

THE ROLES OF SELENIUM AND VITAMIN E IN HEMATOLOGICAL DYNAMICS OF TRANSPORTED BEEF CATTLE

N. Mayasari^{1*}, J.D. Ramadhan¹, M.R. Ismiraj²

¹Faculty of Animal Husbandry, Universitas Padjadjaran, Jatinangor, Indonesia

²Faculty of Animal Husbandry, PSDKU Pangandaran, Universitas Padjadjaran, Indonesia

Abstract

This study aimed to investigate the hematological changes in cattle subjected to transportation stress and the efficacy of selenium (Se) and vitamin E supplementation to mitigate these effects. Four treatments were evaluated: P1 (Control with a placebo solution), P2 (Injection of Se 5 ppm), P3 (Injection of vitamin E 36 ppm), and P4 (Injection of Se 5 ppm + vitamin E 36 ppm). Various blood parameters, such as leukocytes, red blood cells, platelets, and hemoglobin levels, were examined both pre- and post-transportation. Results indicated a significant alteration in these parameters due to transportation, underscoring the physiological stress experienced by the cattle. Among the treatments evaluated, the P2 treatment, containing Se and vitamin E supplementation, maintains relatively the same level of leukocyte although significant transportation effect. Cattle under this treatment demonstrated the most stable hematological profile, with values closely resembling pre-transportation levels. The protective roles of selenium and vitamin E, as evidenced in existing literature, are believed to contribute to this stability. These findings shed light on the potential of targeted nutritional interventions in safeguarding the health and welfare of transported cattle.

Key words: beef cattle, road transportation, blood profile, selenium, vitamin E

INTRODUCTION

The transportation of livestock, particularly beef cattle, is an essential component of the agricultural industry, facilitating the movement of animals from breeding sites to markets and processing facilities. While transportation is vital for the livestock industry, it often induces physiological and psychological stressors that can affect the health, welfare, and productivity of the animals. Elevated cortisol levels, alterations in blood parameters, and disruptions in immune function are among the physiological responses observed in cattle during and after transportation [1,2].

One area of focus has been the potential benefits of nutritional supplementation, specifically with selenium (Se) and vitamin E, to mitigate these negative impacts.

Studies have reported significant differences in white blood cell counts, hemoglobin levels, and other hematological parameters following Se and vitamin E supplementation [3]. Such supplements have been linked to immunostimulatory effects, enhancing the phagocytosis index and NK cell cytotoxicity, which are crucial for the animal's defense against infections [4]. Furthermore, the antioxidant properties of these supplements can support physiological processes under stressful conditions, thereby potentially improving the blood profile and overall health of transported cattle [5].

Beyond the immediate physiological benefits, the broader implications of transportation stress extend beyond the health of the animals. For instance, elevated stress levels can impact meat quality,

*Corresponding author: novi.mayasari@unpad.ac.id

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manifesting in issues such as increased carcass bruising and alterations in meat pH [6]. Additionally, stressed animals might be more susceptible to diseases, leading to increased veterinary costs and potential economic losses for producers [7].

Given the significance of these challenges, there is a pressing need for strategies and interventions to mitigate the adverse effects of transportation on cattle. While various treatments and management practices have been proposed, the efficacy of selenium and vitamin E supplementation in preserving cattle health and welfare during transit remains a topic of ongoing research and debate. This study aims to contribute to this critical area, evaluating the potential of selenium and vitamin E supplementation to alleviate transportation-induced hematological changes in cattle.

MATERIALS AND METHOD

Animals and Transportation Arrangement

This study utilized 28 Brahman Cross (BX) beef feeder cattle (23 ± 1 months old), each weighing approximately 450 ± 30 kg, with a coefficient of variation of 8.65%. These cattle were sourced from PT. Agrisatwa Jaya Kencana (Subang, West Java, Indonesia) and were transported to PT. Lintas Nusa Pratama (Tasikmalaya, West Java, Indonesia), a journey that spanned 6 hours on the mountainous road and route.

The transportation was facilitated using trucks equipped with an open-air ventilated box measuring 586 cm in length, 240 cm in width, and 230 cm in height (Mitsubishi Fuso Fighter X 270 PS, Krama Yudha Tiga Berlian Motors, Jakarta, Indonesia). The box's total volume was 32 m^3 , and it had a load weight capacity of 10 tons. This allowed for the transportation of up to 15 cattle, each weighing around 450 ± 30 kg.

