, lactalbumin, fat, and lactose. These effects may be attributed to the negative effects of heat stress on dry matter intake resulting in nutritional restriction, reduced mammary blood flow resulting in an impaired transfer of IgG and nutrients from the blood stream to the udder. In addition, **Zarei *et al.* (2017)** indicated that stress is among the factors that suppress the immune system, which leads to increasing cases of the disease. It is well known that, the secretion of glucocorticoids due to stress in late pregnancy might be associated with the decreasing ability of the immune system to produce the required antibodies. They hypothesize that one reason for the decreasing level of IgG is the stressful conditions, including weather

conditions and the temperature, in which the cows spent their late pregnancy period. **Godden (2008)** recommend that producers should adopt the similar heat-abatement strategies for prepartum cows and heifers as are routinely used for lactating cows.

Effect of lactation number:

It was found that there was a trend of higher total protein concentration with increasing parity (Table 1). Also, there was a trend towards an increasing the percentage of cows that had IgG ≥50 g/L in colostrum with increasing parity (Table 1). This result agrees with that of **Moore *et al.* (2005)** who reported that cows in their third or greater lactation had mean colostral IgG concentrations (132 g/L) that were greater than the first and second lactation cows (mean, 95 and 100 g/L, respectively). **Godden (2008) and Puppel *et al.* (2019)** hypothesized that the higher colostrum IgG concentration observed in more mature cows could be explained by their longer exposure to pathogens and incidence of disease. **Puppel *et al.* (2019)** indicated that multiparous cows produce more colostrum with a greater concentration of total solids and total proteins, including immunoglobulins. The highest immunoglobulin levels can be found in the colostrum of cows that are in the 3^{rd} –5^{th} lactation, and the lowest in the primiparous cows. Therefore, it can be stated that colostrum production is usually lesser in the first lactation, suggesting weaker mammary development and potentially reduced transport capacity for immunoglobulins into the mammary gland.

However, we have observed that percentage of primiparous cows that had IgG ≥50 g/L in colostrum is higher (56%) than percentage of primiparous cows that had IgG <50 g/L (44%) in colostrum.This supports the idea maintained by previous researchers**(Reschke *et al.*,2017 and Zarei *et al.*,2017)** that all the colostrum of first parity cows should not be discarded.

Effect of time to first milking postpartum:

The concentration of total solid and total protein and the percentage of cows that had IgG ≥50 g/L in colostrum are highest immediately after calving and begin to decrease over time if milking is delayed. The concentration of total solid and total protein and the percentage of cows that had IgG ≥50 g/L in colostrum were higher when cows were milked within two hours after calving (24.31%, 15.08%, and 82.14%), while they were 22.07%, 13.01%, and 75.86% when cows were milked between 2-6 h and they decreased to 19.46%, 10.20%, and 64.71% when the time to harvest colostrum was longer than 6 h, respectively (Table 1). These results are similar to the results obtained by **Moore *et al.* (2005)**, who reported that

colostrum collected 6, 10, and 14 h after calving had significantly lower IgG concentrations than did colostrum collected 2 h after calving. Means colostral IgG concentration were 113, 94, 82, and 76 g/L at 2, 6, 10, and 14 h after calving, respectively. **Reschke *et al.* (2017)** indicated that prolonged time lag until the first milking postpartum (>6 h) was significantly associated with low colostrum quality. Therefore, reducing the time from calving to first milking might be a good management practice to improve the quality of the colostrum. **Godden (2008)** reported that the concentration of Ig in colostrum is highest immediately after calving but begins to decrease over time if milking is delayed. Delaying harvest of colostrum for 6, 10, or 14 h after calving resulted in a 17%, 27%, and 33% decrease in colostral IgG concentration, respectively. **Lorenz *et al.* (2011)** reported that colostral IgG concentration decreases by 3.7% during each subsequent hour post calving. **Puppel *et al.* (2019)** concluded that colostrum has composition changes with each hour and its biological and nurturing value decrease over time. To collect the highest quality colostrum, farmers should aim to milk the cow within 1 to 2 h after calving to improve colostrum management.

Table (1): Factors affecting colostrum composition and quality in Friesian cows.

