Effects of a completely pelleted diet on growing performance of Holstein heifers.

INTERPRETIVE SUMMARY

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Bonfante

The pelletizing process, along with several potential benefits causes an important reduction of fiber particle size compared to a common total mixed ration. The aim of the current study was to evaluate the effects on rumen conditions (temperature and pH), fiber digestibility and animal performance of a pelleted diet fed to growing heifers. The results show that a pelleted diet, well designed to guarantee an adequate amount of NDF, could be fed to growing ruminants without causing dietary dysfunction.

COMPLETE DIET IN PELLET FOR GROWING ANIMALS

Effects of a completely pelleted diet on growing performance of Holstein heifers

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ABSTRACT

The attributes of forage NDF content and particles size are recognized as important factors that impact rumen function. The aim of the current study was to evaluate the effects of pelleting a forage-based diet on rumen health, NDF digestibility, and animal performance. Eight Holstein heifers (age 336±30d, BW 346±35kg) were randomly assigned to a repeated cross-over design. Animals were housed in tie-stalls and fed for ad libitum intake. The study last four 3-wk periods, having the initial 2-wk for adaptation to the diet and 1 for data collection. Diets had the same ingredients but differed in physical forms: total mixed ration (treatment TMR) and PELLET (Ø=8mm; treatment P). The physically effective NDF (peNDF) differed among the two treatments (39.8 and 11.8% of NDF in TMR and P, respectively). During the trial DMI, water intake, rumination time, rumen temperature and pH were evaluated daily. Fecal samples were collected on the third wk of each period for total tract digestibility of the potential digestible NDF (TTdpdNDF). Average daily gain (ADG) and feed conversion ratio (FCR) were calculated at the end of each period. The DMI, DMI/BW, and water consumption were higher during the feeding of pelleted diet. There was no significant difference in ADG and FCR. Rumination time was lower for P than TMR treatment (241 vs. 507 min/d, respectively). Diet had no effect on rumen temperature and rumen pH. TTdpdNDF was greater in the TMR compared to P treatment (90.25 vs. 86.82 %pdNDF, respectively). The results of the current study suggest that a complete-feed pelleted diet was well accepted by the animals, as demonstrated by higher DMI. Rumination time was reduced with the P diet treatment, but rumen pH was not different than with the TMR diet treatment. The pdNDF digestibility was high for both diet treatments, with the TMR treatment significantly higher. Given the similar animal performance between the two treatments, which differed in DMI and apparent extent of fiber digestion, we might hypothesize different retention times of the two diets, related to their respective physical form. In conclusion, a complete-feed pelleted diet formulated to provide a sufficient level of NDF from forages, could be fed to growing ruminants without any apparent negative impact on rumen health and animal productivity, at least for a short period of time. More

52 researches considering a longer growing period are needed before recommending this feeding
53 strategy for growing heifers.
54 **Key Words**: pellet, fiber particle size, pdNDF digestibility.
55

INTRODUCTION

Fiber particle size and its NDF content have been considered important factors influencing rumen health (Allen, 1997; Krause et al., 2002b; Kononoff et al., 2003). This lead to a new concept introduced by Mertens (2000) who estimated the physically effective NDF (**peNDF**) to be the product of NDF concentration and physical effectiveness factor (**pef**). The latter represents the percentage of particles retained on a 1.18-mm sieve, considered highly resistant to passage out of the rumen (Poppi et al., 1985).

Fiber particle size influences chewing time and saliva secretion thereby affecting ruminal pH. It might also impact the retention time of particles in the rumen, and the extent of rumen fermentation and fiber degradation (Kaske et al., 1992; Teimouri Yansari et al., 2004; Kammes and Allen, 2012).

