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Do High-Magnesium Cool-Season Grasses Contemplate Grass Tetany Risk?

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Abstract

Economic losses associated with grass tetany either of death or poor growth performance of livestock are a growing concern. Breeding of high magnesium (high-Mg) cool-season forage grass (C_3) has been a challenging process to minimize the hazards of grass tetany. For appraising the breeding of high Mg-containing cultivars in Japanese Andisol, agronomic properties and grass tetany risk of high-Mg cultivars were compared with commercial cultivars. The high-Mg cultivars of Italian ryegrass (*Lolium multiflorum* L.), tall fescue (*Festuca arundinacea* Schreb.), and orchardgrass (*Dactylis glomerata* L.) were "Magnet", "HiMag" and "Mgwell", respectively. The commercial cultivars were viz., Ace, Tachiwase, and Waseyutaka of Italian ryegrass, Hokuryo, Kentucky-31 (Ky-31), and Fawn of tall fescue, and Akimidori and Okamidori of orchardgrass, respectively. Grasses were grown in temperate Andisol under field conditions with standard management practices and were harvested four times during the year. The average plant heights of the high-Mg containing cultivars were lower than the commercial cultivars with the relative range of −7.4%, −3.7%, and -1.5% for Italian ryegrass, tall fescue, and orchard grass species, respectively. The seasonal yield of high-Mg cultivars was ranked as Magnet > HiMag > Mgwell. The Mgwell orchard grass had lower potassium (K) content compared with their respective commercial cultivars with a relative range of −2.2%. Across four harvests, a significantly higher calcium (Ca) and magnesium (Mg) content, and lower grass tetany potential were recorded in high-Mg cultivars when compared to commercial cultivars, irrespective of species. Across four harvests, the lowest grass tetany index, [K/(Ca + Mg)] of 1.36 was recorded in Mgwell orchard grass. The high-Mg cultivars showed the lowest [K/(Ca + Mg)] across four harvests compared to commercial cultivars, promoting the effectiveness of breeding cool-season grass species to control grass tetany in temperate regions (Andisol) and climatic conditions.

Keywords

Andisol, Temperate, Italian Ryegrass, Tall Fescue, Orchardgrass

1. Introduction

Optimization of mineral nutrition is important to maximize crop productivity. Mineral nutrients especially Mg protect crops against various abiotic stresses, including heat [1], high light radiation [2], drought, secondary salinity, soil acidity, and aluminum (Al) toxicity [3]. Optimum Mg nutrition is also essential for the healthy growth of animals but is a challenge, especially for grazing livestock. Adequate amounts of available Mg are required in soils and in the grass to fulfill the demand of grazing animals to prevent grass tetany [4]. Grass tetany is a non-infectious metabolic disorder in cattle and sheep when grazing on cool-season grasses having low Mg concentration or reduced absorption of Mg [5]. This metabolic disorder is caused by a deficiency of available Mg as well as associated available Ca and K contents in forages [6]. Thus, the economic losses due to grass tetany either death or poor growth performance of livestock is a growing concern. Annual losses due to grass tetany in the United States are estimated at $50 to $150 million [7]. Likewise, the grass tetany epidemic in dairy and beef cattle was also responsible for considerable losses in the United Kingdom and other European countries. McCoy *et al.* [8] reported that grass tetany affected 34% of the animals annually in Ireland. Losses due to grass tetany epidemics were also reported in New Zealand [9] and Australia [10]. In northern Japan, grass tetany incidence is prevalent with cattle grazing on orchardgrass during the autumn and spring seasons [11].

Grass tetany occurs throughout the temperate regions of the world, predominantly while animals are grazing on immature pastures. Several attempts to control or eliminate the incidence of grass tetany have been focused on selective breeding of forages with higher contents of Mg in forage biomass [12]. Among the mineral elements related to grass tetany, Sleper *et al.* [5] reported that heritability estimates were highest for Mg, intermediate for Ca, and lowest for K contents in most of the C_3 forage grass species. Genetic variations of mineral elements and associated mineral ratio, within forage grass species, associated with grass tetany were observed in reed canary grass [13], Italian ryegrass [14] tall fescue [15], wheatgrass [16], orchardgrass [17] [18], and perennial ryegrass [19], respectively.

Tall fescue is one of the major cool-season grasses in American pastureland. Plant breeders developing cultivars to minimize the grass tetany hazards in the United States are concentrating largely on increasing herbage Mg content in tall fescue, and the breeding programs have succeeded in producing "HiMag" tall fescue species [7]. Research efforts led to the breeding of "Magnet" Italian ryegrass [14]. Moseley *et al.* [20] reported that the use of "Magnet" Italian ryegrass proved effective to minimize the grass tetany incidence (hypomagnesemia) in grazed cattle. Orchardgrass is a commonly grown cultivar in Japan and to minimize grass tetany through forage breeding in Japan, there is a primary requirement and with this view, breeding programs have been conducted to increase Mg content in orchardgrass and "Mgwell" orchardgrass as a high-Mg cultivar was bred by Saiga *et al.* [21]. Besides, Italian ryegrass, tall fescue, and orchardgrass are popular grasses grown in the Andisol of Japan. However, there is a lack of information on field-based studies associated with the agronomic characteristics and mineral content of high-Mg cultivars compared with commercially available cultivars of Italian ryegrass, tall fescue, and orchardgrass grown in temperate regions of northern Japan. Sabreen *et al.* [22] reported a higher Mg but lower K uptake in high-Mg cultivars of Italian ryegrass and tall fescue in solution culture, which is further confirmed by this current study under field conditions.

We hypothesized that in Japanese Andisol, all cultivars bred for high-Mg content are expected to balance K optimization with high Mg and Ca uptakes. Our research study was aimed to evaluate the growth, nutrient content, and grass tetany risk of high-Mg cultivars of Italian ryegrass, tall fescue, and orchardgrass grown in Andisols of northern temperate regions of Japan.

2. Materials and Methods

2.1. Experimental Plot, Climate and Soil

A field experiment was conducted at the Iwate University Uadai research farmin Morioka (latitude 39˚42'12" and longitude 141˚09'09"; altitude 141 m from sea level), Japan. While the climate is cold and temperate with uniformly distributed total rainfall (1266 mm) during the year, the winter is freezing cold and partly cloudy (**Table 1**). Highest precipitation falls from May to September of the year. Mean monthly temperature of more than 23˚C was recorded in August and the lowest of −1.9˚C in January. Relative humidity ranged between 67% and 77% during the growing season.

The field soil is characterized as Umbric Andosol (USDA; Andisol: FAO), with a sandy loam texture, pH 5.8, soil organic matter 18.4 g kg^{-1}, total N 0.39%, available P 6.1 mg·kg^{-1}, and exchangeable K, Ca, and Mg were 25.8, 410, and 47.0 mg·kg^{-1}, respectively.

2.2. Test Plant

The test plants were selected as one high-Mg cultivar and three commercial cultivars from Italian ryegrass (*Lolium multiflorum* L.), tall fescue (*Festuca arundinacea* Schreb.), and two commercial cultivars from orchardgrass (*Dactylis glomerata* L.) species, respectively. The "Magnet", "HiMag" and "Mgwell" were cultivars of Italian ryegrass, tall fescue, and orchardgrass were succeeded by Hides

Table 1. Climatic data of the experimental area*.

