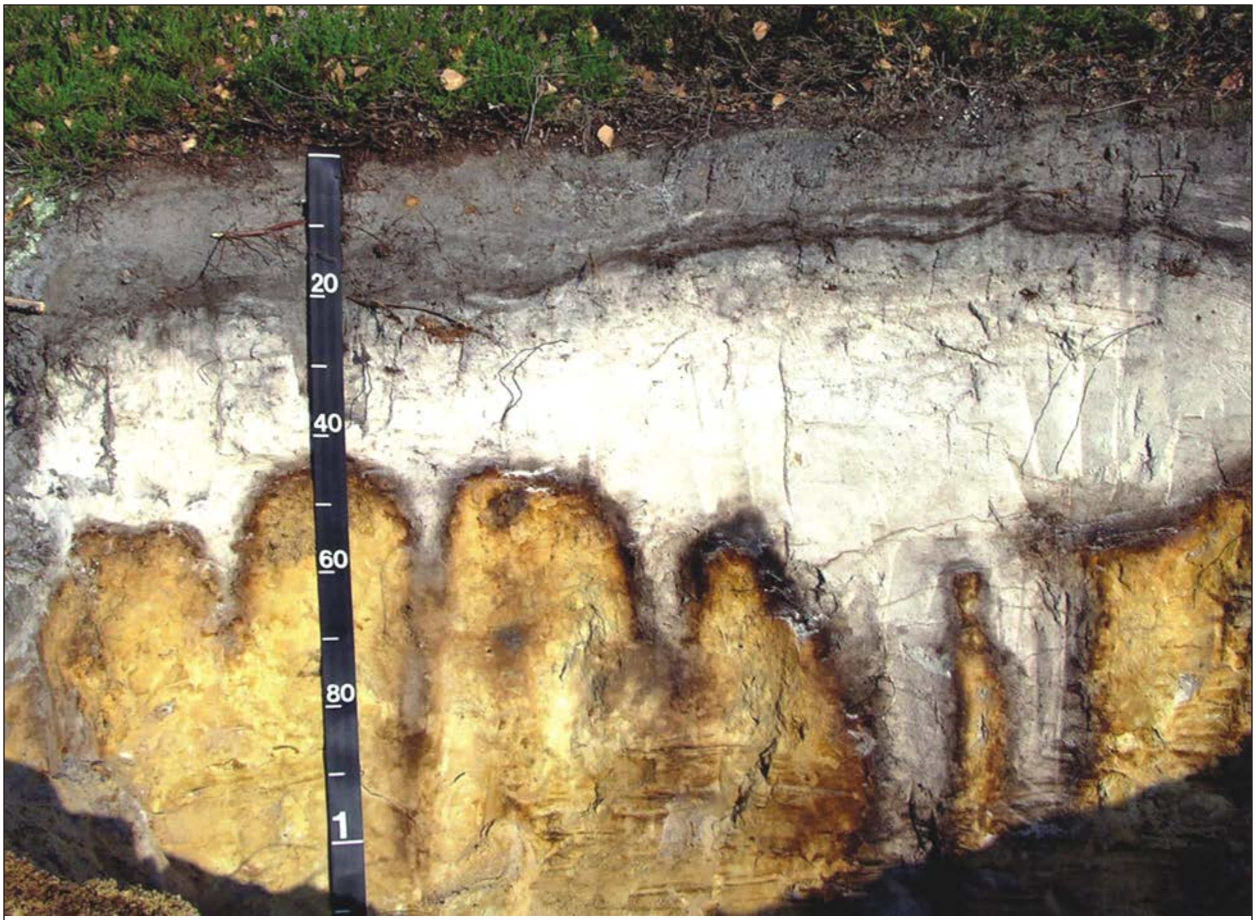

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Genetic Variability in Tetany Potential of Orchradgrass as Influenced by Application of Dairy Manure and Chemical Fertilizer