Variable	No.	Fat (%)	Protein (%)	Lactose (%)	Total Solid (%)	IgG	
						IgG ≥50 g/L	IgG <50 g/L
Overall mean	**102**	**5.50±0.98**	**13.68±2.74**	**2.48±0.49**	**22.86±3.12**	**(77.45%)**	**(22.55%)**
Colostrum volume							
≤3 kg	**17**	**5.61±0.85**	**13.92±2.89**	**2.36±0.45**	**23.10±3.20**	**(76.47%)**	**(23.53%)**
>3-6 kg	**51**	**5.56±0.96**	**13.75±2.91**	**2.52±0.46**	**23.04±3.20**	**(78.43%)**	**(21.57%)**
>6 kg	**34**	**5.37±1.08**	**13.45±2.46**	**2.47±0.55**	**22.49±3.02**	**(76.47%)**	**(23.53%)**
Calving season							
Winter	**38**	**5.75±1.12**	**15.22±1.61^{A}**	**2.27±0.48^{B}**	**24.44±2.31^{A}**	**(84.21%)**	**(15.79%)**
Spring	**22**	**5.34±1.03**	**13.35±2.84^{B}**	**2.69±0.41^{A}**	**22.58±3.345^{B}**	**(77.27%)**	**(22.73%)**
Summer	**16**	**5.73±0.84**	**11.05±2.71^{C}**	**2.41±0.37^{AB}**	**20.39±2.79^{C}**	**(62.50%)**	**(37.50%)**
Autumn	**26**	**5.15±0.68**	**13.33±2.66^{B}**	**2.64±0.52^{A}**	**22.32±3.06^{B}**	**(76.92%)**	**(23.08%)**
Lactation number							
1st	**25**	**5.46±0.86**	**12.84±3.05^{B}**	**2.38±0.44^{B}**	**21.88±3.31**	**(56.00%)**	**(44.00%)**
2nd	**16**	**5.63±1.08**	**13.41±3.25^{AB}**	**2.51±0.52^{AB}**	**22.74±3.43**	**(81.25%)**	**(18.75%)**
3rd	**17**	**5.36±1.06**	**13.56±2.97^{AB}**	**2.71±0.40^{A}**	**22.84±3.22**	**(82.35%)**	**(17.65%)**
≥4th	**44**	**5.54±1.01**	**14.30±2.16^{A}**	**2.44±0.52^{AB}**	**23.48±2.80**	**(86.36%)**	**(13.64%)**
Time to first milking postpartum							
<2 h	**56**	**5.57±0.95**	**15.08±1.72^{A}**	**2.46±0.49**	**24.31±2.16^{A}**	**(82.14%)**	**(17.86%)**
2-6 h	**29**	**5.37±1.17**	**13.01±2.24^{B}**	**2.48±0.53**	**22.07±2.98^{B}**	**(75.86%)**	**(24.14%)**
>6 h	**17**	**5.53±0.75**	**10.20±2.82^{C}**	**2.53±0.43**	**19.46±3.01^{C}**	**(64.71%)**	**(35.29%)**

$^{A, B, C:}$ Means in the same column within the same factor with different superscripts are significantly different (P <0.05).

2. Factors affecting serum IgG and total protein concentrations in newborn calves:

To assess the passive transfer of immunity for calves, both the IgG and TP concentrations were measured in serum samples at 24 h after colostrum feeding. IgG <10 g/L or TP concentrations ≤5.2 g/dL determined photometrically in the serum of the calves were considered to be indicative of a failure of passive transfer of immunity **(Stanek *et al.*, 2019; Cuttance*et al.*,2017;Osaka *et al.*,2014;Deelen*et al.*,2014andGodden,2008).**

The percentage of calves affected by the failure of passive transfer of immunity was 29.41%. The mean serum IgG concentration was 24.46±13.56 g/L (range: 4.62 to 46.13 g/L), while the mean serum TP concentration was 6.09±1.44 g/dL (range: 3.86 to 8.36 g/dL). This percentage is lower as compared with that cited by both **Stanek *et al.*, 2019** (34.6% in Czech dairy herds), **Vogels *et al.*, 2013** (38% in Australian dairy herds), and **Reschke *et al.*, 2017** (43.5% in Swiss dairy herds) and greater than that reported by both **Beam *et al.*, 2009** (19.2% in the US dairy herds) and **MacFarlane *et al.*, 2015** (26 % in the United Kingdom dairy farms). Although the importance of passive transfer has been studied extensively, dairy farms continue to struggle with failure of passive transfer and associated economic and welfare costs **(Atkinson *et al.*, 2017).** Because the failure of passive transfer of maternal immunity is connected with the negative effect on health, morbidity, and mortality of the calves and long-term negative effects of FPT on the future productivity of dairy cows **(Stanek *et al.*, 2019).**

Effect of colostrum quality:

The serum IgG and TP concentrations in calves were significantly related to the quality of the colostrum fed (Table 2). Where the serum IgG and TP concentrations were significantly higher (27.54±12.93 g/L and 6.44±1.21 g/dL, respectively) in calves fed colostrum included IgG ≥50 g/L from the calves fed colostrum included IgG <50 g/L (18.01±13.16 g/L and 5.35±1.65 g/dL, respectively).

This result corresponds to **Reschke *et al.* (2017)** that indicated that the colostrum of poor quality was the main risk factor associated with the failure of passive transfer of immunity. Consequently, 15.5% of the calves in their studies were in an unfavorable situation from the beginning because they received the colostrum of their dams containing IgG less than 50 g/L at first feeding. They indicated that this adverse effect can be compensated by other factors in the management of the calves (for example, good hygiene) or be aggravated by additional management deficits.

