



## Serum Total Protein Concentration as a Protective Factor against Pneumonia in Calves

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### Abstract

Bovine Respiratory Disease (BRD) is one of the most significant health and economic challenges during the pre-weaning period, contributing substantially to calf mortality, reduced growth, and impaired future performance. Calves are born without maternal immunoglobulins due to the epitheliochorial structure of the bovine placenta, making passive immune transfer via colostrum essential for early protection. Serum Total Protein (STP) concentration is widely used as a simple and cost-effective indicator to assess the success of passive immune transfer. This study aimed to investigate the association between STP and the incidence of pneumonia in pre-weaning Holstein calves. The study was conducted on 183 calves under 15 days of age on a dairy farm in Alborz province from August to October 2024. Blood samples were collected at 48 hours of age, and STP was measured using a handheld optical refractometer. Respiratory symptoms were monitored, and data were analyzed using the Cox proportional hazards model. Results showed that higher STP was significantly associated with a reduced risk of pneumonia ( $P = 0.049$ ). Each unit increase in STP corresponded to an approximately 39% reduction in disease risk. These findings highlight the importance of proper colostrum management and support the use of STP as a predictive tool for identifying high-risk calves. Monitoring STP can be an effective strategy to reduce BRD incidence and improve calf health and productivity.

**Keywords:** Calf pneumonia, Serum Total Protein, Passive immune transfer, Colostrum.

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## Introduction

Pneumonia is one of the most common infectious respiratory diseases in dairy cattle and is considered an economic problem, particularly in calf production (Asgharpour *et al.*, 2021). Bovine Respiratory Disease (BRD), as a multifactorial and prevalent disease in young dairy and beef calves, is one of the most significant issues in the livestock industry. In one of the latest studies on 400 pre-weaning calves from 32 dairy farms, the prevalence of respiratory pathogens was 84.4% at the farm level and 45.5% at the calf level, with *Pasteurella multocida* reported as the most common bacterial agent associated with the disease (Lee *et al.*, 2022). This disease typically arises from a combination of several pathogens (bacterial and sometimes viral or mycoplasmal) and is influenced by management and environmental factors. Commonly reported pathogens include *Mannheimia haemolytica*, *Mycoplasma bovis*, and *Histophilus somni* (Nobrega *et al.*, 2021; Hashem *et al.*, 2022; Lee *et al.*, 2022).

The consequences of this disease for the livestock industry are significant: beyond mortality and treatment costs, affected calves experience reduced growth, delayed reproduction, decreased future milk yield, and diminished economic returns (Lee *et al.*, 2022; Nobrega *et al.*, 2021). Recent epidemiological studies indicate that in addition to microbial factors, calf feeding and management methods—particularly the timing and quality of colostrum, type of feeding (milk, waste

milk), and farm conditions—play an important role in the incidence of BRD. For example, in the aforementioned Taiwanese study, delayed colostrum intake and feeding with waste milk were associated with an increased likelihood of carrying respiratory pathogens (Lee *et al.*, 2022).

Thus, effective prevention of calf pneumonia requires an understanding of the multifactorial nature of the disease, reducing stress and density on the farm, improving respiratory hygiene, implementing proper feeding management, and especially providing high-quality colostrum in a timely manner. These measures can significantly reduce the prevalence and complications of BRD.

Due to the impermeable nature of the bovine placenta to maternal immunoglobulins, newborn calves are born without immunoglobulins (agammaglobulinemia), making them completely dependent on colostrum immunoglobulins for protection against diseases (El-Sissi *et al.*, 2020). Inadequate colostrum intake leads to failure of passive transfer (FPT), subsequently increasing disease incidence and mortality in the pre-weaning period. Therefore, proper management to ensure sufficient colostrum intake is of utmost importance. Measuring serum total protein—as the most practical on-farm method—can provide assurance regarding successful passive immune transfer in calves and, consequently, the health status of newborn calves (Cortese

*et al.*, 2020; Choi *et al.*, 2021, Immler *et al.*, 2022).

The bovine placenta is of the epitheliochorial type, which, due to its six layers, prevents the passage of antibodies (Milandarzadeh and Azimpour, 2016). Because of this inhibitory characteristic, newborn calves are devoid of immunoglobulins (agammaglobulinemia), creating a state of complete dependence on colostrum immunoglobulins for protection against diseases (El-Sissi *et al.*, 2020). Inadequate colostrum intake results in failure of passive transfer, leading to increased disease incidence and mortality in the pre-weaning period. Hence, proper management to ensure adequate colostrum volume is critically important (Sadeghi Nasab, *et al.*, 2012). By measuring serum total protein—considered the most practical method at the farm level—successful passive immune transfer in calves and, consequently, the appropriate health status of newborn calves can be confirmed (Cortese *et al.*, 2020; Choi *et al.*, 2021).

In this study, serum total protein concentration was measured using refractometry, a simple, rapid, and low-cost method with minimal equipment requirements.