Experimental Design

The study employed an experimental method, using a Completely Randomized Design (CRD) comprising four treatments,

each replicated seven times. The treatments were as follows:

P1 = Control (placebo solution)

P2 = Injection of Se 5 ppm

P3 = Injection of vitamin E 36 ppm

P4 = Injection of Se 5 ppm + vitamin E 36 ppm

The collected data was subjected to a variance analysis to discern the responses to the given treatments. The mathematical model employed was:

$$Y_{ij} = \mu + \tau_i + \epsilon_{ij}$$

Where:

Y_{ij} = represents the observation value from the j -th replication receiving the i -th treatment.

μ = denotes the general mean

τ_i = signifies the effect of the i -th treatment.

ϵ_{ij} = corresponds to the error effect arising in the j -th replication receiving the i -th treatment.

i = indicates the number of treatments (1,2,3,4).

j = represents the number of replications (1,2,3,4,5,6,7).

Mineral and Vitamin Preparation and Administration

The preparation phase involved the procurement of the Se mineral and vitamin E. Following this, the Se mineral and vitamin E were weighed using an analytical balance according to the calculation for each treatment. The Se mineral was then mixed with physiological NaCl, while the vitamin E was dissolved in glycerol (food grade). A Se mineral and vitamin E combination was prepared using physiological NaCl and glycerol (food grade). This mixture was then transferred into syringes, with each syringe containing a 3 ml dose intended for each cattle.

The treatment phase encompassed the administration of Se mineral and vitamin E injections in predetermined doses. This was performed at the cattle crush facility of PT. Agrisatwa Jaya (Subang, West Java, Indonesia). Prior to transportation, cattle were injected with Se mineral and vitamin E

in the dorsal region. Subsequently, their body weight was measured using a scale with a capacity of 1500 kg and a precision of 500 grams. Blood samples were also drawn at this stage (pre-transportation). Following this, the cattle were transported to PT. Lintas Nusa Pratama (Tasikmalaya, West Java, Indonesia) using livestock trucks. Upon arrival at PT. Lintas Nusa Pratama, the cattle were re-weighed, and additional blood samples (post-transportation) were taken at the cattle crush facility.

Data Collection and Processing

Data were collected from Brahman Cross subjects subjected to four different treatments; each treatment was replicated seven times. The dataset consisted of several measured parameters, such as Hemoglobin concentration, Erythrocyte Count, and Leukocyte Count, Packed Cell Volume, Platelets, Basophils, Eosinophils, Staphylococci, Segmented Neutrophils, Lymphocytes, and Monocytes. Each of these measurements was recorded both before and after transportation. Using SAS's PROC DATA and PROC SORT steps, the dataset was organized and filtered to ensure each treatment had an equal number of replications for paired analyses.

Data Visualization

For each measured parameter, box plots were generated using PROC SGPLOT to visually compare the distributions across different treatments, both before and after transportation. All plots were rendered in grayscale for clarity and consistency. Each box plot was annotated with significance indicators derived from the respective statistical tests.

Statistical Analyses

To determine the effects of transportation, paired t-tests were conducted comparing the measurements of each parameter before and after transportation for each treatment. This was achieved using PROC TTEST with the PAIRED statement. Significance was established at $p < 0.05$.

To evaluate the impact of the treatments on the measured parameters, one-way ANOVAs were performed using PROC GLM. These analyses were conducted for data before transportation, after transportation, and for the delta values (difference between post- and pre-transportation values). The CLASS statement was used to specify the treatment groups, and the MODEL statement was used to define the dependent variable and its relation to the treatment. A significance threshold of $p < 0.05$ was maintained.

For a comprehensive understanding of treatment effects, the least squares mean (LSM) for each parameter across treatments was calculated using PROC GLM with the LSMEANS statement, where the treatment was the main effect in the model. All data handling, visualizations, and statistical analyses were conducted using SAS Statistics ver. 9.4.

RESULTS

This study sought to elucidate the nuanced impacts of supplementation of Se and Vitamin E treatments and transportation on select hematological indices, employing the method of least square mean evaluations (Table 1).

Treatment-Induced Hematological Responses

The hemoglobin (gr/dl), treatments P1 through P4 yielded means of 14.225, 13.421, 13.421, and 13.8, respectively. Intriguingly, these treatments did not confer significant alterations in hemoglobin concentrations across transportation phases, with p-values of 0.625 and 0.506 before and after, respectively.

