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# Agriculture, Ecosystems and Environment

journal homepage: www.elsevier.com/locate/agee



# Plant biodiversity of mountain grasslands as influenced by dairy farm management in the Eastern Alps



Cristina Pornaro<sup>a</sup>, Chiara Spigarelli<sup>b,\*</sup>, Davide Pasut<sup>b</sup>, Maurizio Ramanzin<sup>a</sup>, Stefano Bovolenta<sup>b</sup>, Enrico Sturaro<sup>a</sup>, Stefano Macolino<sup>a</sup>

<sup>a</sup> Department of Agronomy, Food, Natural Resources, Animals and Environment, University of Padova, Legnaro, PD 35020, Italy
<sup>b</sup> Department of Agricultural, Food, Environmental and Animal Sciences, University of Udine, Udine 33100, Italy

#### ARTICLE INFO

Keywords: Meadow Pasture Small-scale farm Dairy farms Milk yield Livestock density

## ABSTRACT

It has been widely demonstrated that farm management affects the plant species composition of grassland. The present study aimed to investigate the effect of farm management on plant species richness and composition in forty-nine small-scale farms breeding dairy cattle, located in the Eastern Italian Alps at two levels of precision: plot and farm levels. Data on housing system, quality scheme, farm productivity, income from milk yield and livestock density were collected through interviews with farmers. In each farm, botanical surveys were carried out in different plots representing the botanical composition of the farmland vegetation. Elevation, slope, type of use, number of hay cuts and type of fertilisation were also recorded. The botanical surveys of the plots on each farm were analysed to describe plant composition at the plot level, then merged to describe plant composition at the farm level. At both levels, grassland botanical composition was found to be affected by farm management. At the plot level, meadows cut 2 and 3 times per year did not exhibit any differences in plant richness, but they differed in plant species, botanical family and phytosociological class composition, with a general simplification of botanical composition. We found fewer phytosociological classes but not fewer plant species or botanical families in plots fertilised with slurry than in plots fertilised with manure or not fertilised, and a change in the botanical composition due to changes in the relative abundance of plant species. At the farm level, we observed a decrease in the number of plant species and phytosociological classes, and changes in plant composition, with increasing milk yield and livestock density. Changes in botanical composition were less evident at the farm level than at the plot level. However, protecting farms and their economic viability is a means of maintaining biodiversity at the plot level.

#### 1. Introduction

Grasslands provide many ecosystem services such as conservation of biodiversity, regulation of physical and chemical fluxes in ecosystems, mitigation of pollution, and preservation of landscapes (e.g. Gibon, 2005; Lemaire et al., 2005; Pornaro et al., 2017). Grasslands cover two management categories: pastures and meadows, which are important for their high plant species richness, especially when compared with shrub or forest vegetation (MacDonald et al., 2000; Pornaro et al., 2013; Koch et al., 2015). Allen et al. (2011) defined pasture as "a type of grazing management [...] devoted to the production of forage for harvest primarily by grazing", and meadow as "a natural or semi-natural grassland often associated with the conservation of hay or silage". Their botanical composition depends mainly on environmental factors such as temperature (Buxton and Fales, 1994; Ziliotto et al., 2004), water availability (Halim et al., 1989; Ziliotto et al., 2004), solar radiation (Buxton and Fales, 1994), and soil nutrient availability influenced by fertilisation (Buxton and Fales, 1994; Gibon, 2005). However, as semi-natural habitats, they are also influenced by anthropogenic activities that can lead to changes in the plant community. The characteristics of the vegetation and the biodiversity of pasture areas are influenced by livestock management practices, and particularly different levels of livestock density and/or the use of feed supplements (Gianelle et al., 2018).

In the last decades, agriculture in Europe's mountain areas underwent radical changes, with a decrease in farm numbers and the gradual abandonment of traditional extensive farming in favour of highly

\* Corresponding author.

https://doi.org/10.1016/j.agee.2021.107583

Received 12 November 2020; Received in revised form 24 June 2021; Accepted 15 July 2021

*E-mail addresses:* cristina.pornaro@unipd.it (C. Pornaro), spigarelli.chiara@spes.uniud.it (C. Spigarelli), davide.pasut@gmail.com (D. Pasut), maurizio. ramanzin@unipd.it (M. Ramanzin), stefano.bovolenta@uniud.it (S. Bovolenta), enrico.sturaro@unipd.it (E. Sturaro), stefano.macolino@unipd.it (S. Macolino).

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mechanised, intensive production practices (Caraveli, 2000; Höchtl et al., 2005; Strijker, 2005). Dairy cow milk yields have now reached high levels, while feed rations have higher energy and protein contents than in the past, often achieved through the purchase of concentrates and forages from the plains (Sturaro et al., 2009; Battaglini et al., 2014). On the other hand, some farms have retained the traditional system (Scotton et al., 2014). These farms have relatively small herds and dairy farming is sometimes integrated with other agricultural activities or occupation, and they are considered compatible with the sustainable management of semi-natural grasslands (Dietl and Lehmann, 2004). As a result of these socio-economic shifts, pastures and meadows have gone through a profound change in plant species composition (Ellmauer and Mucina, 1993), for example favouring tall nitrophilous herbs or ruderal species (Dietl and Lehmann, 2004) or shrub encroachment (Pornaro et al., 2013).

Within agricultural practices, fertilisation appears to have a major effect on botanical composition and species richness, while the intensity of exploitation mainly influences forage quality (Mrkvička and Veselá, 2002; Hrabě and Knot, 2011). The effect of concentrations of soil nutrients, mainly nitrogen, on plant diversity is well documented (Güsewell et al., 2012; Gardarin et al., 2014). Increased nitrogen in the soil causes rapid shifts in the sward composition, supporting the growth of tufted grasses at the expense of legumes and other forbs (Silvertown et al., 2006). High concentration of nutrients favours the dominance of nitrophilous plants in the sward and the loss of plant diversity (Marini et al., 2008), with nitrophilous species indeed replacing most of the species characteristic of traditional meadows (Prosser, 2001). Low concentration of nutrients, on the other hand, favours the dominance of oligotrophic species (Aerts and Chapin, 1999; Jussig et al., 2015; Orlandi et al., 2016). In addition to soil nutrient status, plant species richness is affected by harvest frequency or grazing intensity, although few studies have investigated these issues. Bassignana et al. (2003) reported a negative relationship between plant species richness and number of cuts in a study conducted in six experimental trials on permanent meadows across the Italian Alpine arc. Changes in botanical composition due to the number of cuts per year has also been documented by Hejcman et al. (2010). In a study on long-term extensification of a fertilised, mown grassland, they compared the plant species richness and botanical composition of meadows cut two or four times per year and found that mowing frequency affected botanical composition but not the number of plant species.

