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Research Article

Economic Feasibility and Growth Performance of Holstein Friesian Calves Fed Whey-Dangke Fortified Green Calf Starter

Zyahrul Ramadan, Renny Fatmyah Utamy, Hasbi Hasbi, Ambo Ako, Fatma Maruddin, Viterah Niode, Alif Rahmadi and Indarwati Bua Putri

Department of Animal Production, Faculty of Animal Science, Hasanuddin University, Makassar, South Sulawesi, Indonesia

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Corresponding Author:
Renny Fatmyah Utamy
Department of Animal Production,
Faculty of Animal Science,
Hasanuddin University, Makassar,
South Sulawesi, Indonesia
Email:
rennyfatmyahutamy@unhas.ac.id

Abstract: This research evaluated the impact of feeding green calf starter fortified with whey-dangke on Holstein Friesian dairy calves' economic feasibility, growth performance, and health parameters. Experimental diets were applied to 18 Holstein Friesian dairy calves using a randomized complete factorial design with two factors: type of calf starter and level fortification of whey-dangke. The results showed that calves fed different types of calf starter, the level of whey-dangke fortification, and the interaction between the two factors had no significant effect ($p>0.05$) on physiological, hematological, and blood biochemical. However, the alanine aminotransferase (ALT) parameter had an interaction; the ALT values in the green calf starter treatment fortified with 5% whey-dangke exhibited a highly significant difference ($p<0.01$) compared to the green calf starter treatment fortified with 2.5% whey-dangke, as well as the conventional green calf starter fortified with both 2.5 and 5% whey-dangke. The level of whey-dangke fortified significantly affected ADG, FCR, and feed cost per unit of gain ($p<0.05$). The use of whey-dangke was able to increase ADG and decrease FCR and feed cost per unit of gain. In addition, physiological, hematological, and blood biochemical profile values of Holstein Friesian Friesian dairy calves remained within the normal range when fed whey-dangke-fortified green calf starter.

Keywords: Blood Biochemical, Green Calf Starter, Hematological, Holstein Friesian Dairy Calves, Physiological, Whey-Dangke

Introduction

In Indonesia, many dairy cows belong to the Holstein Friesian breed, known for their adaptability to the local environment (Ginantika *et al.*, 2021). Their performance as calves influences the milk production performance of lactating Holstein Friesian cows. Good calf performance leads to the production of superior replacement stock. From birth to 3 months old, calves' nutritional needs are met with 60% milk and 40% starter feed (calf starter) (Maharani *et al.*, 2015). The 2-month-old phase is critical as calves transition from consuming milk to solid feed/calf starters to maintain their performance (Parsons *et al.*, 2021). Feeding high-quality, low-fiber calf starter stimulates the development of the calf's reticulo-rumen (Maharani *et al.*, 2015). Good quality calf starter should contain Crude Protein (CP) at 16-20%, Crude Fat (CF) at 3%, Total Digestible Nutrient (TDN) at 80%, Calcium (Ca) at 0.6% and Phosphorus (P) at 0.4% (National Research Council, 2001). While commercial calf starters typically offer high nutrition and digestibility (Khan *et al.*, 2011), they are often expensive, not cost-effective, and not readily available. Additionally, commonly used commercial and conventional calf starters contain fishmeal as a protein source, which can reduce their palatability. Making it imperative to develop alternative calf starters using easily accessible ingredients that provide proper nutrition, particularly in terms of protein sources and high palatability.

An alternative calf starter that shows promise is green concentrate (green calf starter). This concentrate primarily comprises leguminous plants, offering high-quality forage and great palatability (Ako *et al.*, 2023).

Indigofera (*Indigofera zollingeriana*) containing CP 30.5%, Crude Fiber (Cf) 19.0% (Gunun *et al.*, 2022), and gliricidia (*Gliricidia sepium*) containing CP 24.22%, Cf 38.2% (Soleh *et al.*, 2022). Fortification can enhance the nutritional quality and palatability of both conventional and green calf starters. Fortification involves adding specific substances/ingredients to the calf starter to improve its quality. Whey-dangke is a suitable material for fortification, as its high lactose content can enhance the consumption and digestibility of calf feed (Gallardo-Escamilla *et al.*, 2007).

Dangke is a processed product made from solidified milk using papain enzymes to resemble a local cheese (soft cheese) typical of Enrekang Regency. The processing of dangke results in a by-product known as whey-dangke, which contains 55% of the nutrients found in milk, including lactose, protein, fat, vitamins, and minerals (Meale *et al.*, 2017). Research has shown that mixing whey-dangke into calf starter feed does not significantly impact calf growth performance (Parsons *et al.*, 2021). Fortified whey in milk replacer formulations demonstrated comparable value to commercial milk replacers (Syaikhullah *et al.*, 2021). This suggests that whey has great potential as a protein source for dairy calves. By utilizing whey, producers can lower feed costs by relying on this by-product as an alternative protein source. By using green concentrate and whey-dangke fortification, it is possible to reduce reliance on commercial concentrates without any adverse effects on dairy calves. A study evaluation of whey-dangke fortified green calf starter on Holstein Friesian calves' health parameters. This technology can potentially lower feed production costs and increase farmers' profits.