Quigley (2002) and **Osaka *et al.* (2014)** reported that the relationship between serum IgG and colostral IgG intake is linear and positive in most experiments, so the concentration of IgG in colostrum influences the apparent efficiency of IgG absorption. They concluded that large amounts of colostrum containing a low concentration of IgG would not be absorbed adequately; instead, limited amounts of high IgG colostrum may be more important. The ability of the intestine to extract Ig from colostrum may be improved when more concentrated (higher Ig) colostrum is fed.

Effect of time of first feeding:

The time lag between birth and first feeding plays an important role in the successful passive transfer of immunity in calves. The serum IgG and TP concentrations in calves were significantly associated with the timing of the first colostrum meal (after birth). Table 2 shows that the calves fed for the first time within 2 h after birth showed serum IgG and TP concentrations significantly higher (29.63±13.67 g/L and 6.55±1.02 g/dL, respectively) than calves fed at 2-6 h after birth (25.85±14.58 g/L and 6.31±1.85 g/dL, respectively). They were also higher than the calves fed after more than 6 h after birth (17.02±9.84 g/L and 5.34±1.27 g/dL, respectively).

In a study of **Reschke *et al.* (2017)** they reported that the high percentage of calves were diagnosed with failure of passive transfer of immunity (43.5%) because a large percentage of calves (49.1%) were fed for the first time after more than two hours after birth combined with a high percentage of calves (40.3%) that received less than 2 L of colostrum at first feeding. **Puppel *et al.* (2019)** indicated that it is so important to provide colostrum immediately after birth (0.5–1 h). The ability to absorb immunoglobulins from colostrum decreases by 1/3 as soon as 6 h after birth and by 2/3 after 12 h, and an intestinal barrier appears after 24 h. So, time constitutes a critical element for feeding calves colostrum. **Fidler *et al.* (2011)** noted that absorption of IgG is dependent on the quantity and quality of the colostrum administered and the time elapsed between birth and ingestion.

The time of first feeding is more properly classified as a loss of efficiency of absorption rather than a loss of IgG concentration per se. Where calves fed the same mass of IgG will be less efficient in absorbing that IgG if they are fed at a later age. Current theories suggest that intestinal epithelial cells lose their ability to absorb intact macromolecules after about 24 h because of maturation of the cells and development of the intracellular digestive apparatus, in

addition to the secretion of digestive enzymes (proteolytic) may also contribute to lower apparent efficiency of IgG absorption by degrading IgG before absorption. Also, the presence of bacteria and the establishment of microbial populations in the intestine with time after birth may increase the rate of intestinal closure, thereby reducing the apparent efficiency of IgG absorption and acquisition of passive immunity **(Quigley, 2002)**.

Effect of colostrum volume at first feeding:

There was a strong correlation between both the serum IgG and TP concentrations in calves and the volume of colostrum ingested at first feeding (Table 2). Calves that received more than 2 L of colostrum at first feeding after birth, the serum IgG and TP concentrations were significantly higher (29.41±13.31 g/L and 6.35±1.25 g/dL, respectively) than calves that received less than 2 L of colostrum (18.46±11.65 g/L and 5.77±1.63 g/dL, respectively).

In a study of **Reschke *et al.* (2017)** they reported that the high percentage of calves were diagnosed with failure of passive transfer of immunity (43.5%) because a high percentage of calves (40.3%) received less than 2 L of colostrum at first feeding after birth combined with a large percentage of calves (49.1%) fed for the first time after more than two hours. To achieve the successful passive transfer of immunity, **Puppel *et al.* (2019)** suggested that a calf needs to receive at least 150–200 g of Ig within 2 h of birth. This can normally be achieved by feeding 3 to 4 L of high-quality colostrum with Ig ≥50 g/L. **Quigley (2002)** indicated that the concentration of Ig in colostrum from the first milking may be inadequate to ensure the transfer of an adequate mass of Ig when ≤2 liter are fed. He cited a suggestion that the prevalence of failure of passive transfer in dairy herds could be minimized by artificially feeding calves large volumes (3 to 4 L) of colostrum within the first 24 h.

Effect of birth season:

There was an effect of the birth season on serum IgG and TP concentrations in calves (Table 2). Calves born during the summer season had the lowest serum IgG and TP levels (15.28±11.52 g/L and 5.21±1.48 g/dL, respectively), while the winter season was the best, where the calves recorded the highest levels (33.11±10.73 g/L and 6.91±0.69 g/dL, respectively). This may be due to the poor quality of the colostrum produced during the summer season compared to other seasons or due to the calves be exposed to heat stress during the summer season.

