



## Effects of mixing a high-fat extruded pellet with a dairy calf starter on performance, feed intake, and digestibility

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### ABSTRACT

During weaning, withdrawal of milk replacer is not directly compensated for by an increase in solid feed intake. Therefore, greater fat inclusion in the starter might mitigate this temporary dietary energy decline. However, fat inclusion in solid feeds may generally limit rumen fermentability and development. To address these potentially conflicting outcomes, we conducted 2 experiments to evaluate the effect of supplementing a high-fat extruded pellet mixed with a calf starter on feed intake, performance, and nutrient digestibility in calves. In experiment 1, 60 Holstein bull calves were blocked by serum IgG ( $2,449 \pm 176$  mg/dL) and date of arrival ( $2.5 \pm 0.5$  d of age). Within each block, calves were randomly assigned to 1 of 3 treatments: a standard control calf starter (CON; 3.1% fat) and mixtures of CON with 10% inclusion of 1 of 2 different high-fat extruded pellets containing 85% of either hydrogenated free palm fatty acids (PFA, 7.1% fat) or hydrogenated rapeseed triglycerides (RFT, 6.7% fat). Calves were offered milk replacer up to 920 g/d until 42 d of age, followed by a gradual weaning period of 7 d. Calves had ad libitum access to the starter diets, straw, and water. No differences were observed between CON, PFA, and RFT calves on body weight (BW) or average daily gain (ADG) until 49 d of age. From weaning (50 d) until 112 d, PFA calves had a greater BW and ADG than RFT and CON animals. Moreover, PFA calves had the highest intakes of starter, straw, calculated metabolizable energy, and crude protein after weaning. Overall, no differences were present in blood  $\beta$ -hydroxybutyrate and glucose concentrations between treatments; however, calves in the RFT treatment had a higher concentration of insulin-like growth factor-1. In experiment 2, 24 Holstein bull calves at 3 mo of age were assigned to 1 of 8 blocks based on arrival BW and

age. Within each block, calves were randomly assigned to 1 of the 3 treatments previously described for experiment 1. Calves on the RFT treatment had the lowest total-tract apparent dry matter and fat digestibility, potentially explaining the differences in performance observed between PFA and RFT calves. Inclusion of the PFA pellet at 10% with a calf starter improved BW, solid feed, and energy intake after weaning. However, these benefits were conditioned by fat source and its digestibility.

**Key words:** calf weaning, fat inclusion, solid feed

### INTRODUCTION

Over the last decade, the industry has been progressively changing to higher liquid feed allowances (~20% of birth BW by volume; Khan et al., 2010). This feeding strategy has been reported to increase growth rate, decrease age at first calving, and increase first-lactation milk yield (Hill et al., 2006; Soberon et al., 2012; Leal et al., 2021). However, greater preweaning milk allowances have been associated with reduced preweaning solid feed intake and delayed rumen development (Khan et al., 2007; Terré et al., 2007; Weary et al., 2008), making weaning more challenging and affecting calf growth and development around the weaning period (Cowles et al., 2006; Hill et al., 2010; Davis Rincker et al., 2011).

Weaning is stressful for calves due to factors such as rapid microbial changes and gastrointestinal structural adaptations (Meale et al., 2017); therefore, the method of weaning can influence calf performance and gastrointestinal development. The immature state of the rumen to ferment solid feeds, combined with the transition from a high-energy-density diet, such as whole milk (~5.4 Mcal of ME/kg of DM; Drackley, 2008) or milk replacer (MR; ~4.5 Mcal of ME/kg of DM) to a relatively low-energy-density solid diet (~2.48 Mcal/kg of DM) can result in lesser growth performance around this period, which is also known as the weaning gap (Leal et al., 2021). It has been suggested that total ME intake can decrease by as much as 39% due to a decline in DMI from MR, which is not compensated for by an

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immediate increase in starter consumption (Leal et al., 2021). Nondairy calves combine solid intakes with milk, resulting in a gradual exchange between these 2 nutrient sources, thus gradually reducing their intake of milk fat. When given the choice, this transition from milk to solid feed can take over 6 mo (Webb et al., 2014). In contrast, traditional rearing programs for dairy calves abruptly reduce their fat intake at weaning. Therefore, nutrient intake depends on total feed intake and diet composition. The above factors suggest that the relatively low energy intake during the weaning and early postweaning periods could be mitigated by increasing the dietary energy density of the starter feed through fat, as previously demonstrated by Araujo et al. (2014) and Berends et al. (2018).

The opportunities and limitations of inclusion of rumen-stable fat to increase energy density in dairy and feedlot diets are well established (Fluharty and Loerch, 1997), but information regarding rumen-inert fat inclusion in calf starters is limited. Gardner and Wallentine (1972) reported growth responses by veal calves to the addition of tallow to starter rations. Waldern and Fisher (1978) found no effect on feed conversion ratios with unprotected tallow in calf diets, whereas Fisher (1980) reported an increase in weight gain for calves receiving diets containing 20% versus 10% (of DM) of a protected lipid source. Berends et al. (2018) reported greater performance in calves fed a mixture of a traditional low-fat pellet (90%) with an extruded high-fat pellet containing hydrogenated fatty acids (FA) from palm oil (10%), compared with the low-fat control pellet. Despite the benefits reported in the previous study, palm oil use in animal nutrition is questioned because of issues related to environmental sustainability, which has led to animal feed manufacturers wanting to reduce its inclusion in feeds. To evaluate alternative sources of fat to increase raw material flexibility in feed production, in this study, we evaluated a 90:10 mixture of a calf starter and a high-fat extruded pellet containing either hydrogenated FA from palm oil, as described in Berends et al. (2018), or an alternative formulated with hydrogenated triglycerides (TG) from rapeseed oil.

