Composition of herbage consumed in mixed sequential grazing of cows with ewes as followers

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ABSTRACT
The aim of this study was to evaluate whether botanical, morphological and nutritional compositions of the herbage ingested by the cows are affected by age of pastures and grazing system. Mixed sequential grazing of dairy Holstein cows with Pelibuey breeding ewes as followers (MixG) and single species cow grazing (CowG) were the systems evaluated. Animals grazed on pastures of alfalfa (Medicago sativa L) and orchard grass (Dactylis glomerata L) in their first and second year (young) or third to fifth year (mature). During spring-summer, the proportion of alfalfa in herbage ingested by the cows was 20% higher (P < 0.01) in young than in mature pastures. Conversely, that of orchard grass was 43% higher (P < 0.05) in mature pastures. The NDF and ADF contents were 9 and 13% lower (P < 0.05) under MixG than CowG. During autumn-winter, CP content was not affected by grazing system or age of pastures. During spring-summer, CP content was 8% higher in MixG than CowG. Sequential mixed grazing of dairy cows and breeding ewes improved the composition of herbage ingested by the cows.

Abbreviations: ADF, acid detergent fiber; CowG, cow grazing; CP, crude protein; DM, dry matter; LW, live weight; MixG, mixed sequential grazing; NDF, neutral detergent fiber

1. Introduction
Extensive low-production breeding systems limit the growth of sheep production in Mexico, while low milk prices endanger the feasibility of small-scale Mexican dairy systems (Améndola et al. 2006). To combat such situations of narrowing profit margins, the development of grazing management guidelines aimed at achieving a more efficient use of grassland resources is required (Fraser et al. 2007).

In this regard, Wright et al. (2001) asserted that the adoption of sequential grazing enables the manipulation of sward structure and composition via the selection of an appropriate sward height and species of grazer, which can be used to increase the performance of the species involved in the subsequent grazing cycle. Whereas the nutrient requirements of grazing breeding ewes are low for around seven months of the year (Nicol and Brookes 2007), it is well known that highly productive grazing dairy cows require high intake rates of excellent quality herbage. Thus, it is possible to hypothesize that a mixed sequential grazing system involving breeding ewes as followers could provide an opportunity to increase the sustainability of small-scale dairy and sheep breeding systems.

Indeed, Putfarken et al. (2008) reported foraging habits of cattle and sheep to be quite complementary. Sormunen-Cristian et al. (2012) concluded that mixed cattle-sheep grazing improved pastures by harvesting to a shorter level and providing better quality forage. Concerning this, Jiménez-Rosales et al. (2018) found benefits of mixed sequential grazing of dairy cows with breeding ewes as followers due to positive changes in the sward; these changes resulted in the attainment of breeding sheep production without negative effects on herbage intake of cows. Dairy cow performance depends on pasture nutritive value; thus, cows may profit from ewes grazing as followers in the previous cycle on the creation of a short vegetation and hence a dense and digestible sward (Wright et al. 2001).

The objective of the present study was to evaluate, under small-scale Mexican farm conditions, the hypothesis that the mixed sequential grazing of dairy cows and breeding ewes is beneficial based on the latter grazing to a shorter stubble, thereby improving the composition of the herbage consumed by the cows during the subsequent grazing cycle.

2. Material and methods
The experiment took place during spring–summer 2013 and autumn–winter 2013–2014 at Chapingo University, México, located at 19° 29′ N, 98° 54′ W and an altitude of 2240 m, under a temperate sub-humid climate with summer rains. The general aspects of the methodology used here, have been previously reported by Jiménez-Rosales et al. (2018).
2.1. Pastures and animals

Alfalfa (Medicago sativa L.) and orchard grass (Dactylis glomerata L.) mixed pastures were used. A total of six 0.53 ± 0.03 ha paddocks were employed in spring-summer (total area 3.2 ha), and nine 0.49 ± 0.03 ha in autumn-winter (total area 4.4 ha). Given the negative effect of age on the productivity of alfalfa-based pastures (Dear et al. 2007), paddocks were classified as "young" pastures, in their first and second year (3 paddocks in spring-summer and 6 paddocks in autumn-winter), or as "mature" pastures in their third to fifth year (3 paddocks in spring-summer and 3 paddocks in autumn-winter). The experimental animals comprised 12 (spring-summer) and 16 (autumn-winter) New Zealand Holstein Friesian cows with live weights (LW) of 500 ± 77 (spring-summer) and 537 ± 64 (autumn-winter) kg, and 24 yearling gestating (spring-summer) and lactating (autumn-winter) Pelibuey ewes of 43 ± 7 kg initial LW as followers.

