University of Reading

Particle size distribution of forages and mixed rations, and their relationship with ration variability and performance of UK dairy herds

Article

Accepted Version

Tayyab, U., Wilkinson, R. G., Reynolds, C. K. and Sinclair, L. A. (2018) Particle size distribution of forages and mixed rations, and their relationship with ration variability and performance of UK dairy herds. Livestock Science, 217. pp. 108-115. ISSN 1871-1413 doi: https://doi.org/10.1016/j.livsci.2018.09.018 Available at http://centaur.reading.ac.uk/79818/

Particle size distribution of forages and mixed rations, and their relationship with ration variability and performance of UK dairy herds

Usama Tayyab[a], Robert G. Wilkinson[a], Christopher K Reynolds[b], Liam A. Sinclair[a*]

[a]Department of Animal Production, Welfare and Veterinary Sciences, Harper Adams University, Edgmond, Newport, Shropshire, TF10 8NB, UK.

[b]School of Agriculture, Policy and Development, University of Reading, PO Box 237, Earley Gate, Reading, RG6 6AR, Berkshire, UK.

U. Tayyab: utayyab@harper-adams.ac.uk

R. G. Wilkinson: rgwilkinson@harper-adams.ac.uk

C. K. Reynolds: c.k.reynolds@reading.ac.uk

L. A. Sinclair: lsinclair@harper-adams.ac.uk

*Corresponding author. Liam A. Sinclair

E-mail address: lsinclair@harper-adams.ac.uk

ABSTRACT

The particle size of the ration has been proposed as a key factor, along with its fibre and non-forage carbohydrate concentration, to ensure healthy rumen function and optimal performance of dairy cows. The current particle size distribution recommendations for forages and rations are primarily based on lucerne-haylage and maize silage (MS) and may not be suitable for the wetter grass silage (GS) based rations typically fed in Northern Europe. In order to characterize the particle size distribution of forages and rations in the UK, fifty commercial dairy herds feeding a range of GS and MS based rations were sampled during the winter of 2015/2016. The particle size distribution of the fresh forages and mixed rations (MR; total and partial mixed rations) were analysed using a modified Penn State Particle Separator with six screens of hole size 60, 44, 26.9, 19, 8, and 4 mm. The fresh MR was collected at 5-equally-spaced locations along the length of the feed-face for each herd within 5-min of feeding to determine the consistency of ration mixing, and again from the same locations 4h post-feeding. Grass silage was the main forage fed on 50 herds, with 80.3% of the dry matter (DM) being retained above the 19 mm sieve, which is considerably higher than the North-American recommendations for lucerne-haylage. The particle size distribution of MS followed the general recommendations for North American forages, however, the 8-19 mm fraction was higher and the <4 mm lower. The >60 mm fraction of the MR had the lowest (0.1% DM) DM retention, and the 8-19 mm fraction the highest (34.9% DM). The MR had a higher proportion of particles retained on the 26.9 mm sieve when GS was the sole forage. Fifty eight % of herds were considered to have either moderately or poorly mixed rations, whilst 66% had evidence of diet selection (either preferential consumption or selective refusals). Particle size of the MR accounted for 33% of the variance in the milk fat content and 12% of milk yield. In conclusion, the particle size distribution of the GS and MR fed on UK dairy herds is different from the current recommendations, suggesting that the particle size of UK dairy rations is too long or new guidelines using additional sieves with larger pore sizes are required. There is also a high proportion of herds with poor mixing and/or evidence of diet selection.

Key words:
Dairy cows, ration variability, diet selection, particle size distribution

1. Introduction

Feeding dairy cows with a mixed ration (MR; either total or partial mixed ration) is an effective way to provide a homogeneous and balanced diet throughout the day (Coppock et al., 1981). The composition of MR can vary considerably but ryegrass (GS) and maize silages (MS) are the main forages used in the MR fed to dairy herds in Northern Europe (Johansen et al., 2018; March et al., 2014). In order to maintain animal performance and promote a healthy rumen function the inclusion of forages with an adequate particle size and dietary concentration of non-forage carbohydrate (fibre) in the MR are required (Zebeli et al., 2012). The physically effectiveness of a ration has been proposed as the product of the particle size multiplied by its neutral detergent fibre (NDF) content, defined as physically effective fibre (*peNDF*; Mertens, 1997). Achieving the correct particle size and *peNDF* in a ration can enhance rumen function leading to an increase in the production of rumen microbes, more efficient degradation of fibre and increased milk fat content (De Brabander et al., 1999; Zebeli et al., 2012). A short forage particle size is associated with improved compaction in the bunker and can result in reduced aerobic spoilage at feed out (McDonald et al., 1991) and may increase dry matter (DM) intake, due to reduced rumen fill and increased fibre digestibility (Thomson et al., 2017). However, too short a forage particle length can increase the rate of volatile fatty acid production in the rumen, reduce rumination time, and decrease the production of saliva (Tafaj et al., 2007), with the consequence of inhibiting cellulolytic bacteria activity and increasing the risk of sub-acute ruminal acidosis (SARA; Tafaj et al., 2007). In a review of the literature, Zebeli et al. (2012) concluded that too short a particle size (and *peNDF),* increases the passage rate of digesta and rate of fibre degradation due to a higher surface area for microbial attachment. In contrast, too long a forage particle size may promote ration sorting and result in some cows receiving excess concentrates and others insufficient (Kononoff and Heinrichs, 2003).