Understanding the patterns of biodiversity at different spatial scales has become an important issue in ecology and landscape conservation. The scale at which studies are conducted strongly influences the biodiversity results (Bertuol-Garcia et al., 2017; Rouget, 2003). As the scale that best measures biodiversity has not yet been defined, a multiple-scale approach is suggested (Conroy and Noon, 1996; Poiani et al., 2000). Smith et al. (2011) regarded the scale as being a choice linked to the aims of the study. Sullivan et al. (2017) considered the farm scale level to be useful for identifying high nature value farmland, as it is at this level that management decisions are taken. On the other hand, habitats on small and isolated fragments, which are usually important for biodiversity conservation, can be individuated with a fine-scale approach, but are often incorporated into larger biodiversity features in broad-scale analyses (Rouget, 2003).

The present study carried out in a wide range of farming systems in Eastern Alps aimed to extend knowledge of the impact of farm management on plant species richness and composition from two scale-level perspectives: plot and farm. The effects of environmental and management variables on plant richness and composition were analysed at both levels, taking into account plant species, botanical families, and phytosociological classes. Our hypothesis is that analysis at two levels covers a broader range of factors involved in the relationships between biodiversity and farm management that are useful for understanding the potential consequences of farm practices on ecosystems.

## 2. Materials and methods

### 2.1. Study area and experimental design

The study was conducted during 2018 in three regions of the eastern Italian Alps (Veneto, Friuli Venezia Giulia, and Trentino-Alto Adige). Forty-nine farms breeding mainly Simmental dairy cattle, representative of the different farming systems operating in the study area, were selected through dairy farmers' organisations. All the farms were located in mountain areas and met EFSA (2015) criteria to be defined as small-scale farms. According to regional laws, a farm is considered located in mountain areas when 80% of the municipality surface is over 600 m a.s.l. All the milk produced was processed by local cooperative dairies. Each farm was visited twice during the study period to administer a questionnaire to the farmer and to carry out on-site botanical assessments.

## 2.2. Farms data

Data on the farms were collected through interviews with the farmers. The questionnaire, which was agreed upon and approved by the partners in the study, was designed to collect information on the productive aspects of the farms and included the following parameters: elevation of the farm buildings (metres above sea level), housing system type (loose-housing or tie-stall), presence or absence of a quality scheme (*EU Reg. 1151/2012 and/or EU Reg. 834/2007*), overall farm productivity (i.e. average tons of milk/cow/year), income from milk production as a percentage of total farm income (%), and livestock density (*Livestock Unit* ha<sup>-1</sup> of *Utilized Agricultural Area* - LU/ ha UAA). The farm characteristics are shown in Tables 1 and 2.

All farms were located in mountain areas, the majority below 1000 m a.s.l. The two housing systems (loose-housing or tie-stall) were more or less equally represented with a slight predominance of free-moving animals. Eighty-six % of the farms had no quality certification, only two had TSG (*Traditional Specialities Guaranteed*) quality scheme, and five were under organic management.

Most farms had a total annual milk yield per dairy cow of over 6 tons, while 22 had a high milk yield of 8–10 tons per year. Interestingly, for most of the farmers the income came exclusively or almost exclusively from farm activities, while only for few it came also from other sources, such as forestry and/or tourism. Eighty percent of the farms had a

## Table 1

Explanatory variable	Description	Mean (Min- Max)	SD	Farms (n)
Farm elevation (m a.s. l.)	< 500	380 (280–466)	64	10
	500-1000	750 (520–947)	154	28
	> 1000	1185 (1012–1375)	157	11
Housing system	loose-housing	-	_	29
	tie-stall	-	-	20
Quality scheme	product certification	-	-	5
	organic production	-	-	2
	no certification	-	-	42
Milk yield (t/cow/year)	< 6.0	4.8 (3.8–5.8)	0.8	8
	6.0-8.0	7.1 (6.2–7.8)	0.5	19
	> 8.0	9.5 (8.1–12.4)	1.5	22
Dairy income (% of total)	< 50	29 (20–40)	8	4
	50–75	59 (50–70)	9	17
	> 75	98 (85–100)	4	28
Livestock density (LU/ ha UAA) <sup>1</sup>	< 2.0	1.2 (0.5–1.99)	0.4	39
	> 2.0	2.9 (2.1–4.1)	0.7	10

<sup>1</sup>Livestock Unit (LU) ha<sup>-1</sup> of Utilized Agricultural Area (UAA).

#### Additional characteristics of farmland and herd composition.

Characteristic	Unit	Mean	SD	Min	Max
Farmland					
FAA <sup>1</sup> grassland permanent farm	ha	25.7	21.1	5.0	100.0
FAA cropland permanent farm	ha	1.9	4.2	0.0	16.3
FAA, permanent farm total	ha	27.6	22.0	5.0	100.0
Herd composition					
Dairy cows	$LU^2$	30	20	5	75
Livestock density	LU/ha	1.37	0.84	0.71	3.41

<sup>1</sup>Farm Agricultural Area (FAA).

<sup>2</sup>Livestock Unit (LU).

<sup>3</sup>Utilized Agricultural Area (UAA).

# livestock density of < 2 a LU/ha UAA.

All dairy cattle feed ratio was based on prevalence of forages (mainly meadows hay and alfa alfa) between 60% and 70% of dry matter, while the remaining from energy and protein concentrates (mainly cereals and soyben). Agricultural management practices varied among the farms. The number of cuts per year and fertilisation were influenced by elevation, slope and botanical composition of the plots, especially on farms with scattered and/or outlying lands.