2.2. Treatments and experimental design

Two treatments were applied: single-species cow grazing (CowG) and mixed sequential grazing (MixG) with ewes as followers. The experimental design was completely randomized with two replicates. During spring-summer, the experimental units of CowG were two groups of three cows; while those of MixG were two groups of three cows and twelve ewes. During autumn-winter, the experimental units of CowG were two groups of four cows; while those of MixG were two groups of four cows and twelve ewes. The experimental units included the respective grazed areas. In both seasons all animals grazed in the same paddock, which in each case was divided into four equally sized plots, two plots were grazed by two groups of cows (CowG) and the remaining two were grazed by two groups of cows followed by two groups of ewes (MixG); as above stated, allotment of groups to plots within paddocks was at random.

2.3. Grazing and animal management

Cows were milked at 06:30 and 15:30 h, after which they received 1.6 kg cow⁻¹ of concentrate and remained in the paddocks. During autumn-winter, the cows were also fed with 2.1 kg dry matter (DM) cow⁻¹ d⁻¹ of maize silage. Ewes grazed between 08:00 and 17:00 h and (for security reasons) were penned overnight with access to water and mineral supplement. During autumn-winter the ewes were lactating and hence were supplemented with 0.4 kg DM ewe⁻¹ d⁻¹ of maize silage.

The grazing management was rotational and took place per paddock, i.e. it occurred simultaneously in the four plots of each paddock. Grazing areas were allotted based on daily herbage allowance to cows, and daily areas within the plots were assigned using polywire temporary electric fences. In the MixG treatment, ewes grazed as followers one day after grazing by cows. Prior to the experiment, one adaptation grazing cycle took place in all paddocks. Afterward, measurements were carried out during the second grazing cycle of all paddocks between 18th July and 23rd August (spring-summer). After 23rd August and until 2nd December, grazing management continued as described above; and thereafter up to 21st February, the second measurement cycle (autumn-winter) took place. Because areas of the paddocks were not exactly the same, there were slight differences in grazing and rest periods, with on average six (spring-summer) and four (autumn-winter) d grazing, and 37 (spring-summer) and 45 (autumn-winter) d resting.

2.4. Variables measured or calculated

The estimated composition of herbage consumed by cows and ewes was based on analyses of samples obtained by an adaptation of the hand-plucking (Bonnet et al. 2011) method. Samples were taken in each plot during grazing of all paddocks, during the spring-summer and autumn-winter sampling periods. Huge variations in the composition of intake and intake rate that occur in the strip-grazing method which was used here (Amendola 2002), preclude the utilization of the method as described by Bonnet et al. (2011). The adaptation consisted of sampling during the third day of grazing, carefully observing the composition of the residual herbage on the second day of grazing and mimicking it by taking approximately twenty subsamples in the ungrazed area; when sampling for the composition of herbage ingested by ewes, the “ungrazed” area to be sampled was the residual left by the cows. The composed samples were divided each in two subsamples for the estimation of the chemical and the botanical plus morphological compositions.

Herbage botanical and morphological compositions were estimated based on hand separation and drying of components (Whalley and Hardy 2000) in a forced air oven at 100°C until constant weight. Subsamples used to estimate herbage chemical composition were initially dried until constant weight in a forced air oven at 55 °C, and then milled in a Wiley® 4 mill (Arthur H. Thomas, Philadelphia, PA, USA) with a 1 mm mesh. Ash content was estimated following the AOAC official method, modified by Thiex et al. (2012). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were sequentially determined via the filter bag technology described by Ferreira and Mertens (2007) in an ANKOM 200 Fiber Analyzer (Macedon NY, USA). Crude protein (CP = N*6.25) was determined using a 2400 Series II CHNS/O Elemental Analyzer (PerkinElmer® Waltham, MA, USA). Daily milk production per cow was electronically measured (DeLaval MM27BC Milk Meter®, Stockholm, Sweden) once weekly.

Herbage intake of cows per ha and d of the grazing cycle on both treatments was calculated as the product of average intake of the group (measured using chromium oxide and acid insoluble ash as reported by Jiménez-Rosales et al. (2018) by the area allotted, considering the total length of the grazing cycle. Herbage intake of ewes in MixG was calculated as the difference of residual herbage of cows and residual herbage of ewes multiplied by the area allotted. Total intake in MixG was obtained by adding intake of cows and ewes.