Materials and Methods

Research Site

This study was conducted from January to April 2024 in Lebang Village, Cendana Subdistrict, Enrekang District, South Sulawesi Province, Indonesia. A hematological test was conducted at Dika Sidrap Clinic, Sidrap District, South Sulawesi Province, Indonesia. Blood biochemical analysis was conducted at the Makassar Health Laboratory Center, South Sulawesi Province, Indonesia.

Research Procedures

Producing green calf starter involves harvesting indigofera and gliricida leaves, which are dried using a dehydrator for 7 h at 70°C. The dried leaves are then ground into a meal using a disc mill. Other feed ingredients such as bran, coconut kernel cake meal, ground corn, fish meal, and molasses are also prepared. Feed ingredients for conventional and green calf starters are formulated based on the protein and TDN adequacy for at least 2 starter calves (Indonesian National Standard, 2024). The green concentrate is produced by mixing indigofera and gliricida meal with other feed ingredients. On the other hand, conventional calf starters are made without adding indigofera and gliricida meals, following the typical feed fed to calves by farmers. For detailed composition and nutritional content of the calf starter, refer to Table (1).

Table 1: Composition and nutritional value of calf starter before fortified whey-dangke

Feedstuff	Composition (DM %)	Nutritive Value (%)						
		CP	CF	TDN	Cf	Ca	P	Ash
Conventional Calf Starter (CS0)								
Rice Bran	36	4.64	4.10	25.22	4.68	0.03	0.08	0.77
Coconut Kernel Cake Meal	29	6.61	3.36	25.51	4.56	0.06	0.20	1.45
Corn Meal	27	2.40	0.59	21.60	1.05	0.01	0.09	1.60
Fishmeal	7	3.35	0.10	5.39	0.57	0.18	0.11	1.40
Molasses	1	0.04	0.08	0.81	0.00	0.01	0.00	0.04
Total	100	17.05	8.24	78,52	10.86	0.27	0.48	5.27
Green Calf Starter (CS1)								
Indigofera Meal	10	2.83	0.96	7.00	0.64	0.12	0.03	0.64
Gliricidia Meal	12	3.09	1.23	8.06	0.66	0.24	0.03	2.03
Rice Bran	37	4.77	4.22	25.92	4.81	0.03	0.08	0.80
Coconut Kernel Cake Meal	20	4.56	2.32	17.59	3.15	0.04	0.14	1.00
Corn Meal	20	1.78	0.44	16.00	0.78	0.00	0.07	1.19
Molasses	1	0.04	0.08	0.81	0.00	0.01	0.00	0.04
Total	100	17.08	9.25	75.38	10.04	0.44	0.34	5.69

CP = Crude Protein; CF = Crude Fat; TDN = Total Digestible Nutrient; Cf = Crude Fiber; Ca = Calcium; P = Phospor

A formulated calf starter was administered to eighteen Holstein Friesian dairy calves, aged 2-3 months and weighing between 80 and 120 kg. Due to the limited availability of calves that meet these criteria simultaneously, it was challenging for us to obtain more dairy cows under the same conditions. Consequently, we conducted only three repetitions of the treatment combination. Three replicates are statistically considered strong enough to show the mean value of the treatment and its level of significance. Although a larger sample size can reduce the standard error of the data. To enhance the accuracy of the data collected, duplicate blood samples were analyzed.

The calves were fed an experimental diet comprising 2.5% of their body weight, 50% calf starter concentrate, and 50% forage. The calf starter concentrates, including both conventional and green varieties, were enriched with whey-dangke as a feed supplement at varying levels. The feed given to the calves had a liquid texture to facilitate the consumption of the calf starter concentrate. The study spanned 50 days, including a 7-day animal adaptation period. The research employed a complete randomized design with 2 factors: type of calf starter concentrates (conventional calf starter (CS0) and green calf starter (CS1)) and level of whey-dangke fortification (0% (L0), 2.5% (L1) and 5% (L2) of the calf's body weight). The following details of the whey-dangke's nutritional value are presented in Table (2).

Table 2: Nutritive of whey-dangke

Nutritive	Value
Proximate analysis (%)	
Water	91.71
Crude Protein	29.9
Crude Fat	3.42
Crude Fiber	0
NFE	54.59
Ash	12.09
TDN*	77.4
Essential amino acids (mg/g)**	
Histidine	17.68
Isoleucine	45.65
Leucine	97.25
Lysine	83.98
Methionine	25.89
Phenylalanine	25.32
Threonine	48.89
Tryptophan	20.45
Valine	45.64

*NFE = Nitrogen-free extract; TDN = Total digestible nutrient; *was calculate according Moran (2005); **Yasmin et al.(2013)*

Physiological parameters were monitored weekly to evaluate calf health. On day 50, blood samples were collected to assess hematological and blood biochemistry. Each blood sample, totaling 3 ml, was drawn from the jugular vein using a project needle and project holder and divided into plain vacutainer tubes for blood biochemistry and EDTA vacutainer tubes for hematological analysis. For blood biochemistry testing, the blood serum was obtained by allowing the blood sample to stand for 30 min, followed by centrifugation at 3000 rpm for 10 min to separate the serum. The serum was then transferred to microtubes and stored in cooler boxes for transportation to the laboratory for analysis. The procedure of this study is shown in Figure (1).