Month	Highest temp (˚C)	Lowest temp (˚C)	Mean temp (˚C)	Rainfall (mm)	Snowfall (mm)	Relative humidity (%)	Sunshine (hrs)
Jan	1.8	−5.6	−1.9	53.1	85	73	116.9
Feb	2.9	−5.2	−1.2	48.7	74	70	127.5
Mar	7	−2.2	2.2	80.5	46	67	160.4
Apr	14.4	3	8.6	87.5	4	65	173.7
May	19.7	8.5	14	102.7	0	69	185.4
Jun	23.5	13.8	18.3	110.1	0	75	154.7
Jul	26.4	18.1	21.8	185.5	0	80	128.5
Aug	28.3	19.6	23.4	183.8	0	79	149.1
Sep	23.6	14.6	18.7	160.3	0	80	123.7
Oct	17.6	7.3	12.1	93	0	77	145.8
Nov	10.6	1.5	5.9	90.2	10	75	116.9
Dec	4.6	−2.4	1	70.8	53	74	101.6
Year	15.0	5.9	10.2	1266.2	272	69	1684.2

*Japan Meteorological Agency.

and Thomas [14] and bred by Mayland and Sleper [7] and Saiga *et al.* [21], respectively, as high-Mg containing cultivars. In contrast, commercial cultivars of Italian ryegrass were Ace, Tachiwase, and Waseyutaka, and tall fescue cultivars were Fawn, Hokuryo, and Ky-31, and orchardgrass were Akimidori and Okamidori as those cultivars are commonly cultivated in the northern region of Japan. The morphological and chemical features of all test plants were documented in **Table 2**.

2.3. Experimental Design and Plant Growth

Prior to establishing the field experiment, one metric ton of dolomite, 140 kg N as urea, 280 kg P_2O_5, and 140 K_2O kg·ha^{-1}, respectively were applied during the land preparation. The experiment was set up in a split-plot arrangement of randomized complete block design with three grass species as the main plots and the high/low Mg cultivars were split plots with three replications for each treatment combination. Grass seeds were obtained from the Seed Bank of Iwate University, Japan. The seeding rate was 3 gm^{-2}, and the seeds were allowed to lay dormant in the field during the winter months (below freezing point) and the grasses were allowed to grow in the spring throughout autumn. The grasses were reared in the field by following standard pasture management practices.

The plant height of the grasses was recorded each time before the harvest. Grasses were harvested in randomly selected 1 m^2 sub-plots in each replicated plot at 6 cm cutting height. Harvesting was performed four times, in May (15 May), July (15 July), September (15 September), and October (15 October), respectively.

Table 2. Morphological features of cultivars of the forage species*.

Species	Cultivar	Type	Origin	Maturity	Growth habit	Others
Italian ryegrass	Magnet	HighMg	UK, 1981	Late	Erect	High Mg containing
Lolium multiflorum L.	Ace	Commercial	Japan, 1984	Late	Erect	Broad leaf
	Tachiwase	Commercial	Japan, 1985	Early	Erect	Loading resistance
	Waseyutaka	Commercial	Japan, 1972	Early	Erect	High productivity
Tall fescue	HiMag	HighMg	USA, 1993	Medium	Erect	High Mg containing
Festuca arundinacea Schreb.	Fawn	Commercial	USA, 1964	Early	Erect	High productivity
	Hokuryo	Commercial	Japan, 1972	Late	Intermediate	Winter hardiness
	Ky-31	Commercial	USA, 1943	Medium	Intermediate	Wide adaptability
Orchardgrass	Mgwell	HighMg	Japan, 2002	Early	Intermediate	High Mg containing
Dactylis glomerata L.	Akimidori	Commercial	Japan, 1976	Very early	Erect	High productivity
	Okamidori	Commercial	Japan, 1972	Medium	Intermediate	High digestibility

*Forage data are generalized from Hides and Thomas [14]; Hides and Lovatt [23]; Kenneth *et al.* [24]; Mayland and Grunes [6]; Mayland and Sleper [7]; Mayland (personal communication) 2002; Saiga (personal communication) 2000; Saiga *et al.* [21].

The harvested ground cover was visually assessed for weed species and separated from the grasses. Weeds were then sampled (composite) and their weights were recorded after oven-drying at 60˚C for a 48-h period to calculate weed pressure.

2.4. Nutrient Analysis

After fresh grass samples from each randomly selected microplot (1-m × 1-m) were collected, approximately 500-g samples were taken, oven-dried at 60˚C for 48-h in a forced-air oven. The oven-dried samples were ground to pass thru a 1-mm screen with a cyclone mill followed by making pellets [17] [25], and analyzed for K, Ca, and Mg contents with a live time of 100-sec by energy reflectance X-ray fluorescence spectrometry [26] [27]. The grass tetany index (GTI = [K/(Ca + Mg)] was computed following Kemp and T'Hart [28]. The relative range of each parameter was calculated as:

$$\text{Relative range}(\%) = \frac{\text{High Mg} - \text{Average of Control}}{\text{Average of Control}} \times 100 \qquad (1)$$

2.5. Statistical Analysis

Analysis of variance (ANOVA) was performed for each grass species and harvested with the JMP procedure of the SAS system (SAS Inst., Cary, NC). Treatment means were separated using Tukey's Honestly Significant Difference (HSD) test when the F-value in the ANOVA was significant at $P \leq 0.05$. To determine statistical differences among cultivars, a further ANOVA was performed as a split-plot with the subplots representing the repeated measurements.

While variability within species is anticipated as well as among cultivars of respective forage species, the cultivar/species specific correlation coefficients provide evidence-based knowledge to producers with suitable environmental and site conditions for the specific cultivar to grow. Therefore, we performed species and cultivar specific correlations among the phenotypic traits. To the best of our knowledge, this is the first statement to establish this sort of correlation among morphological and/or physiological traits and nutrient concentrations and their ratio. Pearson phenotypical correlation coefficients were performed (JMP software 4.0, SAS Institute Inc., USA).

3. Results

3.1. Weed Pressure

The dominant weeds were in an order of *Rumex obutusifolius* ≥ *Romexacetosella* > *Plantago asiatica* > *Carex lanceolata* ≥ *Carexnervata*, *Artemisia princeps* > *Ixeris dentata* > *Potentilla freynian* > *Weigela hortensis*, irrespective of grass species or cultivars (Not shown in Table). Among all the cultivars, the highest and lowest infestations of weeds were observed during the July and September growing seasons (Table 3). The weeds such as *Rumex obutusifolius* and/or *Romexacetosella* were dominated by Tachiwase, Fawn, and Okamidori species of Italian ryegrass, tall fescue, and orchardgrass, respectively. The average weed pressure among Italian ryegrass cultivars is Tachiwase > Waseyutaka > Ace ≥ Magnet.

Table 3. Weed percentage of different grass cultivars across four harvests[1,2,3].

Species	Cultivar	Type	May	July	September	October	Mean[4]
Italian ryegrass	Magnet	HighMg	14.2bA	1.8bC	15.1bA	6.2bB	9.4
	Ace	Commercial	13.9bA	1.2bC	14.1bA	4.3bB	8.4
	Tachiwase	Commercial	30.5aA	4.9aD	20.9aB	12.9aC	17.3
	Waseyutaka	Commercial	12.8bA	3.7aC	14.1bA	7.3bB	9.5
Relative range (%)							−19.9
Tall fescue	HiMag	HighMg	8.1bB	2.0bC	15.5aA	8.6bB	8.6
	Fawn	Commercial	9.9abB	2.1bC	15.9aA	13.7aA	10.4
	Hokuryo	Commercial	10.9aB	3.4aC	17.3aA	7.9bB	9.9
	Ky-31	Commercial	9.9abA	2.5bC	12.9aA	7.3bB	8.5
Relative range (%)							−9.5
Orchardgrass	Mgwell	HighMg	8.1bB	1.9bC	15.1aA	6.6bB	7.9
	Akimidori	Commercial	9.9abB	2.3abC	16.0aA	8.9aB	9.3
	Okamidori	Commercial	10.9aA	4.1aB	16.9aA	9.3aA	10.3
		Relative range (%)					−13.6

[1]Values for average of 3 transects (1 m × 1 m) from each plot. [2]Values within column and species with the same letter(s) are not significantly different at P ≤ 0.05. [3]Values within row and harvest with the same uppercase letters are not significantly different at P ≤ 0.05. [4]Mean of May to September harvests.