The reduction of fiber particle size in feeds has been used as an effective strategy to increase dry matter intake (**DMI**). Several studies demonstrate the difference between two or more feed chop lengths on animal performance (Yang and Beauchemin, 2006, 2009; Kammes and Allen, 2012); however, few trials have focused on pelletizing as a strategy to achieve this effect (McCroskey et al., 1960; Cullison, 1961; Burt, 1966). Through this method, controlled amounts of pressure and heat are applied to the combined aggregate of feed to increase its density (Mani et al., 2006). Pelleting offers many technical advantages including improved stability (owing to very low moisture content), easier handling management, storage and transportation.

Pelleting reduces fiber particle size and thus it might promote an increased rate of passage out of the rumen, and a subsequent decrease in fiber digestibility (Van Soest, 1994). Conversely, reduced particle size might serve to increase the surface area available for bacterial attachment (Miron et al., 2001), thereby increasing fiber digestibility. This reduction of fiber particle size could impact rumen fermentation and promote the development of subactute rumen acidosis (**SARA**; Khafipour et al., 2009).

The objective of this study was to evaluate the effects of a complete pelleted diet, formulated for growing heifers, on eating behavior, rumen fermentation, fiber digestibility, and animal performance. The hypothesis was that peNDF is not the only factor able to maintain an healthy rumen, but a diet high in NDF content can overcome the risk related with a low pH due to a lack in coarse forages.

MATERIALS AND METHODS

Animals and Treatments

The experimental procedures were approved by the Scientific Ethical Committee for animal experimentation at Bologna University. Eight Holstein heifers were used in a repeated cross-over design. The duration of the study was 12 wk, with four 3-wk periods. Heifers were adapted to the diet during the first two wk, and samples and data were collected during the last wk (experimental wk) of each period. The heifers had similar characteristics of age (336 ± 30 d) and body weight (346 ± 35 kg) at the beginning of the experiment, and were divided in two homogeneous groups.

Diet composition was the same for both treatments, but different in physical form (Table 1 and 2). Diet for treatment one (**TMR**) was prepared as total mixed ration with a horizontal auger, trailer-type TMR feed-mixer (Zago 13-m^3, ZAGO srl, PD, Italy). Treatment two (**P**) was produced as a complete-feed pelleted diet with the forages (grass hay and barley straw) chopped at 12-mm theoretical length of cut and then incorporated with the other ingredients (corn meal, sunflower meal, NaCl), mixed and pelleted (8-mm diameter).

Data and Samples Collection

Throughout the duration of the experiment, heifers were housed in tie-stalls bedded with sawdust, and fed their respective diets once daily in the morning (0830 h). The amount of feed offered and the refusals (orts) were weighed daily for each heifer. Feed was given for ad libitum intake based on orts quantity (10% of the DMI of the day before). Feed samples and orts were collected twice a wk for chemical and physical composition analysis. Daily water consumption was also recorded.

Rumination time was recorded for each heifer daily with an acoustic sensor collar (RuminAct®, SCR Heatime, Israel).

Reticulorumen pH and temperature values were monitored continuously via an indwelling pH and temperature sensor (SmaXtech®, Animal Care, Austria) instilled in the reticulorumen region of the stomach. Data were transmitted to an external receiver via a Wi-Fi signal every 10 min.

Heifers were weighed at the beginning of the study and at the end of each 3-wk period.

Fecal samples were collected every 6 h at d 5 (starting at 0000 am) and d 6 (starting at 0300 am) of the experimental wk so that eight samples were taken for each heifer, each period, representing every 3 h of a 24-h period to account for diurnal variation. In each period fecal samples belonging to the same heifer were composited and then analyzed for nutrient chemical composition.

Samples Analysis and Calculations

Feed and fecal samples were dried in a forced-air drying oven (M700-VF, MPM instrument, Bernareggio, IT) at 65°C for 48 h to determine DM content. Particle size distribution of the dried diet was determined using a sieve-type shaker (Ro-Tap®; WS Tyler, Mentor, OH) consisting of six sieves having 6.70, 4.75, 3.35, 2.36, 1.18, 0.15 mm apertures plus a bottom pan. The fraction of DM retained on the 1.18-mm screen, or larger, was used to calculate the physical effectiveness factor (**pef**) of the diets.