These results are in agreement with the findings of **Reschke *et al.* (2017)** who found the

highest prevalence of passive transfer failure in calves born during the spring and summer months (35.4 % – 43.8 %). These findings correspond to the hypothesis that high ambient temperature during late pregnancy is associated with poorer colostrum composition and therefore worse maternal immunity transfer to the calves **(Godden, 2008)**. In contrast, **Stanek *et al.* (2019)** found in Czech dairy herds that the significantly lowest level of serum IgG of calves born in winter (mean 10.3 g/L) compared to calves born in spring, summer and autumn (mean IgG level 14.8, 14.9 and 14.4 g/L, respectively). Also, **Gulliksen *et al.* (2008)** reported that Norwegian cows produce lower quality colostrum during the winter months (cold stress), perhaps explaining the increased mortality in the winter months. **Quigley (2002)** reported that the absorption of Ig may be affected by the environment in which the calf is born. Extreme cold but not moderate cold reduces the absorption of Ig by calves. The effects of ambient temperature outside the thermoneutral range for calves may involve direct effects on intestinal absorption and transport.

Effect of dam parity:

Table (2) shows that calves born to dams of the first parity have the lowest levels of serum IgG and TP (14.71±9.55 g/L and 5.47±1.67 g/dL, respectively) than calves born to dams at the second parity or more. This may be due to that heifers have a lower volume, concentration, and quality of colostrum than have mature cows. **Waldner and Rosengren (2009)** reported that low serum IgG concentrations have been associated with birth to a heifer, where the calving period is lengths for a heifer. This increases the risk of acidosis and the calves may be weak to consume adequate volumes of colostrum. In addition to acidotic calves may absorb immunoglobulins less efficiently.

Effect of calf gender:

The concentration of serum IgG and TP were higher in the female calves (25.61±13.15 g/L and 6.38±1.47 g/dL, respectively) than the male calves (23.08±14.41 g/L and 5.74±1.37 g/dL, respectively), but this difference was not significant (Table 2).

In a study by **Stanek *et al.* (2019)** the influence of sex of calves on the serum IgG level was evaluated in Czech dairy herds, this variable was not found to be statistically significant (P = 0.28), where the mean IgG level of 13.7 g/L was found in heifers and bulls. The same conclusion was reached by **Trotz-Williams *et al.* (2008)**, who found no statistically significant difference in passive transfer status in one day old or older bull and heifer calves

based on the serum TP levels examination. **Quigley (2002)** reported that heifer calves generally have higher serum IgG concentrations than do bull calves. He indicated that it is not clear whether the calf gender may be related more to blood volume than to apparent efficiency of IgG absorption, or the larger size of bull calves influence the metabolic state of the calves, thereby affecting Ig absorption.

Table (2): Factors affecting serum IgG (g/L) and total protein (g/dL) concentrations in Friesian calves at 24 h after colostrum feeding.

Variable	Serum IgG (g/L)	Serum total protein (g/dL)
Overall mean	**24.46±13.56**	**6.09±1.44**
Colostrum quality		
IgG ≥50 g/L	**27.54±12.93^{A}**	**6.44±1.21^{A}**
IgG <50 g/L	**18.01±13.16^{B}**	**5.35±1.65^{B}**
Time of first feeding		
<2 h	**29.63±13.67^{A}**	**6.55±1.02^{A}**
2–6 h	**25.85±14.58^{AB}**	**6.31±1.85^{AB}**
>6 h	**17.02±9.84^{B}**	**5.34±1.27^{B}**
Colostrum volume at first feeding		
≥2 L	**29.41±13.31^{A}**	**6.35±1.25**
<2 L	**18.46±11.65^{B}**	**5.77±1.63**
Birth season		
Winter	**33.11±10.73^{A}**	**6.91±0.69^{A}**
Spring	**25.83±11.07^{AB}**	**6.48±1.47^{AB}**
Summer	**15.28±11.52^{B}**	**5.21±1.48^{B}**
Autumn	**24.21±15.47^{AB}**	**5.86±1.66^{AB}**
Dam parity		
1st	**14.71±9.55^{B}**	**5.47±1.67^{B}**
2nd	**25.41±15.84^{A}**	**5.85±1.43^{AB}**
3rd	**30.56±10.93^{A}**	**6.68±1.11^{A}**
Calf gender		
Male	**23.08±14.41**	**5.74±1.37**
Female	**25.61±13.15**	**6.38±1.47**

$^{A,B:}$ Means in the same column within the same factor with different superscripts are significantly different (P <0.05). Data presented as Mean ± SD.

3.The relationshipbetween serumIgGandTP concentrations during the first week of age:

Traditionally, determination of successful transfer of passive immunity has been by measuring the concentration of IgG in the serum of the calf at 24 to 48 h after birth, if the serum IgG concentration exceeds some critical level, then the calf is thought to be relatively well protected against pathogens **(Quigley,2002)**. Our study aimed to monitor changes of IgG and TP concentrations in serum of calves from birth until 7 days of age to know the extent of the correlation between them and also to provide a basis for recommendations for when the passive transfer of immunity in calves can be measured. Concentrations of serum IgG and TP were measured before colostrum feeding, at 24 h after colostrum feeding and daily from day 2 to day 7 of age.