2.5. Statistical analyses

Analyses of variance regarding the composition of the herbage ingested by cows were carried out using models that included
the fixed effects of treatment (CowG and MixG), paddock and their interaction. Analyses of the composition of herbage eaten by ewes were performed using models that included the fixed effect of the paddock. Independent analyses were carried out per season using the GLM procedure of SAS (SAS 2004). For both traits, a contrast test was used to compare treatments (composition of the herbage ingested by cows), and young and mature pastures (composition of the herbage ingested by cows and ewes).

The analysis of milk production was performed with the MIXED procedure, considering as a fixed effect treatment and the random effect of cow within the interaction among day and treatment. Herbage intake per hectare was analyzed using a model with fixed effects of age of pastures (young and mature), species and grazing method (cows plus ewes, cows in MixG and CowG), and their interaction using the GLM procedure.

3. Results

3.1. Composition of herbage consumed by cows

In the companion paper, Jiménez-Rosales et al. (2018) reported a higher proportion of alfalfa in the herbage consumed by cows under MixG than in CowG, and conversely a higher proportion of orchard grass under CowG than in MixG. Least-square means of the proportions of species in herbage ingested by cows on pastures of different age are shown in Table 1; Jiménez-Rosales et al. (2018) addressed the effect of grazing system on the botanical composition of herbage consumed by cows. Alfalfa levels were 20% higher ($P < 0.001$) in young than in mature pastures, while the opposite occurred with orchard grass, whose content was 30% higher ($P < 0.001$) in mature than in young pastures.

The morphological composition of the herbage eaten by cows (Table 2) corresponded, in general, with its botanical composition. During spring-summer, alfalfa leaf and stem levels were 25 and 27% higher ($P < 0.001$) under MixG; conversely, orchard grass leaf content was 63% higher ($P < 0.001$) under CowG than MixG. In autumn-winter, alfalfa leaf content was 16% higher ($P < 0.001$) in young than in mature pastures. Grazing system did not affect the proportion of alfalfa stems in young pastures, in mature pastures the levels were 63% higher under MixG than under CowG. There was no effect of grazing system on the proportion of orchard grass leaves in young pastures; orchard grass levels in mature pastures were 58% higher under CowG than under MixG.

### Table 2. Morphological composition (% dry matter) of herbage consumed by cows under two grazing systems, single-species cow grazing (CowG) and mixed sequential grazing of dairy cows with breeding ewes as followers (MixG), on alfalfa and orchard grass pastures of different ages, young (one and two years old) and mature (three to five years old).

| Season          | Component     | Grazing system | MixG        | CowG       | SE$^3$ | $P^p$ |
|-----------------|---------------|----------------|-------------|------------|--------|-------|
| Spring–summer   | Leaves Alfalfa| MixG           | 36.3        | 29.3       | 0.92   | 0.002 |
|                  | Stem Alfalfa  | MixG           | 22.6        | 36.9       | 1.25   | <0.001|
|                  | Leaves Orchard grass | MixG | 37.6        | 29.6       | 1.22   | 0.005 |
|                  | Stem Orchard grass | MixG | 2.6         | 3.6        | 0.58   | 0.238 |
| Autumn–winter   | Leaves Alfalfa| MixG           | 50.3        | 43.0       | 1.11   | 0.002 |
|                  | Stem Orchard grass | MixG | 14.1        | 25.8       | 1.23   | <0.001|
|                  | Inflorescence Alfalfa | MixG | 0.4         | 0.4        | 0.18   | 0.981 |
|                  | Stem Orchard grass | MixG | 31.1        | 26.1       | 1.06   | 0.004 |

Note: $^3$ SE, standard error of means; $^p$ $P$, probability of differences.

The effects of grazing system and pasture age on the chemical composition of herbage consumed by cows are reported in Table 3. During spring-summer, CP content was 8% higher in MixG than under CowG ($P = 0.027$), while NDF and ADF contents were respectively 10% ($P < 0.001$) and 9% ($P < 0.001$) lower under MixG than under CowG; however, grazing system did not affect the ash content ($P = 0.187$). During autumn-winter there was no effect regarding CP and ash contents ($P = 0.128$ and 0.510), NDF and ADF contents were respectively 10% and 9% ($P < 0.001$) lower under MixG than under CowG.