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Abstract: The objectives of this study were to compare K, Ca and Mg utilization and grass tetany potential among the high and low magnesium containing strains and commercial cultivars of orchardgrass in response to application of dairy manure and chemical fertilizer. The study was conducted from 2002 through 2003 in northern Honshu Island, Japan on sandy loam Andisol. Highest plant dry matter production was recorded with the application of chemical fertilizer. Soil properties varied with application of manure. Potassium concentration in shoot tissue increased from 2002 to 2003 in all the treatments irrespective of strains and cultivars. However, calcium concentration in shoot tissue decreased from 2002 to 2003 in all the treatments irrespective of strains and cultivars. High magnesium containing strains almost showed low potassium and high magnesium and calcium concentrations in all the treatments. Concentrations of K, Ca and Mg in shoot tissue were highest as a result of dairy manure application and lowest by chemical fertilizer application. The grass tetany potentials were higher in 2003 than 2002. The grass tetany potential was lowest in all the cultivars and strains during fertilization with chemical fertilizer. High magnesium containing strains were less grass tetany prone than the others irrespective of treatments. The correlations between equivalent ratio and K were significantly positive; the correlations involving equivalent ratio, Ca and Mg were negative, however, regardless of treatments and years.

Key words: Andisol, genetic variability, tetany potential, orchradgrass, dairy manure and chemical fertilizer

INTRODUCTION

Incidence of grass tetany, a metabolic disorder of ruminants has been increased in all over the world. The reason for the greater incidence of grass tetany is the increased use of N and K fertilizers or dairy manure on forage production. Increased levels of K and N depress Mg uptake by forage that affects the availability of Mg to ruminants. Tetany occurred more frequently when the temperature is low but the meteorological conditions are favorable for good plant growth ('t Hart, 1960). Grass tetany prevention and treatment is based on the use of Mg supplements. However, Mg supplementation is often not commenced until symptoms are severe and it may be too late to prevent mortality (Harris *et al*., 1983). There is increasing evidence that ion uptake by plants is under genetic control (Clark, 1983; Saric, 1983). Genetic variability for K, Mg, Ca or K/(Ca+Mg) has been reported in several temperate grass species including perennial ryegrass (Cooper, 1973; Crush, 1983; Easton *et al*., 1997), tall fescue (Nguyen and Sleper, 1981), prairie grass (Rumball *et al*., 1972), orchardgrass (Stratton and Sleper, 1979) reed canarygrass (Hovin *et al*., 1978), Italian ryegrass (Hides and Thomas, 1981), crested wheatgrass (Mayland and Asay, 1989; Vogel *et al*., 1989; Asay *et al*., 1996), Russian wild ryegrass (Asay and Mayland, 1990) and festulium (Buckner *et al*., 1981). In this

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context, plant-breeding programs have been started to increase available Mg on Italian ryegrass (Moseley and Griffiths, 1984; Moseley and Baker, 1991), tall fescue (Sleper *et al.*, 1989; Mayland and Sleper, 1993), perennial ryegrass (Binnie *et al.*, 1996) and orchardgrass (Saiga *et al.*, 2002).

Orchardgrass (*Dactylis glomerata* L.) is the major perennial grass species sown in temperate region of Japan caused grass tetany in cattle with high mortality rate. The K/(Ca+Mg) ratio on the mole equivalent basis has been used to ascertain the possibility of grass tetany (hypomagnesaemia) in high producing dairy cattle and in the diets, values>2.2 is appear to be critical for the onset of grass tetany (Kemp and t'Hart, 1957; Butler, 1963; Grunes *et al.*, 1970; Ritter *et al.*, 1984). Management practices and climatic conditions may affect on grass tetany potential (K/(Ca+Mg)) as large environmental effects on herbage mineral concentration were detected (Rabinson *et al.*, 1989; Smith *et al.*, 1999) and nutrient uptake is governed by the interplay of the nutrient supplying power of a soil with the nutrient demand exerted by the plant root (Barber, 1984). Application of fresh farmyard manure to maize (*Zea mays* L) and Italian ryegrass (*Lolium multiflorum* Lam.) grown in Andisols resulted in equivalent ratios above 2.2 (Ito and Miyazawa, 1984). Long-term application of fertilizer and fertilizer/barnyard manure to forage maize grown in sandy loam soil resulted in K/(Ca+Mg) ratio as high as 4.0 (Kagata *et al.*, 1999). However, the effects of dairy manure applications to orchardgrass cultivars grown in Andisols in terms of grass tetany potential are not elucidated. So, the main objectives of this study were to compare K, Ca and Mg utilization and grass tetany potential of orchardgrass strains and cultivars fertilized with dairy manure and chemical fertilizer in Andisol of northeast Honshu Island of Japan.