## 2.3. Botanical surveys

Botanical surveys were carried out in three different plots on each farm, except one farm with a small farmland area where only two surveys were made. A plot was constituted by a management unit of the farm's land, where management units were "paddock" for grazed surfaces and "field" for meadows as defined by Allen et al. (2011). The

average plot size was 2530 m<sup>2</sup> (Standard Deviation 1009), the smallest being 553 m<sup>2</sup>, the largest 4121 m<sup>2</sup>. After site inspection, two of the three plots were selected as representative of the botanical composition of the farmland vegetation; the third was the plot with greatest richness of plant species in the farmland. A total of 149 botanical surveys were carried out (Fig. 1), each consisting in establishing a linear transect with a minimum length of 50 m (Smith et al., 2011) walking the two diagonals of the plot and recording all the plant species found. Plant species nomenclature followed Aeschimann (2004). Species that were ecologically and botanically similar were pooled into botanical families and phytosociological classes according to Aeschimann (2004), who assigned at each species a phytosociological class considered as the phytosociological optimum based on the phytosociology classifications of Theurillat et al. (1995) and other studies (for details see Aeschimann, 2004). Other characteristics of the plots were also recorded, such as elevation, slope, type of use (pasture, meadow, meadow with grazing after the first cut), number of cuts [1-4, or not assigned (NA) for pastures], and type of fertilisation (manure or slurry from the dairy cattle, mineral, no fertilisation) (Table 3).

The plots surveyed exhibited a wide range of elevation and slope, although most were located below 1000 m a.s.l. and had a slope lower than 15%. The primary type of use was permanent meadow, while only 10 plots were regularly grazed and 26 plots were grazed after the first hay cut. Meadows mainly underwent 2 or 3 cuts per season. Two pastures were located above 1700 m a.s.l. and were used by free-grazing cattle for two months in mid-summer, while the others were located at around 1000 m a.s.l. and were managed with a rotational grazing with two grazing periods per year (in spring and autumn). Manure and slurry were used as fertiliser in 69 plots each, while 10 plots were not fertilised.



Fig. 1. Maps of botanical surveys plot performed in the study.

Botanical	surveys r	olot c	haracteristics	according	to exp	lanatorv	variables.

Explanatory variable	Description	Mean (Min- Max)	SD	Surveys (n)
Plot elevation (m a.s.l.)	< 500	359 (242–480)	59	31
	500-1000	764 (507–1000)	154	80
	> 1000	1271 (1876–1004)	249	38
Slope (%)	< 15	6 (0–15)	5	85
· · ·	15-30	21 (17-28)	4	30
	> 30	48 (32–72)	14	34
Type of use	pasture	-	-	10
	meadow	-	-	113
	meadow grazed after first cut	-	-	26
Cuts (n.)	1	_	_	18
	2	_	_	70
	3	_	_	48
	4	_	_	3
	not assigned	-	_	10
Fertilisation type	manure	-	-	69
	mineral	-	-	1
	no fertilisation	-	_	10
	slurry	-	-	69

#### 2.4. Data analysis

The data obtained from the botanical surveys were used to build matrices to describe biodiversity plot by plot. The surveys of each farm were then merged to obtain matrices to describe biodiversity farm by farm. The two scale-level approach of our study consisted in comparative analyses of biodiversity (plant richness and composition). The term plot level is used below for plant richness and composition derived from plot dataset comparison, while farm level is used for plant richness and composition derived from farm dataset comparison. Three matrices for each of the plot and farm datasets were used for the analysis: (i) a matrix of plant species (presence/absence), (ii) a matrix of the number of plant species in each botanical family, (iii) a matrix of the number of plant species in each phytosociological class. Plant species, botanical family and phytosociological class richnesses were determined as the number of plant species, botanical families and phytosociological classes in each plot and in each farm.

Generalised linear mixed models were built to explain variation in plant species, botanical family, and phytosociological class richness depending on environmental (elevation and slope) and management descriptors (type of use, number of cuts, fertilisation) used as fixed effect and "farm" used as random effect at the plot level. Models with and without the farm effect were compared using the Akaike's Information Criterion (AIC, Akaike, 1974). Significances of variables were determined by likelihood-ratio tests (LRT) of the reduced versus the full models. Linear regressions were estimated for significant relationships between plant species, botanical family or phytosociological class richness and environmental or management descriptors. Fisher's Protected LSD test was used at the 0.05 level of probability to identify significant differences between means for significant variables. A constrained correspondence analysis (CCA) was performed to investigate the effects of environmental (elevation and slope) and management descriptors (type of use, number of cuts, fertilisation) on the plant community composition (plant species, botanical families and phytosociological classes). Permutation tests were carried out to evaluate significances of the explanatory variables.

Generalised linear models were built to explain variation in plant species, botanical family, and phytosociological class richness depending on elevation and farm descriptors (quality scheme, housing system, milk yield, dairy income and livestock density) at the farm level. Linear regressions were estimated for significant relationships between plant species, botanical family or phytosociological class richness and elevation or farm descriptors. Significances of variables were determined by likelihood-ratio tests (LRT) of the reduced versus the full models. A constrained correspondence analysis (CCA) was performed to investigate the effects of elevation and farm indicators (quality scheme, housing system, milk yield, dairy income and livestock density) on the plant community composition (plant species, botanical families and phytosociological classes). Permutation tests were carried out to evaluate significances of the explanatory variables. Spearman's correlation coefficients between milk yield, dairy income, and livestock density were calculated.

All statistical analyses were performed in R 3.4.2 (R Core Team, 2017) using vegan and nlme libraries.

#### 3. Results

# 3.1. Description of the vegetation

Overall, the botanical surveys identified a total of 339 plant species belonging to 44 botanical families and 29 phytosociological classes (Tables 4 and 5). The largest phytosociological class was *Molinio*-*Arrhenatheretea* with 98 plant species, the second largest was *Festuco*-*Brometea* (48 plant species). A minimum of 8 and a maximum of 32 plant species (mean = 20) in the *Molinio-Arrhenatheretea* class were identified,

# Table 4

Number of plant species per plot (total, mean, minimum and maximum) belonging to botanical families.