For purposes of the analysis, dried diets, individual feed ingredients, and fecal samples were each ground separately in a Cyclone mill (1-mm screen; model SM100; Resch GmbH, Haan, Germany). Feed samples were analyzed for ash, determined after 4 h combustion at 550°C in a muffle furnace (Vulcan 3-550, Dentsply Neytech, Burlington, NJ); aNDFom, in according with Mertens (2002), with addition of sodium sulfite; ADF, ADL (AOAC, 1990; method 973.18); and CP (AOAC, 1990; method 976.06 and 984.13).

In vitro NDF digestibility at 24h and 240h (**IVNDFD24h** and **IVNDFD240h**) was performed using the Tilley and Terry modified technique (Robertson and Van Soest, 1981; Tilley and Terry, 1963). Rumen fluid was collected from two lactating cows fed a hay-based diet (milk production = 33.2 ± 1.7 kg/d. DIM: 251 ± 2) through the rumen cannula, mixed and placed in a thermally controlled bottle (PYREX, SciLabware, Staffordshire, UK). Rumen contents were filtered through four layers of cheese cloth under constant O_2-free CO_2. 10 ml rumen fluid was added to each 150-ml Erlenmeyer flasks that had been placed in a heated (39.3°C) water bath under CO_2 positive pressure to ensure anaerobiosis. 0.5 g of ground sample was weighed into each flask before the addition of 40 ml of the buffer, as described by Goering and Van Soest (1970). Each sample was analyzed in triplicate, in two separate in vitro incubations. Sample preparation, donor cows and their diets were the same for both assays. At the end of the fermentation, the contents of each flask was analyzed to determine the aNDFom content of the residue, and filtered through crucibles (40-µm porosity) with the addition of microfiber glass filters. Residues were then treated following the procedure described by Goering and Van Soest (1970), after a 3 h drying in a forced – air oven (105°C), and hot weigh recorded of crucibles. Ash correction was made after incineration of the residue at 495°C for 3 h, followed by a second crucible hot – weigh.

Digestibility was then calculated as described in equation 1:

$$\text{IVNDFD, \% aNDFom} = [1 - (aNDFom_r - aNDFom_b) \ / \ aNDFom_i] * 100 \quad [1]$$

where $aNDFom_r$ is the residual aNDFom, $aNDFom_b$ is the blank correction, and $aNDFom_i$ represents the initial NDF. All the described terms are expressed in grams. The unavailable NDF fraction was determined after 240h in vitro fermentations, and calculated as expressed in equation 2:

$$uNDF_{240}, \%DM = (100 - IVNDFD240h) * aNDFom / 100 \quad [2]$$

where *aNDFom* is the aNDFom content of the sample, on DM basis. Potentially digestible NDF was calculated as the difference between aNDFom and $uNDF_{240}$, on DM basis. Total tract digestibility of the pdNDF was then computed as described in the equation 3:

TTdpdNDF, %pdNDF = *[100 – (uNDF$_{240}$ diet / feces) * (pdNDF feces / diet)] * 100* [3]

where *uNDF$_{240}$ diet / feces* is the ratio among dietary and fecal uNDF$_{240}$, and *pdNDF feces / diet* represents the ratio between fecal and dietary pdNDF.

In addition to that described above, length of fermentations was based on previous studies indicating 240h as the maximum extent of fiber digestion in an anaerobic environment in vitro (Fox et al., 2004; Raffrenato and Van Amburgh, 2011; Palmonari et al., 2014, 2016). For these fermentations, both rumen fluid and buffer were re-inoculated after 120h to preserve the microbial activity during the whole process, as described by Palmonari et al., (2014). A final volume of 100 ml was treated for aNDFom determination as described above.

Rumination time data (rumination/DMI, rumination time/aNDFom intake, rumination time/forage aNDFom intake, rumination/peNDF intake) were used to calculate the average daily rumination time in the period.