In tall fescue cultivars, the weed pressure was ranked as Fawn > Hokuryo > Ky-31 ≥ HiMag, while in orchardgrass the weed dominance was Okamidori > Akimidori > Mgwell. Across harvests, weed pressure was consistently lower within the high-Mg cultivars than within the commercial cultivars with the relative range of −19.9%, −9.5%, and −13.6% for Italian ryegrass, tall fescue, and orchardgrass species, respectively, suggesting weeds could be controlled by introducing high-Mg grass cultivars in temperate Andisols.

3.2. Plant Development and Biomass Production

The plant height was shorter in high-Mg cultivars compared to commercial cultivars, irrespective of grass species (**Table 4**). The plant height between high-Mg cultivars and commercial cultivars ranked as orchardgrass > Italian ryegrass > tall fescue. The plant height was tallest in the July harvest and the lowest in the October harvest, regardless of grass species or cultivars.

While the dry-matter yield of grass cultivars varied among the harvests (**Table 5**) and recorded the lowest seasonal yield in high-Mg containing cultivars when compared to commercial cultivars with the relative range −10%, −5.2%, and −8.7%, for Italian ryegrass, tall fescue, and orchardgrass respectively. Results indicated that when compared with both Magnet Italian ryegrass and Mgwell orchardgrass, the HiMag tall fescue responded better in dry-matter production,

Table 4. Plant height (cm) of different grass species across four harvests[1,2,3].

Species	Cultivar	Type	May	July	September	October	Mean[4]
Italian ryegrass	Magnet	HighMg	91.5aA	96.4aA	83.0aB	64.7aC	83.9
	Ace	Commercial	96.9aA	102aA	91.8aA	71.0aB	90.4
	Tachiwase	Commercial	96.9aA	106aA	89.2aB	68.0aC	90.0
	Waseyutaka	Commercial	98.0aA	106aA	92.9aB	69.0aC	91.5
Relative range (%)							−7.41
Tall fescue	HiMag	HighMg	88.4aB	98.8aA	89.8aB	57.9aC	83.7
	Fawn	Commercial	95.0aAB	98.5aA	91.8aB	70.5aC	88.9
	Hokuryo	Commercial	91.9aB	99.3aA	85.9aB	66.5aC	85.9
	Ky-31	Commercial	90.8aB	101aA	91.2aB	60.4aC	85.9
Relative range (%)							−3.68
Orchardgrass	Mgwell	HighMg	105.6aA	107aA	99.1aA	90.3aB	100.5
	Akimidori	Commercial	106.7aA	112aA	102aA	91.6aB	102.9
	Okamidori	Commercial	108.3aA	110aA	102aA	90.8aB	102.8
		Relative range (%)					−1.54

[1]Values for average of 10 plants from each plot. [2]Values within column and species with the same lowercase letters are not significantly different at P ≤ 0.05. [3]Values within row and harvest with the same uppercase letters are not significantly different at P ≤ 0.05. [4]Mean of May to September harvests.

Table 5. Dry matter yield ($t ha^{-1}$) of different grass species across four harvests[1,2,3].

Species	Cultivar	Type	May	July	September	October	Seasonal[4]
Italian ryegrass	Magnet	HighMg	4.15aA	3.59aA	2.35aB	3.50aA	13.58
	Tachiwase	Commercial	4.46aA	3.47aAB	3.16aB	3.69aAB	14.77
	Ace	Commercial	4.13aA	3.25aA	3.76aA	3.76aA	14.90
	Waseyutaka	Commercial	4.78aA	3.12aB	3.34aAB	4.33aA	15.58
Relative range (%)							−9.97
Tall fescue	HiMag	HighMg	3.03aA	2.98aA	3.40aA	3.38aA	12.81
	Fawn	Commercial	3.56aA	3.52aA	3.05aA	3.65aA	13.80
	Hokuryo	Commercial	3.43aA	3.61aA	3.12aA	3.12aA	13.30
	Ky-31	Commercial	3.34aA	2.89aA	3.81aA	3.35aA	13.40
Relative range (%)							−5.16
Orchardgrass	Mgwell	HighMg	2.04aA	2.12aA	2.30aA	1.49aB	7.95
	Akimidori	Commercial	2.53aA	2.20aA	2.71aA	1.74aB	9.19
	Okamidori	Commercial	2.23aA	2.31aA	2.64aA	1.79aB	8.98
		Relative range (%)					−8.66

[1]Values within column and species with the same lowercase letters are not significantly different at $P \le 0.05$. [2]Values within row and harvest with the same uppercase letters are not significantly different at $P \le 0.05$. [3]Values for Italian ryegrass includes reseeding plants from the original plants. [4]Total of May to September harvests.

while the seasonal yields ranked as Magnet > HiMag > Mgwell. Among the grasses, the total forage yield was highest in Italian ryegrass followed by tall fescue, and the lowest in orchardgrass, irrespective of cultivars. Likewise, the highest dry-matter yield was recorded in the May harvest for Italian ryegrass and in the October harvest for orchardgrass. For cultivars of tall fescue, the yield decreased from May to the July harvests and then continued to increase in the September harvest.

3.3. Potassium, Calcium and Magnesium Concentration

The highest K content for Italian ryegrass was observed in September which can be attributed to the low dry-matter yield for that harvest (Table 6). Regardless of the cultivars of tall fescue and orchardgrass species, the highest K content was recorded in October harvests which can be partially associated with the low dry-matter yield for that harvest. Among the Italian ryegrass species, the highest K content was determined in Waseyutaka and the lowest was in Tachiwase over four harvests. Likewise, the highest K content was observed in Ky-31 and the lowest was in Hokuryo cultivars in tall fescue species across the harvests. For orchardgrass species, the cultivar Okamidori showed the highest and Mgwell showed the lowest K content across harvests. Tall fescue cultivar Hokuryo showed a lower K content than the other three cultivars across the harvests. Irrespective of grass

Table 6. Potassium content (% dry matter) of different grass species across four harvests[1,2,3].

Species	Cultivar	Type	May	July	September	October	Mean[4]
Italian ryegrass	Magnet	HighMg	3.12aB	2.87aB	4.86aA	4.04aA	3.72
	Ace	Commercial	2.87aB	3.06aB	5.18aA	3.51bB	3.65
	Tachiwase	Commercial	3.04aB	2.32bC	4.67abA	4.33aA	3.59
	Waseyutaka	Commercial	3.08aC	2.68abC	5.64aA	4.15ab	3.89
Relative range (%)							0.34
Tall fescue	HiMag	HighMg	3.82aB	3.86aB	4.05bA	4.21aA	3.98
	Fawn	Commercial	3.59abB	4.04aAB	4.45aA	4.32aA	4.10
	Hokuryo	Commercial	3.12bA	3.92aA	4.37aA	4.40bA	3.86
	Ky-31	Commercial	3.86aA	4.08aA	4.38aA	4.64aA	4.24
Relative range (%)							−1.99
Orchardgrass	Mgwell	HighMg	2.30aB	2.64aB	3.92aA	3.39aA	3.06
	Akimidori	Commercial	2.66abB	3.79aA	3.94aAB	4.26aA	3.66
	Okamidori	Commercial	2.73bB	3.99aAB	3.70aA	4.25aA	3.67
		Relative range (%)					−2.24

[1]Values within column and species with the same lowercase letters are not significantly different at $P \leq 0.05$. [2]Values within row and harvest with the same uppercase letters are not significantly different at $P \leq 0.05$. [3]Values for Italian ryegrass includes reseeding plants from the original plants. [4]Mean of May to September harvests.

species and cultivars, the mean K content was lowest in Okamidori (2.72% DM) while the highest was in Ky-31 (4.42% DM). Among all cultivars, Mgwell cultivars accumulated less K than that of other high-Mg cultivars regardless of the species. Across the harvests, tall fescue HiMag and orchardgrass Mgwell showed lower K content when compared to their commercial cultivars with a relative range of −1.99% and −2.24%, respectively. The Waseyutaka showed the highest K content among all cultivars of Italian ryegrass; however, over the four harvests, Magnet showed higher K content than that of the commercial cultivars with a relative range of 0.34%.