The estimation of the particle size of forages and MR is problematic, and various methods have been proposed to characterise feed particle distribution using different sieving techniques, with no universally accepted standard. Maulfair and Heinrichs (2012) concluded that the Penn State Particle Separator was the most useful method and proposed dietary guidelines for use on-farm. These recommendations are primarily based on North American rations that consist of MS and lucerne haylage (Eastridge, 2006), and may therefore not be suitable for the typically wetter (e.g. less than 30% DM) MS and GS commonly fed in Northern Europe (Møller et al., 2000).

Heinrichs et al. (1999) reported that processing by the mixer wagon prior to feeding can also have a large effect on the consistency of the mix, and affect the particle size and *peNDF* concentration of the ration subsequently consumed. Mixing protocols have been shown to affect feed intake and milk yield, particularly in rations containing longer chop lengths (Humphries et al., 2010; Maulfair and Henrichs 2010). Consideration should therefore also be given to the effect of particle size and consistency of mixing on the degree of diet selection by dairy cows.

The primary objective of the present study was to characterise the particle size distribution and *peNDF* content of GS, MS and MR fed on UK dairy herds using a modified Penn State Particle Separator, and to compare the observed particle size distribution with current guidelines. The secondary objective of the study was to evaluate the consistency of mixing of MR and extent of sorting of GS and GS/MS based MR, and to determine the relationship between particle size and cow performance on UK dairy herds.

2. Material and methods

2.1. Herd characteristics

Fifty commercial dairy herds located throughout the UK (32 in the Midlands of England, 9 in the South of England and 9 in Southwest Scotland) that were feeding GS and/or MS were visited between January and June, 2016. The herds were randomly selected from a database supplied by the Agricultural and Horticultural Development Board, the levy body covering England, Scotland and Wales, with the provision that they were using a MR (partial or total) feeding system and had a high yielding group that contained at least 50 cows. Herds were enrolled onto the study through an initial telephone contact and questionnaire survey to determine suitability and willingness to participate. On the day of the visit a second questionnaire was completed to collect details of herd characteristics, performance levels and frequencies of fresh feed delivery, feed push up and orts removal. In addition, feeding space per cow, feed mixer make and model, forage harvester make and model, and mixing protocol were recorded. The ingredient composition of MR fed to the target group and the mean concentrate fed in the parlour was also recorded.

Out of the 50 herds, 50 fed GS, with 34 using MS in the MR. Other sources of forage being fed were; whole-crop wheat (19), wheat straw (15), fodder beet (5), grass haylage (2), whole-crop triticale (1), whole-crop barley (1), lucerne (1), pea silage (1) and oat silage (1). Forty-four of the herds had an all year around calving pattern, 4 were autumn block calving and 2 spring block calving. Holstein-Friesian was the major breed on 36 herds, with the predominant breed on the remaining herds being Ayrshire (2), Jersey (1), Brown Swiss (1), or (10) having a mixture of Holstein with other breeds (Brown Swiss, New Zealand Friesian, and Jersey) or crossbred. The main feeding system was total MR which was used on 28 herds, while the remaining 22 herds fed a partial MR with additional concentrate fed in the milking parlour. Twenty-four herds used a “tub” type mixer wagon, 18 a “barrel” type, 7 an “auger” design (vertical or horizontal) and one used a forage box.

Total herd size ranged from 75 to 2220 animals, with a mean of 354 (Table 1). The number of lactating cows ranged from 67 to 1770 cows/herd, with a mean and median of 310 and 277, respectively. The annual milk yield ranged from 6000 to 12500 kg/cow, with a mean of 9199 kg/cow (median = 9200). Annual energy corrected milk yield (corrected for milk fat and protein; Sjaunja et al., 1991) ranged from 7248 to 13209 kg/cow, with a mean of 10011 kg/cow. All herds delivered fresh feed either once or twice daily, with a mean of 1.3 times/d. Of the 50 herds, 20 were feeding the MR in a trough where there was no push up the feed. The average frequency of feed push up in the remaining 30 herds was 4.7 times/d. The mean orts removal frequency was 4.4 times/wk, with a range from 0.25 (monthly) to 7 (daily) times/wk. Feed space per cow ranged from 0.30 to 0.76 m/cow, with a mean of 0.56 m/cow. Length of feed mixing was either manually recorded or provided by the farmer, and ranged from 5 to 60 min. The number of chews per bolus was manually counted for three full bouts for 10 cows randomly selected from the feeding group sampled (Kononoff et al., 2002).

2.2. Determination of particle size and peNDF distribution of forages and MR

Where more than one feeding group was present, data were collected from the high yielding group in each herd (n = 40). Where feed was delivered more than once (n = 15), the first (morning) feed was sampled. The feed face of the high yielding group of cows (or all cows if no subdivision was present) was divided into five equal sections to determine the consistency of mixing (Sova et al., 2014). Within each feed face section, a 30 cm × 30 cm quadrat was randomly placed over the MR within 5 mins of fresh feed-out, and all material removed and thoroughly mixed by hand (0hMR; Endres and Espejo, 2010). To determine the level of diet selection (feed sorting), the MR was sampled using the quadrat from the same locations along the feed fence again four hours post feeding (4hMR; Leonardi et al., 2005). Prior to fresh feed delivery, refusals, where available, were also sampled (n = 33).