Although most commercially available fat supplements for ruminants are based on hydrogenated free fatty acids (FFA), hydrogenated TG are also available. However, TG are generally considered less digestible (Elliott et al., 1999; Weiss and Wyatt, 2004). Esterification and FA profile of fat supplements are important factors affecting FA digestibility. Based on differences in melting point, FA profile, and chemical structure, a C16:0-rich hydrogenated FFA product could be expected to have higher digestibility than a C:18-rich hydrogenated TG (Doppenberg and van der Aar, 2017). Yet, the magnitude and relevance of this difference is

difficult to predict for such a specific and novel application of hydrogenated fats.

The objective of this study was to evaluate the effects of including a high-fat extruded pellet in a calf starter on performance and feed intake in rearing calves. This study also aimed to evaluate the effects of hydrogenated fat sources differing in esterification and FA profile (palm and rapeseed origin) on feed intake and digestibility. In experiment 1, we hypothesized that increasing dietary fat in calf starter feed could mitigate growth loss during weaning and the postweaning period. In experiment 2, we aimed to measure differences in digestibility between the 2 sources of hydrogenated fats and evaluate their eventual relevance on energy supply and growth in postweaning calves.

## MATERIALS AND METHODS

This study was conducted in 2 experiments, the first between October 2017 and January 2018 and the second between June 2020 and August 2020 at the Calf & Beef Research Facility of Trouw Nutrition Research and Development (Sint Anthonis, the Netherlands). All procedures described in this article complied with the Dutch Law on Experimental Animals, which complies with ETS123 (Council of Europe 1985 and the 86/609/EEC Directive) and were approved by the animal welfare authority (DEC Utrecht, the Netherlands).

### Experiment 1

**Animals and Experimental Design.** A total of 60 Holstein bull calves ( $45.7 \pm 0.87$  kg of BW,  $2.5 \pm 0.5$  d of age) were collected from local dairy farms at a maximum distance of 14 km from the calf research facility. At the farm of origin, a standardized protocol for colostrum management was followed, wherein calves were fed 3 L of colostrum within the first 3 h after birth followed by 2 feedings of 2 L for a total of 3 feedings of colostrum in the first 24 h of life. Upon arrival at the facility, successful application of this protocol was monitored by assessment of blood IgG within 48 to 72 h after birth of the calf. Calves were assigned to 1 of 20 blocks (3 calves per block) based on IgG category (low: 1,000–2,000 mg/dL; high: >2,000 mg/dL; NAHMS, 2007), day of arrival, and BW. Within each block, calves were randomized using the RAND function in Excel (Microsoft Corp.), to 1 of 3 solid feed treatments: (1) calf starter control pellet (CON, 3.1% fat); (2) control pellet mixed with an extruded pellet containing hydrogenated palm free fatty acids (PFA, 7.1% fat in pellet mix; Bewi-Spray 99FA, Bewital agri GmbH & Co); and (3) control pellet mixed with an extruded pellet containing hydrogenated rapeseed tri-

glycerides (RFT, 6.7% fat in pellet mix; Bewi-Spray RS 70, Bewital agri GmbH & Co). Mean serum IgG concentration was  $2.429 \pm 176$  mg/dL for CON,  $2.420 \pm 179$  mg/dL for PFA, and  $2.498 \pm 176$  mg/dL for RFT.

**Housing and Feeding.** Calves were housed indoors in individual pens ( $1.22 \times 2.13$  m) separated by galvanized bar fences and equipped with 50% rubber-slatted floors in the front and 50% lying area, including a mattress covered with flax straw in the back until weaning (wk 7); thereafter, calves were moved to a pen that was twice the initial size ( $2.44 \times 4.26$  m) with the same characteristics until the end of the study period (wk 12). From arrival until 14 d of age, calves were fed a total of 6 L of MR per day (Sprayfo Excellent, Trouw Nutrition) containing 22.9% CP and 18.4% fat (on a DM basis) provided in 2 equally sized meals of 3 L each at 0700 and 1700 h. Meal size was then increased to 3.5 L from 15 d until 42 d of age, after which milk meal size was decreased to 2.0 L until 49 d, when calves were fully weaned. Milk replacer was reconstituted with

water at 150 g/L of MR powder and supplied in a teat bucket at 40°C. Calves had ad libitum access to water, starter feed pellets, and chopped wheat straw (4.2% CP, 71.8% NDF, and 50.6% ADF on a DM basis; 3–7 cm chop length), all provided in separate buckets.

**Experimental Diets.** The CON treatment was designed to be representative of commonly used calf starter pellets. Experimental starter feeds were composed of a mixture of 90% CON pellets and 10% either PFA or RFT pellets. The 90:10 mixing ratio resulted in diets with fat concentrations of 7.1 and 6.7% for PFA and RFT, respectively. Table 1 describes the composition of the 3 pelleted feeds, and Table 2 shows the nutrient composition of the final starter feeds. The feed manufacturing processes followed those described by Berends et al. (2018). Although the treatments could be identified by visual assessment of the feeds, treatments were randomly coded and treatment identity was blinded at the research farm.