In Italian ryegrass Waseyutaka, and all cultivars of tall fescue, Ca content was increased in the July harvest compared to the May harvest and then decreased in the September and October harvests, respectively (Table 7). In other words, the lowest Ca content in tall fescue was recorded in the September harvest when compared to the highest Ca content was recorded in the October harvest. The Ca content increased gradually over time in all cultivars of orchardgrass species and in the Ace cultivar of tall fescue. The Magnet Italian ryegrass, HiMag tall fescue, and Mgwell orchardgrass cultivars showed a significant increase in Ca content than others. Mean forage Ca content varied from 0.35% to 0.53% across harvests, irrespective of grass cultivars. The Ca content averaged across harvests for tall

Table 7. Calcium content (% dry matter) of different grass species across four harvests[1,2,3].

Species	Cultivar	Type	May	July	September	October	Mean[4]
Italian ryegrass	Magnet	HighMg	0.50aAB	0.45bB	0.59aA	0.50abB	0.51
	Ace	Commercial	0.32bB	0.34cB	0.40cAB	0.42bA	0.37
	Tachiwase	Commercial	0.49aAB	0.47bAB	0.39cB	0.56aA	0.48
	Waseyutaka	Commercial	0.48aB	0.57aA	0.48bB	0.51abAB	0.51
Relative range (%)							11.5
Tall fescue	HiMag	HighMg	0.39aB	0.45aAB	0.36aB	0.59aA	0.44
	Fawn	Commercial	0.27cB	0.37bB	0.34aB	0.40cA	0.35
	Hokuryo	Commercial	0.31bcB	0.39abAB	0.32aB	0.42bcA	0.36
	Ky-31	Commercial	0.32bB	0.42abAB	0.35aCB	0.50bA	0.40
Relative range (%)							19.8
Orchardgrass	Mgwell	HighMg	0.29aB	0.51abA	0.56aA	0.59aA	0.48
	Akimidori	Commercial	0.25bB	0.44bA	0.46bA	0.49aA	0.41
	Okamidori	Commercial	0.25bB	0.53aA	0.50bA	0.54aA	0.46
				Relative range (%)			7.16

[1)]Values within column and species with the same lowercase letters are not significantly different at $P \leq 0.05$. [2)]Values within row and harvest with the same uppercase letters are not significantly different at $P \leq 0.05$. [3)]Values for Italian ryegrass includes reseeding plants from the original plants. [4)]Mean of May to September harvests.

fescue was 0.35% to 0.44% and for orchardgrass 0.41% to 0.48%, respectively, while Italian ryegrass ranged between 0.51% to 0.68%.

Across all harvests, the HighMg cultivars showed a significantly higher Mg content when compared with the commercial cultivars of respective species (**Table 8**). Cultivars of Italian ryegrass had highest Mg content in the September harvest while having the lowest in the May harvest. For tall fescue, the highest Mg values were observed in the October harvest while the lowest values were recorded in the May harvest. However, there was no significant difference in content Mg among the tall fescue cultivars between the July and September harvests. Among the orchardgrass cultivars, the lowest and highest Mg content in forages were determined in the May and October harvests, respectively.

3.4. Grass Tetany Index [K/(Ca + Mg)]

The K/(Ca + Mg) was calculated on an equivalent molecular weight basis to evaluate the balance of K content against Ca and Mg contents and these values indicate the potential risk of grass tetany as caused by forage grasses (**Table 9**). The values for the equivalent ratio of tetany risk were highest in the September harvest and lowest in the July harvest for all cultivars of Italian ryegrass species. The lowest grass tetany ratio was recorded in October for tall fescue and July for orchardgrass, irrespective of cultivars. While considering different species for

Table 8. Magnesium content (% dry matter) of different grass species across four harvests[1,2,3].

Species	Cultivar	Type	May	July	September	October	Mean[4]
Italian ryegrass	Magnet	HighMg	0.20aC	0.24aBC	0.35aA	0.29aB	0.27
	Ace	Commercial	0.13bB	0.18bB	0.26cA	0.28aA	0.22
	Tachiwase	Commercial	0.15abC	0.17bC	0.31bA	0.25aB	0.23
	Waseyutaka	Commercial	0.17aC	0.24aBC	0.29bcA	0.26aB	0.25
Relative range (%)							16.5
Tall fescue	HiMag	HighMg	0.35aB	0.37aB	0.37aB	0.52aA	0.40
	Fawn	Commercial	0.19bB	0.33aA	0.36aA	0.36cA	0.32
	Hokuryo	Commercial	0.19bB	0.32aA	0.33aA	0.39bcA	0.31
	Ky-31	Commercial	0.18bC	0.31aB	0.32aB	0.41bA	0.31
Relative range (%)							27.2
Orchardgrass	Mgwell	HighMg	0.35aB	0.40aA	0.42aA	0.45aA	0.40
	Akimidori	Commercial	0.21bB	0.36aA	0.35bA	0.36bA	0.32
	Okamidori	Commercial	0.20bB	0.40aA	0.37bA	0.39abA	0.34
		Relative range (%)					13.2

[1]Values within column and species with the same lowercase letters are not significantly different at P ≤ 0.05. [2]Values within row and harvest with the same uppercase letters are not significantly different at P ≤ 0.05. [3]Values for Italian ryegrass includes reseeding plants from the original plants. [4]Mean of May to September harvests.

Table 9. Equivalent ratio of K/(Ca + Mg) of different grass species across four harvests[1,2,3].

Species	Cultivar	Type	May	July	September	October	Mean[4]
Italian ryegrass	Magnet	HighMg	1.94bAB	1.74bB	2.13bA	2.12aA	2.00
	Ace	Commercial	2.74aAB	2.44aB	3.13aA	2.01aB	2.57
	Tachiwase	Commercial	2.15abAB	1.55bcB	2.62bA	2.25aAB	2.14
	Waseyutaka	Commercial	2.07bB	1.41cC	2.95abA	2.22aB	2.16
Relative range (%)							−12.9
Tall fescue	HiMag	HighMg	2.07bA	1.88aAB	2.16aA	1.48bB	1.90
	Fawn	Commercial	3.14aA	2.21aB	2.39aB	2.19aB	2.48
	Hokuryo	Commercial	2.64bA	2.16aAB	2.53aA	1.92aB	2.31
	Ky-31	Commercial	3.17aA	2.19aB	2.51aB	1.98aB	2.46
Relative range (%)							−21.3
Orchardgrass	Mgwell	HighMg	1.37bA	1.15bA	1.60aA	1.31bA	1.36
	Akimidori	Commercial	2.36aA	1.86aB	1.92aB	1.99aAB	2.03
	Okamidori	Commercial	2.44aA	1.17bB	1.69aB	1.81aB	1.91
Relative range (%)							−22.9

[1]Calculated on an equivalent per kilogram basis. [2]Values within column and species with the same lowercase letters are not significantly different at P ≤ 0.05. [3]Values within row and harvest with the same uppercase letters are not significantly different at P ≤ 0.05. [4]Mean of May to September harvests.

grazing it seems that orchardgrass has less tetany risk throughout the season. For high Mg containing cultivars of Italian ryegrass (Magnet) and tall fescue (HiMag), the equivalent ratios fluctuated with harvests; however, the values were lower than the threshold values of grass tetany risk compared to the commercial cultivars. In the autumn harvest (September), the K/(Ca + Mg) values posed a risk of grass tetany in the HiMag cultivar of Italian ryegrass and tall fescue, respectively.

The Ace Italian ryegrass showed lower tetany values than the critical level of equivalent ratio (2.2) only in the October harvest. In contrast, the Tachiwase and Waseyutaka (commercial cultivars) which are commonly used as forages in Japan showed lower tetany values than the critical level of equivalent ratio only in May and July harvests. The Fawn, Hokuryo and Ky-31 cultivars of tall fescue species showed grass tetany risk in May and September harvests. The values of relative range for grass tetany risk were recorded in orchardgrass (−22.9%) followed by tall fescue (−21.3%) and the lowest in Italian ryegrass (−12.9%), respectively.