### Table 3. Chemical composition (% dry matter) of herbage consumed by cows under two grazing systems, single-species cow grazing (CowG) and mixed sequential grazing of dairy cows with breeding ewes as followers (MixG), on alfalfa and orchard grass pastures of different ages, young (one and two years old) and mature (three to five years old).

| Season          | Component     | Grazing system | MixG        | CowG       | SE$^3$ | $P^p$ |
|-----------------|---------------|----------------|-------------|------------|--------|-------|
| Spring–summer   | Crude Protein | MixG           | 25.64       | 23.80      | 0.52   | 0.027 |
|                  | Neutral Detergent Fiber | MixG | 41.13       | 45.55      | 0.64   | 0.004 |
|                  | Acid Detergent Fiber | MixG | 23.24       | 25.52      | 0.27   | <0.001|
| Autumn–winter   | Crude Protein | MixG           | 10.81       | 11.18      | 0.19   | 0.187 |
|                  | Neutral Detergent Fiber | MixG | 25.83       | 24.60      | 0.54   | 0.128 |
|                  | Acid Detergent Fiber | MixG | 34.36       | 38.30      | 0.65   | 0.005 |
|                  | Ash            | MixG           | 18.33       | 20.10      | 0.31   | 0.007 |
|                  |               | MixG           | 11.00       | 11.11      | 0.13   | 0.510 |

Note: $^3$ SE, standard error of means; $^p$ $P$, probability of differences.

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### Table 1. Botanical composition (% dry matter) of herbage consumed by cows on alfalfa and orchard grass pastures of different ages, young (one and two years old) and mature (three to five years old).

| Season          | Component     | Pasture age | Young | Mature | SE$^3$ | $P^p$ |
|-----------------|---------------|-------------|-------|--------|--------|-------|
| Spring–summer   | Alfalfa       | Young       | 72.9  | 60.8   | 1.49   | <0.001|
|                  | Orchard grass | Young       | 27.1  | 38.7   | 1.47   | 0.001 |
|                  | Weeds         | Young       | 0.0   | 0.5    | 0.29   | 0.227 |
|                  | Alfalfa       | Mature      | 78.2  | 69.5   | 1.49   | 0.007 |
|                  | Orchard grass | Mature      | 19.0  | 30.3   | 1.45   | <0.001|
|                  | Weeds         | Mature      | 2.8   | 0.2    | 0.72   | 0.019 |

Note: $^3$ SE, standard error of means; $^p$ $P$, probability of differences.
During spring and summer, the milk production (kg milk cow\(^{-1}\) d\(^{-1}\)) was respectively 22.2 in MixG and 21.3 in CowG, with no difference among treatments (\(P = 0.209\)); however, during autumn and winter, milk production in MixG was 19.0, higher (\(P = 0.002\)) than that attained in CowG (14.7).

### 3.2. Composition of herbage consumed by ewes

Least-square means of the botanical composition of herbage eaten by ewes in pastures are reported in Table 4. Alfalfa content was 57% (\(P = 0.006\)) and 49% (\(P < 0.001\)) lower in mature than in young pastures during spring-summer and autumn-winter, inversely, orchard grass content was higher in mature pastures by 56% (\(P = 0.006\)) during spring-summer and by 74% (\(P < 0.001\)) during autumn-winter.

The results obtained regarding the morphological composition (Table 5) corresponded largely with herbage botanical composition. Pasture age had no effect on the content of alfalfa leaves throughout the year (\(P = 0.859\) in spring-summer and \(P = 0.374\)) in autumn-winter), or orchard grass leaf levels in spring-summer (\(P = 0.320\)). However, in autumn-winter, herbage orchard grass leaf content was 81% higher (\(P = 0.001\)) in mature than in young pastures. In young pastures, alfalfa stem content was 70 and 68% lower (\(P = 0.002\)) in spring-summer and \(P < 0.001\) in autumn-winter) in mature than in young pastures; in contrast, the content of orchard grass stems tended to be lower (\(P = 0.067\)) in spring-summer and \(P = 0.065\) in autumn-winter) in young than in mature pastures.

During spring-summer, while no effect of pasture age was observed on CP and ADF (\(P = 0.484\) and 0.102), NDF content tended (\(P = 0.065\)) to be 19% higher and ash content 23% higher (\(P = 0.003\) in mature than in young pastures (Table 6). In autumn-winter, NDF and ADF were 8 and 10% higher (\(P = 0.041\) and 0.048) in young than in mature pastures, while CP and ash were 14 and 18% higher (\(P = 0.028\) and 0.009) in mature than in young pastures.