The particle size distribution of the forage (GS and MS) and MR samples were analysed on both a fresh and dried basis. A modified Penn State Particle

Separator with four screens of 26.9, 19, 8, and 4 mm was used to determine the particle size of GS and GS/MS based MR, and three screens of 19, 8 and 4 mm for MS according to the manual shaking procedure described by Kononoff et al. (2003). Perennial ryegrass (*Lolium perenne*) and MS (*Zea mays* L.) were sampled from first, second or third cut GS and MS silage bunkers as described by Sinclair (2006) and the particle size measured using the modified Penn State Particle Separator described above. The particle size distribution (%) was calculated by dividing the weight of each fraction by the sum of all fractions and multiplying by 100.

The on-farm particle size distribution analysis using one additional Penn State Particle Separator sieve screen (26.9 mm) was found to be inadequate to accurately determine the geometric mean particle size (X_m) of GS and GS based MR. Consequently, two larger sieve screens of size 44 and 60 mm were used to reanalyse particle size of 0hMR and GS using frozen and defrosted samples. The frozen samples were thawed at room temperature for 6h prior to analysis.

2.3. Chemical analysis

The DM content (AOAC, 2012; 988.05) of each fraction of 0hMR, 4hMR, refusals, GS and MS for each herd was determined by oven drying at 105°C to constant weight. Forage and MR samples were then milled in a hammer mill (Crompton Control Series 2000, Wakefield West Yorkshire UK) fitted with a 1 mm screen. The crude protein (988.05; Dumas method [N × 6.25]), ash (942.05; at 550°C for 6 h) and ether extract (920.39) was analysed as described by AOAC (2012). The NDF (using sodium sulphite and heat stable amylase, and expressed residual of ash) and acid detergent fibre (ADF) content was analysed according to Van Soest et al. (1991). The starch content of the 0hMR was analysed by Trouw Nutrition (Blenheim House, Blenheim Road, Ashbourne, Derbyshire, UK) using the procedure described by McCleary et al. (1997).

2.4. Calculations and statistical analysis

Energy corrected milk yield (kg) was calculated as: milk yield (kg) × [(38.3 × fat (g/kg) + 24.2 × protein (g/kg) + 15.71 × lactose (g/kg) + 20.7)/3,140], as described by Sjaunja et al. (1991). The geometric mean particle size (X_m) was calculated using the method described by ANSI (1992). The physical effectiveness factor (*pef*) was determined as the DM proportion of particles longer than 8 mm ($pef_{>8mm}$) or 4 mm ($pef_{>4mm}$, Lammers et al., 1996; Maulfair and Heinrichs, 2010). The $peNDF_{>4mm}$ was calculated by multiplying the NDF content (% DM) of the MR by the $pef_{>4mm}$, and $peNDF_{>8mm}$ by multiplying the NDF content (% DM) of the MR by the $pef_{>8mm}$ (Lammers et al., 1996; Mertens, 1997).

The consistency of ration mixing of each herd was calculated using the co-efficient of variation (CV%) of each particle size fraction of the 0hMR (Buckmaster et al., 2014; Oelberg and Stone, 2014; Sova et al., 2014), with a CV of >5% considered significant (Silva-del-Rio and Castillo, 2012). The CV of each fraction was weighted for the respective percentage particle size distribution and then the corrected CV summed. Herd-level diet selection was calculated for each fraction by dividing the proportion (DM basis) at 0hMR by the corresponding proportion at 4hMR and refusals, and presented as a percentage. A sorting value of 100% indicated no sorting, <100% indicated preferential consumption, and >100% indicated selective refusal.

All data were summarised by herd and tested for normality using the general descriptive statistics component of GenStat 17.1 ® (VSN International Ltd., Oxford, UK). Associations between measures of productivity (energy corrected milk yield, milk fat g/kg, milk protein g/kg), feeding management and ration characteristics were analysed using a standard linear model (i.e. ANOVA) with forage source and shaking technique as fixed effects and herds and location as random effects. A linear regression model was used to determine the association between X_m and energy corrected milk yield and milk fat using GenStat 17.1 ® (VSN International Ltd., Oxford, UK). For multiple comparisons, all fractions of the mixed ration were analysed by general ANOVA followed by a Tukey test, with the significant level set at $P < 0.05$.

3. Results

3.1. Forage proximate and physical characteristics

The mean DM of the GS was 23 g/kg lower ($P = 0.022$) and the CP 54 g/kg DM higher than the MS (Table 2). The NDF and ADF content were also 65 and 64 g/kg DM higher in the GS than the MS ($P < 0.001$). The highest % DM retention of GS was the 26.9-44 mm fraction (51.6%, $P < 0.001$), with the majority of the DM (80.3%) being longer than 19 mm. In contrast, the highest retention of DM for MS was between 8-19 mm (73.2%, $P < 0.001$). The X_m, $peNDF_{>4mm}$ and $peNDF_{>8mm}$ content was higher ($P < 0.001$) in GS than MS (mean values of 42.6 and 10.5 mm, 48 and 40%, and 47 and 34% for X_m, $peNDF_{>4mm}$ and $peNDF_{>8mm}$ for GS and MS respectively).