MATERIALS AND METHODS

Experimental Field and Climatic Conditions

The field was established in 1999 in sandy loam Andisol with pH 6.03, exchangeable K_2O, CaO and MgO were 48, 443 and 53 mg 100 g^{-1} soil, respectively and a soil organic matter content of 183.97 g kg^{-1} in the top 15 cm. The experiment was started in March and continued until October for 2002 and 2003 at the Uwadai field of Iwate University, Japan. The climatic conditions during the experiment were shown in Table 1.

Plant Materials

Orchardgrass (*Dactylis glomerata* L.) was used in this experiment, because it is the most productive forage and widely cultivated in northeast Japan. Orchardgrass including two commercial

Table 1: Meteorological data of the field area during the experiments

	Total precipitation (mm)		Temperature (°C)				Wind speed (m s^{-1})				Mean relative humidity (%)		Total daylight (h)	
			Minimum		Maximum		Minimum		Maximum					
Month	2002	2003	2002	2003	2002	2003	2002	2003	2002	2003	2002	2003	2002	2003
January	103	100	10.0	4.5	-10.8	-2.2	14.2	12.8	2.6	2.8	77	76	119	98
February	28	33	11.1	9.8	-13.7	-0.9	9.0	9.8	2.4	2.6	71	71	144	147
March	134	51	15.6	16.0	-8.0	1.7	10.2	13.4	3.1	3.5	69	67	139	189
April	47	105	25.9	25.0	-0.3	9.3	11.6	11.7	3.6	3.5	62	68	182	173
May	99	38	26.8	29.0	1.4	15.1	9.8	9.7	3.1	3.6	71	66	188	211
June	69	85	30.3	31.0	8.7	18.8	9.0	10.8	3.3	3.2	74	74	172	149
July	382	188	32.8	28.2	14.2	18.8	10.2	11.1	2.8	3.1	83	81	102	75
August	346	251	32.3	31.2	12.3	21.8	11.3	9.8	2.8	2.9	85	82	77	78
September	105	121	31.3	28.3	5.7	18.5	7.9	12.1	2.5	2.8	80	79	137	104
October	153	63	25.6	22.2	2.1	11.7	13.7	9.3	2.7	2.5	81	74	124	168
November	125	104	13.1	21.3	-6.3	7.1	10.6	13.4	2.6	2.6	80	75	94	129
December	24	67	12.5	11.0	-10.4	2.9	16.8	12.4	2.7	3.2	71	71	105	82

cultivars viz. Okamidori and Akimidori and four experimental strains viz. HighMgE, HighMgM, LowEq and LowMg which differed in morphological and chemical features as well as in genetic variability (Saiga *et al*., 2002) were used in this experiment. Okamidori, HighMgE, HighMgM and LowEq were bred for high Mg containing plants.

Treatments

Dairy manure (DM), chemical fertilizer (CF) and dairy manure and chemical fertilizer (DMCF) were broadcasted in each year on 26 March at the rate of 1 t/0.1 ha, 10 kg/0.1 ha and 1/2 t+5 kg/0.1 ha, respectively. The N, P, K composition of dairy manure was 1, 2and 1.4%, respectively. The experimental site had no previous history of dairy manure application and we conducted experiment during 2002 and 2003. At neither plot were weeds a significant problem during the experiment. Hand weeding was used, if and where necessary to control spot outbreaks.

Plant Harvest and Soil Analysis

Forage was harvested (6 cm cutting height) four times at 27th April, 26th June, 25th August and 26th October maintaining 60 days growing period during 2002 and 2003. Soil samples (0-15 cm) were collected at the day of last harvest in each year using core sampler and analyzed for soil bulk density (Blake and Hartge, 1986), hardness, porosity (Danielson and Sutherland, 1986), moisture content (Gardner, 1986) at field condition. The pH of soils was determined in a 1:2.5 soil to water suspension (Jackson, 1973) by a digital pH meter. Soil organic matter estimated according to Ball (1965).