**Measurements and Chemical Analysis.** Milk replacer was sampled (200 g per MR) for analysis im-

**Table 1.** Formulated ingredients and analyzed nutrient composition of the 3 pelleted solid feeds used to compose the 3 solid feed treatments

Item	Pellet feed		
	Calf starter	Extruded hydrogenated rapeseed triglyceride	Extruded hydrogenated palm free fatty acid
Ingredient (% of DM)			
Soybean meal	25.0	—	—
Corn	21.0	21.3	21.3
Wheat	—	38.7	38.7
Hydrogenated palm fatty acids	—	—	40
Hydrogenated rapeseed fatty acids	—	40	—
Soy hulls	15.0	—	—
Alfalfa	10.5	—	—
Beet pulp	10	—	—
Barley	10	—	—
Sugar cane molasses	5	—	—
Limestone	1	—	—
Monocalcium phosphate	1	—	—
Mineral and vitamin premix <sup>1</sup>	1.5	—	—
Nutrient composition, DM basis			
CP (%)	19.3	5.8	5.5
Ether extract (%)	3.1	39.5	43.1
Ash (%)	7.4	1.2	1.2
NDF (%)	18.8	6.3	6.3
ADF (%)	11.4	1.8	1.6
Starch (%)	21.0	37.6	43.7
ME <sup>2</sup> (Mcal/kg)	2.47	5.07	5.43
Part of fatty acids (%)			
Free fatty acids	—	15	85
≤C14:0	—	1	1
C16:0 (palmitic acid)	—	4	44
C18:0 (stearic acid)	—	92	54
≥C20:0	—	3	1
Melting point (°C)	—	70	58

<sup>1</sup>Premix (provided per kg of concentrate): 24,000 IU/kg vitamin A, 5,000 IU/kg vitamin D<sub>3</sub>, 90 mg/kg vitamin E, 1.5 mg/kg Co (CoCO<sub>3</sub>), 377 mg/kg Fe (FeSO<sub>4</sub>), 88 mg/kg Mn (MnO), 87.7 mg/kg Zn (ZnO), 31.8 mg/kg Cu (CuSO<sub>4</sub>), and 0.30 mg/kg Se (NaSe).

<sup>2</sup>Calculated following NRC (2001).

**Table 2.** Analyzed and estimated nutrient composition of starter feed treatments used in the study

Item	Starter treatment <sup>1</sup>		
	CON	RFT	PFA
Ingredient (% of total starter feed)			
Calf starter	100	90	90
High-fat extruded pellet	—	10	10
Nutrient, DM basis			
CP (%)	19.3	18.0	17.9
Ether extract (%)	3.1	6.7	7.1
Starch (%)	21.0	22.7	23.3
Ash (%)	7.4	6.8	6.8
Gross energy <sup>2</sup> (Mcal/kg of DM)	3.93	4.04	4.21
ME <sup>3</sup> (Mcal/kg)	2.61	2.51	2.90

<sup>1</sup>CON = calf starter; RFT = control pellet mixed with an extruded hydrogenated rapeseed triglyceride; PFA = control pellet mixed with an extruded hydrogenated palm free fatty acid.

<sup>2</sup>Gross energy = CP content (g/kg) × 0.0057 Mcal/g + crude fat content (g/kg) × 0.0095 Mcal/g + (starch + sugars + fiber) × 0.0042 Mcal/g.

<sup>3</sup>Calculated following the Quigley et al. (2019) formula for ME with the calculated digestible energy coefficient as determined in experiment 2.

mediately after production in the factory. Starter feed (200 g) and straw (100 g) were sampled upon arrival on the farm. Individual intakes of MR, water, concentrate, and straw were weighed and recorded daily during the entire experiment. Feed refusals were collected daily and sampled 3 times per week to monitor for potential feed sorting. The feeds and pooled refusals were analyzed for DM content by drying to a constant weight in a 103°C stove for 4 h (EC 152/2009; EC, 2009). Crude protein was analyzed by combustion, according to the Dumas method (Etheridge et al., 1998). Crude fat content was determined after acid hydrolysis according to ISO Standard 6496 (ISO, 1999). Starch content was determined enzymatically as described by Rijnen et al. (2001). Crude ash content was determined by incineration in a muffle furnace by combustion at 550°C (EC, 1971; 71/250/EEG 1971).

Body weight and body measurements [hip height, chest girth, withers height, and body barrel (circumference around the abdominal area of the animal)] were measured on the day of arrival and weekly thereafter. Blood samples were obtained at 4, 6, 7, 8, 10, and 16 wk at 1300 h. Blood samples were taken into one 10-mL EDTA tube for serum, and one 6-mL sodium fluoride (NaF) evacuated tube containing a glycolysis inhibitor for plasma glucose (Becton Dickinson). All blood samples were centrifuged immediately at  $2,800 \times g$  for 30 min at room temperature, and 1.5 mL of plasma was pipetted into 2-mL cryotubes and stored immediately at -20°C. Blood samples were analyzed for BHB, glucose, and IGF-1 at Synlab.vet (Leipzig, Germany). Plasma

concentrations of BHB were determined using a kinetic UV test, glucose was determined using the enzymatic UV test-hexokinase method, and IGF-1 by RIA.

Health was monitored daily by caretakers, and a standard veterinary protocol was followed in case of disease. A veterinarian was consulted if the symptoms were not described in the standard protocol. Photos of feces were taken daily for all calves, and fecal scores were evaluated at the end of the study by one single observer blinded to treatments. The scoring system included 4 categories, where 0 was considered normal and 3 extremely loose. Fecal scores  $\geq 2$  were considered abnormal. Medical treatments were recorded daily.