3.2. Mixed ration proximate and physical characteristics

The mean forage to concentrate ratio across the 50 herds was 77:23 on a fresh weight basis, and 57:43 on a DM basis, with a GS to MS ratio on the 34 herds that fed both forages of 50:50 (fresh weight basis) or 48:52 (DM basis; Table 3). The DM concentration of the MR ranged from 213 to 544 g/kg, with a mean value of 373 g/kg across the 50 herds, whilst the mean CP ranged from 116 to 205 g/kg DM, with a mean value of 160 g/kg DM. The mean and median NDF concentration of the MR was 391 and 381 g/kg DM respectively. For the MR, the lowest proportion of DM was retained on the 60 mm fraction ($P < 0.001$), with the 8-19 mm fraction having the highest proportion ($P < 0.001$), and there was no difference ($P > 0.05$) between the 44-60 and 19-26.9 mm fractions. The $peNDF_{>4mm}$ concentration of the MR ranged from 22 to 47% with a mean of 33%, and the mean $peNDF_{>8mm}$ was 73%. The mean X_m of the MR was 19.5 mm, ranging from 6.2 to 44.9 mm. The starch concentration of MR ranged from 63 to 237 g/kg DM with a mean value of 138 g/kg DM. The mean DM of the 0h, 4h and refusals did not differ ($P = 0.10$) between sampling times, and the DM concentration of the various fractions of MR did not change over time ($P > 0.05$; data not shown).

Herds that fed GS as the main forage had a higher ($P < 0.01$) proportion of the DM retained on the 26.9-44 mm fraction of the 0hMR compared to those that used a mixture of GS and MS (Table 4). In contrast, herds that used a mixture of both forages had a higher ($P < 0.01$) proportion of the DM retained on the 8-19 mm fraction. The type of mixer wagon (barrel, tub or auger) had no effect ($P > 0.05$) on the particle size distribution of any fraction of the 0hMR (data not shown). When the partial or total MR were considered separately, the proportion of longer fractions (26.9-44 and 44-60 mm) was higher ($P < 0.05$) when in the partial MR, while the shorter fractions (8-19, 4-8 and <4 mm) were highest ($P < 0.05$) when fed as a total MR (Supplementary Table S1).

3.3. Variability in mixed ration mixing

The coefficient of variation of mixing of MR was highest for the 19-26.9 and >26.9 mm fractions at 15 and 13.7% respectively, while the minimum CV of 6.4% was for the 8-19 mm fraction (Table 5). The type of wagon mixer, forage source, total MR or partial MR, and X_m had no effect ($P > 0.05$) on ration variability across all five fractions (data not shown).

3.4. Particle size distribution of mixed rations post-feeding and diet selection

Diet selection calculated between 0-4h, 4-24h and 0-24h, demonstrated that there was selective refusal of the >26.9 and 19-26.9 mm fractions and a preferential consumption of the 8-19, 4-8 and <4 mm fractions between 0-24h period (Table 6), although there was considerable variation between herds. Sorting activity calculated between 0 and 4h showed preferential consumption ($P < 0.001$) for the 4-8 and 8-19 mm fraction of the MR while the >26.9, 19-26.9 and <4 mm fractions were selectively refused. The inclusion of whole-crop wheat (n = 19) and straw (n = 15), the mixer wagon type or X_m had no effect ($P > 0.05$) on the level of feed sorting (data not shown).

3.5. Association between particle size and production

There was a positive relationship ($R^2 = 0.33$; $P = 0.004$) between X_m and mean milk fat content (g/kg) across all herds (Figure 1). The relationship was improved when Holstein-Friesian and Holstein-Friesian crosses were analysed separately (R^2 = 0.36; P < 0.001), with the R^2 being highest when Holstein-Friesian herds were analysed alone, with almost 50% of the variation in milk fat content between herds being accounted for by X_m (R^2 = 0.47; P < 0.001). In contrast, there was a negative relationship between X_m and energy corrected milk across the 50 dairy herds, accounting for 16% of the variation (P < 0.001).

3.6. Fresh vs dried particle size distribution

When dried prior to separation there was a difference in particle size distribution, with less long material and more short material than when measured fresh and then dried (Table 7 and Supplementary Table S2). For GS the >26.9 mm fraction decreased (P < 0.001), while the 8-19, 4-8 and the <4 mm fractions increased (P < 0.001) when analysed in a dried form. Similarly, the 4-8 and <4 mm fractions of the MS increased (P < 0.001) when analysed in a dried compared to a wet form. For the MR, the proportion of the >26.9 mm decreased (P < 0.001), while the proportion of the 4-8 and the <4 mm fractions increased (P < 0.01) when analysed in a dried form compared to fresh and then dried.

4. Discussion

4.1. Herd characteristics and proximate analysis

The mean annual milk yield and herd size recorded in the current study were higher than the values reported for the UK (yield of 8180 kg and 143 cows/ herd, respectively; AHDB, 2016). This difference may be due in part to the selection criteria for the current study, with all herds recruited feeding MR and using GS, MS or a mixture as the main forage source. As a consequence, spring calving, grazed grass based herds that have a lower mean milk yield (AHDB, 2016; Garcia and Holmes, 1999) were not used, although the trend in the UK is for more continuous housing, indoor feeding rather than grazing (March et al., 2014).