Mineral Analysis

After harvesting, the sample was dried at 80°C for 24 h in a forced-air oven. After drying, the samples were ground to pass a 0.5 mm screen with a cyclone mill and 0.5 to 1.0 g sample was pressed (with a coherent disc of 2.5 cm) by applying 15.0 tons pressure to make a pellet with a uniform surface. After that the concentrations of magnesium (Mg), potassium (K) and calcium (Ca) of the both sites of the pellet were measured with a live time of 100s by energy reflectance x-ray fluorescence analyzer (ERF; JEOL Co., JSX-3220, Element Analyzer) as described by Hutton and Norrish (1977) and Norrish and Hutton (1977). Each plant sample was replicated three times. The K/(Ca+Mg) was computed on a mole equivalent basis. In this study the average values for four harvests in dry matter production and nutrients content were evaluated and the values for changes in soil properties were evaluated over two years.

Experimental Design

The experiment was set up as a split plot design with two dairy manure rates and one fertilizer treatment (main plots) and grass genotypes (subplots) as random effects replicated three times with an individual plot area of 8.6 m^2.

Statistics

Analysis of variance was used to detect significant differences between the main experimental factors, namely (1) effect of treatment and (2) effect growing season. The relative proportions of sum of square (%SS) due to treatments, growing seasons and their interactions to the total SS were calculated to clarify the effects of treatments, growing seasons and interactions of these variables, respectively. Duncan's Multiple Range Test (Duncan, 1955) was conducted to compare results with the variables at a 5% level of significance. The Least Significant Differences (LSD) test was also used to determine whether differences between growing seasons were statistically significant ($p<0.05$). Pearson correlation coefficients among nutrients were performed. All statistical analyses were conducted using lease square ANOVA procedure of SAS at the Computer Center of Iwate University, Japan.

RESULTS AND DISCUSSION

Selected Soil Properties and Dry Matter Yield

Table 2 showed that soil properties and dry matter yield was affected by treatment, growing season and interaction of those variables. Treatment had a predominant effect on soil bulk density, hardness and porosity explained 57.9, 81.2 and 64.0%, of the total variations, respectively, as compared with growing season and treatment x growing season. Soil pH was affected by treatment, growing season and treatment x growing season accounting for 16.2, 58.0 and 25.8% of the total variance in the data, respectively. Growing season had a predominant effect on soil organic matter explained 79.7% of the total variations, as compared with treatment and treatment x growing season. Soil bulk density decreased and as a consequence porosity increased with the application of manure although differences in porosity were not statistically significant (Table 3) among the treatments. In our experiment soil bulk density significantly decreased with application of dairy manure but Eghball (2002) reported that soil bulk density was found unaffected by application of manure or compost. Significantly higher soil hardness was found in CF treatment than the other treatments. Highest soil pH and organic matter was recorded in DM treatment. It is resulted that the organic matter content of composted municipal solid waste exceeds 25% and its addition to most soils increases the organic matter content (Schrader, 1967), total pore space (Pagliai, 1981) and pH of acid soils (Sanderson, 1980; Scanlon *et al.*, 1973) and decreases soil bulk density (Mays and Giordano, 1989; Guidi and Petruzzelli, 1989). Magdoff and Amadon (1980) stated that the effect of inorganic-N fertilizer on lowering soil pH could be counteracting by application of manure. They observed that application of manure increased organic matter as well. In our experiment soil pH decreased causes by application of chemical fertilizer. Dairy manure raised the soil pH when no chemical fertilizer was added to soil. A large amount of bases related to N was probably responsible for the influence of dairy manure on soil pH. Eghball (2002) observed that soil surface pH significantly increased with N-based manure or compost application but decreased with NH_4-N fertilizer application as compared with check.