Rumen pH data were used to evaluate mean pH, area under the curve (the area between the observed pH and a line draw at pH 5.8 and 5.5), and time (min) under pH 5.8 and 5.5. Rumen pH of 5.8 was chosen as threshold for a subacute rumen acidosis (SARA) status and 5.5 for sever SARA. The duration (min/d) and total area (pH x min, area under the curve: AUC) that pH was below each SARA threshold were calculated to evaluate the severity of rumen acidosis. AUC was calculated by adding the absolute value of negative deviations in pH from 5.5 or 5.8 for each 10-min interval. (Dohme et al., 2008)

Body weight (BW) was used to calculate average daily gain (ADG) using the formula reported below:

(final weight (kg) – initial weight (kg))/period length (d)

the calculation was made at the end of all four periods.

Feed efficiency was computed as feed consumption adjusted for differences in gain (feed conversion ratio, FCR).

Statistical Analysis

Data recorded in the third wk of each period were analyzed using the statistical program JMP-12 software (SAS Institute Inc., Cary NC). DMI, water intake, rumination time, rumen pH and temperature, and NDF digestibility were carried out according with a mixed effects model for repeated measures. Treatment (T), period (P), day (D), treatment x period (TP), treatment x day (TD) were used as fixed effect and heifers (H) as random. The following model was used:

$$Y_{ijkl} = \mu + T_i + P_j + D_k + H_l + TP_{ij} + TD_{ik} + e_{ijkl}$$

ADG and FCR were analyzed using a post hoc Tukey's adjustment.

Data were considered significant if $P < 0.01$.

RESULTS AND DISCUSSION

Diet Characteristics, Fiber Particle Size, and Intakes

The two diets used in this trial were similar in chemical composition (Table 1). The CP (% of DM) resulted lower compared to NRC (2001) suggestions for 300 kg heifers. However, diets in the current study were formulated according to the Cornell Net Carbohydrate and Protein System (CNCPS; Higgs et al., 2015 and Van Amburgh et al., 2015) software, in which the MP and ME requirements were covered (656.4 g/d and 16.1 Mcal/d, respectively with a DMI of 8.4 kg/d). The two diets had different distribution of fiber particles (Table 2). The amount of particles retained by a 1.18-mm screen was greater in the TMR ration compared with the pelleted one (66.12% and 20.12% respectively). We used the threshold of 1.18 mm to distinguish the particles that are highly resistant to passage and consequently are able to stimulate rumination (Cardoza, 1985; Poppi et al., 1985; Mertens, 2000). PeNDF was 39.78 and 11.82 % of DM in the TMR diet and in the P diet respectively. Measurement of peNDF is important to determine the size of particles that are retained in the rumen. The minimum peNDF recommendation is 21% of the ration DM (Mertens, 2000) with consideration to the previous study in which approximately 19.7% peNDF was needed to maintain milk fat percentage of Holstein cows at 3.4%, and 22.3% peNDF was needed to maintain an average rumen pH of 6.0 (Mertens, 1997). The values of peNDF recorded in the TMR ration of this study were more than adequate to guarantee a good chewing activity, saliva production and rumen

health. Conversely, P diet was created intentionally, to have a low peNDF (11.82%) compared to recommendations. Since the study involved only the use of pre-primiparous growing animals, observations were limited primarily to rumination time and rumen pH mainly.

A treatment effect was noted on DMI (Table 3). Differences were observed in DMI at 3 h post feeding (2.70 vs. 3.25 kg in P and TMR treatment, respectively. $P < 0.01$). Greater daily DMI was noted during the consumption of the P treatment (10.80 vs. 8.40 kg; $P < 0.01$). This difference was still significant (2.88 vs. 2.23 % of BW; $P < 0.01$) even when the DMI was normalized for animal BW. The DMI resulted higher compared with values suggested by NRC (2001) for 300 kg growing heifers. This is not in line with literature, in which a low protein content could negatively affect DMI (Tedeschi et al., 2000). This could partly compensate the lower protein content in the diet, allowing similar or higher consumption of grams of protein per day compared to the NRC (2001) guidelines (756 vs. 972 g of CP/d in TMR and P diet respectively).