## Experiment 2

**Animal and Experimental Design.** A total of 24 weaned Holstein bull calves at 3 mo of age (different calves from those in experiment 1) were used in this study. Calves were assigned to 1 of 8 blocks based on arrival BW and age. Within each block, calves were randomly assigned to 1 of the 3 treatments described in experiment 1 (n = 8 calves/treatment). Similar to experiment 1, animals had ad libitum access to water, straw, and treatment feed.

**Housing.** During the adaptation period (14 d) to the experimental diets, calves were housed in individual pens (5.4 m<sup>2</sup>). Pens were equipped with 50% rubber-slatted and 50% mattress covered with flax. After the adaptation period, calves were moved into metabolic cages with rubber-slatted floors, where they stayed for a 72-h total collection period. In the metabolic cages, calves were tethered to the front of the pen with enough space to lie down. Immediately after the end of the 72-h collection period, calves were moved back to their individual pens.

**Measurements and Chemical Analysis.** Individual intakes of concentrate, water, and straw were weighed and recorded daily during the entire experiment. Feces were collected for a total of 72 h using fecal bags glued to the hindquarters of the animal. Once the fecal bags were full, the bags were removed, weighed, frozen, and then replaced with a new bag. After 3 d of total collection, feces were thawed and homogenized. Two samples of 450 g were taken from the pooled feces and frozen for later analysis. Diets and feces were analyzed for DM content by drying to a constant weight in a 103°C stove for 4 h (EC, 2009; EC 152/2009). Crude protein was analyzed by combustion, according to the Dumas method (Etheridge et al., 1998). Crude fat content was determined after acid hydrolysis according to ISO Standard 6496 (ISO, 1999). Starch content was determined enzymatically as described by Rijnen

et al. (2001). Crude ash content was determined by incineration in a muffle furnace by combustion at 550°C (EC, 1971; 71/250/EEG 1971). Fecal VFA content was determined by HPLC, using the method described in Canale et al. (1984), and a pH meter was used to measure fecal pH.

**Calculations and Statistical Analysis.** The ME content of the MR was calculated as follows: ME (Mcal/kg) =  $[0.057 \times \text{CP} (\%) + 0.092 \times \text{crude fat} (\%) + 0.0395 \times \text{lactose} (\%)] \times 0.93$  (NRC, 2001). The ME content of starter feed was calculated based on NRC (2001) ME values for the raw materials and applying the equation for calf starter. The ME content of the starter treatments was estimated based on the energy digestibility coefficient calculated in experiment 2, using gross energy intake from feed and fecal energy output. Gross energy intake (Mcal/kg) was calculated as feed intake (feed supply minus refusals) multiplied by the calculated gross energy content of each feed component  $[\text{CP content (g/kg)} \times 0.0057 \text{ Mcal/g} + \text{CF content (g/kg)} \times 0.0095 \text{ Mcal/g} + (\text{starch} + \text{sugars} + \text{fiber}) \times 0.0042 \text{ Mcal/g}]$ . Fiber was estimated as  $(\text{DM} - \text{CP} - \text{ash} - \text{crude fat} - \text{starch} - \text{sugars})$ . Fecal energy output (Mcal/g) was calculated as  $[\text{fecal DM output (g of DM/d)} \times \text{fecal energy content in DM (Mcal/g of DM)}]$ . Feed conversion was calculated by dividing the total ME (Mcal/kg) by ADG (kg).

A classical power analysis was conducted to determine the number of experimental units needed. The power  $(1 - \beta)$  was chosen to be equal to 80%, and the  $\alpha$ -level was 0.05. Growth performance was considered the most reliable parameter to determine power. Based on a previous study conducted at the same research facility (Calf & Beef Research, Sint Anthonis, the Netherlands) for 57 d after arrival at 0 to 3 d of age, a standard deviation (SD) of 100 g/d was assumed. The minimal meaningful difference in ADG was considered to be 90 g/d. The minimal sample size was 20 calves per treatment, accounting for a maximum mortality rate of 10%.

All statistical analyses were performed using SAS Studio (version 3.2, SAS Institute Inc.). Data on growth, intakes, blood parameters, fecal pH, and fecal contents were analyzed with a mixed-effects model procedure (PROC MIXED). Proportional data were analyzed (apparent total-tract digestibility) with a  $\beta$  distribution using a generalized linear mixed model (PROC GLIMMIX). In all analyses, the individual calf was considered the experimental unit, with treatment and block as fixed and random effects, respectively. Time entered the model as a repeated statement in case of repeated measurements, and then the interaction between time and treatment and the SLICE command from SAS Studio (version 3.2, SAS Institute Inc.) to control type

I error were included. For both experiments, treatment averages were presented as least squares means and standard errors of the mean. The normal distribution of the residuals was checked to verify model assumptions. Significance was declared when  $P \leq 0.05$ , and trends were declared when  $P < 0.10$ .

## RESULTS

### Experiment 1

In total, 5 calves were removed from the study based on pre-established exclusion criteria for health or feed refusals. Data collected from these calves before removal were excluded from the analyses; therefore, the data set included 55 successful observations (CON = 19, PFA = 18, and RFT = 18 calves).