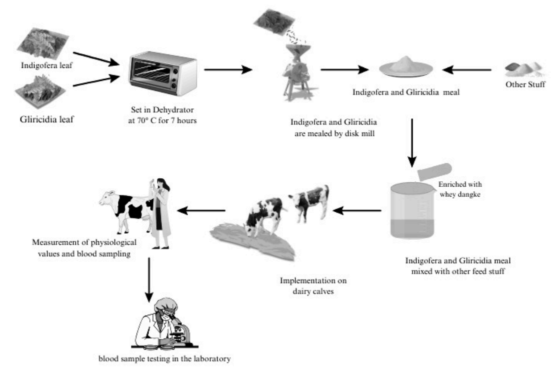

Fig. 1: The procedure of this study

Parameters

The study measured environmental parameters such as temperature, humidity, and the Temperature-Humidity Index (THI). The THI was calculated using the temperature and humidity measurements. The microclimate conditions of the cage were assessed three times a day: In the morning (6:00 am), midday (12:00 pm), and late afternoon (5:00 pm). The THI provides insight into the comfort level of the dairy cows in their environment and was calculated using the formula developed by Thompson and Dahl (2012):

$$THI = (1.8 \times Ta + 32) - [(0.55 - 0.0055 \times RH) \times (1.8 \times Ta - 26)]$$

Ta = Ambient Temperature (°C) and RH = Relative Humidity (%).

Body temperature was calculated based on McLean *et al.* (1983) as follows:

$$Tb = 0.86\,Tr + 0.14\,Ts$$

Tb = Temperature of the body; Tr = Temperature of the rectum; Ts = Temperature of the skin

According to Suherman *et al.* (2017), the temperature of the skin surface (Ts) was measured at four measurement locations, i.e., the back (A), chest (B), upper limbs (C), and lower limbs (D). Mean skin surface temperature was calculated based on the formula of McLean *et al.* (1983):

$$Ts = 0.25\,(A + B) + 0.32C + 0.18D$$

The rectal (Tr) temperature is measured by inserting a rectal thermometer into the cow's rectum as deep as ± 10 cm until the thermometer sounds. Measurement of physiological values was carried out 3 times a day, i.e., in the morning (06.00 am), midday (12.00 pm), and late afternoon (05.00 pm).

Observation of the respiratory rate involved monitoring the symmetrical movement of the flank and ribs during inspiration. Heart rate measurements were taken using a stethoscope placed on the left axilla for one minute, with three replications of each measurement using a stopwatch. Hematological parameters were measured using a Prokan model PE-6100 hematology analyzer in China to assess Red Blood Cell (RBC), White Blood Cell (WBC), hematocrit values, and hemoglobin levels. Blood biochemistry testing, which included glucose, urea, cholesterol, ALT, and AST, was carried out using the spectrophotometric method with the Thermo Scientific Indiko clinical chemistry blood equipment brand made in Finland.

The growth parameters of calves analyzed included: (1) Average Daily Gain (ADG), calculated using the equation (final body weight - initial body weight (kg)) / experimental period (days); (2) Feed consumption; and 3) Feed Conversion Ratio (FCR), which is determined by dividing the total feed consumption by the ADG. Additionally, a cost-economic benefit analysis was conducted with dangke whey fortification, which encompassed the following: (1) Feed input costs, expressed in dollars per dry matter kilogram (dollars/DM kg) for conventional calf starters, were determined based on prevailing market prices for concentrates. For green concentrates, the calculation relied on the market price of feed raw materials. Although farmers cultivate forage, there are still associated costs in the cultivation process, such as purchasing fertilizer and transporting the forage from the pasture to the cattle house. Similarly, while dangke whey may be readily available, there are still preparation costs to consider. (2) Daily feed costs (dollars/day/head) were calculated using the equation: feed price (dollars/DM kg) multiplied by the amount of feed consumed (DM kg/day/head). (3) Feed cost per unit of gain was computed as feed cost per unit (dollars/DM kg) multiplied by feed per unit gain (DM kg/1 kg ADG).

Statistical Analysis

Research data were Analyzed Using Variance Analysis (ANOVA) using the General Linear Model-Univariate (descriptive statistics and homogeneity tests) with a significance level of 5% using SPSS software version 25. Additionally, a normality analysis was performed to ascertain whether the data followed a normal distribution. Only data that met the normal distribution criteria were subjected to ANOVA analysis and in the event of a significant effect, the Duncan multiple range test would be applied.

Results

Microclimate Conditions at the Research Site

The microclimate conditions within the cage are an external factor that can significantly impact dairy cows' production performance and physiological well-being. According to the data presented in Table (2), it is evident that the microclimate within the cage at the research site is notably affected by the time of measurement. Table (3) shows a highly significant difference (p<0.01) in ambient temperature, relative humidity, and THI measurements. The ambient temperature is notably higher in midday compared to the morning and late afternoon and it's also significantly higher in midday than in the morning. Additionally, the morning relative humidity is significantly higher than in the midday and late afternoon, while the midday relative humidity is significantly higher than in the late afternoon. Moreover, the THI value is significantly higher in the midday and late afternoon than in the morning.

Effect of Whey-Dangke Fortified Green Calf Starter on Holstein Friesian Calves' Health Parameters

The study on the impact of whey-dangke fortified green calf starter on the health parameters of Holstein Friesian calves found that the type of calf starter (conventional versus green), the level of whey-dangke fortification and their interactions did not have a significant effect (p>0.05) on physiological measures (body temperature, respiratory rate and heart rate), hematologic values (RBC, WBC, hemoglobin and hematocrit), or blood biochemical levels such as glucose, urea, AST, cholesterol and ALT. However, there was an observed interaction between the type of concentrate and the level of whey-dangke fortification concerning the ALT level. Notably, the ALT value in the group receiving the green calf starter fortified with 5% whey-dangke showed a highly significant result (p<0.05), exceeding the values observed in both the green calf starter fortified with 2.5% whey-dangke and the conventional calf starter fortified with 2.5 and 5% whey-dangke.