The concentrations of serum IgG in relation to the age of the calves showed no significant changes during the first week of age Fig. (1). Calves showed the highest values (peak) at 24 h after colostrum feeding (24.46±13.56 g/L). Thereafter, the values insignificantly decreased gradually until the end of the first week of age (19.24±9.05 g/L). This decrease was at a rate of 0.87±0.29 g/L per day.

Similarly, the concentrations of TP in relation to the age of the calves showed no significant changes during the first week of age Fig. (2). the lowest mean concentration was observed at birth (4.01± g/dL), which was followed by a significant increase one day after colostrum intake (6.09±1.44 g/dL; P <0.05). Thereafter, the values insignificantly decreased gradually until the end of the first week of age (5.37±1.03 g/dL; P >0.05). This decrease was at a rate of 0.12 ±0.11 g/dL per day.

These results were similar to those found by **Wilm *et al.* (2018)** where calves showed low values of serum TP and IgG concentrations before colostrum feeding and an initial peak at 24 h after colostrum feeding (5.83±0.73 g/dL and 22.2±9.6 g/L, respectively). The IgG concentrations then decreased over time relative to IgG measured at 24 h at a rate of 0.69±0.05 g/L per day. Also, **Tóthová *et al.*(2016)** found that the concentration of TP showed the highest mean at the age of one day after colostrum intake (8.52 g/dL). Thereafter, the values significantly decreased gradually until the end of the first month of age (6.71 g/dL; P <0.05). Furthermore, **Villarroel *et al.*(2013)** found that serum IgG and TP concentrations decreased over time by 0.74 g/L and 0.07 g/dL per day, respectively within the first week of age. **Hammon *et al.* (2002)** explained the high increase in the serum protein levels during the

first 24 h of life reflects the intestinal absorption of proteins (particularly immunoglobulins) from colostrum due to the enhanced intestinal permeability.

Fig.(3) shows the correlation coefficients between concentrations of serum IgG and also serum TP measured from day 2 to 7 and the value recorded for both of them after 24 h of colostrum feeding. Concentrations of serum IgG measured from day 2 to 7 were very highly correlated with the reference value recorded 24 h after colostrum feeding ($r \geq 0.905$; $P < 0.0001$). Also, concentrations of serum TP at 24 h after colostrum feeding were highly correlated with those recorded up to day 7 of age ($r \geq 0.765$; $P < 0.0001$). This indicates that all necessary measures must be taken within the first 24 h of age for the calf to obtain sufficient total proteins, including immunoglobulin. This agrees with **Wilm *et al.* (2018)** who reported that concentrations of IgG measured from days 2 to 10 were very highly correlated with the reference value recorded 24 h after colostrum feeding ($r \geq 0.98$). Correlations were also high between the reference (24 h) value for serum TP and values recorded through day 2 to 10 of age ($r \geq 0.76$).

Fig. (4) shows the measures of serum IgG and TP concentrations that were highly correlated ($r = 0.842$, $P < 0.0001$) during the first week of age. This finding agrees with that of **Villarroel *et al.* (2013)** who found that serum TP and IgG concentrations were moderately correlated, up to 30 days of age ($r = 0.67$), and they concluded that serum TP concentration up to one week of age could be used to evaluate FPT. Also, **Deelen *et al.* (2014) and Morrill *et al.* (2013)** found that serum TP was positively correlated with IgG ($r = 0.93$ and $r = 0.87$, respectively). This indicates that determination of serum TP concentration up to at least day 7 of the colostrum feeding can provide a reliable estimate of immunity passive transfer. Now, there are many tools (digital or optical refractometers) available through which it is possible to determine the concentration of serum TP in less than 15 seconds. The use of these tools has many advantages compared with measuring IgG using a RID assay that has a long incubation time (18 to 24 h) and expensive, also it needs a special laboratory for this analysis, all of which limit its on farm application. This proposal supports what was indicated by **Deelen *et al.* (2014)** and **Stanek *et al.* (2019)** to use serum TP estimation as an alternative method for rapid and simple monitoring of immunity passive transfer in calves.