**Intake and Growth Performance.** Treatment effects on feed intake and performance are presented in Tables 3 and 4, respectively. No differences in MR intake were observed during the preweaning period ( $P = 0.66$ ). Similarly, no significant differences were detected for solid feed intake and straw intake during the preweaning and postweaning periods among treatments (Table 3); however, after weaning, PFA calves tended to consume more starter feed ( $P = 0.08$ ) than CON and RFT calves (Table 3). During the entire experimental period (112 d), PFA calves consumed 146 g/d more solid feed than CON calves, and 40 g/d more than RFT calves ( $P = 0.03$ ; Table 3). Intakes of ME ( $P = 0.61$ ) and CP ( $P = 0.38$ ) did not differ between the 3 treatments during the preweaning period. However, as a result of the high intake of the PFA treatment and the low digestibility of the RFT diet, calves in the PFA group had the highest ME intake after weaning ( $P < 0.001$ ) compared with CON and RFT calves (Table 3 and Figure 1). Initial BW and body measurements were similar among all animals. No differences among treatments were observed for BW ( $P = 0.41$ ; Table 4) or ADG ( $P = 0.77$ ; Table 4) during the preweaning period. Calves consuming the PFA treatment had greater ADG than CON and RFT calves during the weaning period ( $P = 0.05$ ). After weaning and until the end of the study (112 d), calves in the PFA treatment had greater BW ( $P = 0.01$ ), being 8.8 and 9.3 kg heavier than CON and RFT calves, respectively (Table 4). During the postweaning period, PFA calves had greater ADG than the other groups, with this difference being significant between d 50 and 84 ( $P = 0.05$ ), and showing a tendency for a treatment effect thereafter ( $P = 0.07$ ). Over the total period, the ADG of PFA calves was the greatest compared with that in the CON and RFT calves ( $P = 0.03$ ). Furthermore, no differences were observed for withers

**Table 3.** Effect of dietary treatments on intakes of young calves before (0–49 d of age) and in 2 periods after weaning (50–84 d of age) and (85–112 d of age) (experiment 1)

Item	Treatment <sup>1</sup>			SEM	P-value <sup>2</sup>	
	CON	RFT	PFA		T	T × t
MR intake (g/d)						
0–49 d	913.5	918.7	911.6	5.7	0.66	0.87
Starter intake (g/d)						
0–49 d	324	378	351	46.4	0.76	0.81
50–84 d	2,534	2,615	2,903	125.9	0.08	0.75
85–112 d	3,966	4,047	4,322	173.1	0.31	0.91
Total period	1,892 <sup>b</sup>	1,998 <sup>a</sup>	2,038 <sup>a</sup>	39.7	0.03	0.99
Straw intake (g/d)						
0–49 d	12	16	16	4.6	0.78	0.11
50–84 d	82	88	127	16.2	0.11	0.02
85–112 d	162	177	241	28.7	0.12	0.99
Total period	73.0 <sup>b</sup>	79.7 <sup>a</sup>	107.2 <sup>a</sup>	9.4	0.03	0.99
Water intake (kg)						
0–49 d	1.4	1.6	1.7	0.1	0.44	0.79
50–84 d	7.9	8.1	8.8	0.2	0.38	0.76
85–112 d	12.1	12.2	12.8	0.3	0.78	0.97
Total period	6.1	6.2	6.5	0.3	0.53	0.93
ME intake <sup>3</sup> (Mcal/d)						
0–49 d	4.47	4.59	4.63	0.13	0.61	0.87
50–84 d	6.71 <sup>b</sup>	6.63 <sup>b</sup>	8.07 <sup>a</sup>	0.30	0.01	0.41
85–112 d	10.35 <sup>b</sup>	10.19 <sup>b</sup>	12.29 <sup>a</sup>	0.52	<0.01	0.92
Total period	6.62 <sup>b</sup>	6.60 <sup>b</sup>	7.56 <sup>a</sup>	0.10	0.01	0.52
CP intake (g/d)						
0–49 d	262	272	266	5.2	0.38	0.95
50–84 d	404	419	437	21.3	0.54	0.62
85–112 d	624	644	670	29.5	0.55	0.84
Total period	397	411	420	14.2	0.31	0.92
Feed conversion (kg of gain/Mcal)						
0–49 d	0.145	0.145	0.139	0.045	0.24	0.80
50–84 d	0.150	0.150	0.140	0.005	0.27	0.56
85–112 d	0.127	0.125	0.119	0.005	0.31	0.09
Total period	0.145	0.145	0.139	0.003	0.24	0.79

<sup>a,b</sup>Means that do not share a common letter differ ( $P < 0.05$ ).

<sup>1</sup>CON = calf starter; RFT = CON mixed with an extruded hydrogenated rapeseed triglyceride; PFA = CON mixed with an extruded hydrogenated palm free fatty acid.

<sup>2</sup>T = effect of treatment; T × t = interaction between treatment and time.

<sup>3</sup>Calculated following the Quigley et al. (2019) formula for ME with the calculated digestible energy coefficient as determined in experiment 2.

height, hip height, chest girth, or body barrel for the main effect of treatment (Table 4).

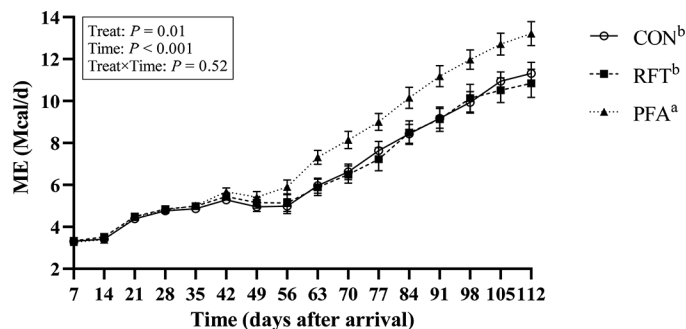
**Blood Parameters.** Results for blood parameters are summarized in Table 5. No differences in blood BHB and glucose concentrations were observed across treatments in any period (Table 5). Nevertheless, IGF-1 concentration differed during the preweaning period ( $P = 0.02$ ), with RFT calves having greater concentrations than CON and PFA calves. No differences in fecal scores were observed during the experimental period (data not shown).