3.5. Phenotypic Correlation

The plant height of tall fescue positively and significantly correlated with the grass tetany index (GTI). Likewise, the HighMg cultivar dry matter positively correlated with the GTI. Moreover, a species-specific correlation was observed between K content and the GTI. In Italian ryegrass species, K content positively correlated with Mg($r = 0.735$) and the GTI ($r = 0.682$). However, in tall fescue, K content positively correlated with Mg content ($r = 0.630$) but negatively correlated with the GTI ($r = -0.337$). In commercial cultivars, K content positively correlated with Mg content but poorly correlated with the GTI ($r = 0.284$). While in high Mg cultivars, K content significantly correlated with the GTI ($r = 0.620$), it did not correlate significantly with Mg content ($r = 0.279$). In orchardgrass, K content moderately correlated with Ca ($r = 0.654$) and the Mg ($r = 0.409$). However, the correlation between K content was very weak and insignificant with GTI ($r = 0.052$) in orchardgrass. In commercial cultivars, K content was significantly correlated with Mg content ($r = 0.629$) but not with the GTI ($r = 0.284$). In contrast, K content significantly and positively correlated with the GTI ($r = 0.620$) but not with Mg content ($r = 0.279$) in the high Mg cultivars. Likewise, Ca content significantly correlated with the GTI among all species and cultivars except the High Mg cultivar. Regardless of the grass species and cultivars, a positive correlation was observed between Ca content and Mg content. Grass tetany index negatively correlated with Mg and Ca regardless of species (Table 10).

4. Discussion

4.1. Weed Pressure

A significantly reduced weeds pressure has shown how undesirable and highly invasive weeds could be controlled by introducing highMg grass cultivars in

Table 10. Correlation among phenotypic traits of cool season grasses from pooled data over four harvests*.

	Grass	Plant height	Dry matter	K	Ca	Mg	GTI
Weed	Italian ryegrass	0.033	0.029	0.294	0.072	−0.078	0.361
	Tall fescue	−0.214	0.045	0.229	−0.368	−0.069	0.328
	Orchardgrass	−0.277	0.118	0.229	−0.057	−0.186	0.404
	Commercial	−0.110	0.246	0.165	−0.041	−0.150	0.309
	HighMg	−0.163	0.210	0.472	0.114	−0.004	0.416
	Combined	−0.104	0.070	0.235	−0.031	−0.174	0.331
Plant height	Italian ryegrass		0.002	−0.462	−0.271	−0.560	−0.113
	Tall fescue		0.052	−0.451	−0.553	−0.625	0.468
	Orchardgrass		0.063	−0.430	−0.527	−0.439	0.071
	Commercial		−0.351	−0.456	−0.185	−0.270	−0.137
	HighMg		−0.379	−0.634	−0.419	−0.216	−0.346
	Combined		−0.101	−0.485	−0.242	−0.230	−0.146
Dry matter	Italian ryegrass			−0.050	−0.005	−0.049	0.013
	Tall fescue			−0.123	0.172	−0.106	−0.058
	Orchardgrass			0.034	−0.013	−0.027	0.096
	Commercial			−0.077	−0.066	−0.498	0.334
	HighMg			0.194	−0.084	−0.534	0.595
	Combined			0.029	−0.001	−0.119	0.130
K	Italian ryegrass				0.126	0.735	0.682
	Tall fescue				0.325	0.630	−0.338
	Orchardgrass				0.654	0.409	0.052
	Commercial				0.210	0.629	0.284
	HighMg				0.455	0.279	0.620
	Combined				0.174	0.399	0.394
Ca	Italian ryegrass					0.398	−0.544
	Tall fescue					0.685	−0.835
	Orchardgrass					0.901	−0.610
	Commercial					0.355	−0.651
	HighMg					0.330	−0.163
	Combined					0.380	−0.599
Mg	Italian ryegrass						0.109
	Tall fescue						−0.871
	Orchardgrass						−0.834
	Commercial						−0.422
	HighMg						−0.492
	Combined						−0.537

*Shaded indicates statistically significance at P ≤ 0.05.

temperate Andisols. Weeds especially docks (*Rumex* sp.) are one of the highly adaptive ones predominant in intensive grassland management systems globally [29]. Haggar [30] reported that docks were most prevalent and associated with the frequent usage of chemical fertilizers. As our grass management systems were based on chemical fertilizers, *i.e.*, increasing infestations of docks were judged [31].

4.2. Plant Height and Biomass Production

A significantly lower amount of dry matter produced by High Mg cultivars when compared to commercial cultivars was reported in other countries as the herbage yield depends on nutrient availability to support adequate nutrition and growth [32] [33]. Irrespective of cultivars, the annual dry-matter production of Italian ryegrass (14.7-ton·ha^{-1}), tall fescue (13.3-ton·ha^{-1}), and orchardgrass (5.3-ton·ha^{-1}) seemed lower in Andisol than other soils regardless of species or cultivars when compared with dry-matter production of tall fescue [34], Italian ryegrass, and orchardgrass [35], respectively. Rahman and Saiga [17] reported 6.4-to-14.1-ton·ha^{-1}·y^{-1} biomass production of orchardgrass grown in Andisol under different management practices. The total annual dry matter production of forage mixtures was about 13 to 16-ton·ha^{-1} in red brown soil under slurry application [36]. In Andisol of New Zealand, up to 20-tondry dry-matter, ha^{-1} of pasture yield was recorded under suitable climatic and site conditions [37]. Both biotic- and abiotic stresses, as well as the microclimate, may be responsible for the lower production of aboveground biomass of cool-season grasses in the temperate region of Japan. A supplement of Mg significantly increased the yield in grass species, concurrently declining concentrations of crude protein, Ca, sodium (Na), manganese (Mn), and K/Mg and Ca/Mg as the dilution effect of biomass increased with increased Mg content [38].

4.3. Crown Nutrients

While the Mg contents among the cultivars of different species were similar, the HiMg tall fescue and Mgwell orchardgrass had lower K content than with their respective commercial cultivars. It is reported that the absorption of Ca and Mg by ruminants decreased with increased K levels in forage [33]. Smith *et al.* [39] suggested that when forage K content is ≥25 g·kg^{-1} dry-matter, the Mg content decreased to 1.9 g·kg^{-1}. Similarly, when the forage K content increased to 65 g·kg^{-1}, the Ca content decreased to 6 g·kg^{-1}. Sleper *et al.* [5] indicated that the correlations between herbage yield and K, Ca, and Mg contents were low in tall fescue. In support of our results, Wilkinson and Mayland [40] reported that the total yield of HiMag tall fescue was 11% less than that of Ky-31. When the high Mg Italian ryegrass cultivar Magnet was compared with other cultivars, its dry-matter yield was between 8% and 10% lower than the higher yielding cultivar [41]. Then one significant difference in yield between high Mg and commercial cultivars indicated that the cultivars bred for high Mg contents did not compensate their yield under temperate climates. While mean K content varied between 2.72 (Okamidori) to 4.42% (Ky-31), which is accepted as adequate for sheep nutrition [42], but inadequate for cattle [43] and deer [44] nutrient requirements. The values are low to support adequate vegetative growth of grasses [33] [32]; however, are nontoxic for herbage quality [45]. Among all cultivars, HighMg cultivars showed a low relative range suggesting that HighMg cultivars accumulated lower K than other commercial cultivars which was the main focus of the breeding program.