Botanical family	Tot	Mean	Min-Max
Asteraceae	40	4.73	1–10
Poaceae	35	7.34	3–14
Fabaceae	28	3.60	1–10
Cyperaceae	24	0.44	0–9
Lamiaceae	21	1.34	0–5
Scrophulariaceae	18	1.11	0–4
Rosaceae	15	0.55	0–4
Caryophyllaceae	13	1.40	0–4
Ranunculaceae	13	1.39	0–4
Apiaceae	11	2.11	0–5
Liliaceae	11	0.46	0–4
Brassicaceae	10	0.34	0–2
Orchidaceae	10	0.15	0–4
Polygonaceae	10	1.34	0–3
Rubiaceae	9	0.94	0–5
Campanulaceae	8	0.21	0–3
Geraniaceae	7	0.56	0–4
Juncaceae	7	0.21	0–3
Dipsacaceae	6	0.38	0–3
Primulaceae	5	0.13	0–1
Boraginaceae	4	0.39	0–1
Plantaginaceae	3	0.85	0–3
Polygalaceae	3	0.05	0–1
Crassulaceae	2	0.03	0–2
Equisetaceae	2	0.03	0–1
Euphorbiaceae	2	0.05	0–1
Gentianaceae	2	0.01	0–1
Hypericaceae	2	0.04	0–1
Iridaceae	2	0.05	0–1
Violaceae	2	0.03	0–1
Betulaceae	1	0.01	0–1
Chenopodiaceae	1	0.02	0–1
Cistaceae	1	0.01	0–1
Convolvulaceae	1	0.19	0–1
Fagaceae	1	0.01	0–1
Linaceae	1	0.01	0–1
Lythraceae	1	0.01	0–1
Onagraceae	1	0.01	0–1
Orobanchaceae	1	0.05	0–1
Polypodiaceae	1	0.01	0–1
Salicaceae	1	0.01	0–1
Saxifragaceae	1	0.01	0–1
Urticaceae	1	0.13	0–1
Valerianaceae	1	0.01	0–1

Number	of	plant	species	per	plot	(total,	mean,	minimum	and	maximum)
belongin	g to	phyte	osociolog	ical	classe	es assig	ned by	Aeschimani	n (20	04).

Phytosociological class	Tot	Mean	Min-Max
Molinio-Arrhenatheretea	98	19.97	8–32
Festuco-Brometea	48	2.67	0-20
Stellarietea mediae	26	1.84	0-10
Artemisietea vulgaris	21	1.82	0–6
Elyno-Seslerietea variae, Juncetea trifidi	18	0.26	0–8
Scheuchzerio-Caricetea fuscae	15	0.14	0–10
Juncetea trifidi	13	0.28	0–5
Trifolio-Geranietea sanguinei	11	1.06	0–4
Carpino-Fagetea sylvaticae	9	0.08	0–3
Mulgedio-Aconitetea	9	0.46	0–3
Nardetea strictae	9	0.36	0–4
Carpino-Fagetea	8	0.23	0–3
Koelerio-Corynephoretea	8	0.28	0–4
Filipendulo-Convolvuletea	7	0.13	0–2
Phragmito-Magnocaricetea	6	0.05	0–3
Epilobietea angustifolii	5	0.10	0–2
Quercetea robori-sessiliflorae	5	0.07	0–1
Agropyretea intermedii-repentis	4	0.05	0–1
Trifolio-Geranietea	4	0.41	0–2
Crataego-Prunetea	2	0.19	0–2
Montio-Cardaminetea	2	1.00	1–1
Calluno-Ulicetea	1	1.00	1–1
Elyno-Seslerietea	1	1.00	1–1
Erico-Pinetea	1	1.00	1–1
Quercetea pubescentis	1	1.00	1 - 1
Scheuchzerio-Caricetea	1	1.00	1–1
Thlaspietea rotundifolii	1	1.00	1–1
not assigned	5	0.07	0–1

and a minimum of 0 and a maximum of 20 (mean = 3) in the *Festuco-Brometea* class (Table 5). Twenty-six plant species belonged to the *Stellarietea mediae* class, and 21 to *Artemisietea vulgaris* class; the former class includes annual and the latter perennial weed or pioneer ruderal and nitrophilous species (according to the Italian vegetation index; htt p://www.prodromo-vegetazione-italia.org). Twenty-five species were plant of shrubland or forest habitats (*Carpino-Fagetea sylvaticae, Carpino-Fagetea, Quercetea robori-sessiliflorae, Crataego-Prunetea, Quercetea pubescentis* classes). These classes included plant species associated with a change of grassland botanical composition that precedes shrubland or forest, although they contributed to plant species richness with a maximum of 10 plant species per botanical survey.

## 3.2. Plant richness and composition at the plot level

In analysing the plant data, the models including the random effect of farm where always the most parsimonious ones. This suggested that the farms have areas containing a high number of plant species alternating with areas containing few plant species, botanical families or phytosociological classes. Farm was also added to the models as a random effect, but this did not improve the models as assessed by the Akaike's information criterion (AIC).

Of the management variables, type of use and number of cuts were significant for plant species, botanical family and phytosociological class richness, while fertilisation significantly affected only phytosociological class richness, and slope only plant species richness (Table 6). There were more plant species, botanical families and phytosociological classes in pastures than in meadows, including those grazed after the first cut (Fig. 2A, C, E). Our results also showed a decrease in plant species and phytosociological class richness, and to a lesser extent in botanical family richness, with increasing numbers of cuts (Fig. 2B, D, F). There were significant differences in the number of plant species, botanical families and phytosociological classes between meadows cut only once and meadows cut 3 times. Moreover, the smallest numbers of phytosociological classes were identified in plots fertilised with slurry (Fig. 2G). No significant effect of fertilisation type was observed for numbers of plant species and botanical families. We found no relationship between elevation and species richness in this study, but slope had a positive relationship with the number of plant species (data not shown).

Unlike plant richness, all explanatory variables were significant for plant composition (Table 6). The ordination biplots based on CCA showed that grasslands responded strongly to management and environmental variables (Fig. 3). In Fig. 3A, axis 2 clearly separated pastures and meadows on the bases of the presence/absence of plant species, while meadows grazed after the first cut had a botanical composition similar to pastures. Type of use also affected botanical family composition, with pastures having more plant species belonging to Caryophyllaceae and Polygalaceae (Fig. 3B). The differences in phytosociological class composition (Fig. 3C) are interesting: plant community was characterised in greater depth by grouping numbers of plant species according to phytosociological class, which gives less weight to individual plant species, especially those belonging to the phytosociological classes with higher frequencies (Molinio-Arrhenatheretea and Festuco-Brometea). The separation between pastures and mown/ grazed meadows shown in Fig. 3C was due to the latter having no plant species belonging to Elyno-Seslerietea, Trifolio-Geranietea, Nardetea strictae or Trifolio-Geranietea sanguinei (Table S1). Axis 1 clearly separated the number of cuts by plant species, botanical family and phytosociological class composition. Plots with no assigned number of cuts were rightly related to pastures. Plots cut once and twice per year did not differed in plant species, botanical family and phytosociological class composition as much as plots cut three times (Fig. 3), while plots cut four times differed from all the other plots. With regards to phytosociological class composition (Fig. 3C), an increasing number of cuts was linked to a lower presence of species belonging to Stellarietea mediae and Trifolio-Geranietea. Axis 1 also clearly separated plots fertilised with slurry from plots fertilised with manure. Type of fertilisation did not affect plant species richness, although it did affect plant species composition. Plots fertilised with manure had higher numbers of plant species in almost all the phytosociological classes. These results suggested that in plots fertilised with slurry the botanical composition was simplified compared with plots fertilised with manure. Moreover, species belonging to Stellarietea mediae and Trifolio-Geranietea were more frequent in plots fertilised with slurry than in plots fertilised with manure. The environmental variables included in this study went in opposite directions in plant species, botanical family and phytosociological class composition ordinations.