Water intake (L/d) was higher for the P diet (55.0 vs. 45.0 L; $P < 0.01$), but the difference disappeared when corrected for DMI. This result suggests that water intake wasn't a treatment effect, but rather, was related to the amount of DMI.

The aNDFom intake (kg/d and % of BW) were greater in P treatment compared with TMR (6.34 vs. 5.03 kg/d, 1.70 vs. 1.35 % of BW; $P < 0.01$); as were the corresponding uNDF intake (kg/d and % of BW; 1.33 vs. 1.18 kg/d, 0.36 vs. 0.32 % of BW; $P < 0.01$). The peNDF intake, consistent with previous findings, was higher in the TMR treatment (3.45 vs. 1.21 kg/d and 0.92 vs. 0.32 % of BW $P < 0.01$). Results from this trial confirm that an increase in fiber particle size in the diet has a negative impact on DMI, as reported in other studies (Allen, 2000; Kononoff et al., 2003; Kammes and Allen, 2012). The reduction of dietary fiber particle size could be considered as a strategy to decrease the DMI limiting fill effect in the reticulum-rumen when diet fiber composition could, otherwise, prevent animals from attaining an adequate DMI to reach their energy requirements (Montgomery and Baumgardt, 1965).

The ratio between $uNDF_{240h}$ intake and BW (0.36 vs. 0.32 % of BW in P and TMR treatment, respectively) was similar to values reported by other authors (Cotanch et al., 2014). In this study done with dairy cows, the ratio was 0.36 for grass hay based diet and 0.48 with alfalfa hay. Based on these data it is possible to hypothesize a minimum requirement of $uNDF_{240h}$ to assure rumen health and function.

Animal Performance

Average daily gain (ADG) was similar in the two treatments (1.1 vs. 1.0 kg in the P and TMR diet, respectively; $P = 0.94$); which was considered within the range of normality for breed, age, sex, and size of the cattle used in this study, even if it was referred to a short period of time. The optimal ADG for growing heifers is 0.8 kg/d (NRC, 2001). Higher values are associated with the delay of age at first conception and calving. Conversely, other authors (Gardner et al., 1977) reported that high ADG (1.1 kg/d) is not associated to reproductive problems, but with a reduction in milk production, primarily, in the first lactation. Still other studies reported no negative effect on milk production with an ADG higher than 0.9-1 kg/d (Gardner et al., 1988; Van Amburgh et al., 1998).

Feed conversion ratio (FCR) was similar in the two treatments (11.0 vs. 10.6 kg in P and TMR diet, respectively; $P = 0.33$).

Rumination Time and Rumen pH

Rumination time and rumen attribute data are reported in Table 4. Cows fed with P and TMR treatment ruminated 241 and 507 min/d ($P < 0.01$), respectively. Rumination time decreased during P treatment administration (-52%), as expected with the reduction of fiber particle size. This effect on rumination was observed when related to DMI, aNDFom, or forage–aNDFom (23.3 vs. 58.5 min/kg; 41.0 vs. 94.0 min/kg; 23.0 vs. 58.5 min/kg in P and TMR respectively; $P < 0.01$). Rumination time is a parameter closely related to physical and chemical characteristics of the diet (Grant et al., 1990). The difference in rumination time observed in the current study could be related to different **pef** of the diets (20.12 and 66.12% for P and TMR treatment, respectively). This

relationship was also reported in other studies (Woodford and Murphy, 1988; Mertens, 2000; Krause et al., 2002). Other authors (Teimouri Yansari et al., 2004) evaluated the impact of reduced fiber particles on chewing, rumination time, and rumen pH; however that study was conducted on dairy cows fed alfalfa hay – based diets. In the current study the reduction in rumination time had no effects on rumen pH. Recorded values were similar among the two treatments (6.10 vs. 6.11 in P and TMR, respectively; $P = 0.79$). According to other authors this effect may be related to diet composition being low in starch, high in fiber, and with an adequate uNDF intake (Yang and Beauchemin, 2007; Cotanch et al., 2014).