Table 3: Average temperature, humidity, and THI at the research site

Parameters	Time factor of measurement			*p*-value
	Morning	Midday	Late Afternoon	
Ambient Temperature (°C)	26.14±0.49^{a}	33.20±1.27^{c}	30.26±1.67^{b}	0
Relative Humidity (%)	87.57±2.43^{c}	68.00±5.85^{a}	81.42±6.62^{b}	0
THI	77.67±0.68^{a}	85.79±1.84^{b}	84.27±1.75^{b}	0
Environmental comfort categories*	Mild Stress	Moderate Stress	Moderate Stress	-

^{abc}*Different superscripts in the same row indicate significant differences (P<0.01); THI= Temperature Humidity Index; *Wiersma (1990)*

Effect of Whey-Dangke Fortified Green Calf Starter on Holstein Friesian Calves' Calf Growth Parameters and Cost-Economic Benefit

The study examining the influence of whey-dangke fortified green calf starter on the growth parameters and cost-economic benefit of Holstein Friesian calves revealed that the level of whey-dangke fortified significantly affected ADG, FCR, and feed cost per unit of gain (p<0.05). However, neither the type of calf starter (conventional versus green) nor the interaction between the type of calf starter and the level of whey-dangke fortified had a significant impact (p>0.05) on ADG, feed consumption, FCR, daily feed cost, or feed cost per unit of gain.

Discussion

Microclimate Conditions at the Research Site

An increase in the microclimate value of the cage can lead to heat stress in livestock. Heat stress occurs when livestock cannot maintain their normal body condition due to increased environmental temperature (Giannone *et al.*, 2023). Heat stress can also lead to decreased production performance as livestock use energy to adapt to environmental changes rather than for production purposes (Zeng *et al.*, 2023). At the research location, the microclimate conditions of the cage showed a fairly high value, with a temperature range of 26.14–33.20˚C and humidity of 68.00–87.57%. This resulted in a range of THI from 77.67–85.79, indicating that the calves experienced mild to moderate stress (Table 3). The THI measures livestock's comfort level in an environment influenced by temperature and humidity (Sukandi *et al.*, 2023). The microclimate conditions in Indonesia are unsuitable for Holstein Friesian dairy cows, which are accustomed to a sub-tropical climate. According to Asmarasari *et al.* (2023), Holstein Friesian calves perform best at a temperature of 13–25˚C with 50%–60% humidity.

This study demonstrates that the highest ambient temperature occurs at midday due to peak solar radiation (Giannone *et al.*, 2023). As the day progresses, the temperature gradually rises until it reaches its zenith when the sun is directly overhead, then decreases in the late afternoon. Concurrently, humidity levels decrease throughout the day, indicating an inverse relationship with ambient temperature (Sukandi *et al.*, 2023). It is worth noting that humidity plays a pivotal role in determining the comfort of dairy cows. The most critical heat stress conditions arise when ambient temperature and humidity are high (Song *et al.*, 2023). High THI values can induce heat stress in dairy cows, potentially reducing feed consumption and consequently impacting the growth performance of dairy calves. This diverts energy that should be utilized for growth towards heat regulation, triggering physiological and metabolic responses, such as increased blood insulin and protein catabolism (Wang *et al.*, 2020).

The microclimate conditions within the cages, particularly the THI, play a crucial role in influencing physiological values and blood profiles. Measuring these microclimate conditions ensured that the data from this study reflected the effects of the treatment rather than environmental factors. Elevated ambient temperatures and humidity levels can significantly impact health parameters. Sukandi *et al.* (2023) demonstrated variations in hematological status across different microclimate conditions. An increase in THI, coupled with higher body heat in livestock from feed intake, can lead to severe heat stress, potentially resulting in mortality among calves. In response to this stress, livestock tend to reduce their feed consumption.

Effect of Whey-Dangke Fortified Green Calf Starter on Holstein Friesian Calves' Health Parameters

Dairy cows are able to maintain their body condition despite environmental changes, including feed variation. When they encounter heat stress, calves utilize homeostatic mechanisms to counter the effects of external heat and increased internal body temperature. This process leads to physiological adjustments aimed at removing excess heat generated by feed metabolism. Indicators such as body temperature, heart rate, and respiratory rate can signal the presence of heat stress in dairy cows. Feeding high-energy feed can contribute to the calorigenic effect, increasing body heat production due to energy metabolism and consequently causing heat stress (Utamy *et al.*, 2024). Elevated body temperature can cause heat stress in dairy cows, resulting in health complications and reduced performance, which can be monitored through physiological parameters. In response to heat stress, dairy cows may adopt various strategies, such as reducing feed intake and increasing water consumption. In addition, heat stress can adversely affect feed efficiency, resulting in significant costs for farmers (Chen *et al.*, 2024). Research by Ako *et al.* (2023) reported that green concentrate contains high protein and energy. In addition, dangke whey contains high enough energy to potentially cause a feed calorigenic effect that will increase body temperature. Body temperature serves as a key indicator to assess the impact of the caloric effect of feed. Increased body temperature is an early warning sign of heat stress associated with feed nutrition, leading to increased metabolic heat production in dairy cows (Suherman *et al.*, 2013). This study found that body temperature ranged from 38.26 to 38.97ºC (Table 4). This is in line with the findings of Saputra *et al.* (2022), who noted that dairy calves receiving protein supplements had body temperatures ranging from 38 to 39ºC. The normal body temperature indicated that feeding fortified dangke whey to green calf starter did not cause feed calorigenic effects for FH dairy calves (Utamy *et al.*, 2024).