In terms of Erythrocyte Count (million/mm³), the treatments showcased means spanning 8.491, 8.508, 8.249, to 8.563. Across the board, these treatment-induced variations remained consistent and statistically non-significant through transportation phases.

Leukocyte Count (/mm³) offered a more varied landscape. The treatments P1 to P4 elicited responses ranging from 12216.67, 10578.57, 11507.14, to 11025. While post-

transportation figures retained statistical homogeneity ($p = 0.559$), pre-transportation data intimated potential differentials, albeit not statistically significant ($p = 0.158$).

Table 1 Effects of supplementation of Selenium and Vitamin E on transported beef cattle

Treatment	Least Square Means						p-values		
	P1	P2	P3	P4	Before Transport	After Transport	Treatment before transportation	Treatment After transportation	Transportation
Hemoglobin (gr/dl)	14.23	13.42	13.42	13.80	13.7	13.7	0.63	0.51	0.42
Erythrocyte Count (million/mm ³)	8.49	8.51	8.25	8.56	8.5	8.5	0.71	0.66	0.40
Leukocyte Count (/mm ³)	12217	10579	11507	11025	9807 ^a	12896 ^b	0.16	0.56	0.00
Packed Cell Volume (%)	50.17	47.29	46.00	48.58	47.9	48.0	0.45	0.44	0.46
Platelets (/mm ³)	1,128,417	1,382,929	1,248,643	1,119,083	1,262,703 ^b	1,196,296 ^a	0.37	0.82	0.02
Eosinophils (%)	7.08	6.36	6.29	6.75	7.1	6.2	0.37	0.17	0.86
Staphylococci (%)	2.50	2.43	2.43	2.50	2.3	2.7	0.87	0.99	0.17
Segmented Neutrophils (%)	48.92	42.93	45.50	46.25	37.4 ^a	54.0 ^b	0.09	0.80	0.00
Lymphocytes (%)	32.58	40.29	37.64	36.83	44.7 ^b	29.3 ^a	0.01	0.70	0.00
Monocytes (%)	8.92	8.00	8.64	7.67	8.6	8.1	0.31	0.25	1.00
Neutrophils to Lymphocytes Ratio	1.65	1.23	1.44	1.37	0.9 ^a	2.0 ^b	0.03	0.80	0.00

^{a,b}Least Square Mean (LSM) values with different superscripts in the same row are significantly different ($P < 0.05$)

While these parameters displayed differential sensitivities to treatments, a unifying theme was the resilient nature of many hematological indices, which predominantly remained impervious to external interventions.

Transportation Effects

Transportation, a notable external variable in our study, unveiled its distinct hematological imprints. For instance, Hemoglobin (gr/dl) subtly transitioned from a mean of 13.715 before transportation to 13.674 after. This modest shift, however, evaded statistical significance ($p = 0.418$).

In regard to Erythrocyte Count (million/mm³), the figures rested at 8.476 pre-transportation, adjusting slightly to 8.455 post-event. This minor alteration remained statistically insubstantial ($p = 0.400$).

Conversely, Leukocyte Count (/mm³) painted a more dynamic picture, marking a robust transition from 9,807 pre-transportation to a heightened 12,896 post-transportation. This pronounced alteration spotlighted the transportation's profound influence and clinched statistical significance with a p-value of 0.001 (Figure 1).

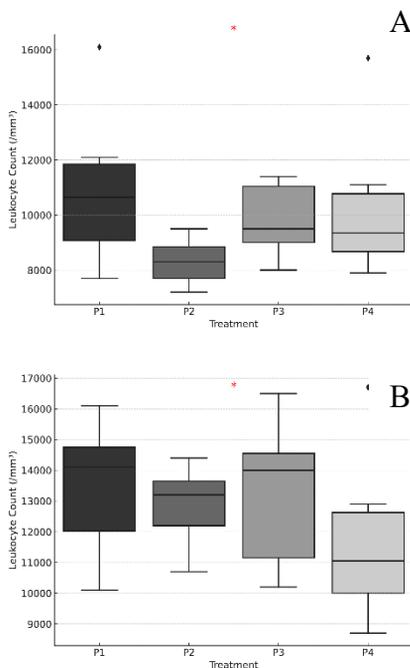


Figure 1. Leukocyte count of transported cattle across all treatments ($p < 0.01$). A: Before transportation; B: after transportation. P1 = Control (placebo solution); P2 = Injection of Se 5 ppm; P3 = Injection of vitamin E 36 ppm; P4 = Injection of Se 5 ppm + vitamin E 36 ppm



DISCUSSION

The physiological effects of transportation on cattle have been a focal point of numerous studies, consistently underscoring the myriad of hematological changes that can be detrimental to the health and welfare of the animals. Transportation has been associated with a pronounced increase in cortisol levels [1], which can influence various hematological parameters, including blood glucose levels and overall immune function.