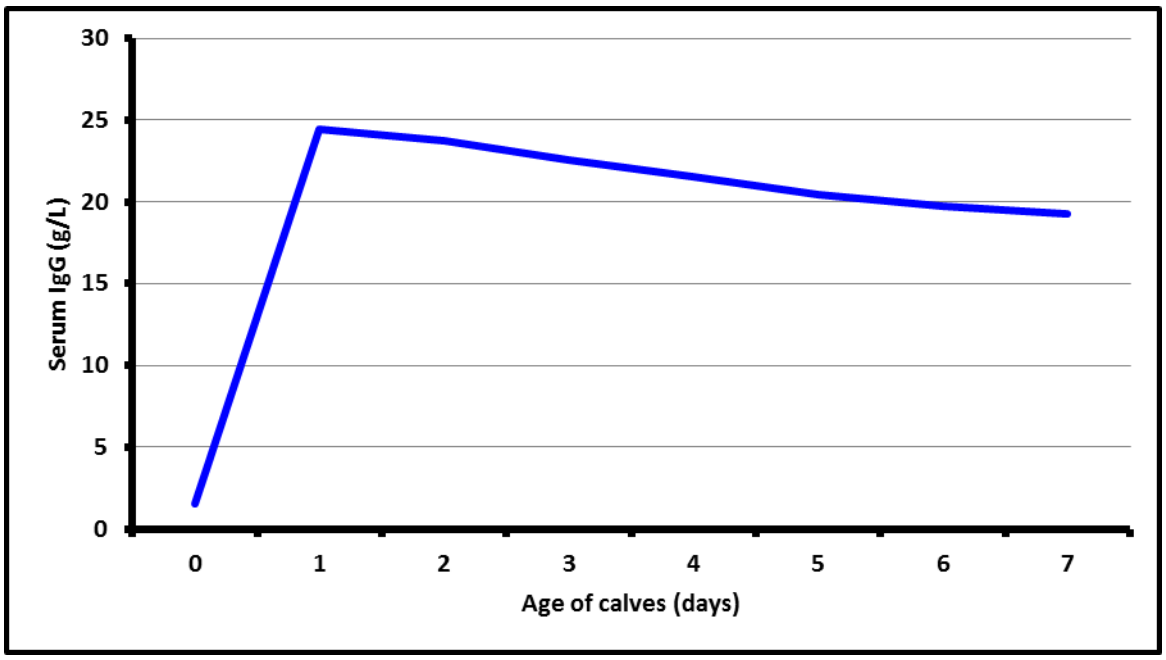

Fig. (1): The changes in serum IgG concentrations (g/L) in Friesian calves during the first week of age.

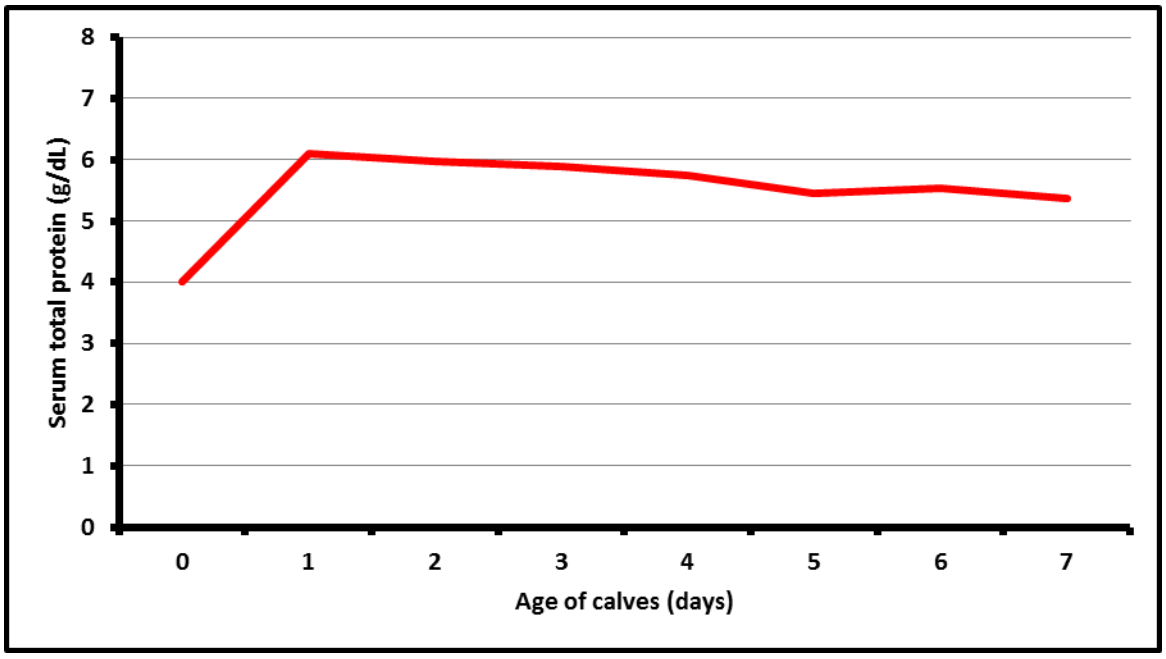

Fig. (2): The changes in serum total protein (TP) concentrations of (g/dL) in Friesian calves during the first week of age.

