

AN ABSTRACT OF THE THESIS OF

Sabam Oloan Manurung for the degree of Master of Science
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Title: GROWTH AND FORAGE QUALITY OF TALL FESCUE (FESTUCA
ARUNDINACEA SCHREB.) AND PERENNIAL RYEGRASS (LOLIUM PERENNE L.)
AS AFFECTED BY MEFLUIDIDE

Abstract approved:

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Dr. D.O. Chilcote

Field study with 5 tall fescue genotypes treated at the vegetative growth stage, with 0, 0.28, 0.56, and 0.84 kg/ha of mefluidide was initiated in July, 1980. The regrowth of these plants were again treated at the booting growth stage in April, 1981, with the same rates of mefluidide as in the first experiment. In addition, three rates of mefluidide (0, 0.28, and 0.56 kg/ha) were applied at the floral initiation stage of perennial ryegrass cv. Linn in March, 1981.

Four varieties of tall fescue were transplanted from the field into 6-inch pots in February, 1981, and grown under 50/60°F and 70/80°F night/day temperature regimes in growth chambers. Mefluidide at the rate of 0, 0.28, and 0.56 kg/ha were applied at the pre-floral or at floral initiation growth stage. The same varieties were again transplanted from the field into 6-inch pots in April, 1981, and grown under the same temperature regimes and treated at pre-booting or at booting growth stage with the same rate of mefluidide as those

in the first growth chamber experiment.

Mefluidide interacted with tall fescue genotypes and also resulted in a reduction in dry matter yield, ADF (Acid Detergent Fiber) and increased CP (Crude Protein) content in both tall fescue trials in the field. The WSC (Water Soluble Carbohydrate) concentration was reduced by mefluidide applied at the vegetative growth stage in the first experiment, but it was increased at the booting growth stage application of mefluidide on regrowth of these plants. Late applications of mefluidide did maintain forage quality of tall fescue with some elimination of dry matter yield reduction caused by earlier application.

In perennial ryegrass cv. Linn, mefluidide reduced stem dry matter production, but did not affect leaf dry matter yield. This was accompanied by high leaf/stem ratios and retardation of plant height. Fertile tiller production was inhibited, accompanied by reduction in ADF, increased stem CP and increased WSC of leaf and stem tissue.

Under the 70/80°F night/day temperature regime, all varieties produced a higher dry matter yield than plants under a 50/60°F night/day temperature regime. However, WSC content of leaf and stem were higher in the low temperature regime. There were indications that mefluidide reduced ADF and WSC content of leaf and stem tissues, but increased leaf and stem CP under both temperature regimes in both experiments. This was accompanied by high leaf/stem ratios of treated plants.

GROWTH AND FORAGE QUALITY OF TALL FESCUE
(FESTUCA ARUNDINACEA SCHREB.) AND PERENNIAL RYEGRASS
(LOLIUM PERENNE L.) AS AFFECTED BY MEFLUIDIDE

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Sabam Oloan Manurung

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Redacted for Privacy

Professor of Crop Science in charge of major

Redacted for Privacy

Head of Department of Crop Science

Redacted for Privacy

Dean of Graduate School

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Typed by Gloria M. Foster for Sabam Oloan Manurung

TO THE MEMORY OF

My Father, late Cornelius Manurung and my Mother
late Juliana Simorangkir. They lived, worked, suffered
and died in Christianity.

(. . . I am the resurrection and the life: he that
believeth in me, though he were dead, yet shall he live
. . . St. John 11:25, New Testament)

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INTRODUCTION

Poorer forage quality of tall fescue as well as perennial ryegrass is noted soon after the initiation of reproductive growth, where the *in vitro* dry matter digestibility starts to decline rapidly (10, 15, 16, 18). As the season progresses, the apparent digestibility of grasses for ruminants decreases with the advancing stage of growth. Rapid decline in digestibility following the emergence of the reproductive structure is mainly associated with increased fiber, cell wall material, and lignin which are negatively correlated with dry matter digestibility (1, 2, 10, 17, 18). In addition, both the amount and digestibility of protein in forage grasses decline as growth approaches maturity (7, 14, 15, 16, 21).

In contrast, carbohydrate digestibility depends on its chemical nature. The group of readily soluble and readily hydrolysable carbohydrates are regarded as being potentially fully digestible. Cellulose and hemicellulose that are free of lignin protection are also regarded as potentially fully digestible. Lignin, silica, cuticle, and higher molecular carbohydrates which are fully protected by encrustations of lignin are completely indigestible (13, 22).

The percentage of total nonstructural carbohydrates (TNC) is influenced greatly by the ratio of stem to leaf tissue. The TNC concentration appears to increase with advancing maturity. Higher concentrations usually are found at mature rather than young growth stages, at cool rather than warm temperatures, and at low instead of high soil

nitrogen levels (19).