Dry matter yield was significantly affected by treatment and growing season, accounting for 47.6 and 38.6% of the total variance in the data, respectively (Table 2). Dry matter yield was also significantly affected by treatment×growing season interaction indicated that there were treatment-

Table 2: Overall effects of treatment and growing season on different variables using ANOVA

	Treatments			Growing season			Treatment×growing season		
Parameters	SS (%)	F	Pr>F	SS (%)	F	Pr>F	SS (%)	F	Pr>F
Dry matter yield	47.58	67.70	0.0001	38.62	109.90	0.0001	13.79	19.62	0.0001
Bulk density	57.86	37.92	0.0001	35.76	46.88	0.0001	6.38	4.18	0.0250
Soil hardness	81.17	55.56	0.0001	14.99	20.52	0.0001	3.84	2.63	0.0889
Porosity	64.02	47.78	0.0001	29.46	43.97	0.0001	6.53	4.87	0.0147
pH	16.20	526.94	0.0001	57.96	3771.44	0.0001	25.84	840.66	0.0001
OM	15.29	138.22	0.0001	79.68	1440.43	0.0001	5.03	45.44	0.0001

Table 3: Dry matter yield and some selected soil properties as a function of dairy manure and chemical fertilization application over two years

	Treatments		
Properties	DM	CF	DMCF
Dry matter yield ($g\ m^{-2}$)	63.64b[1]	141.16a	111.46a
Bulk density ($Mg\ m^{-3}$)	0.67b	0.75a	0.73a
Soil hardness ($kg\ cm^{-2}$)	2.58c	4.39a	3.84b
Porosity (%)	72.41a	69.24a	69.96a
pH	6.89a	5.70b	5.74b
OM (%)	18.62a	17.55a	17.84a

[1]Means in within treatments followed by same letter(s) are not statistically different ($p<0.05$)

specific differences in dry matter yield to growing season. Dry matter yield increased with growing season for DM treatments and decreased for other treatments (Table 3). Present results were in good agreement with those of Kagata *et al*. (1999).

Potassium, Calcium and Magnesium Concentration

The analysis of variance for K, Ca and Mg of this study is listed in Table 4 through 6. The fit of the model (R^2) was very good for all the variables (K>Ca>Mg). The effects of treatment, strain/cultivar and interaction of these variables were statistically significant with the exception of treatment effect on Mg in 2002 growing season. The coefficients of variation (CV) over two growing season were 3.43, 4.01 and 7.24% for K, Ca and Mg, respectively. Herbage K concentration increased after first season in all the treatments (Table 4). Low Mg containing plants showed greatly higher K than high Mg containing plants. Application of compost showed highest concentration of K in all the plants than the other treatments. Calcium concentration of plants decreased with season progressed in all the strains and commercial cultivars (Table 5). Low Mg containing plants showed greatly lower Ca than high Mg containing plants. Application of dairy manure showed highest concentration of Ca in all the plants than the other treatments. Calcium content in forage may increase by applying nitrogenous fertilizer. Slowing of pasture growth by any means or seasonal decline soil temperature increased forage Ca level (Underwood and Suttle, 1999). Magnesium concentration of plants decreased with advancement of growing season in all the strains and commercial cultivars for CF and DMCF

Table 4: Effect of dairy manure and chemical fertilizer on K concentration of orchardgrass

		Potassium (%DM)			
Treatments	Strain/Cultivar	2002	2003	Mean	LSD[2]
DM	HighMgE	2.20bc[1]	2.76b	2.48b	0.299
	HighMgM	1.95d	2.56c	2.26c	0.321
	LowEq.	2.19bc	2.71bc	2.45b	0.278
	LowMg	2.66a	3.16a	2.91a	0.267
	Okamidori	2.31b	2.73bc	2.52b	0.221
	Akimidori	2.18c	2.86b	2.52b	0.359
	Av.	2.25	2.80	2.52	0.291
CF	HighMgE	1.73e	2.08bc	1.90c	0.184
	HighMgM	1.89d	2.05c	1.97c	0.084
	LowEq.	1.98cd	2.08bc	2.03bc	0.052
	LowMg	2.19a	2.23ab	2.21a	0.022
	Okamidori	2.17ab	2.21abc	2.19a	0.023
	Akimidori	2.04bc	2.29a	2.17ab	0.137
	Av.	2.00	2.16	2.08	0.084
DMCF	HighMgE	2.07b	2.25b	2.16b	0.095
	HighMgM	2.12b	2.20b	2.16b	0.040
	LowEq.	2.32a	2.52a	2.42a	0.111
	LowMg	2.38a	2.60a	2.49a	0.121
	Okamidori	2.42a	2.54a	2.48a	0.065
	Akimidori	2.11b	2.66a	2.39a	0.294
	Av.	2.24	2.46	2.35	0.121
ANOVA					
Source of variation					
Treatment		125.30***[3]	289.00***	287.37***	
Strain/Cultivar		87.33***	31.28***	63.30***	
Treatment×strain/cultivar		12.59***	5.33***	8.93***	
R^2		0.9004	0.8975	0.9160	
Mean square ($\times 10^{-2}$)		27.37	59.07	36.41	
CV (%)		3.50	4.56	3.43	