In this study, respiration rates were observed to range from 51.21 to 60.58 breaths per minute (Table 4). This is in line with the findings of Bohlen and Rollin (2018), who noted that the common respiration frequency in healthy dairy cows ranges from 30 to 60 breaths per minute. High respiration frequency is often associated with high protein and energy intake, which affects the calorigenic effect of feed because the metabolic process of these nutrients requires oxygen and will produce heat, the release of heat can be done through breathing frequency. This increase in energy and protein in the feed increases oxygen demand, which must be met by a corresponding increase in respiration frequency (Syaikhullah *et al.*, 2021). Anggoro *et al.* (2023) found that cattle fed feed containing 14% CP showed a higher respiration frequency compared to cattle fed 12% CP.

In this study, heart rate values were in the normal range of 68.88-75.55 beats per minute (Table 4). It is noted that the heart rate of dairy calves is generally about twice that of lactating cows, with a normal range of 55-115 beats per minute (Tanuwiria *et al.*, 2023). Heart rate is influenced by the livestock's comfort with their environment. When livestock experience stress due to environmental changes or nutritional deficiencies, their heart rate will increase. In addition, an increase in heart rate can be caused by disease disorders that can be caused by microorganisms or metabolic diseases including nutritional deficiencies (Clapp *et al.*, 2015). Specifically, physiological values of dairy calves fed conventional calf starter and those fed green calf starter fortified with whey-dangke showed no significant differences. These findings suggest that green calf starter can be a viable replacement for conventional calf starter.

Table 4: Physiological of Holstein Friesian dairy calves

Parameter	Type of calf starter	Level fortified whey-*dangke* (% of body weight)			Average	p-value		
		L0	L1	L2		CS	L	CS*L
Temperature of Body (°C)	CS0	38.97±0.53	38.50±0.44	38.28±0.30	38.59 ±0.48	0.97	0.64	0.07
	CS1	38.10±0.04	38.87±0.19	38.78±0.16	38.58±0.38			
	Average	38.54±0.54	38.68±0.37	38.52±0.33				
Respiratory Rate (breath/min)	CS0	58.70±3.34	57.66±1.00	54.55±5.87	56.97±3.89	0.73	0.77	0.23
	CS1	51.21±3.59	55.96±5.72	60.58±12.83	55.92±8.23			
	Average	54.95±5.14	56.81±3.78	57.57±9.41				
Heart Rate (beats/min)	CS0	74.03±13.02	68.88±6.37	73.70±9.29	72.28±8.34	0.69	0.6	0.98
	CS1	74.88±5.85	71.22±9.71	75.55±6.55	73.88±6.35			
	Average	74.46±9.04	70.05±7.46	74.62±7.26				

CS Type of calf starter; L = Level fortified whey-dangke (% of body weight); CS0= conventional calf starter; CS1= and green calf starter; L0 = 0% Fortified Whey-dangke; L1 = 2.5% Fortified Whey-dangke; and L2 = 5% Fortified Whey-dangke of fresh weight of body weight

Table 5: Hematological of Holstein Friesian dairy calves

Parameter	Type of calf starter	Level fortified whey-*dangke* (% of body weight)			Average	p-value		
		L0	L1	L2		CS	L	CS*L
Red Blood Cells ($1\times10^6/\mu l$)	CS0	5.43±1.65	5.82±0.16	5.07±1.01	5.44±0.96	0.85	0.74	0.94
	CS1	5.36±1.31	5.47±0.95	5.18±1.29	5.34±1.04			
	Average	5.40±1.33	5.64±0.64	5.12±1.04				
White Blood Cells ($1\times10^3/\mu l$)	CS0	7.43±0.86	7.83±1.30	6.66±0.55	7.31±0.91	0.34	0.57	0.52
	CS1	6.73±0.55	9.43±4.14	9.30±4.22	8.48±3.25			
	Average	7.08±0.75	8.63±2.88	7.98±3.05				
Hemoglobin (mg/dl)	CS0	8.43±0.83	8.63±1.15	8.30±0.69	8.45±0.76	0.29	0.82	0.96
	CS1	8.73±0.76	9.00±0.60	8.83±0.40	8.85±0.53			
	Average	8.58±0.73	8.81±0.84	8.56±0.85				
Hematocrit (%)	CS0	26.06±3.61	26.26±2.84	26.50±2.81	26.27±2.54	0.17	0.71	0.78
	CS1	28.53±0.05	27.13±1.40	30.13±6.10	28.63±3.39			
	Average	27.30±2.65	26.70±2.06	28.31±4.69				

CS = Type of calf starter; L = Level fortified whey-dangke (% of body weight); CS0 = conventional calf starter; CS1 = and green calf starter; L0 = 0% Fortified Whey-dangke; L1 = 2.5% Fortified Whey-dangke; and L2 = 5% Fortified Whey-dangke of fresh weight of body weight