The MS being fed in the current study had a lower DM content at 300 g/kg compared to the 395 g/kg reported by Lammers et al. (1996) in the northeast of the United States of America (USA). The nutrient composition of the GS used in the current study was, however, typical of European ryegrass silage (Møller et al., 2000), with a mean CP of 136 g/kg DM and NDF of 492 g/kg DM. The mean forage to concentrate ratio of the MR in the current study (57:43 DM basis) was higher than that reported for 50 herds in Minnesota (52:48, Endres and Espejo, 2010). A higher forage to concentrate ratio is more likely to maintain an efficient rumen function and should minimise the risk of SARA (Zebeli et al., 2012). However, twenty four out of the 50 herds fed a lower proportion of forage in the MR than the minimum of 56% proposed by Zebeli et al. (2012), and may subsequently have been at risk of SARA.

The average DM of the MR in the current study of 373 g/kg was lower than that reported by Eastridge (2006) and Sova et al. (2013) for typical North American rations. In similar cross-sectional studies, Sova et al. (2013) reported a mean total MR DM of 477 g/kg in 22 Canadian herds, while Endres and Espejo (2010) reported a mean of 523 g/kg DM in the total MR of 50 herds in Minnesota, USA. Rations with a high DM content may increase DM intake, but may also encourage cows to sort (Leonardi et al., 2005). The CP content of the MR in the current study was also lower compared to that of 50 herds in the USA (175 g/kg DM; Endres and Espejo, 2010) or 22 herds in Canada (165 g/kg DM; Sova et al., 2013). This difference may be due to the greater use of concentrates and lower use of forages in North American rations as reflected in the lower forage to concentrate ratio (Endres and Espejo, 2010). The average NDF content of the MR in the current study was approximately 90 g/kg DM higher than that reported in the USA (298 g/kg DM; Endres and Espejo, 2010) or Canadian rations (313 g/kg DM; Sova et al., 2013). This was probably due to the greater use of forage in the current study, especially GS, which has a higher NDF concentration than MS or lucerne haylage (Hoffman et al., 1993), but may also be affected by maturity at harvesting which increases NDF concentration (Dawson et al., 2002). The higher concentration of NDF in the MR along with a sufficient particle size are associated with a more efficient rumen function for fibre degrading microbiota by resisting a depression in rumen pH (Zebeli et al., 2012). Similarly, the ADF content was approximately 50 g/kg DM higher in the current study compared to that fed in the USA (198 g/kg DM; Endres and Espejo, 2010) or Canadian rations (205 g/kg DM; Sova et al., 2013), but was typical of Northern European rations (Johansen et al., 2018).

4.2. Ration physical characteristics

The particle size distribution of MS followed the general guidelines suggested by Heinrichs (2013) of 3-8% above 19 mm, 45 to 65% between 8-19 mm, 20 to 30% between 4 and 8 mm, and <10% below 4 mm although the 8-19 mm fraction of MS in the current study was higher than that reported by Maulfair et al. (2010). This difference may be due to the higher moisture content of MS used in the UK that promotes the adherence of shorter particles, but may also reduce sorting (Leonardi et al., 2005). Overall, the particle size distribution of MS in the UK was similar to the current guidelines for MS based on North America rations, and consequently, there is little requirement for additional research or separate recommendations for UK and northern European MS. Out of the 50 herds used in the current study, the minimum % DM of GS retained on the >19 mm sieve was 49%, considerably higher than the 10-20% guidelines for lucerne haylage in the USA (Heinrichs, 2013). Feeding a longer particle size may result in a higher rumen pH and avoid SARA, but is also associated with a reduction in feed intake due to a greater rumen fill (Tafaj et al., 2007; Zebeli et al., 2012).

The mean particle size distribution of the 0hMR in the current study differed from the guidelines based on North American rations (Heinrichs, 2013), with the long (>19 mm) particle size distribution being 38%, approximately 50% higher than that reported by Sova et al. (2013), DeVries et al. (2011) or Hosseinkhani et al. (2008), and approximately 4 times higher than that reported by Heinrichs (2013), Endres and Espejjo (2010), Miller-Cushon and DeVries (2009), or Heinrichs and Kononoff, (1996) (Supplementary Table S3). The difference in particle size distribution of MR in the current study reflected the high inclusion of GS that contained a very long particle size (>19 mm = 80% DM, X_m = 42.6 mm). The use of other forages (e.g. whole-crop wheat, wheat straw, fodder beet) in the MR in the current study did not significantly affect the particle size distribution of the MR, and supports that the high proportion of GS in the ration was the major factor causing the differences. The higher proportion of the 26.9-44 and 8-19 mm particle fractions in the MR may also be explained by the high moisture content, as 4-8 and <4 mm particles may have adhered to longer particles (Leonardi et al., 2005). However, the considerably longer particle size of GS than lucerne haylage based MR suggests that either the particle size of UK dairy rations is too long or the need for more specific particle size measurement methods and distribution recommendations when wetter GS is the major forage in the MR.