Average daily weight gain of ewes was 0.040 ± 0.002 kg d\(^{-1}\) during spring-summer and 0.111 ± 0.018 kg d\(^{-1}\) during autumn-winter. Average production of weaned lambs was 20.3 kg per ewe, receiving 36 kg DM of maize silage and minerals as supplementary feed. The maize silage was produced in-farm at a cost of US$ 58 per ton DM, resulting in a feeding cost per kg weaned lamb of US$ 0.10.

### 3.3. Herbage intake per unit of area

During spring and summer, the herbage intake per ha (kg DM ha\(^{-1}\) grazing cycle\(^{-1}\), an indirect estimate of net herbage production) was higher (\(P < 0.001\)) in young (2282) than in mature pastures (1767). The herbage harvested (total herbage intake per ha) in MixG, i.e. cows plus ewes, was higher (\(P < 0.001\)) than that of only cows in MixG (2744 vs 1753), which tended to be (\(P = 0.063\)) higher than that of cows in CowG (1576). In autumn and winter, herbage intake per area was higher (\(P < 0.001\)) in young pastures (1241) than in mature pastures (508), and under MixG (1112) was higher (\(P < 0.001\)) than by cows in MixG (737), which was similar (\(P > 0.05\)) to that of cows in CowG (774).

### 4. Discussion

#### 4.1. Composition of herbage consumed by cows

Jiménez-Rosales et al. (2018) attributed the higher proportion of alfalfa and lower level of orchard grass under MixG than CowG to the different response of these species to high harvest intensity. In spring-summer, alfalfa levels were higher in young than in mature pastures. This pattern is likely related to the lack of persistence of alfalfa, as also reported by Dear et al. (2007). Given the importance of alfalfa in mixed pastures as both a N source and a high herbage producing species, the results of the present study emphasize the need of avoiding high proportions of three to five years old pastures within the pasture and fodder crops rotation.

Overall, an average of 56% of alfalfa was eaten as leaves and the remaining 44% as stems (Tables 1 and 2). This pattern is
related to the vertical distribution of components, with leaves concentrated in the upper half of the canopy and stems dominating the lower half (Cangiano et al. 2008). In orchard grass, an average of 90% was eaten as leaves and the remaining 10% as pseudostems. These proportions are related to sward canopy structure, with the presence of pseudostems in the lower strata inhibiting deeper bite prehension by cattle, as detected by Griffiths et al. (2003). The differences in morphological composition of the two species also reflect general differences in morphological composition recorded among pasture ages, with higher proportions of alfalfa (Table 1) resulting in a lower proportion of leaf consumption in young than in mature pastures (63% vs 68%, Table 2).

Fiber content (NDF and ADF) in herbage ingested by cows under MixG (Table 3) was comparable to the three-year average estimated for alfalfa by Andrzejewska et al. (2014). Despite the lower proportions of leaves, these contents were lower than under CowG in both seasons. The CP content was higher in MixG than under CowG during spring-summer, lower than under CowG in both seasons. The CP content was despite the lower proportions of leaves, these contents were lower than under CowG in both seasons. The CP content was higher in MixG than under CowG during spring-summer, lower than under CowG in both seasons. The CP content was

4.3. Herbage intake per unit of area

The higher herbage intake in young pastures can be explained by the greater preference of cows for alfalfa over grass (Villalba et al. 2015), because in young pastures there is a higher proportion of alfalfa (Dear et al. 2007). The larger total herbage harvested (intake of cows plus ewes) achieved in the mixed grazing system corroborates that increasing the efficiency of forage harvesting might be the main benefit of adopting such a grazing system. This advantage of mixed grazing concurs with the higher herbage intake by the cows in MixG and the acceptable herbage intake of the ewes as followers such as reported by Jiménez-Rosales et al. (2018).

4. Conclusions

Concurring with the expressed hypothesis, the sequential mixed grazing of dairy cows with breeding ewes was beneficial, improving the composition of herbage consumed by the cows; additionally, it increased the total amount of herbage harvested. Other benefits of adopting such a grazing system derive from higher milk production per cow during autumn and winter and the production of weaned lambs at a feeding cost of US$ 0.10 per kg.

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