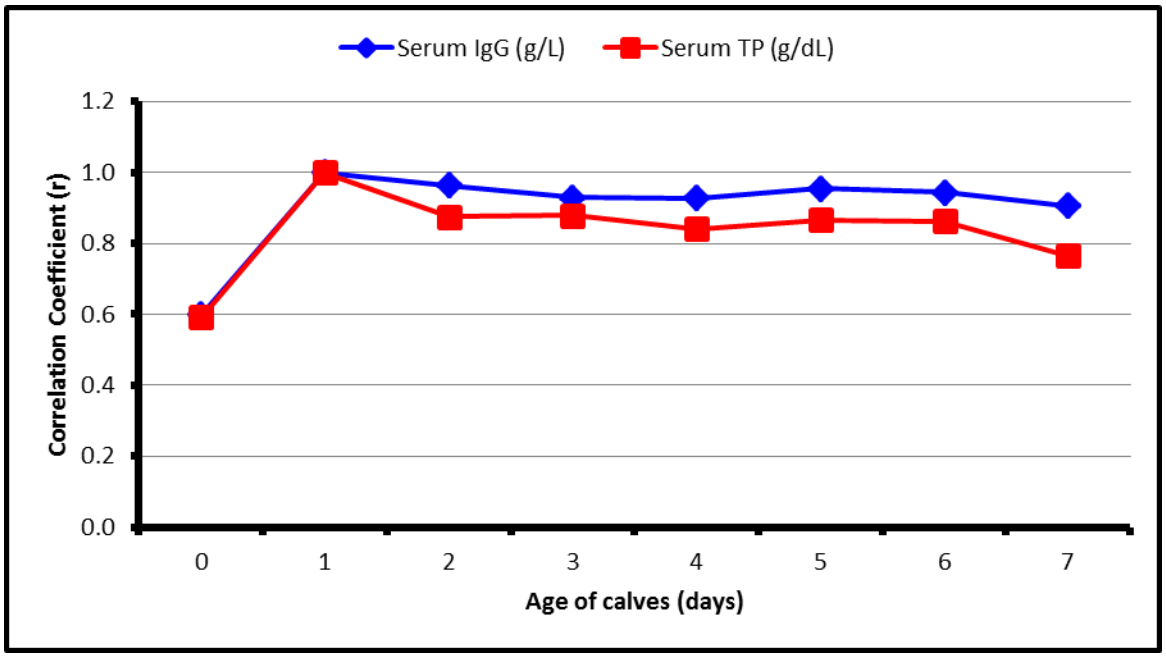

Fig. (3): Pearson correlation coefficients between values of either serum IgG (g/L) or serum TP (g/dL) concentrations measured 24 h after colostrum feeding and measured in subsequent days of Friesian calves.

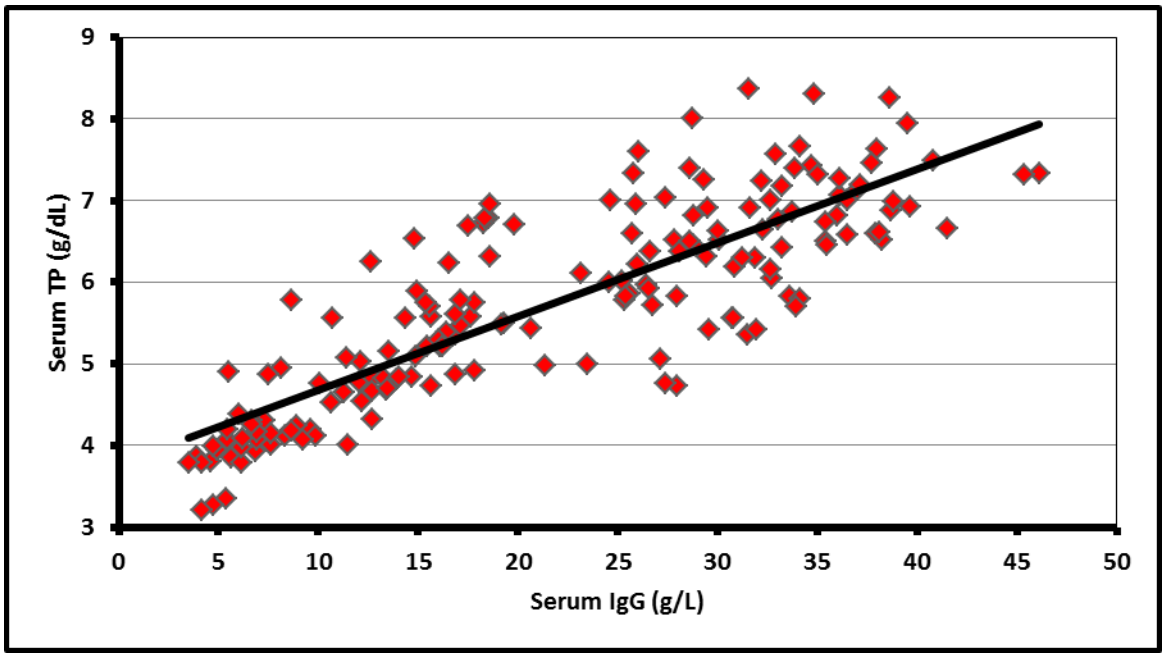

Fig. (4): Association between serum IgG (g/L) and TP (g/dL) concentrations in Friesian calves during the first week of age.