When GS was the sole forage in the MR, rations had a higher proportion of the 26.9-44 and 44-60 mm fractions which may promote ration sorting (DeVries et al., 2007), although in the current study there was no relationship between Xm and degree of sorting after 4 or 24 h. The additional 26.9, 44 and 60 mm pore size sieves used in the Penn State Particle Separator in the current study allowed a more even distribution of particle size for GS and MR samples than the traditional Penn State Particle Separator. However, as a very small proportion of particles was retained on the 19-26.9 mm screen, a screen larger than 26.9 mm may be more appropriate.

4.3. Variability in ration mixing

Feeding MR is an effective method to provide all the required nutrients to dairy cows, and a properly mixed ration ensures a uniform delivery of all feed ingredient to the animal (Coppock et al., 1981). Mixer wagons and mixing protocols can however, influence particle size distribution and result in differences in feed intake and milk yield, particularly for rations with longer chop lengths (Humphries et al., 2010). Heinrichs et al. (1999) also reported that processing by the mixer wagon prior to feed-out can have a large effect on the particle size and *peNDF* subsequently fed and the consistency of the mix. In a survey of Iranian herds, Esmaeili et al. (2016) reported a high variability (CV >10%) in particle size distribution of MR with the highest variation recorded for the >19 mm fraction, a finding in agreement with the current study. There were 42% of herds that had a CV ≤5% (indicating a well-mixed ration), 26% that had a CV of between 5-10% (moderately mixed), and 32% that had a CV >10% (poorly mixed ration). There was no effect of mixer model on overall ration variability across all herds. In contrast, Heinrichs et al. (1999) reported that MR processing by the mixer wagon can have a significant effect on the ration consistency, particle size and *peNDF* concentrations of the ration subsequently consumed.

4.4. Herd level diet selection

Herd level diet selection was calculated as the proportional change in each fraction of the MR over time post-feeding. Feed sorting activity is usually associated with the preferential consumption of fine starch or protein rich particles in the ration (DeVries et al., 2007). However, in the current study, there were selective refusals for the >19 mm fraction and preferential consumption for the <8 mm fraction. To more easily determine the variability of diet selection across herds, the long fractions (>60, 44-60, 26.9-44 and 19-26.9 mm) were summed (>19 mm), and the short (4-8 and <4 mm) fractions summed (<8 mm), while assuming that a sorting value of 100% ± 5 indicated no sorting, >105% indicated selective refusal and a sorting value of <95% indicates preferential consumption. Of the 50 herds, 82% had either selective refusal or did not show preferential consumption for the >19 mm fraction which may be associated with the inclusion of long particles of GS. There was no sorting activity observed for the <8 mm fraction in 46% of the herds. As discussed previously, this may have been due to the comparatively high moisture content of the MR in the current study that caused the cohesion of smaller particles to larger particles making it more difficult to sort (Beauchemin, 1991; Fish and DeVries, 2012; Leonardi et al., 2005).

4.5. Associative effects of particle size and production

Several authors have reported a relationship between *peNDF* and milk performance (Tafaj et al., 2007; Zebeli et al., 2012). In the current study there was also a positive relationship between $peNDF_{>4mm}$ or $peNDF_{>8mm}$ and milk fat content ($R^2 = 0.14$ and $R^2 = 0.16$; $P < 0.01$, respectively), but these were not as strong as with X_m, although due to the nature of the data caution should be exercised when interpreting the results. The positive relationship between X_m and milk fat content, and the negative relationship with milk yield is in agreement with De Brabander et al. (1999). A long fibrous particle size is associated with an increase of acetic acid production in the rumen that can subsequently lead to a higher milk fat content (Merten, 1997). Alternatively, a higher fibre ration may increase rumen pH and reduce the ruminal production of *trans*-10, *cis*-12 conjugated linoleic acid that has been associated with milk fat reduction (Harvatine and Bauman, 2011). Contrary to our findings, Tafaj et al. (2007) reported no correlation between particle size and milk yield or milk components and suggested that any effect of particle size on milk yield mainly depends on its influence on DM intake, which was not measured in the current study.

4.6. Comparison of fresh and dry separation

Compared with when measured fresh, the particle size distribution of dried forages and MR differed, with the proportion of longer fractions decreasing while short fractions increased after drying of samples (Kononoff et al., 2003). This difference may be attributed to the wetter forages and rations used resulting in adherence of short particles to larger particles, or the physical reduction in particle size due to the shaking when undertaken dry. It is therefore recommended to partially or completely dry the forages and MR before analyses in order to overcome the moisture variation (Heinrichs, 2013). However, this may not be a practical way of measuring particle size of wetter forages and MR on-farm.

5. Conclusions

The particle size distribution of GS and MR based on GS in UK dairy herds was found to be considerably higher than current guidelines that are based on North American forages and rations. This suggests that the particle size of UK dairy rations is either too long, or that new guidelines or methods of particle size evaluation for GS and GS/MS based MR in Northern Europe are required. The poor consistency of mixing and high degree of selection recorded on the majority of herds is of concern, and further research into reasons for this variation and its impact on cow performance is required. Finally, the high use of concentrates by 50% of the herds in the current study is a potential threat to SARA and reiterates the need for more appropriate means of particle size characterisation and guidelines for wetter, GS based dairy rations, with further controlled studies required to determine the optimal particle size distribution of these rations.