## Experiment 2

Data collected during the digestibility assessment period are shown in Tables 6 and 7. Calves on PFA consumed more starter, straw, and water ( $P = 0.01$ )

than those in the CON and RFT groups. The greatest total-tract apparent DM digestibility was observed in calves fed the CON diet, and the lowest was reported for calves fed the RFT diet ( $P = 0.01$ ). Differences in total-tract apparent digestibility of crude fat were observed among all treatments. Fat digestibility was 31.1 and 6.8 percentage points higher in PFA calves ( $P = 0.01$ ) than in RFT and CON calves, respectively (Table 6). The RFT treatment resulted in an overall lower nutrient digestibility than the CON treatment, even though intakes of nutrients in RFT were higher ( $P = 0.01$ ). Dietary treatment also affected fecal pH, with PFA calves having the lowest pH ( $P = 0.01$ ) compared with CON and RFT calves (Table 7). No differences were observed in total fecal VFA between treatments; however, the molar proportion of fecal butyrate was lower in the CON treatment ( $P = 0.02$ ) but comparable

## DISCUSSION



**Figure 1.** Intake of ME (Mcal/d) corrected for the digestible energy coefficient determined in experiment 2 as affected by treatment. CON = calf starter; RFT = CON mixed with an extruded hydrogenated rapeseed triglyceride; PFA = CON mixed with an extruded hydrogenated palm free fatty acid. Error bars indicate SE. Treatments that do not share a common letter (a–c) differ ( $P < 0.05$ ). Treat = treatment.

between RFT and PFA calves. Total fecal fat was affected by dietary treatment ( $P = 0.01$ ): total fecal fat in RFT calves was 3-fold higher than in PFA calves and 5-fold higher than in the CON group (Table 7).

The objective of this study was to evaluate the effects of increasing the energy levels of calf starter by increasing the fat content (from different sources) on feed intake, performance, and digestibility in rearing calves during the weaning transition. In the current study, during the preweaning period, the same MR formulation was fed in similar amounts to calves in all treatments, resulting in similar daily consumption of liquid feed. It has been suggested that the main disadvantage when feeding high amounts of milk or MR is the reduction of starter intake during the preweaning period (Jasper and Weary, 2002; Terré et al., 2006; Khan et al., 2007; Kristensen et al., 2007; Borderas et al., 2009). However, in more recent studies, it has been demonstrated that, under adequate weaning practices, feeding high-energy MR at high levels (7 L, 150 g/L; Amado et al., 2019) or even ad libitum (Echeverry-Munera et al., 2021) is possible without negative effects on solid feed consumption or growth postweaning. During the preweaning period of the current study, solid feed intake was similar

**Table 4.** Effect of dietary treatments on performance and body measurements of young calves until 112 d of age (experiment 1)

Item	Treatment <sup>1</sup>			SEM	P-value <sup>2</sup>	
	CON	RFT	PFA		T	T × t
BW (kg)						
Initial <sup>3</sup>	45.4	46.2	45.5	1.1	0.84	—
49 d	78.8	81.6	81.3	2.3	0.62	—
84 d	114.2	116.6	121.1	2.4	0.12	—
112 d	153.3 <sup>b</sup>	152.8 <sup>b</sup>	162.1 <sup>a</sup>	2.5	0.01	—
Total period gain	93.5	95.2	97.6	2.2	0.41	0.29
ADG (kg/d)						
49 d	0.66	0.69	0.68	0.04	0.77	—
84 d	0.99 <sup>b</sup>	0.98 <sup>b</sup>	1.13 <sup>a</sup>	0.06	0.05	—
112 d	1.28	1.23	1.40	0.06	0.07	—
Total period	0.88 <sup>b</sup>	0.90 <sup>b</sup>	0.98 <sup>a</sup>	0.03	0.03	0.48
Body measurements (cm)						
Withers height						
Initial	78.5	79.4	78.7	0.57	0.49	—
Final	102.7	102.9	102.4	1.17	0.97	—
Hip height						
Initial	80.8	81.6	80.6	0.63	0.51	—
Final	106.9	107.2	107.3	1.81	0.96	—
Chest girth						
Initial	82.5	82.8	81.8	0.68	0.52	—
Final	118.2	118.1	119.2	1.98	0.92	—
Body barrel						
Initial	83.2	83.7	82.5	0.71	0.47	—
Final	148.1	147.0	149.5	3.28	0.86	—

<sup>a,b</sup>Means that do not share a common letter differ ( $P < 0.05$ ).

<sup>1</sup>CON = calf starter; RFT = CON mixed with an extruded hydrogenated rapeseed triglyceride; PFA = CON mixed with an extruded hydrogenated palm free fatty acid.

<sup>2</sup>T = effect of treatment; T × t = interaction between treatment and time.

<sup>3</sup>Initial BW was used as a covariable in the model.

**Table 5.** Effect of dietary treatments on plasma concentrations of BHB, glucose, and IGF-1 in growing calves preweaning (4, 6, and 7 wk) and postweaning (8, 10, and 16 wk; experiment 1)

Item	Treatment			SEM	P-value <sup>2</sup>
	CON	RFT	PFA		
BHB (mmol/L)					
Preweaning	0.09	0.09	0.10	0.01	0.96
Postweaning	0.34	0.33	0.36	0.02	0.64
Glucose (mmol/L)					
Preweaning	5.46	5.89	5.30	0.15	0.29
Postweaning	4.91	5.23	4.97	0.15	0.30
IGF-1 (ng/mL)					
Preweaning	121.3 <sup>ab</sup>	139.6 <sup>a</sup>	106.9 <sup>b</sup>	7.92	0.02
Postweaning	190.6	191.3	174.0	28.59	0.89

<sup>a,b</sup>Means that do not share a common letter differ ( $P < 0.05$ ).