Short particle size, as well as reduced rumination time and saliva production, is usually associated to metabolic disorders such as sub-acute rumen acidosis (SARA). The definition of SARA is based upon rumen fluid pH (Plaizier et al., 2008). For purposes of this study we considered two different thresholds of suboptimal ideal pH: a pH below 5.8 as an indicator of fibrolytic bacteria depression, and a pH below 5.5 as the cutoff value for SARA determination, in accordance with Kleen et al., (2003). In our study, the average daily pH values (recorded every ten minutes over the experimental wk) were > 6.0 throughout the entire experimental wk. Furthermore, the pH value, expressed either as min under the critical pH thresholds (5.5 and 5.8) or the corresponding areas under the curve, did not demonstrate any significant differences between the two treatments or indicate any risk of SARA, defined to be likely to occur when rumen pH remain below 5.5 for at least 180 min/d (Plaizier et al., 2008; Kleen et al., 2003).

Neutral Detergent Fiber Digestibility

Data reported in Table 5 specify chemical composition of feces and the corresponding calculation of fiber digestibility in the gastrointestinal tract.

Fecal chemical composition of the two diets, show a similar aNDFom, ADF and ADL content; however $uNDF_{240}$ content was higher in TMR compared to P treatment (52.12 vs. 47.38 % of DM; $P < 0.01$). PdNDF results were lower in the TMR compared with the P diet (17.14 vs. 22.18 % of DM; $P < 0.01$).

In vitro NDF digestibility (**IVNDFD**) was conducted at two different time points (24 and 240 h). The IVNDFD24h was not different among the treatment (11.41 vs. 10.70 % of aNDFom in P and TMR, respectively; $P = 0.51$), while IVNDFD240h was higher in diet P compared to TMR diet (31.82 vs. 24.72 % of aNDFom; $P < 0.01$). Considering the fecal IVNDFD24h as the aNDFom fraction with potential rapid digestibility, the difference between treatments observed in the IVNDFD240h rates could hypothetically, be assigned to a slowly degradable fraction. This result suggests that fiber particle size influenced digestibility of the slowly digestible aNDFom, while having no effects on the rapidly digestible aNDFom.

Fecal slowly digestible fraction represents that fibrous material not digested in the gastro – intestinal tract. Given that this fraction was lower in the TMR treatment, a higher total tract digestibility would be expected. Total tract digestibility of the potentially digestible aNDFom (TTdpdNDF) was indeed higher in the TMR compared to the P treatment (90.25 vs. 86.82 % of pdNDF; $P < 0.01$). Data observed in the current study are consistent with those recorded by Kammes and Allen (2012). In that study, the animals were fed forage based diet chopped at two different lengths (19 vs. 10 mm). The calculated TTdpdNDF in the cited study was 90.6 and 88.7% for long and short particles based diets, respectively. However, no treatment effect was observed in that experiment. As reported in the current study, the pelletizing process could have had an effect on particle structure and density, increasing their respective passage rate.

Fiber particle size influences many aspects of rumen function and digestion kinetics. The passage rate of particles is related to their reduction in size and increase in density. The dynamic relationship between these factors define the egress from the forage mat and flow out from the rumen (Sutherland, 1988). By experimental design, the fiber particle size was higher in the TMR diet. This size difference could have resulted in an increase in rumen retention time, thereby improving de facto fiber digestion (Sejrsen et al., 2006). While the shorter particles of the P treatment could theoretically have increased surface area available for microbial attachment, and consequently more extensive rumen degradation, the same attribute of size may have also increased

313 the escape rate from the rumen, limiting potential degradation. (Lammers et al., 1996; Kaske et al.,
314 1992).