Acknowledgments

This research was financed by AHDB. The authors would like to thank S. Parsons and G. Vince for helping in the modification of Penn State Particle Separator and A. Harrison for assistance with data collection.

6. References

AHDB, 2016. Statistics of UK dairy industry. http://dairy.ahdb.org.uk/market-information/#.V4zZrfkrKUk (accessed 18 July 2016).

ANSI, (American National Standards Institute), 1992. ANSI/ASAE S424.1 MAR1992 (R2012): Method of determining and expressing particle size of chopped forage materials by screening. St. Joseph, MI: ASAE.

AOAC. 2012. Official methods of analysis, 19th Ed. AOAC Int., Washington DC.

Beauchemin, K.A., 1991. Ingestion and mastication of feed by dairy cattle. Vet. Clin. N. Am-Food A. 7, 439-463.

Buckmaster, D.R., Wang, D., Wang, H., 2014. Assessing uniformity of total mixed rations. Appl. Eng. Agric. 30, 693-698.

Coppock, C.E., Bath, D.L., Harris, B., 1981. From feeding to feeding systems. J. Dairy Sci. 64, 1230-1249.

Dawson, L.E.R., Kirkland, R.M., Ferris, C.P., Steen, R.W.J., Kilpatrick, D.J., Gordon, F.J., 2002. The effect of stage of perennial ryegrass maturity at harvesting, fermentation characteristics and concentrate supplementation, on the quality and intake of grass silage by beef cattle. Grass and Forage Sci. 57, 255-267.

De Brabander, D.L., De Boever, J.L., Vanacker, J.M., Boucque, Ch.V., Botterman, S.M., 1999. Evaluation of physical structure in dairy cattle nutrition. In: Garnsworthy, P.C., Wiseman, J. (Eds.), Recent Advances in Animal Nutrition. Nottingham University Press, pp. 111–145.

DeVries, T.J., Beauchemin, K. A., Von Keyserlingk, M.A.G., 2007. Dietary forage concentration affects the feed sorting behavior of lactating dairy cows. J. Dairy Sci. 90, 5572-5579.

DeVries, T.J., Holtshausen, L., Oba, M., Beauchemin, K.A., 2011. Effect of parity and stage of lactation on feed sorting behavior of lactating dairy cows. J. Dairy Sci. 94, 4039-4045.

Eastridge, M.L., 2006. Major Advances in Applied Dairy Cattle Nutrition. J. Dairy Sci. 89, 1311-1323.

Endres, M.I., Espejo, L.A., 2010. Feeding management and characteristics of rations for high-producing dairy cows in freestall herds. J. Dairy Sci. 93, 822-829.

Esmaeili, M., Khorvash, M., Ghorbani, G.R., Nasrollahi, S.M. and Saebi, M., 2016. Variation of TMR particle size and physical characteristics in commercial Iranian Holstein dairies and effects on eating behaviour, chewing activity, and milk production. Livest. Sci. 191, 22-28.

Fish, J.A., DeVries, T.J., 2012. Short communication: Varying dietary dry matter concentration through water addition; Effect on nutrient intake and sorting of dairy cows in late lactation. J. Dairy Sci. 95, 850-855.

Garcia, S.C., Holmes, C.W., 1999. Effects of time of calving on the productivity of pasture-based dairy systems: A review. New. Zeal. J. Agr. Res. 42, 347-362.

Harvatine. K.J., Bauman, D.E., 2011. Characterization of the acute lactational response to trans-10, cis-12 conjugated linoleic acid. J. Dairy Sci. 94, 6047-6056.

Heinrichs, A.J., Buckmaster, D.R., Lammers, B.P., 1999. Processing, mixing and particle-size reduction of forages for dairy cattle. J. Anim. Sci. 77, 180-186.

Heinrichs, J., 2013. The Penn State Particle Separator. Penn State University. http://extension.psu.edu/animals/dairy/nutrition/forages/forage-quality-physical/separator/extension_publication_file (accessed 19 July 2016).

Hoffman, P.C., Sievert, S.J., Shaver, R.D., Welch, D.A., Combs, D.K., 1993. In situ dry matter, protein, and fiber degradation of perennial forages. J. Dairy Sci. 76, 2632-2643.

Hosseinkhani, A., DeVries, T.J., Proudfoot, K.L., Valizadeh, R., Veira, D.M., von Keyserlingk, M.A.G., 2008. The effects of feed bunk competition on the feed sorting behavior of close-up dry cows. J. Dairy Sci. 91, 1115-1121.

Humphries, D.J., Beever, D.E., Reynolds, C.K., 2010. Adding straw to a total mixed ration and the method of straw inclusion affects production and eating behaviour of lactating dairy cows. Proceedings of the British Society of Animal Science and Agricultural Research Forum, page 95.

Johansen, M., Lund, P., Weisbjerg, M.R., 2018. Feed intake and milk production in dairy cows fed different grass and legume species: a meta-analysis. Animal 12, 66-75.

Kononoff, P.J, Lehman, H., and Heinrichs, A., 2002. Technical note- A comparison of methods used to measure eating and ruminating activity in confined dairy cattle. J. Dairy Sci. 85, 1801-1803.