### Materials and methods

This study was conducted on 183 Holstein calves under 15 days of age on a dairy farm in Alborz province from August to October 2024. Of these, 100 were male and 83 were female. From all calves, a blood sample was collected

aseptically from the jugular vein 48 hours after colostrum feeding and placed in plain (without anticoagulant) centrifuge tubes. After transfer to the laboratory and clotting, the blood samples were immediately centrifuged at 4000 rpm for 5 minutes. To assess passive immune status, serum total protein was measured using a handheld optical refractometer, Kruss HRB90-T 0-90 model (manufactured by Kruss, Germany) (Image 1). All calves underwent clinical examination and were assessed for diarrhea symptoms.

### Statistical analysis

The Cox proportional hazards model in IBM SPSS Statistics (version 26) was used to evaluate the association between serum total protein concentration and pneumonia incidence. Results are presented as Hazard Ratio (HR) with a 95% Confidence Interval (CI). A P-value less than 0.05 was considered statistically significant (Fig. 1).



**Figure 1:** Handheld optical refractometer, Kruss HRB90-T 0-90 model, manufactured by Kruss, Germany.

## Results

Calves affected by pneumonia showed clinical signs of weakness and lethargy. The incidence of pneumonia was associated with serum total protein concentration ( $P=0.049$ ). The Hazard Ratio (HR) was 1.389 (95% CI: 1.002–1.926). This result indicates that for each 1 g/dL increase in total protein concentration, the relative risk of developing pneumonia decreases by 39%.

## Discussion

Current evidence suggests that calf serum total protein (STP) can serve as a useful indicator for evaluating the success of passive immune transfer (Passive Transfer of Immunity; TPI) and subsequently predicting the risk of pre-weaning diseases, including pneumonia (Raboisson *et al.*, 2016; Altvater-Hughes *et al.*, 2022; Sutter *et al.*, 2023). Various studies have defined reference values for STP; for example, values less than 5.2–5.5 g/dL are considered indicative of Failure of Passive Transfer (FPT) (Aghakhani *et al.*, 2023; Crannell and Abuelo., 2023).

In a cross-sectional study on surplus dairy calves, approximately 24% of samples had "poor" STP ( $<5.1$  g/dL) (Wilson *et al.*, 2023). Analytical studies have also reported a significant association between low STP and increased incidence of pneumonia and mortality. In one report that classified TPI status into four categories—'poor', 'fair', 'good', and 'excellent'—based on STP, calves with poor TPI (STP $<5.1$  g/dL) had approximately double the risk

(HR $\approx 2.00$ ) of developing pre-weaning pneumonia, along with increased overall morbidity and mortality (Crannell and Abuelo., 2023; Sutter *et al.*, 2023). Proper nutrition, growth, and development of calves are among the most prominent aspects of dairy herd management. Furthermore, the relationship between colostrum quality, feeding management, and STP in calves has been well-documented. For instance, a study on 551 cow-calf pairs showed that appropriate timing, sufficient quantity, and high quality of colostrum (e.g., high Brix) were strongly correlated with increased STP and serum immunoglobulin (IgG) in calves. Therefore, measuring STP with a refractometer—a simple and feasible on-farm method—is a practical approach for assessing the success of passive immune transfer (Poulsen *et al.*, 2010; Altvater-Hughes *et al.*, 2022).

However, it should be noted that the association between STP and disease or mortality is not necessarily linear or absolute. In some studies, no significant difference in the incidence of diarrhea or pneumonia was observed between calves with low and high STP (Villarroel *et al.*, 2013; Altvater-Hughes *et al.*, 2022). From a practical standpoint, to reduce the risk of pneumonia and other diseases in calves, colostrum management must be carefully implemented: prompt feeding after birth, provision of adequate volume and quality of colostrum, and the use of refractometers for monitoring TPI at the farm level. This approach can provide

adequate immune coverage for calves and contribute to a significant reduction in the incidence of BRD and other diseases.

Based on these findings, it is recommended that in herds with a high prevalence of respiratory disease, STP be used as a primary screening indicator to identify calves at risk. For calves with low STP, supportive measures (such as colostrum re-feeding, plasma therapy, or closer monitoring) should be considered.

### Conclusion

The results of this study indicate that serum total protein concentration can be a reliable indicator for evaluating passive immune transfer and predicting the risk of pneumonia in calves. The significant association between higher STP and reduced pneumonia incidence suggests that calves receiving sufficient, high-quality colostrum in the first hours of life benefit from better immune protection and are less susceptible to respiratory diseases.

Given the simplicity, speed, and low cost of measuring STP, the use of this indicator as a screening tool at the farm level is recommended. Monitoring STP can help identify at-risk calves and provide a basis for timely and effective interventions to reduce pneumonia incidence and improve calf health.

### Conflict of Interest

The authors declare no conflict of interest.

### Authors' Contributions:

Supervision: AA, AJ; Design: AJ, BB; Data collection: PB; Analysis: SHA, Drafting: AA, MMM; Revision: AJ; Funding: AJ, AA.

### Declarations:

This project was completed with personal funding

### Conflict of interest

The authors declare no conflict of interest.

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