Crude protein content of the leaf is higher than that of stem parts, so the decline in average crude protein content as grasses mature can, in part, be attributed to the decreasing proportion of leaf, as well as to the fall in protein content of the leaf itself (11). An increase in protein is often offset by a depression in soluble carbohydrate (13). Low temperature and low soil fertility, mainly nitrogen, result in low protein content (11).

If the onset of reproductive growth is responsible for a rapid decline in digestibility, better forage quality could be obtained if flowering could be prevented or delayed. Frequent cutting is a classical way to keep high forage quality in pasture species. This mechanical method is laborious and time consuming.

Endogenous growth substances normally control plant growth, but modifications of growth can be produced by applications of exogenous growth substances. Chemical inhibition of reproductive growth in grasses is possible through growth manipulation by mefluidide (N-[2,4-dimethyl-5-[[trifluoromethyl]-sulfonyl]amino]phenyl]acetamide), a plant growth regulator previously known as MBR 12325 or Embark. This compound is reported to inhibit seed head production in tall fescue (3), accompanied by increased protein content, suppression of cellulose percentage, and increased digestibility based on acid detergent fiber (ADF) and neutral detergent fiber (NDF) determinations (4, 5). However, dry matter production is reduced for a 2-week period after mefluidide application on April 1 and April 15 (4).

In Oregon, forage production comprises about 250,000 acres of irrigated tall fescue, perennial ryegrass, orchardgrass, and subclover mixtures, and about 500,000 acres of non-irrigated tall fescue, perennial ryegrass, and subclover mixtures (12). Income from cattle and calves provides nearly 20% of the annual total agricultural income in the Pacific Northwest and constitutes approximately one-half of the livestock products income of the region (9).

Because of reduction of dry matter yield by mefluidide applied early, later applications under Oregon conditions were considered. The goal, of course, was to maintain higher forage quality without losing dry matter yield and attempting to balance quantity and quality factors. In addition, if improvement in quality of forage in the absence of reproductive development can be obtained, benefits of mefluidide could be further extended. Genetic interaction with mefluidide can be expected and, therefore, some information on this aspect, as well as environmental effects, are needed.

Objectives of studies reported here were (1) to examine grass forage quality in terms of crude protein, acid detergent fiber, and water soluble carbohydrate content in selected genotypes of tall fescue and perennial ryegrass as affected by rate of mefluidide application at various growth stages; (2) to evaluate the proper rate of mefluidide application, as well as effect of growth stage; (3) to evaluate the possibility of forage quality enhancement on tall fescue in the absence of reproductive growth; and (4) to compare forage quality of various tall fescue cultivars treated under different temperature regimes and at various growth stages under controlled environment.

MATERIALS AND METHODS

Field experiments were conducted in two successive years (1980 and 1981) on five selected genotypes of tall fescue and one field experiment was conducted in 1981 on perennial ryegrass. Two controlled environment experiments were conducted on four varieties of tall fescue in growth chambers under two different night/day temperature regimes.

A. Field Trials

Experiment 1. Five well established (planted in 1978) genotypes of tall fescue (Festuca arundinacea Schreb.) were selected in June, 1980, at the Oregon State University Field Laboratory Hyslop Farm. These genotypes, selected from the breeding program of Dr. R.V. Frakes, were entries 315-434, 229-692, 111, 292-851, with Ky 31 as a standard variety. First growth was harvested in mid-May, 1980, which removed the reproductive structure. On July 31, 1980, when plants were in a vegetative growth stage, mefluidide was applied to individual plants at the rate of 0 (control), 0.28, 0.56, and 0.84 kg/ha. Two other treatments included were DMSO at 10% (of spray volume) and DMSO at 10% plus 0.28 kg/ha mefluidide, to see if DMSO could enhance mefluidide activity. Treatments were in a factorial arrangement with a completely randomized block design and five observations per treatment. On September 24, 1980, all individual clones or plants were harvested at 7.5 cm aboveground level and weighed. Grab samples were obtained for dry matter yield determination and chemical analysis.

Ground samples were kept in air-tight glass jars after passing through a 1-mm screen, for subsequent analysis of acid detergent fiber (ADF), crude protein (CP), and water soluble carbohydrates (WSC).

Experiment 2. The same plants as in experiment 1 were again selected for treatments in 1981. Regrowth of these plants was removed on April 6, 1981. The plants were allowed to regrow until the booting stage of growth before they were treated on April 28, 1981. Chemical treatments, rates and experimental design were the same as those for experiment 1. After untreated plants on each genotype showed an emerged inflorescence, all plants were harvested at 7.5 cm aboveground level on May 28, 1981 through June 3, 1981. Fresh weight was obtained and grab samples were taken for dry matter yield determination and for chemical analysis. Those samples were ground to pass through a 1-mm screen and ground samples were kept in air-tight glass jars for subsequent chemical analysis for ADF, CP, and WSC.