Dairy calves' health and nutrition can be assessed by examining their blood parameters, including RBC, WBC, hemoglobin levels, and hematocrit values, which should ideally remain stable or within normal ranges (Raguati and Rahmatang, 2012). The values for RBC, WBC, hemoglobin and hematocrit in our study (Table 5), align with the findings of Ježek *et al.* (2011), who investigated hematological parameters in dairy calves and reported RBC levels of $4.83–11.33\times10^6/\mu L$, WBC levels of $6.00–15.64\times10^3/\mu L$ and hemoglobin levels of 6.91–12.82 mg/dL. Similarly, Cabral *et al.* (2015) reported 20.0–45.0% hematocrit values. The RBCs are crucial in nutrient transport throughout the body due to their unique structure and composition (Pretini *et al.*, 2019). The membrane of platelets is made up of proteins that bind to lipids. Therefore, the quantity of platelets and WBC is influenced by the availability of dietary proteins (Kutlu *et al.*, 2020).

The assessment of feed quality involves considering various blood cell components such as hemoglobin and hematocrit values, in addition to RBC and WBC. Hemoglobin, a molecule of symmetrical pairs of polypeptide chains, is primarily responsible for transporting oxygen from the lungs to mammalian body tissues (Byers *et al.*, 1952). The blood hemoglobin value of dairy cows is often used to indicate their nutritional status, particularly concerning protein and mineral consumption (Vabushana *et al.*, 2021). Meanwhile, hematocrit represents the ratio of RBC to the entire blood volume (RBC: Plasma). Research has indicated that the increase in hematocrit value is influenced by the protein content of the feed, with supplementary feed containing indigofera and gliricidia showing better results compared to forage feed in the form of grass and straw alone, which is commonly provided by farmers (Raguati and Rahmatang, 2012).

Urea and glucose in the blood result from the feed absorption process. This study showed urea and glucose in the blood levels (Table 6), indicating the absorption process of the feed. A study has revealed that an increase in the fortification of whey in green calf starter, compared to conventional calf starter, led to elevated levels of urea and glucose in the blood (Anin *et al.*, 2022). Blood urea indicates protein utilization efficiency and energy adequacy in feed, as it is a byproduct of protein metabolism. Therefore, higher blood urea levels suggest more efficient protein intake from the feed. According to research by Wichman *et al.* (2022), calves typically exhibit blood urea levels ranging from 6–12 mg/dL. On the other hand, blood glucose, which serves as an energy source for dairy cows, is derived from feed and is mainly obtained from energy-rich feed (Anin *et al.*, 2022). The concentration of blood glucose levels in dairy calves, as indicated by a study conducted by Roadknight *et al.* (2021), ranges from 50–124 mg/dL. While not statistically significant, the blood glucose levels of Holstein Friesian dairy calves fed green concentrate tend to be higher than those fed conventional calf starters. This is attributed to cellulose in green calf starter, derived from forage, which serves as an energy source and contributes to increased blood glucose levels (Santamaría-Fernández & Lübeck, 2020). Furthermore, the fortification level of whey-dangke positively correlates with higher blood glucose levels, as whey contains lactose that can be converted into glucose (Gallardo-Escamilla *et al.*, 2007).

The findings indicate that green calf starters and conventional calf starters have similar effects on blood cholesterol levels, regardless of fortification levels (Table 6). However, green calf starters appear to result in relatively lower blood cholesterol levels compared to conventional calf starters. On the other hand, fortifying with whey-dangke tends to increase blood cholesterol levels. Cholesterol is a lipid substance found in the blood and bile fluids, serving as a precursor for the biosynthesis of steroid hormones. Blood cholesterol levels are affected by DMI and Cf, which provide cholesterol precursors such as Acetyl CoA from glucose and the catabolism of fatty acids and amino acids in mitochondria (Hartati *et al.* 2023). Blood cholesterol levels range from 83.60-160.20 mg/dL (Das *et al.*, 2019).

Table 6: Blood biochemical profile and liver function of Holstein Friesian dairy calves

Parameter	Type of calf starter	Level fortified whey-*dangke* (% of body weight)			Average	p-value		
		L0	L1	L2		CS	L	CS*L
Urea (mg/dl)	CS0	9.50±2.50	10.00±1.73	11.00±2.00	10.16±1.93	0.41	0.28	0.84
	CS1	9.50±0.50	11.33±3.15	12.33±2.08	11.05±2.40			
	Average	9.50±1.61	10.66±2.58	11.66±1.96				
Glucose (mg/dl)	CS0	103.66±16.25	104.00±1.00	114.50±5.50	107.38±10.11	0.41	0.3	0.85
	CS1	104.00±21.51	114.00±19.31	125.00±26.00	114.33±21.46			
	Average	103.83±17.05	109.00±13.40	119.75±17.76				
Cholesterol (mg/dl)	CS0	141.66±42.52	149.00±4.00	151.50±18.50	147.38±23.69	0.72	0.61	0.92
	CS1	130.66±24.98	144.33±34.26	153.00±27.87	142.66±27.18			
	Average	136.16±31.77	146.66±21.96	152.25±21.17				
AST (U/L)	CS0	49.50±1.50	46.00±7.00	44.00±6.24	46.5±5.02	0.08	0.87	0.73
	CS1	52.33±9.45	53.00±11.53	53.50±3.50	52.94±7.67			
	Average	50.91±6.24	49.50±9.35	48.75±6.89				
ALT (U/L)	CS0	19.00±3.60^{ab}	17.00±2.00^{a}	15.00±5.00^{a}	17.00±3.46	0.36	0.19	0.03
	CS1	17.66±2.51^{ab}	14.00±1.00^{a}	24.33±6.03^{b}	18.66±5.61			
	Average	18.33±5.00	15.50±2.16	19.66±7.11				