<sup>1</sup>CON = calf starter; RFT = CON mixed with an extruded hydrogenated rapeseed triglyceride; PFA = CON mixed with an extruded hydrogenated palm free fatty acid.

<sup>2</sup>T = effect of treatment.

among the 3 treatments, with calves consuming, on average, 351 g of calf starter per day. These solid feed intakes could be attributed to the high supply of MR and are comparable to those reported in recent studies (de Carvalho et al., 2021). In the study conducted by de Carvalho et al. (2021), calves were fed a high plane of MR (50 kg of MR during the preweaning phase) and were observed to consume 450 g of calf starter per day until wk 7. Earlier studies evaluating the inclusion of fat in starter feeds have associated high-fat single pellets with reduced intake and depressed weight gain (Doppenberg and Palmquist, 1991). In contrast, a later study by Araujo et al. (2014) showed that when calves were offered either restricted or moderate amounts of

MR (4 or 6 L/d) along with either a low-fat (4.1% fat inclusion) or a high-fat (11.2%) starter feed, calves fed the high-fat calf starter had higher BW gain postweaning, mainly due to higher energy intakes from the solid feed.

In the current study, which aimed to resolve the conflict between energy delivery and rumen function and development, fat inclusion in starters was considered an important factor, following encouraging results in an earlier study by our group (Berends et al., 2018), in which calves were fed a traditional pellet mixed with an extruded high-fat pellet, but this study also evaluated an alternative rumen-inert fat source, hydrogenated rapeseed triglycerides. Confirming our earlier results,

**Table 6.** Effect of dietary treatments on intakes and apparent total-tract digestibility of nutrients from solid feed of 12-wk-old calves (experiment 2)

Item	Treatment <sup>1</sup>			SEM	P-value <sup>2</sup>
	CON	RFT	PFA		
Intake					
Starter intake, kg/d					
Collection period (72 h)	4.07 <sup>c</sup>	4.39 <sup>b</sup>	4.76 <sup>a</sup>	0.20	0.01
Total period (2 wk)	3.44 <sup>c</sup>	3.83 <sup>b</sup>	4.03 <sup>a</sup>	0.14	0.01
Straw intake, g/d					
Collection period (72 h)	148	168	215	46.9	0.12
Total period (2 wk)	115.86 <sup>c</sup>	162.28 <sup>b</sup>	178.14 <sup>a</sup>	38.9	0.01
Water intake, kg					
Collection period (72 h)	15.5	16.9	17.6	0.84	0.05
Total period (2 wk)	14.58 <sup>b</sup>	16.25 <sup>a</sup>	16.84 <sup>a</sup>	0.75	0.01
Apparent total-tract digestibility (%)					
DM	79.4 <sup>a</sup>	75.1 <sup>b</sup>	76.0 <sup>b</sup>	1.36	0.01
OM	81.1 <sup>a</sup>	76.4 <sup>b</sup>	77.1 <sup>b</sup>	1.27	0.03
CP	78.3	75.6	75.4	1.39	0.27
Crude fat	81.0 <sup>b</sup>	56.8 <sup>c</sup>	87.9 <sup>a</sup>	1.55	0.01

<sup>a-c</sup>Means that do not share a common letter differ ( $P < 0.05$ ).

<sup>1</sup>CON = calf starter; RFT = CON mixed with an extruded hydrogenated rapeseed triglyceride; PFA = CON mixed with an extruded hydrogenated palm free fatty acid.

<sup>2</sup>T = effect of treatment.



**Table 7.** Effect of dietary treatments on fecal parameters of 12-wk-old calves (experiment 2)

Item	Treatment			SEM	<i>P</i> -value <sup>2</sup>
	CON	RFT	PFA		
pH	6.2 <sup>a</sup>	6.1 <sup>b</sup>	6.0 <sup>b</sup>	0.06	0.01
VFA, molar proportions (%)					
Acetate	69.9	68.8	67.3	0.98	0.17
Propionate	21.9	21.7	22.9	0.87	0.56
Butyrate	8.1 <sup>b</sup>	9.8 <sup>a</sup>	9.5 <sup>a</sup>	0.43	0.03
Fecal content (g/kg of DM feces)					
CP	702.7	777.2	833.2	44.32	0.15
Crude fat	98.7 <sup>c</sup>	510.4 <sup>a</sup>	157.3 <sup>b</sup>	20.00	0.01
Ash	5.2	5.6	5.6	0.33	0.67

<sup>a-c</sup>Means that do not share a common letter differ ( $P < 0.05$ ).

<sup>1</sup>CON = calf starter; RFT = CON mixed with an extruded hydrogenated rapeseed triglyceride; PFA = CON mixed with an extruded hydrogenated palm free fatty acid.

<sup>2</sup>T = effect of treatment.

PFA calves tended to eat more calf starter during the weaning period and throughout the experiment compared with CON calves. Calves in the PFA and RFT groups consumed, on average, 146 and 106 g more, respectively, than CON calves during the study. Furthermore, calves across all treatments had comparable plasma BHB levels, both before and after weaning. According to Coverdale et al. (2004), BHB could be a good indicator of a shift in nutrient source from liquid feed to solid and an indicator of rumen development and fermentation around the time of weaning (Quigley et al., 1991, 1992). When using BHB as a proxy for rumen fermentation, it is possible that calves were able to access the fermentable pellets despite the high fat content in experimental diets. Similarly, Berends et al. (2018) reported no differences in the concentrations of BHB in starter diets with different inclusion levels of high-fat extruded pellets.