Kononoff, P.J., Heinrichs, A.J., Buckmaster, D.R., 2003. Modification of Penn State forage and total mixed ration particle separator and the effects of moisture content on its measurements. J. Dairy Sci. 86, 1858-1863.

Lammers, B.P., Buckmaster, D.R., Heinrichs, A.J., 1996. A simple method for the analysis of particle sizes of forage and total mixed rations. J. Dairy Sci. 79, 922–928.

Leonardi, C., Giannico, F., Armentano, L.E., 2005. Effect of water addition on selective consumption (sorting) of dry diets by dairy cattle. J. Dairy Sci. 88, 1043-1049.

March, M.D., Haskell, M.J., Chagunda, M.G.G., Langford, F.M., Roberts, D.J., 2014. Current trends in British dairy management regimens. J. Dairy Sci. 97, 7985-7994.

Maulfair, D.D., Heinrichs, A.J., 2010. Technical note: Evaluation of procedures for analyzing ration sorting and rumen digesta particle size in dairy cows. J. Dairy Sci. 93, 3784-3788.

Maulfair, D.D., Heinrichs, A.J., 2012. Methods to measure forage and diet particle size in the dairy cow. Prof. Anim. Sci. 28, 489-493.

Maulfair, D.D., Zanton, G.I., Fustini, M., Heinrichs, A.J., 2010. Effect of feed sorting on chewing behavior, production, and rumen fermentation in lactating dairy cows. J. Dairy Sci. 93, 4791-4803.

McCleary, B.V., Gibson, T.S., Mugford, D.C., 1997. Measurement of total starch in cereal products by amyloglucosidase-a-amylase method: Collaborative study. J. AOAC Int. 80, 571-579.

McDonald P., Henderson, A.R., Heron, S.J.E., 1991. The Biochemistry of Silage. 2nd edition. Chalcombe Publications, Marlow, Bucks, UK.

Mertens, D.R., 1997. Creating a system for meeting the fiber requirements of dairy cows. J. Dairy Sci. 80, 1463-1481.

Miller-Cushon, E.K., DeVries, T.J., 2009. Effect of dietary dry matter concentration on the sorting behavior of lactating dairy cows fed a total mixed ration. J. Dairy Sci. 92, 3292-3298.

Møller, J., Thøgersen, R., Kjeldsen, A., Weisbjerg, M.R., Søegaard, K., Hvelplund, T., Børsting, C.F., 2000. Feedstuff tables. (English version) The NationalCommittee on Cattle Husbandry, Report no. 91.

Oelberg, T.J., Stone, W., 2014. Monitoring total mixed rations and feed delivery systems. Vet. Clin. N. Am-Food A. 30, 721-744.

Silva-del-Rio, N., Castillo, A.R., 2012. Degree of agreement between the ration formulated and the ration fed on seven California dairies. J. Dairy Sci. 95, (E. Suppl. 2):579. (Abstr.).

Sinclair, L.A., 2006. Effect of sample position within a clamp on the nutritive value of fermented and urea-treated whole crop wheat. Proceedings of the British Society of Animal Science, 44.

Sjaunja, L.O., Baevre, L., Junkkarinen, L., Pedersen, J., Setala, J., 1991. Measurement of the total energy content of cow's milk and the energy value of milk fat and milk protein. Eaap Public, 50, 52-155.

Sova, A.D., LeBlanc, S.J., McBride, B.W., DeVries, T.J., 2013. Associations between herd-level feeding management practices, feed sorting, and milk production in freestall dairy farms. J. Dairy Sci. 96, 4759-4770.

Sova, A.D., LeBlanc, S.J., McBride, B.W., DeVries, T.J., 2014. Accuracy and precision of total mixed rations fed on commercial dairy farms. J Dairy Sci. 97, 562-571.

Tafaj, M., Zebeli, Q., Baes, C., Steingass, H., Drochner, W., 2007. A meta-analysis examining effects of particle size of total mixed rations on intake, rumen digestion and milk production in high-yielding dairy cows in early lactation. Anim. Feed Sci. Tech. 138, 137-161.

Thomson, A.L., Humphries, D.J., Jones, A.K., Reynolds, C.K., 2017 The effect of varying proportion and chop length of lucerne silage in a maize silage-based total mixed ration on diet digestibility and milk yield in dairy cattle. Animal*,* 11, 2211-2219.

Van Soest P.J., Robertson J.B., Lewis B.A., 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74, 3583-3597.

Zebeli, Q., Aschenbach, J.R., Tafaj, M., Boghun, J., Ametaj, B.N., Drochner, W., 2012. Review: Role of physically effective fiber and estimation of dietary fiber adequacy in high-producing dairy cattle. J. Dairy Sci. 95, 1041-1056.

Figure captions

Figure 1. Relationship between mean particle size of MR (X_m, mm) and milk fat (g/kg/herd) across 50 herds containing Holstein Friesian (HF; ●=36), Ayrshire (▲=2), Jersey (**+**=1), Brown Swiss (■=1) and Holstein crossbred (HFX; ♦=10).

Figure 2. Relationship between mean particle size of MR (X_m, mm) and energy corrected milk (ECM; Sjaunja et al., 1991) across 50 herds containing Holstein Friesian (HF; ●=36), Ayrshire (▲=2), Jersey (**+**=1), Brown Swiss (■=1) and Holstein crossbred (HFX; ♦=10).