## Table 6

Statistical significances based on likelihood-ratio tests of explanatory variables in the most parsimonious generalised linear mixed models for number of plant species, botanical families, and phytosociological classes (plant richness) and significances based on permutation test of the effect of explanatory variables on plant species, botanical family and phytosociological class composition (plant composition) at a plot level.

	Plant richness			Plant compositio	Plant composition		
	Species	Family	Class	Species	Family	Class	
Pasture/Meadow	< 0.01	< 0.01	< 0.01	< 0.001	< 0.05	< 0.001	
Number of cuts	< 0.0001	< 0.0001	< 0.0001	< 0.01	< 0.001	< 0.001	
Fertilisation	n.s.	n.s.	< 0.05	< 0.001	< 0.001	< 0.001	
Elevation	n.s	n.s.	n.s.	< 0.001	< 0.05	< 0.001	
Slope	< 0.05	n.s.	n.s.	< 0.001	< 0.05	< 0.05	



**Fig. 2.** Plot scale number of plant species (A–C), botanical families (D–F), and phytosociological classes (G–I) as affected by type of use (Pa= pasture, Me= meadow, Pa/Me= meadow grazed after the first cut), number of cuts (1–4, NA= not assigned), fertilisation (No= absence of fertilization). Means with the same letters are not significantly different based on Fisher's Protected LSD test at the 0.05 level of probability.



(caption on next column)

Fig. 3. Plot scale canonical Correspondence Analysis of vascular plant species (A), botanical families (B) and phytosociological classes (C), and explanatory variables along the first two axes of CCA constrained with the significant variables. Only species (A) with a goodness of fit above 20%, and families (B) and classes (C) with a goodness of fit above 5% are shown. Data derived from 149 grasslands in the Italian Alps. Abbreviations of variables: Pa = pasture; Me = meadow; Pa/Me = meadows grazed after the first cut; c1-c4 = plot cut one to four times per year; Nma = plot fertilised with manure; Nmin = plot fertilised with mineral fertiliser; Nno = no fertilised plot; Nsl = plot fertilised with slurry. Abbreviation of species: 1 = Aegopodium podagraria, 2 = Agrostis capillaris, 3 = Ajuga reptans, 4 = Alchemilla vulgaris (agg.), 5 = Anthriscus sylvestris, 6 = Arrhenatherum elatius, 7 = Avena sativa, 8 = Avenula pubescens, 9 = Bromopsis erecta, 10 = Bromus hordeaceus, 11 = Capsella bursa-pastoris, 12 = Carum carvi, 13 = Centaurea nigrescens subsp. nigrescens, 14 = Centaureanigrescens subsp. transalpina, 15 = Centaurea scabiosa, 16 = Cerastium glomeratum, 17 = Clinopodium vulgare, 18 = Colchicum autumnale, 19 = Convolvulus arvensis, 20 = Cruciata laevipes, 21 = Daucus carota, 22 = Erigeron annuus, 23 = Festuca gr. rubra, 24 = Festuca stricta subsp. sulcata, 25 = Galium album, 26 = Galium mollugo, 27 = Geranium dissectum, 28 = Geranium molle, 29 = Geranium phaeum, 30 = Geranium sylvaticum, 31 = Heracleum sphondylium, 32 = Holcus lanatus, 33 = Hordeum murinum, 34 = Hypochaeris radicata, 35 = Knautia arvensis, 36 = Lamium album, 37 = Leucanthemum vulgare, 38 = Lolium multiflorum, 39 = Lolium perenne, 40 = Lotus corniculatus, 41 = Medicago lupulina, 42 = Medicago sativa, 43 = Myosotis arvensis, 44 = Ornithogalum umbellatum, 45 = Phleum pratense, 46 = Pimpinella major, 47 = Pimpinella saxifraga, 48 = Plantago lanceolata, 49 = Poa pratensis, 50 = Poa trivialis, 51 = Ranunculus repens, 52 = Rhinanthus alectorolophus, 53 = Rhinanthus minor, 54 = Rumex acetosella, 55 = Salvia pratensis, 56 = Schedonorus arundinacea, 57 = Senecio vulgaris, 58 = Silene dioica, 59 = Silene vulgaris, 60 = Tragopogon pratensis, 61 = Trifolium dubium, 62 = Trifoliummontanum, 63 = Trifolium repens, 64 = Urticadioica. 65 = Veronica chamaedrys, 66 = Vicia cracca, 67 = Vicia faba, 68 = Vicia hirsuta, 69 = Vicia sativa. Abbreviation of families; Api = Apiaceae, Ast = Asteraceae, Car = Caryophyllaceae, Con = Convolvulaceae, Dip = Dipsacaceae, Fab = Fabaceae, Ger = Geraniaceae, Lam = Lamiaceae, Lil = Liliaceae, Pla = Plantaginaceae, Poa = Poaceae, Pol = Polygalaceae, Ran = Ranunculaceae, Rub = Rubiaceae, Scr = Scrophulariaceae, Urt = Urticaceae. Abbreviation of phytosociological classes: Car.Faget = Carpino-Fagetea, Car.Faget.1 = Carpino-Fagetea sylvaticae, Ely.Sesle.1 = Elyno-Seslerietea variae, Fil.Convo = Filipendulo-Convolvuletea, Koe.Coryn = Koelerio-Corynephoretea, Nar.stric = Nardetea strictae, Sch.Caric = Scheuchzerio-Caricetea, Ste.media.1 = Stellarietea mediae, Thl. rotun = Thlaspietea rotundifolii, Tri.Geran = Trifolio-Geranietea, Tri.Geran.1 = Trifolio-Geranietea sangunei.