CS = Type of calf starter; L = Level fortified whey-dangke (% of body weight); CS0 = conventional calf starter; CS1= and green calf starter; L0 = 0% Fortified Whey-dangke; L1 = 2.5% Fortified Whey-dangke; and L2 = 5% Fortified Whey-dangke of fresh weight of body weight. AST = ; ALT = . The different superscripts in the same row and column of the same parameter indicate a highly significant difference (p<0.05)

The nutritional needs of dairy cows are closely tied to liver performance. When dairy cows have insufficient nutrient intake, it increases the liver's workload in energy metabolism. This leads to the mobilization of energy from fat reserves to energy metabolized in the liver. Continuous energy mobilization causes fat accumulation in the liver, leading to ketosis, which can reduce liver performance (Gershuni *et al.*, 2018). One way to assess liver health and performance is by measuring AST and ALT levels. These parameters can also be used to evaluate the effects of feed toxicity. A study by Yu *et al.* (2019) found that the AST value of dairy calves ranged from 16.0–77.0 IU/l, while ALT ranged from 11–40 IU/l. The study also showed differences in ALT values between treatment combinations, indicating an interaction between calf starter type and whey-dangke fortification level (Table 6). This feeding regimen did not have any adverse effects on the calves.

The highest ALT value was observed in the combination of green concentrate with 5% dangke whey fortification (Table 6). In green calf starters, an increase in ALT value corresponded with a higher level of whey-dangke fortification. Conversely, conventional calves exhibited a decrease in ALT levels. It appears that whey-dangke fortification can enhance the digestibility of green calf starters. Whey dangke is rich in lactic acid bacteria, which can help reduce crude fiber in feed; this reduction, in turn, increases the digestibility of feed nutrients (Liu *et al.*, 2022) and helps normalize the AST value. Conventional calf starters tend to contain higher corn starch levels than green calf starters. Excessive energy levels from corn starch can bind with lactose, forming more complex compounds that impair the digestibility of feed nutrients (Davis, 2015).

Effect of Whey-Dangke Fortified Green Calf Starter on Holstein Friesian Calves' Calf Growth Parameters and Cost-Economic Benefit

The effective performance of dairy calves is crucial for their development into high-quality replacement stock capable of substantial milk production. The growth performance of calves, particularly their ADG, significantly influences their milk production potential during lactation periods. Ideally, dairy calves should attain an average ADG of 0.5-1 kg per day, allowing them to reach a minimum body weight of 300 kg by 15-18 months of age (SNI 2735: 2014). In this study, the incorporation of whey-dangke fortification resulted in an increased ADG in Holstein Friesian dairy calves (Table 7). This enhancement can be attributed to the ability of whey-dangke to balance essential nutrients such as energy, protein, and minerals, thereby optimizing growth performance (Araújo *et al.*, 2020). Moreover, whey-dangke serves as a natural source of Lactic Acid Bacteria (LAB), which can decrease the fiber content in calf starters and enhance feed digestibility (Liu *et al.*, 2022). Additionally, the amino acid composition of whey-dangke can act as a precursor for the synthesis of growth hormones, including Growth Hormone (GH) and Insulin-like Growth Factor 1 (IGF-1). Both of these growth hormones are peptide hormones, with GH consisting of 191 amino acids and IGF-1 comprising 70 amino acids. A notable essential amino acid found in whey-dangke is leucine, which is present at a concentration of 97.25 mg/g (Table 2). Leucine is crucial for the formation of GH and IGF-1 (Lu *et al.*, 2019).

The growth performance of Holstein Friesian dairy calves is significantly influenced by feed consumption and FCR. Research indicates that the use of green calf starter and whey-dangke fortified leads to increased feed consumption, primarily due to the high palatability of green concentrate (Ako *et al.*, 2023). Similarly, whey-dangke is also highly palatable and is a cost-effective option (Sutera *et al.*, 2023). FCR serves as an important indicator for assessing feed quality; a lower FCR signifies more efficient feed utilization. The composition and digestibility of feed are critical factors that determine FCR. Higher feed quality enhances the efficiency of converting feed into 1 kg of ADG (Ahn *et al.*, 2021). Furthermore, the fortification of feed with whey-dangke can improve digestibility and overall feed quality, thus contributing to lower FCR values.

According to Table (7), utilizing green calf starter along with whey-dangke fortified can significantly decrease daily feed expenses. This reduction is largely due to the green calf starter being formulated from locally sourced raw materials that are relatively affordable, including some by-products from agricultural processing. In addition to lowering daily feed costs, whey-dangke fortified contributes to a reduction in feed cost per unit of weight gain. This improvement is connected to the enhanced feed quality and a lower Feed Conversion Ratio (FCR) provided by whey-dangke, ultimately resulting in reduced feed costs per unit of weight gain. High-quality feed can boost farmers' income by increasing efficiency and achieving higher ADG (Erdem and Ağır, 2024).