The mean forage Ca content varied between 0.35% to 0.53% across harvests, irrespective of grass cultivars which are considered inadequate for sheep [42], cattle [32], and deer [46] nutrition. However, the Ca content averaged across harvests was adequate in tall fescue and orchardgrass, respectively, while in Italian ryegrass its content was higher for grasses [47]. The Mg content increased consistently across the season which is below the normal levels of forage grasses [39]. These high Mg-containing cultivars had higher Ca content which had consequently lower K/(Ca + Mg) with minimum risks of grass tetany [48]. A significant variation in Ca content in cool-season grasses across harvests observed in our studies collaborates with the findings of Jones and Tracy [49]. They reported a similar phenomenon of variation in herbage mineral contents throughout the growing season. A significantly higher Ca and Mg content across harvests recorded in high Mg cultivars compared to commercial cultivars, irrespective of species justifies the breeding of high Mg cool-season grass cultivars. The grasses bred for high Mg content are anticipated to balance K optimization with high Mg and Ca uptakes which could minimize grass tetany risks in temperate regions of the world including Japan. The lowest nutrient content was recorded in the May harvest, regardless of species and cultivars may be associated with the rapid vegetative growth of grasses in the spring. The Energy Dispersive X-ray Microanalysis showed that Mg density in xylem had higher Mg content across two harvests and higher Ca and lower K contents for HiMag tall fescue when compared to commercial tall fescue cultivar Ky-31 [50]. The HiMag was able to translocation more Mg from root to shoot than the commercial cultivars. A doubling of the K in nutrient solution decreased Mg content and increased K/Mg in roots but did not significantly affect Mg contents in crowns nor leaves. Increasing the Mg in solution increased Mg content in roots, crowns, and leaves. This may be evidenced associated with several metabolic processes that limit K uptake and possibly favor a greater Mg translocation in all tall fescue cultivars [34]. Sabreen *et al.* [51] reported a similar trend in separate studies with optimized nutrient solutions using increased K concentration in nutrient solutions [52], where three cool-season High Mg cultivars of Italian ryegrass, tall fescue, and orchardgrass were compared with commercial cultivars of respective species.

4.4. Grass Tetany Index [K/(Ca + Mg)]

The grass tetany index, [K/(Ca + Mg)] has shown fluctuations with harvests, and the index of 1.4 was recorded in Mgwell (orchardgrass), 1.90 in HiMag (tall fescue), and 2.00 in Magnet (Italian ryegrass), respectively across harvests. Among the species, the lowest K/(Ca + Mg) was observed for Mgwell which is bred by Saiga *et al.* [21]. It appears that the values of grass tetany ratio [K/(Ca + Mg)]were lower than the threshold values in all high Mg cultivars, suggesting that all cultivars breed for high Mg content are safe for grazing ruminants throughout the seasons in temperate region globally including Japan. Moseley and Baker [41] evaluated the Magnet Italian ryegrass cultivar (Bb2067) for its efficacy in alleviating the incidence of hypomagnesemia in lactating ewes by comparing it with the control pasture cv. RvP, and Magnet (Italian ryegrass) maintained higher Mg contents throughout the season. High levels of hypomagnesemia and fatalities were reported in animals grazing RvP control cultivar than those on Magnet (Italian ryegrass) pasture, which was corroborated by differences in serum Mg concentration. Moseley and Baker [41] concluded that the use of a high Mg grass cultivar consistently proved its effectiveness to reduce hypomagnesemia under intensive grazing. A high Mg Italian ryegrass had a consistently greater Mg content associated with lower K content and grass tetany index than the low Mg cultivar (Bb1276). They reported that the high Mg content was not related to forage yields in Aberystwyth (Wales) and Edinburgh (Scotland). Therefore, the Bb2067 cultivar was assumed a promising cultivar for reducing grass tetany, and the cultivar was enclosed for breeding programs [53]. Binnie *et al.* [19] compared perennial ryegrass (Ramore) bred for high Mg content with a control cultivar, aiming for its ability to reduce the grass tetany incidence. They suggested that under grazing high Mg cultivars provide a proactive management strategy to reduce the grass tetany incidence. All these observations are in collaboration with our results.

Rahman and Saiga [17] studied the impact of dairy manure on mineral nutrient uptake patterns with a special reference to tetany potential risk by orchardgrass grown in Andisol of Japan. They observed that the uptake of grass tetany-related mineral nutrients was greater in dairy manure amendments than that of the chemical fertilizers and the uptake of mineral nutrients in forages was directly proportionate to grass tetany risk. This demonstrated that soil amendments (viz. slurry, compost, vermicompost, biochar) might change grass tetany potentiality as they alter soil oxygen diffusion resulting in impeding gas exchange processes by water-saturation of soil pores. Introducing grass with biological modifications (endophyte infection) showed inconsistent results on the uptake of mineral nutrients with soil types and plant ecotypes [54] [18]. Therefore, breeding grass could be the other possibility to minimize grass tetany risk.

The HiMag tall fescue is a cultivar selected for increased Mg and Ca uptake to reduce the K/(Ca + Mg). Wilkinson and Mayland [40] compared the HiMag with other tall fescue cultivars for mineral concentration and their results showed that the shoot Mg and Ca contents were greater and the K/(Ca + Mg) was lower for HiMag than the Ky-31. Crawford *et al.* [55] reported that cattle grazing of HiMag is likely had a reduced risk of grass tetany. Our results showed the same phenomenon for Magnet Italian ryegrass, HiMag tall fescue, and orchardgrass grown in temperate Andisol. However, the K/(Ca + Mg) for Magnet Italian ryegrass and the HiMag tall fescue were 2.12 in the October harvest and 2.16 in the September harvest, respectively, which is very close to grass tetany risk. It can be inferred that Magnet Italian ryegrass and HiMag fescue could be a grass tetany risk for ruminants during October and September harvests, respectively, while growing in temperate Andisols globally including Japan. The higher shoot Mg content and the lower K/(Ca + Mg) in high Mg-containing cultivars proved that there is significant potential to breed forages with a balanced uptake of nutrients (K, Ca, and Mg) to improve animal health hazards. The relative range values for grass tetany risk between high Mg and commercial cultivars were negative for all species which indicates that the selection of high Mg grasses via breeding is justifiable [56]. Our results suggested that breeding grass species for reducing grass tetany are reliable for temperate regions of Japanese Andisol.

4.5. Relationship among the Plant Traits

As one of the macronutrients essential for plant growth, K plays important physiological roles in photosynthesis and stomatal control of transpiration and thus, participates in other biochemical roles in protein synthesis [1]. Therefore, a significant correlation of K and Mg contents with forage dry matter production would be expected; however, we did not observe any correlations of K with dry-matter production in our experiments. While the Mg content was negatively correlated with dry-matter production in commercial cultivars and high Mg cultivar, the Mg and Ca contents negatively and K positively correlated with the GTI as expected. Similar relations were reported by other studies [16] [57] [58]. In high Mg cultivars, the K content positively and Mg content negatively correlated with the GTI, which suggests that the selection of the High Mg cultivar had a positive effect on grass tetany risk in ruminants.

5. Conclusion

Field research in the USA, Europe, Australia, and New Zealand revealed that high Mg containing Italian ryegrass, tall fescue, and orchardgrass bred by scientists had significantly higher shoot Mg concentrations than commercial cultivars. These high Mg-containing cultivars were also higher in Ca concentration, and consequently reduced K concentration, resulting in a lower K(Ca + Mg) ratio than the commercial cultivars. We evaluated those species of high magnesium-containing cultivars with commercials cultivars for grass tetany risk in the Japanese climate. Results showed that the high Mg Italian ryegrass, tall fescue, and orchardgrass breeds had consistently higher shoot Mg content than that of the commercial cultivars. These high Mg cultivars had a higher Ca content, which was consequently associated with K content with a resulting decrease in grass tetany risk than the commercial cultivars grown in temperate Andisols. It could be conferred that the tetany index in grass species is very much age (harvest) specific and microclimate and site-dependent. While there is a possibility that high Mg cultivars moderately perform to dry-matter productivity, further research on dry-matter productivity of high Mg forage is required.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

[1] Marschner, P. (2011) Marschner's Mineral Nutrition of Higher Plants. 3rd Edition, Academic Press, San Diego.