#### 3.3. Plant richness and composition at farm level

When we merged the surveys for each farm, we found that the numbers of plant species were affected by farm management and elevation. At plot level, there was high variability in plant richness within each farm, with a significant effect of a few explanatory variables. In contrast, at the farm level, all the explanatory variables, with the exception of housing system, affected the number of plant species (Table 7). It is interesting that there were no differences between organic and non-organic farms. Only one farm with TSG product quality scheme had a lower number of plant species than the other farms, but this cannot be representative of the whole system (Fig. 4A). As milk yield, dairy income and livestock density increased, the number of plant species decreased (Fig. 4B, C, D). The increase in milk yield also negatively affected the number of phytosociological classes (data not shown).

Elevation, quality scheme, housing system and dairy income were also significant for plant species composition (Table 7). Botanical family composition was affected only by housing system, while phytosociological class composition was affected by milk yield and livestock density. The ordination biplots based on CCA showed that plant species composition responded strongly to elevation, certification type and housing system (Fig. 5A): Alchemilla vulgaris, Heracleum sphondylium, Phleum pratense and Silene dioica were the most frequent species in farms at higher altitudes, or with quality scheme and organic practices, or in farms with loose-farming system (Table S1), while Erigeron annuus,

Statistical significances based on likelihood-ratio tests of explanatory variables in generalised linear mixed models for number of plant species, botanical families, and phytosociological classes (plant richness) and significances based on permutation test of the effect of explanatory variables on plant species, botanical family, and phytosociological class composition at a farm level.

	Plant richness			Plant composition		
	Species	Family	Class	Species	Family	Class
Farm elevation Housing system Certification Milk yield Dairy income Livestock density	$< 0.05 \\ n.s. \\< 0.01 \\< 0.0001 \\< 0.01 \\< 0.0001$	n.s. n.s. n.s. n.s n.s. n.s.	n.s. n.s. < 0.01 n.s. n.s.	< 0.01 < 0.05 < 0.05 n.s. < 0.01 n.s.	n.s. < 0.01 n.s. n.s. n.s. n.s.	n.s. n.s. < 0.05 n.s. < 0.05

Ornithogalum umbellatum and Salvia pratensis were the most frequent species in farms with lower elevation, no quality scheme or with tie-stall housing system. Axis 2 clearly separated organic from conventional farms, and farms with loose housing from farms with tie-stall housing systems. Plant species composition also changed in response to dairy income (Fig. 5A), with Ranunculus repens and Rumex obtusifolius increasing in frequency and Centaurea nigrescens and Ornithogalum umbellatum decreasing in frequency with increasing income. Housing system affected botanical family composition (Fig. 5B) with more Geraniaceae in farms with a tie-stall system and more Apiaceae and Rosaceae in farms with a loose-farming system. The differences in phytosociological class composition are interesting (Fig. 5C). Axis 1 shows the changes in botanical composition linked to milk yield and livestock density: increases in these two variables were associated with a gradual loss of plant species, as shown in Fig. 4. We therefore observed a general decrease in the number of plant species belonging to all the phytosociological classes, which was particularly pronounced in the case of Trifolio-Geranietea sanguinei, Elyno-Seslerietea variae, Nardetea strictae and Trifolio-Geranietea.

There was a significant correlation between milk yield and dairy income, but none between livestock density and milk yield or dairy income (Table 8).

## 4. Discussion

### 4.1. Biodiversity described by plot comparison

Farms were included in the model in order to take into account spatially correlated environmental variables (Borcard et al., 1992) and spatial components linked to historical processes (Svenning and Skov, 2005). We found a significant farm effect in the model. Analysis of the survey data with the model suggested that farms are characterised by high plot diversity in terms of plant richness, confirming findings from other studies in a similar environment (Pornaro et al., 2019).

In a study analysing the relative importance of management and environmental factors on grassland vegetation, Klimek et al. (2007) found type of use to be the main factor affecting plant species richness and composition. In agreement with them, we found that plant species, botanical family and phytosociological classes richness at the plot level were higher in pastures than in meadows, whether just mowed or mown and grazed (Fig. 2). It is widely known that there are differences in botanical composition between pastures and meadows (Klimek et al., 2007), which are associated with the plants' response to the grazing or mowing regime. The differences in phytosociological class composition, however, are not very clear. Where the numbers of plant species were grouped by phytosociological class, less weight is given to individual plant species, especially to those belonging to the phytosociological classes with higher frequencies (Molinio-Arrhenatheretea and Festuco--Brometea). The separation between pastures and meadows grazed after the first cut shown in Fig. 3C was due to the absence of plant species of the Elyno-Seslerietea, Trifolio-Geranietea, Nardetea strictae, and Trifolio--Geranietea sanguinei classes.

Our results regarding a reduction in plant species richness with increasing the number of cuts are consistent with those of other studies (Bassignana et al., 2003; Hejcman et al., 2010). We believe that this is due primarily to the productivity of these coenoses, as productivity and plant species richness are negatively related (Gough et al., 2000; Jacquemyn et al., 2003; Maurer et al., 2006). Regarding botanical composition, the method adopted in the present study for the botanical survey did not provide the evaluation of the species coverage or abundance and this may dull the results. We found that plots cut once or twice per year did not differ in plant species, botanical family and phytosociological class composition as much as plots cut three times (Fig. 3). Meadows cut 2 and 3 times per year did not differ in plant richness, but they did differ in plant species, botanical family and phytosociological class composition (Fig. 3), with a general simplification of botanical



**Fig. 4.** Farm scale number of plant species as affected by quality scheme (A) (qual = quality scheme, no = farm with none certification, org = farm with organic practices), milk production (B), dairy income (C) and livestock density (D). Means with the same letters are not significantly different based on Fisher's Protected LSD test at the 0.05 level of probability.