Table 7: Calf growth parameters and cost-economic benefit Holstein Friesian calves

Parameter	Type of calf starter	Level fortified whey-*dangke* (% of body weight)			Average	p-value		
		L0	L1	L2		CS	L	CS*L
ADG (Kg/day/head)	CS0	0.28±0.14	0.58±0.06	0.64±0.18	0.50±0.20	0.59	0.03	0.41
	CS1	0.47±0.08	0.54±0.26	0.62±0.11	0.54±0.16			
	Average	0.37±.0.14[a]	0.56±0.17[ab]	0.63±0.14[b]				
Feed consumption (Kg DM/day/head)	CS0	2.18±0.76	2.63±1.12	2.19±0.05	2.33±0.71	0.67	0.84	0.75
	CS1	2.32±0.14	2.44±1.42	2.77±0.85	2.51±0.85			
	Average	2.25±0.49	2.54±1.14	2.48±0.62				
FCR	CS0	8.23±1.37	4.75±1.54	3.63±1.24	5.54±2.39	0.38	0.03	0.14
	CS1	5.35±3.03	4.38±0.44	4.78±1.05	4.81±1.67			
	Average	6.79±2.62[b]	4.57±1.03[a]	4.21±1.20[a]				
Feed input (dollar/DM kg)								
Concentrate	CS0	0.43	0.43	0.43	-	-	-	-
		CS1	0.3	0.3	0.3	-	-	- -
Forage		0.15	0.15	0.15	-	-	-	-
Whey-Dangke		0.01	0.01	0.01	-	-	-	-
Daily feed cost (dollars/day/head)	CS0	0.60±0.26	0.73±0.34	0.61±0.08	0.65±0.23	0.48	0.74	0.66
	CS1	0.51±0.03	0.55±0.30	0.66±0.23	0.57±0.20			
	Average	0.55±0.17	0.66±0.31	0.63±0.15				
Feed cost per unit of gain (dollars/ kg DM/kg ADG)	CS0	2.19±0.16	1.43±0.52	1.10±0.54	1.57±0.61	0.11	0.01	0.18
	CS1	1.53±0.33	1.07±0.18	1.25±0.23	1.28±0.29			
	Average	1.86±0.43[b]	1.25±0.40[a]	1.18±0.38[a]				

CS = Type of calf starter; L = Level fortified whey-dangke (% of body weight); CS0 conventional calf starter; CS1 = and green calf starter; L0 = 0% Fortified Whey-dangke; L1 = 2.5% Fortified Whey-dangke; and L2 = 5% Fortified Whey-dangke of fresh weight of body weight. ADG = average daily gain; FCR = feed conversion ratio. The different superscripts in the same row and column of the same parameter indicate a highly significant difference (p<0.05)

Incorporating whey-dangke fortified into the diets of Holstein Friesian dairy calves offers significant advantages for livestock and positively influences the welfare of farmers, particularly those operating small-scale farms. As a by-product, whey-dangke is readily available at little to no cost. Despite its by-product status, it retains 55% of the nutritional value found in milk (Meale *et al.*, 2017). Research indicates that feeding whey-dangke can enhance growth rates, achieving performance comparable to that of commercial feed (Parsons *et al.*, 2022). Furthermore, Hartadi *et al.* (2019) noted that integrating whey into animal feed can lead to increased daily weight gain, improved palatability, and enhanced feed efficiency. Therefore, the inclusion of fortified whey in dairy cows' diets not only enhances animal performance but also improves the economic well-being of farmers by significantly reducing feed costs.

The use of Fed Whey-Dangke Fortified Green Calf Starter can significantly reduce feed costs for farmers. This product is much cheaper than commercial calf starters, with a price of only $0.30 per kg of Dry Matter (DM), compared to up to $0.62 per kg for commercial options. This cost-effectiveness indicates that using Fed Whey-Dangke Fortified Green Calf Starter is beneficial for farmers. Additionally, Fed Whey-Dangke Fortified Green Calf Starter enhances livestock productivity, leading to improved Average Daily Gain (ADG) without a significant increase in production costs. In fact, it tends to offer lower production costs than conventional calf starters, which typically have a lower ADG. These advantages suggest that Fed Whey-Dangke Fortified Green Calf Starter has strong potential for adoption among farmers. Given its comparative benefits over both conventional and commercial calf starters, farmers are likely to prefer it as their choice for calf nutrition.

Conclusion

Whey-dangke fortified green calf starter has the potential to serve as a viable alternative to conventional calf starters. The use of whey-dangke was able to increase ADG and decrease FCR and feed cost per unit of gain. In addition, physiological, hematological, and blood biochemical profile values of Holstein Friesian dairy calves remained within the normal range when fed whey-dangke-fortified green calf starter. This feeding regimen did not have any adverse effects on the calves. As a result, green calf starter with whey-dangke fortification has the potential to serve as a viable alternative to conventional calf starters.

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Author's Contributions

Zyahrul Ramadan, Renny Fatmyah Utamy, and Hasbi Hasbi: Conceived and designed the experiments, performed the field experiments, analyzed data, and wrote the paper.

Ambo Ako and Fatma Maruddin: Conceived and designed the experiments and performed analyzed the data.

Viterah Niode, Alif Rahmadi, and Indarwati Bua Putri: Performed the field experiments and analyzed data.

Ethics

Research ethics have been granted by the Animal Ethics Commission of the Faculty of Veterinary Medicine, Udayana University, with Number B/104/UN14.2.9/PT.01.04/2024.

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