[2] Cakmak, I. and Marschner, H. (1992) Magnesium Deficiency and High Light Intensity Enhance Activities of Superoxide Dismutase, Ascorbate Peroxidase and Glutathione Reductase in Bean Leaves. *Plant Physiology*, **98**, 1222-1227. https://doi.org/10.1104/pp.98.4.1222

[3] Rengel, Z., Bose, J., Chen, Q. and Tripathi, B.N. (2015) Magnesium Alleviates Plant Toxicity of Aluminium and Heavy Metals. *Crop and Pasture Science*, **66**, 1298-1307. https://doi.org/10.1071/CP15284

[4] White, P.J. and Broadley, M.R. (2009) Biofortification of Crops with Seven Mineral Elements Often Lacking in Human Diets-Iron, Zinc, Copper, Calcium, Magnesium, Selenium and Iodine. *New Phytologist*, **182**, 49-84. https://doi.org/10.1111/j.1469-8137.2008.02738.x

[5] Sleper, D.A., Vogel, K.P., Asay, K.H. and Mayland, H.F. (1989) Using Plant Breeding and Genetics to Overcome the Incidence of Grass Tetany. *Journal of Animal Science*, **67**, 3456-3462. https://doi.org/10.2527/jas1989.67123456x

[6] Mayland, H.F. and Grunes, D.L. (1979) Soil-Climate-Plant-Relationships on the Etiology of Grass Tetany. In: Rendig, V.V. and Grunes, D.L., Eds., *Grass Tetany*, ASA Special Publication No. 35, ASA, Madison, WI, 123-175. https://doi.org/10.2134/asaspecpub35.c6

[7] Mayland, H.F. and Sleper, D.A. (1993) Developing a Tall Fescue for Reduced Grass Tetany Risk. *Proceedings XIV International Grassland Congress*, Palmerston North, Vol. 17, 1095-1096

[8] McCoy, M.A., Goodall, E.A. and Kennedy, D.G. (1993) Incidence of Hypomagnesemia in Dairy and Suckler Cows in Northern Ireland. *The Veterinary Record*, **132**, Article No. 537. https://doi.org/10.1136/vr.132.21.537

[9] Baker, D.O. (1976) A Survey of the Incidence of Deaths from Hypomagnesaemic Grass Tetany and of Some Management Factors Possibly Related to Their Occurrence in Beef Cows in the Wairarapa. *New Zealand Journal of Experimental Agriculture*, **4**, 51-55. https://doi.org/10.1080/03015521.1976.10425844

[10] Lewis, D.C. and Sparrow, L.A. (1991) Implications of Soil Type, Pasture Composition and Mineral Content of Pasture Components for the Incidence of Grass Tetany in the Southeast of South Australia. *Australia Journal of Experimental Agriculture*, **31**, 609-615. https://doi.org/10.1071/EA9910609

[11] Saiga, S. (1997) Possibility of Livestock Poisoning Caused by Endophyte. *The Dairy Situation*, **57**, 14-21.

[12] Asay, K.H., Mayland, F.H., Jefferson, P.G., Berdahl, J.D., Karn, J.F. and Waldron, B.L. (2001) Parent-Progeny Relationships and Genotype x Environment Effects for Factors Associated with Grass Tetany and Forage Quality in Russian Wildrye. *Crop Science*, **41**, 1478-1484. https://doi.org/10.2135/cropsci2001.4151478x

[13] Hovin, A.W., Tew, T.L. and Stucker, R.E. (1978) Genetic Variability for Mineral Elements in Reed Canary Grass. *Crop Science*, **82**, 81-304.

[14] Hides, D.H. and Thomas, T.A. (1981) Variation in the Magnesium Content of Grasses and Its Improvement by Selection. *Journal of the Science Food and Agriculture*, **32**, 990-991.

[15] Buchner, R.C., Burrus II, P.B., Corneliu, P.L., Bush, L.P. and Leggett, J.E. (1981) Genetic Variability and Heritability of Certain Forage Quality and Mineral Constituents in *Lolium-Festuca* Hybrid Derivatives. *Crop Science*, **21**, 419-423. https://doi.org/10.2135/cropsci1981.0011183X002100030016x

[16] Vogel, K.P., Mayland, H.F., Reece, P.E. and Lamb, J.F.S. (1989) Genetic Variability for Mineral Element Concentration of Crested Wheatgrass Forage. *Crop Science*, **29**, 1146-1150. https://doi.org/10.2135/cropsci1989.0011183X002900050009x

[17] Rahman, M.H. and Saiga, S. (2007) Genetic Variability in Tetany Potential of Orchardgrass as Influenced by Application of Dairy Manure and Chemical Fertilizer. *International Journal of Soil Science*, **2**, 29-39. https://doi.org/10.3923/ijss.2007.29.39

[18] Rahman, M.H. and Saiga, S. (2007) Endophyte Effects on Nutrient Acquisition in Tall Fescue Grown in Andisols. *Journal of Plant Nutrition*, **30**, 2141-2158. https://doi.org/10.1080/01904160701700632

[19] Binnie, R.C., Johnstone, D.T. and Chestnutt, D.M.B. (1996) The Effect of a High-Magnesium Perennial Ryegrass Variety on the Magnesium Status of Sheep. *Grass and Forage Science*, **51**, 456-463. https://doi.org/10.1111/j.1365-2494.1996.tb02081.x

[20] Moseley, G., Baker, D.M. and Hides, D.H. (1989) The Efficacy of a High Magnesium Grass in Alleviating Hypomagnesaemia. *Proceedings of XIV International Grassland Congress*, Vol. 16, 395-396.

[21] Saiga, S., Saitoh, H., Sabreen, S. and Tsuiki, M. (2002) Effectiveness of Nutrient Solution Culture for Detecting Genetic Variability in Mg Concentration of Orchardgrass (*Dactylis glomerata* L.). *Journal of the Japanese Grassland Society*, **48**, 209-215.

[22] Sabreen, S., Saiga, S., Saitoh, H., Tsuiki, M. and Kawai, S. (2001) Mineral Absorption of High Magnesium Containing Cultivars of Italian Ryegrass and Tall Fescue. *Grassland Science*, **47**, 312-313.

[23] Hides, D.H. and Lovatt, J.A. (1988) Breeding for Magnesium Content in Italian Ryegrass and Its Effect on Levels of Sodium, Potassium, Calcium and Phosphorus. British Grassland Society Research Meeting, No. 1, Aberystwyth.

[24] Kenneth, J.M., Collins, M., Nelson, C.J. and Redfearn, D.D. (2020) Forages: The Science of Grassland Agriculture. Volume 2, 7th Edition, Wiley-Blackwell, Hoboken, NJ, 968 p.

[25] Sabreen, S., Saiga, S. and Rahman, M.H. (2021) Screening Forage Grasses with Atomic Absorption Spectrometry, X-Ray Fluorescence and X-Ray Microanalysis. *SAARC Journal of Agriculture*, **19**, 245-256. https://doi.org/10.3329/sja.v19i2.57685

[26] Hutton, J.T. and Norrish, K. (1977) Plant Analyses by X-Ray Spectrometry. II Elements of Atomic Number Greater than 20. *X-Ray Spectrometry*, **6**, 12-17. https://doi.org/10.1002/xrs.1300060105

[27] Norrish, K. and Hutton, J.T. (1977) Plant Analyses by X-Ray Spectrometry. I. Low Atomic Number Elements, Sodium to Calcium. *X-Ray Spectrometry*, **6**, 6-11. https://doi.org/10.1002/xrs.1300060104

[28] Kemp, A. and $_T$'Hart, M.L. (1957) Grass Tetany in Grazing Milking Cows. *Netherlands Journal of Agricultural Science*, **5**, 4-17. https://doi.org/10.18174/njas.v5i1.17745

[29] Holm, L.G., PIucknett, D.L., Pancho, J.V. and Herberger, J.P. (1977) The World's Worst Weeds. Distribution and Biology. The University Press of Hawaii, Honolulu.

[30] Haggar, R.J. (1980) Survey of the Incidence of Docks (*Rumex* spp.) in Grassland in Ten Districts in United Kingdom in 1971. *ADAS Quarterly Review*, **39**, 256-270.