CONCLUSIONS

The results obtained from the study suggest that calving season, lactation number, and time of first milking postpartum are the factors that could be exploited in identifying colostrum quality used for feeding newborn calves, while colostrum volume of first milking does not serve as reliable indicator of the expected colostrum quality. Levels of serum IgG and TP in the calves were also significantly associated with the colostrum quality, time of first feeding, colostrum volume at first feeding, birth season, and dam parity. It would be desirable to further focus on management colostrum in dairy cattle.

Serum IgG concentrations of Friesian calves declined at a rate of approximately 0.7 g/L per day beginning from 24 h to 7 days of age, but values on all days were highly correlated with values taken 24 h after colostrum feeding. Also, serum IgG and TP concentrations was highly correlated ($r = 0.842$; $P < 0.0001$) during the first week of age. Thus, serum TP values during the first week of age can be used in the evaluation of passive transfer of immunity, as a predictor of calves' health, and as a monitoring program for colostrum management in farms.

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العوامل المُؤثرة على جودة السرسوب والمناعة السلبية لعجول الفريزيان الرضيعة تحت ظروف دلتا النيل ـ مصر

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الملخص

تم استخدام 102 بقرة فريزيان ونسلهم لتحديد العوامل المرتبطة بجودة السرسوب وعوامل الخطر المرتبطة بفشل نقل المناعة السلبية ولمراقبة التغيرات في تركيزات IgG والبروتين الكلي بسيرم الدم خلال الأسبوع الأول من العمر.تم تسجيل الوقت من الولادة حتى أول حلب ، موسم الأم ، تاريخ الولادة وجنس العجل. تم تحديد حجم وتركيب السرسوب ومستويات IgG به.جمعت عينات دم من كل عجل يومياً حتى عمر 7 أيام وتم قياس تركيزات IgG والبروتين الكلي بسيرم الدم.أعلي تركيزات تم تسجيلها في السرسوب للبروتينات الكلية والجوامد الصلبة الكلية ونسبة الأبقار التي يحتوي إنتاجها من السرسوب على IgG أعلي من 50جم / لتر كانت في فصل الشتاء ، وموسم الحلب الرابع أو أكثر وعندما تم حلب الأبقار خلال ساعتين بعد الولادة.لتقييم نقل المناعة السلبية للعجول ، تم قياس كل من IgG وتركيز البروتين الكلي في سيرم الدم بعد 24 ساعة من تغذية السرسوب. كانت نسبة العجول المتأثرة بفشل نقل المناعة السلبية 29.41٪. أعلي تركيزات تم تسجيلها من IgG والبروتين الكلي في سيرم عجول الفريزيان التي حصلت على أكثر من 2 لتر سرسوب والذي يحتوي على أكثر من 50 جم/لتر من IgG عند التغذية الأولى خلال ساعتين بعد الولادة ، وكذلك العجول المولودة أثناء فصل الشتاء أو أمهاتهم من الموسم الثاني أو أكثر.

لم تظهر تركيزات IgG والبروتين الكلي بسيرم الدم في العجول أي تغيرات معنوية خلال الأسبوع الأول من العمر. كما أظهرت العجول أعلى القيم عند 24 ساعة من تغذية السرسوب (24.46 جم / لتر و 6.09 جم / ديسيلتر ، على التوالي). بعد ذلك ، انخفضت القيم بشكل ضئيل تدريجيًا حتى نهاية الأسبوع الأول من العمر. كانت تركيزات كل من IgG والبروتين الكلي بسيرم الدم من اليوم 2 إلى 7 مرتبطة ارتباطًا وثيقًا بالقيمة المسجلة بعد 24 ساعة من تغذية السرسوب ($r \geq 0.905$ و $r \geq 0.765$ على التوالي). ارتبطت تركيزات IgG بتركيزات البروتين الكلي ارتباطًا وثيقًا ($r = 0.842$، $P<0.0001$) خلال الأسبوع الأول من العمر. هذا يشير إلى أن تحديد تركيز البروتين الكلي في سيرم الدم حتى اليوم السابع يمكن أن يوفر تقديرًا موثوقًا فيه لنقل المناعة السلبية ويعتبر طريقة بديلة للمراقبة السريعة والبسيطة لنقل المناعة السلبية في العجول.