[1]Means in a column within a treatment and year followed by same letter (s) are not statistically different. [2]Least significant different (LSD) at $p<0.05$ between growing seasons (columns). [3]*, **, *** represent statistical significant at 0.01, 0.001 and 0.0001 probability levels, respectively

Table 5: Effect of dairy manure and chemical fertilizer on Ca concentration of orchardgrass

		Calcium (%DM)			
Treatments	Strain/Cultivar	2002	2003	Mean	LSD[2]
DM	HighMgE	0.508b[1]	0.402ab	0.455b	0.056
	HighMgM	0.475c	0.422a	0.448b	0.028
	LowEq.	0.540a	0.428a	0.484a	0.060
	LowMg	0.418d	0.401ab	0.410d	0.009
	Okamidori	0.474c	0.369b	0.421cd	0.056
	Akimidori	0.465c	0.424a	0.444bc	0.022
	Av.	0.480	0.408	0.444	0.039
CF	HighMgE	0.437bc	0.382a	0.409ab	0.029
	HighMgM	0.465b	0.358a	0.412ab	0.057
	LowEq.	0.516a	0.357a	0.436a	0.085
	LowMg	0.418c	0.353a	0.386b	0.035
	Okamidori	0.432bc	0.349a	0.390b	0.044
	Akimidori	0.452bc	0.375a	0.413ab	0.041
	Av.	0.453	0.362	0.408	0.049
DMCF	HighMgE	0.516a	0.366b	0.441ab	0.080
	HighMgM	0.487ab	0.366b	0.426abc	0.065
	LowEq.	0.477b	0.435a	0.456a	0.023
	LowMg	0.461b	0.333b	0.397c	0.068
	Okamidori	0.485ab	0.357b	0.421bc	0.068
	Akimidori	0.478b	0.366b	0.422bc	0.060
	Av.	0.484	0.370	0.427	0.061
ANOVA					
Source of variation					
Treatment		24.20***[3]	37.66***	39.94***	
Strain/Cultivar		30.20***	10.18***	27.55***	
Treatment×strain/cultivar		8.46***	4.73***	0.96	
R^2		0.7594	0.6584	0.7163	
Mean square ($^{-}10^{-2}$)		0.67	0.57	0.39	
CV (%)		4.30	6.23	4.01	

[1]Means in a column within a treatment and year followed by same letter(s) are not statistically different.[2]Least significant different (LSD) at $p<0.05$ between growing seasons (columns).[3]*, **, *** represent statistical significant at 0.01, 0.001 and 0.0001 probability levels, respectively

treatments and increased for DM treatment (Table 6). Significant increases were observed by Saiga *et al.* (1997) as the season progresses for Mg concentration in orchardgrass grown in Andisol. Magnesium concentration was also found higher in all the high Mg containing plants. Application of dairy manure showed highest concentration of Mg in all the plants. On the other hand, application of chemical fertilizer showed lowest concentration of Mg in all the plants. Nitrogen fertilization increased the concentration of higher fatty acids in plants and this may depress the availability of Mg (Grunes, 1973).

The Equivalent Ratio

The tetany potential increased from season 2002 to 2003 and it was below the critical level for all the treatments irrespective of strains and commercial cultivars (Table 7). The lowest values were recorded in case of chemical fertilizer application. Animal consuming low Mg containing orchardgrass would be more tetany prone than those of high Mg containing orchardgrass as a result of dairy manure application.