[31] Sakai, H., Shimada, Y., Sato, T. and Fujiwara, K. (1971) Study on Ecology and Control of *Rumex obtusifolius*, a Perennial Weed in Grassland. *Weed Research*, **12**, 40-45. https://doi.org/10.3719/weed.1971.12_40

[32] National Research Council (1989) Nutrient Requirements of Sheep. 6th Edition, National Academy Press, Washington DC.

[33] Mayland, H.F. and Wilkinson, S.R. (1996) Mineral Nutrition. In: Moser, L.E., *et al.*, Eds., *Cool-Season Forage Grasses*, Agronomy Monograph No. 34, American Society of Agronomy, Crop Science Society of America, Soil Science Society of America, Madison, 165-191. https://doi.org/10.2134/agronmonogr34.c6

[34] Shewmaker, G.E., Johnson, D.A. and Mayland, H.F. (2008) Mg and K Effects on Cation Uptake and Dry Matter Accumulation in Tall Fescue (*Festuca arundinacea*). *Plant and Soil*, **302**, 283-295. https://doi.org/10.1007/s11104-007-9482-3

[35] Jensen, K.B., Asay, K.H. and Waldron, B.L. (2001) Dry Matter Production of Orchardgrass and Perennial Ryegrass at Five Irrigation Levels. *Crop Science*, **41**, 479-487. https://doi.org/10.2135/cropsci2001.412479x

[36] Rahman, M.H. and Saiga, S. (2007) The Use of Slurry for Managing Forage Mixtures in Temperate Brown Forest Soils. *Journal of Applied Science*, **7**, 687-694. https://doi.org/10.3923/jas.2007.687.694

[37] Clark, D.A., Caradus, J.R., Monaghan, R.M., Sharp, P. and Thorrold, B.S. (2007) Issues and Options for Future Dairy Farming in New Zealand. *New Zealand Journal of Agricultural Research*, **50**, 203-221. https://doi.org/10.1080/00288230709510291

[38] Sun, X., Chen, J., Liu, L., Rosanoff, A., Xiong, X., Zhang, H. and Pei, T. (2018) Effects of Magnesium Fertilizer on the Forage Crude Protein Content Depend upon Available Soil Nitrogen. *Journal of Agricultural and Food Chemistry*, **66**, 1743-1750. https://doi.org/10.1021/acs.jafc.7b04028

[39] Smith, G.S., Cornforth, I.S. and Henderson, H.V. (1985) Critical Leaf Concentrations for Deficiencies of Nitrogen, Potassium, Phosphorus, Sulphur, and Magnesium in Perennial Ryegrass. *New Phytologist*, **101**, 393-409. https://doi.org/10.1111/j.1469-8137.1985.tb02846.x

[40] Wilkinson, S.R. and Mayland, H.F. (1997) Yield and Mineral Concentration of Hi-Mag Compared to Other Tall Fescue Cultivars Grown in the Southern Piedmont. *Journal of Plant Nutrition*, **20**, 1317-1331.

https://doi.org/10.1080/01904169709365337

[41] Moseley, G. and Baker, D.H. (1991) The Efficacy of a High Magnesium Containing Cultivar in Controlling Hypomagnesaemia in Grazing Animals. *Grass and Forage Science*, **46**, 375-380. https://doi.org/10.1111/j.1365-2494.1991.tb02397.x

[42] Reid, R.L., Baker, B.S. and Vona, L.C. (1984) Effects of Magnesium Sulfate Supplementation and Fertilization on Quality and Mineral Utilization of Timothy Hays by Sheep. *Journal Animal Science*, **59**, 1403-1410. https://doi.org/10.2527/jas1984.5961403x

[43] Jones, D.I.H. and Thomas, T.A. (1987) Minerals in Pastures and Supplements. In: Snaydon, R.W., Ed., *Managed Grasslands*, Analytical Studies (E*cosystems of the World* 17*B*), Elsevier, Amsterdam, 145-153.

[44] Blaxter, K.L., Kay, R.N.B., Sharman, G.A.M., Cunningham, J.M.M., Eadie, J. and Hamilton, W.J. (1988) Farming the Red Deer. Rowett Research Institute and the Hill Farming Research Organization, Department of Agriculture and Fisheries Scotland HMSO, Edinburgh.

[45] Gough, L.P., Shacklette, H.T. and Case, A.A. (1979) Element Concentrations Toxic to Plants, Animals, and Man. U.S. Geological Survey Bulletin, 1466. U.S. Govt. Print Office, Washington DC.

[46] Brelurut, A., Pingard, A. and Thériez, M. (1990) Le cerf et son élevage. INRA, Paris.

[47] Mayland, H.F. (1983) Assessing Nutrient Cycling in the Soil/Plant/Animal System of Semi-Arid Pasture Lands. *Proceedings Advisory Group Meeting*, Vienna, 10-14 November 1980, 109-117.

[48] Grunes, D.L., Stout, P.R. and Brownell, J.R. (1970) Grass Tetany of Ruminants. *Advances in Agronomy*, **22**, 331-374. https://doi.org/10.1016/S0065-2113(08)60272-2

[49] Jones, G.B. and Tracy, B.F. (2013) Evaluating Seasonal Variation in Mineral Concentration of Cool Season Pasture Herbage. *Grass and Forage Science*, **70**, 94-101. https://doi.org/10.1111/gfs.12094

[50] Sabreen, S., Saiga, S., Tsuiki, M. and Mayland, H.F. (2002) Plant Tissues Suitable for Individual Selection of Mg in Tall Fescue. *Grassland Science*, **48**, 316-317.

[51] Sabreen, S., Saiga, S., Saitoh, H., Tsuiki, M. and Mayland, H.F. (2003) Performance of High-Magnesium Cultivars of Three Cool Season Grasses Grown in Nutrient Solution Culture. *Journal of Plant Nutrition*, **26**, 589-605. https://doi.org/10.1081/PLN-120017667

[52] Sabreen, S. and Saiga, S. (2004) Potassium Level Suitable for Screening High Magnesium Containing Grass Seedlings under Solution Culture. *Journal of Plant Nutrition*, **27**, 1015-1027. https://doi.org/10.1081/PLN-120037533

[53] Penrose, B., Lovatt, J.A., Palmer, S., Thomson, R. and Broadley, M.R. (2020) Revisiting Variation in Leaf Magnesium Concentrations in Forage Grasses for Improved Animal Health. *Plant and Soil*, **457**, 43-55. https://doi.org/10.1007/s11104-020-04716-9

[54] Rahman, M.H. and Saiga, S. (2005) Endophytic Fungi (*Neotyphodium coenophialum*) Affect the Growth and Mineral Uptake, Transport and efficiency Ratios in Tall Fescue (*Festuca arundinacea*). *Plant and Soil*, **272**, 163-171. https://doi.org/10.1007/s11104-004-4682-6

[55] Crawford, R.J., Massie, M.D., Sleper, D.A. and Mayland, H.F. (1998) Use of an Experimental High-Magnesium Tall Fescue to Reduce Grass Tetany in Cattle. *Journal of Production Agriculture*, **11**, 491-496. https://doi.org/10.2134/jpa1998.0491

[56] Sabreen, S., Saiga, S., Islam, R. and Rahman, H. (2022) Phosphorus Fertilization Af-

fects Cation Balance in Cool-Season Grasses Associated with Grass Tetany. *Journal of Plant Nutrition*. https://doi.org/10.1080/01904167.2022.2057330

[57] Berdahl, J.D., Mayland, H.F., Asay, K.H. and Jefferson, P.G. (1999) Variation in Agronomic and Morphological Traits among Russian Wildrye Accessions. *Crop Science*, **39**, 1890-1895. https://doi.org/10.2135/cropsci1999.3961890x

[58] Jefferson, P.G., Mayland, H.F., Asay, K.H. and Berdahl, J.D. (2001) Variation in Mineral Concentration and Grass Tetany Potential among Russian Wildrye Accessions. *Crop Science*, **41**, 543-548. https://doi.org/10.2135/cropsci2001.412543x