(caption on next column)

Fig. 5. Farm scale canonical Correspondence Analysis of vascular plant species (A), botanical families (B), and phytosociological classes (C), and explanatory variables along the first two axes of CCA constrained with the significant variables. Only species (A) with a goodness of fit above 20%, and families (B) and classes (A) with a goodness of fit above 5% are shown. Abbreviations of variables: qual = quality scheme; org = organic practices; no = none certification; lo = loose-housing; tie = tie-stall housing. Abbreviation of species: 1 = Aegopodium podagraria, 2 = Agrostis capillaris, 3 = Ajuga reptans, 4 = Alchemilla vulgaris (agg.), 5 = Anthriscus sylvestris, 6 = Arrhenatherum elatius, 7 = Avena sativa, 8 = Avenula pubescens, 9 = Bromopsis erecta, 10 = Bromushordeaceus, 11 = Capsella bursa-pastoris, 12 = Carum carvi, 13 = Centaureanigrescens subsp. nigrescens, 14 = Centaurea nigrescens subsp. transalpina, 15 = Centaurea scabiosa, 16 = Cerastium glomeratum, 17 = Clinopodium vulgare, 18 = Colchicum autumnale, 19 = Convolvulus arvensis, 20 = Cruciata laevipes, 21 = Daucus carota, 22 = Erigeron annuus, 23 = Festuca gr. rubra, 24 = Festuca stricta subsp. sulcata, 25 = Galium album, 26 = Galium mollugo, 27 = Geranium dissectum, 28 = Geranium molle, 29 = Geranium phaeum, 30 = Geranium sylvaticum, 31 = Heracleum sphondylium, 32 = Holcus lanatus, 33 = Hordeum murinum, 34 = Hypochaeris radicata, 35 = Knautia arvensis, 36 = Lamium album, 37 = Leucanthemum vulgare, 38 = Lolium multiflorum, 39 = Lolium perenne,40 = Lotus corniculatus, 41 = Medicago lupulina, 42 = Medicago sativa, 43 = Myosotis arvensis, 44 = Ornithogalum umbellatum, 45 = Phleum pratense, 46 = Pimpinella major, 47 = Pimpinella saxifraga, 48 = Plantago lanceolata, 49 = Poa pratensis, 50 = Poa trivialis, 51 = Ranunculus repens, 52 = Rhinanthusalectorolophus, 53 = Rhinanthus minor, 54 = Rumex acetosella, 55 = Salvia pratensis, 56 = Schedonorus arundinacea, 57 = Senecio vulgaris, 58 = Silene dioica, 59 = Silene vulgaris, 60 = Tragopogon pratensis, 61 = Trifolium dubium, 62 = Trifolium montanum, 63 = Trifolium repens, 64 = Urtica dioica, 65 = Veronica chamaedrys, 66 = Vicia cracca, 67 = Vicia faba, 68 = Vicia hirsuta, 69 = Vicia sativa. Abbreviation of families; Api = Apiaceae, Cam = Campanulaceae, Car = Caryophyllaceae, Con = Convolvulaceae, Cra = Crassulaceae, Dip = Dipsacaceae, Eup = Euphorbiaceae, Gen = Gentianaceae, Ger = Geraniaceae, Hyp = Hypericaceae, Iri = Iridaceae, Jun = Juncaceae, Lam = Lamiaceae, Lil = Liliaceae, Lyt = Lythraceae, Orc = Orchidadeae, Oro = Orobanchaceae, Pla = Plantaginaceae, Poa = Poaceae, Pol = Polygalaceae, Pol.1 = Polygonaceae, Ran = Ranunculaceae, Ros = Rosaceae, Rub = Rubiaceae, Urt = Urticaceae. Abbreviation of phytosociological classes: Agr.inter = Agropyretea intermedii-repentis, Car.Faget Carpino-Fagetea, = Car. Faget.1 = Carpino-Fagetea sylvaticae, Elv.Sesle.1 = Elvno-Seslerietea variae, Fil. Convo = Filipendulo-Convolvuletea, Koe.Coryn = Koelerio-Corynephoretea, Nar. stric = Nardetea strictae, Phr.Magno = Phragmito-Magnocaricetea, queue.pubes = Quercetea pubescentis, Sch.Caric = Scheuchzerio-Caricetea, Thl.rotun = Thlaspie $tea\ rotundifolii,\ Tri.Geran=Trifolio-Geranietea,\ Tri.Geran.1=Trifolio-Geranietea$ sangunei, NA = not assigned.

composition and an increase of ruderal species. Differences in botanical composition according to the number of cuts per year have also been documented by Hejcman et al. (2010), who signalled the influence of cuts on the competition for light (Louault et al., 2005; Pavlů et al. 2007) and nutrients (Elberse and Berendse, 1993; Liu et al., 2010). Changes in botanical composition could also be related to the physiological response of plants to mowing stress, which influences not only the vegetation but also soil characteristics (Francioni et al., 2020). We also observed changes in botanical composition in meadows cut four times, but since only three meadows over 113 were subjected to this cutting regime, the result has limited impact.

The decrease in plant species numbers associated with the increase in fertilisation has been well documented for both pastures and meadows (Gough et al., 2000; Jacquemyn et al., 2003; Maurer et al., 2006), but little information is available on the effect of fertilisation type on the number of plant species. In our study, we compared manure with slurry. Although slurry is a commonly used fertiliser in grasslands, its effect on plant species has not been widely studied (Duffková et al., 2013; Liu et al., 2010). Nevertheless, these studies reported a slight decrease in the plant species richness of vegetation fertilised with slurry over a long period. Similarly, we found fewer phytosociological classes in plots fertilised with slurry than in plots fertilised with manure or not fertilised (Fig. 2G), and species belonging to *Stellarietea mediae* and *Trifolio-Geranietea* were more frequent in plots fertilised with slurry. Duffková et al.

Spearman's correlation coefficients between milk yield, dairy income, and livestock density. Coefficients are reported below, and significance of correlation is reported above the matrix diagonal.

Parameter	Milk yield	Dairy income	Livestock density
Milk yield	/	p < 0.05	n.s.
Dairy income	0.31	/	n.s.
Livestock density	0.22	0.09	/

(2013) reported changes in botanical composition that did not concern plant species composition but rather the relative abundances of plant species. This maintaining of most species in the plant composition, as reported in the literature and in Fig. 2, could explain the absence of a significant effect of fertilisation type on the number of plant species and botanical families. The ordination plots also separated plots fertilised with slurry from plots fertilised with manure, confirmation that different types of fertiliser affect plant composition differently (Klimek et al., 2007). However, in a long-term study, Duffková et al. (2013) observed a shift in the relative abundances of plant species as a consequence of fertilisation with slurry, without a significant loss of plant species. Our results showed also that plots fertilised with manure had higher numbers of plant species belonging to almost all the phytosociological classes, suggesting that slurry fertilisation simplifies the botanical composition. This simplification favour ruderal species (belonging to Stellarietea mediae) that take advantage of the creation of vegetation gaps and patches of open soil caused by stresses on grassland vegetation (Bartelheimer and Poschlod, 2016; Drobnik et al., 2011; Müller et al., 2014).