315 **CONCLUSIONS**

316 This study demonstrates that reduction of fiber particle size is a potential strategy to increase
317 DMI in young ruminants. The shorter particle size led to a reduction in rumination time, without
318 causing an adverse effect on rumen pH; furthermore the use of a pelleted diet did not affect ADG.

319 The different particle size of the treatments would be expected to impact the rate of passage
320 from the rumen, being faster for the P treatment. Due to this, total tract digestibility of pdNDF was
321 remarkably high in both treatments, although the effect of larger particle size in the TMR diet
322 resulted in a significant increase.

323 We can conclude that a complete pelleted diet, well designed to provide an adequate amount
324 of NDF, could be fed to growing ruminants without any apparent negative impact on rumen health
325 and animal productivity, at least for a short period of time. More researches considering a longer
326 growing period are needed before recommending this feeding strategy for growing heifers.
327 Moreover, future studies are required to evaluate the effectiveness of this strategy on dairy cows, in
328 particular during the transition period, when the low DMI is not sufficient to meet the increasing
329 animal requirement.

330

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332

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Table 1. Ingredients and chemical composition of a pelleted (P) and total mixed ration (TMR) treatment diet fed to heifers for ad libitum intake; the diets were formulated to be similar in chemical composition but different in physical form (evaluated as physical effectiveness factor and physically effective NDF)

	Treatment		
Item	P	TMR	SEM
Ingredients, % AF			
Grass hay	41.8	41.8	-
Barley straw	27.4	27.4	-
Corn grain	16.4	16.4	-
Sunflower meal	13.7	13.7	-
Salt (NaCl)	0.7	0.7	-
Chemical composition, % DM			
DM, % as fed	92.0	88.0	1.02
Crude protein	8.7	9.0	0.36
Ash	9.6	7.9	0.38
aNDFom[1]	58.8	60.2	0.66
ADF	40.7	41.4	0.80
ADL	8.1	8.4	0.42
IVNDFD 24h[2]	45.3	46.2	1.50
IVNDFD 240h[2]	78.4	77.3	0.73
$uNDF_{240}$[3]	12.4	14.1	0.61
Starch	15.7	15.6	1.07

[1]aNDFom = amylase- and sodium sulfite-treated NDF, corrected for ash residue.
[2]IVNDFD = in vitro NDF digestibility.
[3]$uNDF_{240}$ = unavailable NDF estimated via 240-h in vitro fermentation.

Table 2. Physical characteristics and particle size distribution of a pelleted (P) and total mixed ration (TMR) treatment diet fed to heifers for ad libitum intake; the diets were formulated to be similar in chemical composition but different in physical form (evaluated as physical effectiveness factor and physically effective NDF)

	Treatment			
Item	P	TMR	SEM	*P*-value
Particle size distribution, %[1]				
6.70, mm	0	4.86	0.28	<0.01
4.75, mm	0	8.95	0.58	<0.01
3.35, mm	1.19	11.78	0.39	<0.01
2.36, mm	4.29	12.15	0.28	<0.01
1.18, mm	14.62	28.36	0.34	<0.01
0.15, mm	60.66	31.11	0.94	<0.01
Pan	19.23	2.77	0.27	<0.01
Physical effectiveness factor[2]	20.1	66.1	5.90	<0.01
Physically effective NDF[3], % of DM	11.8	39.8	3.58	<0.01

[1]Particle size was measured using the Tyler Ro-Tap (W. S. Tyler, Mentor, OH).
[2]Physical effectiveness factor: determined as the proportion of fiber retained by the sieve with 1.18-mm pore size.
[3]Physically effective NDF: measured as the NDF content of the forages (DM basis) multiply by the physical effective factor.