Relationship Among the Nutrients

Correlation coefficients were calculated for each year for individual treatment as well as for years combined (Table 8). Potassium was significantly and positively correlated with K/(Ca+Mg) irrespective of year and treatment. On the other hand, Ca and Mg was significantly and negatively correlated with K/(Ca+Mg) irrespective of year and treatment. In years combined Ca was more closely

Table 6: Effect of dairy manure and chemical fertilizer on Mg concentration of orchardgrass

		Magnesium (%DM)			
Treatments	Strain/Cultivar	2002	2003	Mean	LSD[2]
DM	HighMgE	0.397a[1]	0.365a	0.381a	0.017
	HighMgM	0.348ab	0.328a	0.338ab	0.011
	LowEq.	0.346ab	0.340a	0.343ab	0.003
	LowMg	0.304bc	0.365a	0.334ab	0.033
	Okamidori	0.291c	0.270b	0.280c	0.012
	Akimidori	0.274c	0.347a	0.311bc	0.039
	Av.	0.327	0.336	0.331	0.005
CF	HighMgE	0.313a	0.316a	0.315a	0.002
	HighMgM	0.338a	0.295ab	0.316a	0.021
	LowEq.	0.335a	0.221c	0.279a	0.063
	LowMg	0.298a	0.255bc	0.277a	0.023
	Okamidori	0.337a	0.282ab	0.310a	0.030
	Akimidori	0.319a	0.306ab	0.312a	0.007
	Av.	0.324	0.279	0.301	0.024
DMCF	HighMgE	0.333ab	0.307a	0.320a	0.014
	HighMgM	0.336a	0.307a	0.322a	0.015
	LowEq.	0.317ab	0.305a	0.311a	0.006
	LowMg	0.285b	0.244b	0.264b	0.022
	Okamidori	0.312ab	0.281a	0.296ab	0.017
	Akimidori	0.304ab	0.290a	0.297ab	0.008
	Av.	0.315	0.289	0.302	0.014
ANOVA					
Source of variation					
Treatment		0.81	44.21***[3]	15.30***	
Strain/Cultivar		7.25***	8.70***	8.29***	
Treatment×strain/cultivar		2.45***	9.20***	4.84***	
R^2		0.4093	0.7133	0.5723	
Mean square ($\times 10^{-2}$)		0.35	0.87	0.36	
CV (%)		9.67	8.54	7.24	

[1]Means in a column within a treatment and year followed by same letter(s) are not statistically different.[2]Least significant different (LSD) at $p<0.05$ between growing seasons (columns).[3]*, **, *** represent statistical significant at 0.01, 0.001 and 0.0001 probability levels, respectively

Table 7: Grass tetany potential in orchardgrass as a result of dairy manure and chemial fertilizer applications

		K/(Ca+Mg)			
Treatments	Strain/Cultivar	2002	2003	Mean	LSD[2]
DM	HighMgE	1.01c[1]	1.45cd	1.23c	0.235
	HighMgM	0.95c	1.35d	1.15d	0.215
	LowEq.	1.01c	1.40cd	1.20cd	0.209
	LowMg	1.48a	1.61b	1.55a	0.068
	Okamidori	1.24b	1.71a	1.48a	0.252
	Akimidori	1.22b	1.46c	1.34b	0.129
	Av.	1.15	1.50	1.33	0.185
CF	HighMgE	0.93d	1.17c	1.05d	0.132
	HighMgM	0.95cd	1.24bc	1.10cd	0.155
	LowEq.	0.95cd	1.49a	1.22b	0.289
	LowMg	1.23a	1.49a	1.36a	0.136
	Okamidori	1.12ab	1.38ab	1.25ab	0.140
	Akimidori	1.07bc	1.34abc	1.20bc	0.145
	Av.	1.04	1.35	1.20	0.166
DMCF	HighMgE	0.99d	1.32c	1.15c	0.172
	HighMgM	1.04d	1.29c	1.16c	0.130
	LowEq.	1.18bc	1.38c	1.28bc	0.104
	LowMg	1.31a	1.81a	1.56a	0.267
	Okamidori	1.24ab	1.58b	1.41b	0.185
	Akimidori	1.10cd	1.61b	1.36b	0.273
	Av.	1.14	1.50	1.32	0.189