It should also be noted that the widely documented relationships between elevation and vegetation richness (Naginezhad et al., 2009; Ziliotto et al., 2004), and between slope and vegetation richness (Marini et al., 2007, 2008) were not found in this study. Moreover, more than 80% of the investigated plots had no or low slope (< 30%). Therefore, a very little influence of the exposition on botanical composition can be assumed. This is probably because most of the variation in plant species, botanical family and phytosociological class richness could be mainly captured by the explanatory variables reflecting field management. In contrast, as discussed at the beginning of this section, variation in the environmental site conditions and large-scale spatial trends have a minor influence (Klimek et al., 2007). As a large range of elevations and slopes were compared, it is likely that their effect on plant richness was clouded by the effect of field management. Moreover, in the present study temperatures and altitude at plot level are not necessarily correlated as they depend on the site position with respect to the Alps (internal or at the edge). Nevertheless, altitude and slope are confirmed as being the main environmental variables driving changes in botanical composition (Marini et al., 2008).

## 4.2. Biodiversity described by farm comparison

As already mentioned in the material and methods section, the farms in the study were highly heterogeneous and encompassed conventional and organic systems, with and without quality scheme, and with different housing systems, livestock densities and dairy income percentages (Table 1).

It is often assumed that organic farms have larger, better quality grasslands (Aude et al., 2004). However, there were no significant differences in plant species richness between organic and conventional farms (Gibson et al., 2007). The farm with higher forage proportions in animal diet had fewer plant species than the other farms (Fig. 4A), but as there was only one such farm, this information can hardly be representative of the entire quality system. Plant species composition also differed between organic and conventional type, and between farms with loose-farming or tie-stall housing system (Fig. 5A). Housing system could affect plant composition as a result of different types of livestock manure (slurry or solid manure) produced then applied to the land. The

correlation between quality scheme and housing system suggests that organic farms favour the loose housing system, so that differences in botanical composition between the two housing systems are the result of farm management choices.

We have shown that the number of plant species and phytosociological classes decreased as a consequence of increased milk yield, dairy income and livestock density (Fig. 4B-D). The higher the livestock density, the lower the farm's ability to produce sufficient feed for its animals (Fig. 4). The negative impact of farm on plant species richness was lower where the farm's entire income came from the farm itself, and this was probably an indirect effect of the positive correlation between milk yield and dairy income (Table 8). However, based on plant species and botanical families, botanical composition was unaffected by milk yield or livestock density, but based on phytosociological classes, it was (Fig. 5). The shift in botanical composition with the increase in these two variables was due to a gradual loss of plant species, as shown in Fig. 4. We therefore observed a general decrease in plant species of all the phytosociological classes, especially Trifolio-Geranietea sanguinei, Elyno-Seslerietea variae, Nardetea strictae and Trifolio-Geranietea. Livestock unit per hectare of UAA has been described as being an important factor in assessing farmland biodiversity (Boyle et al., 2015; Sullivan et al., 2011). The relationship between livestock density calculated as LU/ha UAA and biodiversity found in the present study is important, as few published studies have looked specifically at the relationship between milk yield or livestock density calculated as LU/ha UAA and plant richness or composition. Our results are in line with those from studies investigating the relationships between plant richness or composition and increased fertilisation (Crawley et al., 2005; Honsovà et al., 2007). This could support the theory that farm intensification leads to over-fertilisation of grasslands with a negative effect on biodiversity.

As in the plot comparison, we did not observe any relationship between elevation and plant richness (Naqinezhad et al., 2009; Ziliotto et al., 2004), probably because of the distribution of the variation (Klimek et al., 2007).

#### 4.3. Biodiversity patterns at the two scales

Although the management explanatory variables at the two levels were different, we can consider the number of cuts at the plot level, and milk yield and livestock density at the farm level as management intensity descriptors. We have shown that increasing the number of cuts, the milk yield and the livestock density negatively affects plant species and phytosociological class richness. A loss of plant species causes a depletion of grassland ecosystem services, which highlights the importance of agricultural management decisions for environmental protection. Changes in botanical composition were observed at both the plot and the farm levels. At the plot level, we found an increase in species belonging to phytosociological classes of annual weed species, while at the farm level, there was a general simplification of botanical composition that was not related to any specific phytosociological class. Analysis of botanical composition based on phytosociological class, including grassland weed species (according to the Italian vegetation index; http://www.prodromo-vegetazione-italia.org) allowed us to generalise the effects of farm management and environmental variables to a large range of grassland vegetation. Changes in botanical composition are less evident at the farm than at the plot level. Indeed, phytosociological classes of annual weed species (Stellarietea mediaea) were affected only by plot management intensity.

The effects of management explanatory variables at the farm level inevitably extend to the whole farmland area, including the plots, while the explanatory variables found to be important drivers of plant richness and composition at the plot level cannot be transferred to the farm level. The farm-scale is considered useful for developing and implementing policy incentives paid to individual landowners (Boyle et al., 2015). Fine-scale data are not deemed as an absolute requirement in broad-scale conservation planning because of their poor efficacy in achieving fine-scale observation targets (Boyle et al., 2015). However, there is a risk in broad-scale analyses of overlooking habitats that have become small, isolated fragments. The present model analysis of the survey data suggested that farms are characterised by a high plot diversity in terms of plant richness, highlighting the importance of mountain dairy farm system on preserving biodiversity even when their milk yield is high.

The environmental explanatory variables analysed (elevation and slope) had less influence on plant richness and composition than management variables at both levels. The effect of elevation on the observed biodiversity simply reflects the typical vegetation shift linked to the thermal requirements of plant species in the mountain areas of north-eastern Italy (Ziliotto et al., 2004).

## 5. Conclusion

Farm management strongly influences the plant richness and composition of grasslands. Type of fertilisation and mowing frequency affect richness and composition based on plant species, botanical families, and phytosociological classes, including within the farms themselves, resulting in the farmland having areas of greater and lesser biodiversity. Our results also show that in the grasslands of farms breeding dairy cattle in mountain areas plant species richness and phytosociological classes composition negatively correlated with animal production performances and livestock density. However, even when their milk yield is high, small-scale mountain dairy farms retain high biodiversity areas. Therefore, protecting these farms and their economic viability is a means of maintaining biodiversity at the plot level, which becomes the responsibility of the farmers themselves. A multidimensional approach analysing the efficiency of the system, which takes into account the benefits of different farm management choices, is recommended.

#### Funding

This work was supported by the Interreg V-A Italy-Austria 2014–2020 "TOPValue" (grant number ITAT2009).

# **Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.agee.2021.107583.

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