A deeper exploration of the literature reveals that these stress-induced changes extend beyond just hematological parameters. The onset of transportation can lead to alterations in circulating steroid concentrations, with a classical acute increase in plasma cortisol observed [1]. This, in tandem with a decrease in dehydroepiandrosterone, can profoundly elevate the cortisol: dehydroepiandrosterone ratio, signaling a significant physiological stress response. Additionally, transportation has been found to induce changes in red blood cell count, likely due to dehydration or stress-induced alterations in blood flow [8].

Our research primarily aimed to assess the efficacy of various treatments in mitigating these transportation-induced changes. Notably, P2 emerged as a particularly effective treatment, especially for key parameters such as leukocyte count and the neutrophils to lymphocytes ratio. The importance of maintaining stable leukocyte levels, given their pivotal role in the immune system, cannot be overstated [2,9]. Furthermore, the neutrophils to lymphocytes ratio has been established as an important stress indicator in cattle [10].

The intricacies of transportation stress in cattle are further highlighted by studies indicating that transport can influence triiodothyronine (T3) and thyroxine (T4) concentrations, with variations observed during different transportation phases [10]. Additionally, the stress of transportation can lead to alterations in the balance of blood

electrolytes, potentially disrupting muscle function and overall fluid balance [6].

It is also worth noting that transportation, irrespective of the treatments, can lead to increased levels of creatine kinase in plasma, indicating muscle damage [1]. Such findings emphasize the multifaceted challenges of ensuring cattle welfare during transit.

While our study provides valuable insights, it is essential to recognize that cattle transportation's physiological challenges are intertwined with psychological stressors. Literature suggests that traditional management practices, potentially more stressful, can lead to animals having compromised immune systems [11]. This underscores the importance of holistic practices that consider both the physiological and psychological well-being of the animals.

In the context of supplementation, selenium and vitamin E have emerged as potential agents to combat the adverse effects of transportation on cattle. Studies have shown that these supplements can influence white blood cell counts, hemoglobin levels, and other hematological indicators [3]. For instance, selenium and vitamin E supplementation has been linked to enhancements in phagocytosis index and NK cell cytotoxicity, suggesting an improvement in the animal's immune response [4]. These findings align with our observations, indicating that treatments incorporating these supplements might be particularly effective.

Furthermore, the antioxidant properties of selenium and vitamin E can play a pivotal role in supporting physiological processes under the stress of transportation. Research indicates that these supplements can help maintain electrolyte balance, prevent anemia, and even bolster the immune system's ability to fend off blood parasites [5]. Such multifaceted benefits underscore the potential of selenium and vitamin E in preserving the health and welfare of transported cattle.

With antioxidant properties, Vitamin E, a significant fat-soluble vitamin, counteracts the detrimental effects of free radicals from reactive oxygen species [12]. As a component of the antioxidant enzyme glutathione peroxidase, selenium is essential in breaking down reactive oxygen species like H₂O₂ and lipid hydroperoxides [13]. After experiencing post-birth stress, dairy cows see an improvement in their resistance to oxidation with vitamin E supplementation [14]. Furthermore, when vitamin E and selenium are administered via muscle injection, there was an observed enhancement in the activity of the antioxidant indicator erythrocyte glutathione peroxidase in high-producing dairy cows [15].

CONCLUSION

Our research underscores the multifaceted impact of transportation on cattle hematology, emphasizing the pronounced changes that occur in various blood parameters. The P2 treatment, as highlighted in our findings, offers a promising avenue to maintain hematological stability during transportation, potentially driven by the benefits of ingredients like selenium and vitamin E. These supplements, with their established roles in enhancing immune responses and mitigating oxidative stress, might be integral to preserving cattle health during transportation. While our study provides valuable insights, there remains a scope for future research to delve deeper into the specific mechanisms through which treatments like P2 exert their effects. Additionally, investigating the influence of factors such as cattle breed, age, and individual health statuses could offer a more holistic understanding of treatment efficacy. Ultimately, continued research in this domain is vital to ensure the optimal welfare and health of transported cattle.

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