Table 7: Continued

		K/(Ca+Mg)		
Treatments	Strain/Cultivar	2002	2003	Mean
ANOVA				
Source of variation				
Treatment		29.58***3	26.83***	48.43***
Strain/Cultivar		76.01***	33.88***	84.30***
Treatment×strain/cultivar		6.23***	6.97***	3.59**
R^2		0.8479	0.7649	0.8603
Mean square ($\times 10^{-2}$)		13.94	16.55	13.08
CV (%)		6.18	6.77	4.95

[1]Means in a column within a treatment and year followed by same letter(s) are not statistically different.[2]Least significant different (LSD) at p<0.05 between growing seasons (columns).[3]*, **, *** represent statistical significant at 0.01, 0.001 and 0.0001 probability levels, respectively

Table 8: Linear correlation coefficients of K, Ca, Mg and K/(Ca+Mg) for orchardgrass

			Correlation of coefficients		
Treatments		Variable	2002	2003	Years combined
DM	K	Ca	-0.474**1	0.146NS	-0.440**
	K	Mg	-0.280NS	0.537**	0.023NS
	K	K/(Ca+Mg)	0.845**	0.373*	0.778**
	Ca	Mg	-0.449**	0.632**	0.456**
	Ca	K/(Ca+Mg)	-0.716**	-0.715**	-0.762**
	Mg	K/(Ca+Mg)	-0.712**	-0.550**	-0.575**
CF	K	Ca	-0.118NS	0.215NS	-0.116NS
	K	Mg	0.079NS	0.014NS	-0.018NS
	K	K/(Ca+Mg)	0.512**	0.308NS	0.704**
	Ca	Mg	0.495**	0.421**	0.236NS
	Ca	K/(Ca+Mg)	-0.603**	-0.534**	-0.419**
	Mg	K/(Ca+Mg)	-0.574**	-0.856**	-0.670**
DMCF	K	Ca	-0.193NS	0.089NS	-0.106NS
	K	Mg	-0.229NS	-0.505**	-0.532**
	K	K/(Ca+Mg)	0.808**	0.786**	0.834**
	Ca	Mg	0.312NS	0.476**	0.498**
	Ca	K/(Ca+Mg)	-0.530**	-0.443**	-0.525**
	Mg	K/(Ca+Mg)	-0.718**	-0.889**	-0.867**1 *

[1]*Significant at the 0.05 level; **Significant at the 0.01 level; NSNot significant

correlated with K/(Ca+Mg) for DM treatment and Mg was closely correlated with K/(Ca+Mg) for CF and DMCF treatments. In years combined K was negatively correlated with Ca and Ca was positively correlated with Mg. Mayland and Asay (1989) worked with the genetic variability of K, Ca and Mg in crested wheatgrass and found that Ca significantly and positively correlated with Mg and K and negatively with K/(Ca+Mg). They observed that Mg was significantly and positively correlated with Ca and K and negatively with K/(Ca+Mg). In that study it was also noted that K/(Ca+Mg) was significantly and negatively correlared with Ca and Mg and positively with K.

CONCLUSIONS

Genetic variability in tetany potential was evaluated for orchardgrass when grown in Andisol fertilized with dairy manure and chemical fertilizer. During the two growing seasons no symptom was observed in equivalent ratios of above 2.2. The high Mg containing cultivars showed low equivalent ratio in all the treatments. Although no negative effects were observed in relation to tetany potential due to application of dairy manure but from the view point of soil oxygenation, it should be emphasized that large additions material such as slurry, sewage and/or dairy manure must be applied with care, because while they increase oxygen demand they may simultaneously impede gas exchange by clogging the soil pore. In the above mentioned case, when the soil oxygenation is insufficient other gases such as CH_4, H_2S, N_2O, C_2H_2 and H_2 may occur in the soil air. The presence of any of these gases, despite low concentrations, is an important indicator of the status of the soil quality.

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