513 **Table 3.** Intake characteristics of heifers (daily average) fed for ad libitum intake with diet as pellet
514 (P) or total mixed ration (TMR); the diets were formulated to be similar in chemical composition
515 but different in physical form (evaluated as physical effectiveness factor and physically effective
516 NDF)

	Treatment			
Item	P	TMR	SEM	*P*-value
DMI				
3h post feeding, kg/d	2.70	3.25	0.439	<0.01
24h post feeding, kg/d	10.80	8.40	0.451	<0.01
% of BW	2.88	2.23	0.100	<0.01
aNDFom[1] intake				
kg/d	6.34	5.03	0.267	<0.01
% of BW	1.69	1.34	0.059	<0.01
$uNDF_{240}$[2] intake				
kg /d	1.33	1.18	0.059	<0.01
% of BW	0.36	0.32	0.013	<0.01
peNDF[3] intake				
kg /d	1.21	3.45	0.118	<0.01
% of BW	0.32	0.92	0.024	<0.01
Water intake				
L/d	55.00	45.00	3.229	<0.01
L/kg of DMI	5.01	5.13	0.245	0.31

517 [1]aNDFom = amylase- and sodium sulfite-treated NDF, corrected for ash residue.
518 [2] $uNDF_{240}$ = unavailable NDF estimated via 240h in vitro fermentation.
519 [3]peNDF= physically effective NDF, computed as the NDF content of the forages (DM basis)
520 multiplied by the physical effectiveness factor.

521 **Table 4.** Rumination time and rumen condition of heifers (daily average) fed for ad libitum intake
522 with diet as pellet (P) or total mixed ration (TMR); the diets were formulated to be similar in
523 chemical composition but different in physical form (evaluated as physical effectiveness factor and
524 physically effective NDF)

	Treatment			
	P	TMR	SEM	*P*-value
Rumination				
Time, min/d	241.00	507.00	17.20	<0.01
Time/DMI per d, min/kg	23.30	58.50	1.86	<0.01
Time/NDF intake per d, min/kg	41.00	94.00	1.56	<0.01
Time/forage NDF intake per d, min/kg	23.00	58.50	0.96	<0.01
Rumen condition[1]				
Mean rumen pH	6.10	6.11	0.07	0.79
Mean rumen Temperature, °C	38.87	38.84	0.07	0.34
Time below pH 5.8, min/d	188.00	176.00	124.90	0.33
Time below pH 5.5, min/d	3.40	4.60	5.58	0.67
Area below pH 5.8, minxpH units/d	24.40	22.80	15.60	0.51
Area below pH 5.5, minxpH units/d	0.21	0.31	0.34	0.59

525 [1] pH values evaluated as described by Kleen et al., (2003)

Table 5. Fecal composition and fiber digestibility of heifers fed for ad libitum intake with diet as pellet (P) or total mixed ration (TMR); the diets were formulated to be similar in chemical composition but different in physical form (evaluated as physical effectiveness factor and physically effective NDF)

	Treatment			
Item	P	TMR	SEM	*P*-value
Chemical composition[1], % of DM				
aNDFom	69.59	69.21	0.397	0.26
ADF	57.13	54.94	0.454	0.24
ADL	26.82	27.88	0.707	0.26
$uNDF_{240}$	47.38	52.12	0.748	<0.01
pdNDF	22.18	17.14	0.817	<0.01
NDF digestibility, % of aNDFom				
IVNDFD 24h[2]	11.41	10.70	0.724	0.51
IVNDFD 240h[2]	31.82	24.72	1.128	<0.01
TTdpdNDF[3], % of pdNDF	86.82	90.25	0.652	<0.01

[1]aNDFom = amylase- and sodium sulfite-treated NDF, corrected for ash residue, $uNDF_{240}$= unavailable NDF estimated via 240h in vitro fermentation, pdNDF= potentially digestible NDF.
[2]IVNDFD = in vitro NDF digestibility.
[3]TTdpdNDF = total tract digestibility of the potentially digestible NDF.