The increased solid feed intake observed in PFA and RFT calves, together with the high level of ME in the respective dietary treatments, resulted in greater ME intakes of PFA calves. The increased consumption of energy through solid feed potentially helped to compensate for the shortage of energy provided by the MR and the lower starter intake during the weaning period. We observed that calves in the PFA treatment required 2 wk, on average, to consume the same level of ME they were consuming before the weaning period (wk 6). In contrast, the CON and the RFT groups required 3 and 4 wk, respectively, to match their preweaning ME intake. Notwithstanding, the effect of high-fat extruded pellets (PFA or RFT) on BW and ADG began after the weaning period and was maintained during the next month of observation (112 d), when the PFA calves had gained 8.8 kg more than the CON animals. These results add to those reported by Berends et al. (2018), in which delivering dietary fat (~7% of DM) by mix-

ing an extruded high-fat pellet with a conventional calf starter improved solid feed intake, energy intake, and rate of BW gain compared with a traditional low-fat pellet (~3% of DM).

Usually, rumen-inert fats have a high degree of fatty acid saturation, resulting in high melting points, or are saponified with Ca. These characteristics make rumen-inert fats less likely to interfere with rumen fermentation (Elliott et al., 1999). Additionally, TG forms have previously been shown to have a lower digestibility compared with FFA in dairy cows (Pantoja et al., 1995; Elliott et al., 1999; Weiss and Wyatt, 2004). In a literature review, Naik (2013) reported a range of 82.05 to 89.0% for fat digestibility of a dairy ration supplemented with rumen-inert fats. Similarly, in the current study, fat digestibility of PFA and CON was 87.9 and 81.0%, respectively, but in calves consuming the RFT diet, substantially lower total-tract fat digestibility was observed (56.8%). Elliot et al. (1999) reported that steers fed diets containing hydrogenated TG as supplemented fat showed lower digestibility when the saturated fat source contained more TG (57.7%) than FFA (64.7%). Their data showed that hydrogenated TG are resistant to lipolysis in the rumen and small intestine when TG consist of mostly SFA. The fat digestibility results in the current study might be influenced not only by the degree of esterification, but also by the FA profile of the supplement fed. This has been reported in earlier studies in mature dairy cows, where it was observed that palmitic acid is more digestible and stearic acid less digestible than the average FA mixture in ruminants (Palmquist, 1994). As a result of the reduced digestibility, a larger portion of fat was found in the feces of animals consuming the RFT diet. The poor digestibility of the fat presents a simple explanation for the decreased growth performance observed in calves consuming the RFT diet despite comparable feed intakes.

The principal VFA that result from carbohydrate fermentation in the large bowel are acetate, propionate, and butyrate (Sato and Koiwa, 2008). However, changes in diet alter both substrate flow and disappearance in the bovine large intestine (Karr et al., 1966); therefore, it is likely that fermentation patterns and VFA production rates change accordingly. Results on fecal acetate and propionate in the present study did not differ among treatments, indicating similar fermentation and an adequate transit of digesta and dilution of feces, as stated by Sato and Koiwa (2008). Nevertheless, we found that calves in the RFT (9.8%) and PFA (9.5%) treatments had higher butyrate in feces compared with CON calves (8.1%). Increased fecal butyrate might have been the result of increased hindgut fermentation. High fat concentrations in the intestine might interfere with starch digestion or absorption, increasing the carbohydrate load in the hindgut (Jenkins and Jenny, 1989).

Plasma glucose concentration was also observed to decrease around weaning, which is consistent with previous studies (Khan et al., 2011; Echeverry-Munera et al., 2021). This may be attributed to the physiological shift in the primary energy source from glucose to VFA when the rumen in young calves becomes functional (Hammon et al., 2002). Nevertheless, in the study of Jenkins and Jenny (1989), cows fed with fat supplements tended to have higher plasma glucose. Similarly, Berends et al. (2018) reported that when young calves are provided with high dietary fat levels up to 13.4% of crude fat on a dry basis in the starter, glucose metabolism can be compromised, as suggested by an accumulation of glucose in blood following a glucose tolerance test in the calves fed the high-fat diet. However, in the current study, blood glucose concentration was similar among all groups and within the normal range for plasma glucose previously stated by Egli and Blum (1998). Moreover, calves consuming the RFT diet had the highest MR intake and levels of plasma IGF-1 during the preweaning period. Interestingly, in recent studies, it has been shown that increased nutrient intake from MR stimulates IGF-1 secretion and increases IGF-1 blood concentration in calves (Bartlett et al., 2006; Daniels et al., 2008; Schaff et al., 2016).

## CONCLUSIONS

Energy intake is the main factor limiting health and performance during the weaning process of young dairy calves. In this study, we fed calves a high-fat extruded pellet to sustain energy intake and growth around the weaning period. Our results confirmed that the high-fat supplement was consumed in addition to basal starter feed intake, substantially increasing energy intake. However, this effect was better translated to growth

when the fat source was hydrogenated palm free fatty acids (PFA) because of the wide differences in total-tract digestibility found between PFA and RFT, which likely derived from their different esterification and fatty acid composition. Supplementing fat in the solid feed as a high-fat extruded pellet mixed with a calf starter feed was shown to supply additional energy to dairy calves around and after weaning.

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