Experiment 3. Three rates of mefluidide, 0 (control), 0.28, and 0.56 kg/ha, were applied on March 27, 1981, to an established perennial ryegrass cv Linn. Treatments were arranged in a randomized block design with three replications. Harvest was on May 8, 1981, when untreated plants were in the heading stage. Forage was obtained from each plot by cutting at ground level. Dry matter yield, ADF, CP, and WSC in leaf and stem tissue was determined. Leaf area index (LAI), plant height, and number of tillers producing heads were also determined. The leaf and stem parts were separated, oven-dried, and ground to pass a 1-mm screen. Each ground plant part was kept in air-tight

glass jars for subsequent chemical analysis.

B. Growth Chamber Experiments

Experiment 1. Four varieties of tall fescue, Fortune, Alta, S-170, and Fawn, were removed from the Schmidt Experimental Farm on February 4, 1981, transplanted into 6-inch pots, and kept in the greenhouse for two weeks. They were placed in growth chambers on February 18, 1981, under 50/60 and 70/80°F night/day temperature regimes with a 12-hour photoperiod. Three rates of mefluidide were used (0 or control, 0.28, and 0.56 kg/ha). On February 20, 1981, at the pre-floral initiation growth stage, half of each group of plants were treated. The remaining plants were allowed to reach the floral initiation growth stage before treatments were applied. This group was treated on March 5, 1981. In each chamber, treatments were a factorial arrangement in a completely randomized block design, with three observations per treatment. Forage was harvested at ground level in each pot on April 20, 1981. Leaf and stem parts were separated, oven-dried and ground to pass a 1-mm screen and each plant part was kept in separate air-tight glass jars before chemical analysis for ADF, CP, and WSC.

Experiment 2. The same varieties from the same field as those in experiment 1 were removed and transplanted into 6-inch pots on April 9, 1981. Plants were selected and separated based on their growth stage. A group of plants at the pre-booting growth stage and another group at the booting growth stage were selected for treatment

in each temperature regime (50/60 and 70/80°F night/day under a 12-hour photoperiod). Plants in each chamber were treated with mefluidide at the same rate as those in experiment 1, on April 27, 1981. All treatments in each growth chamber were a factorial arrangement in a completely randomized block design with three observations per treatment. Forage was harvested on June 5, 1981, at ground level for each pot. Leaf and stem samples were separated, oven-dried, and ground to pass a 1-mm screen, and then retained in air-tight glass jars for subsequent analysis of ADF, CP, and WSC.

Chemical and Data Analysis

Acid detergent fiber (ADF), crude protein (CP), and water soluble carbohydrate (WSC) were determined as a percentage of dry matter obtained from ground forage material. ADF was determined by the methods of Goering and Van Soest (6). CP was determined by multiplying the N percentage obtained in micro-Kjeldahl analysis by 6.25 (8). WSC was extracted and determined using the anthrone method (8), and concentration was measured by using a Hitachi Perkin Elmer UV-VIS Spectrophotometer Model 139.

Statistical analysis included F-test at the 5% and 1% probability level, to determine treatment effects. Comparison among treatment means was tested by using the least significant difference (LSD) at 5% and 1% probability level (20).

EXPERIMENTAL RESULTS

A. Field Trials

Experiment 1

Dry Matter Yield

The F-test on the analysis of variance for dry matter yield as affected by mefluidide application at vegetative stage of growth in 1980, is presented in Appendix Table 1. The effect of genotypes, mefluidide, as well as the genotypes x mefluidide interaction is highly significant (P, 0.01). The presence of a first order interaction (genotype x mefluidide) indicates that neither genotype nor mefluidide act independently and genotypes respond differently to mefluidide in terms of dry matter yield.

Table 1 shows the dry matter yield for individual clones. Entry 111 showed the most tolerance to mefluidide and yielded the highest dry matter at all levels of mefluidide treatment. Mefluidide at 0.28 kg/ha with or without DMSO, reduced dry matter production significantly (P, 0.01) for all genotypes except Ky 31. Ky 31 required a 0.56 kg/ha rate of mefluidide to reduce dry matter a significant amount (P, 0.01). By increasing the rate of mefluidide from 0.28 kg/ha to 0.56 kg/ha, dry matter yield was reduced significantly (P, 0.01) for all genotypes except Entry 111. A further yield reduction was observed in all entries except Entry 111. A further yield reduction was observed in all entries except Entry 111 or 292-851, when mefluidide rate was increased from 0.56 to 0.84 kg/ha.

Table 1. Dry matter yield of individual clones of 5 genotypes of tall fescue as affected by mefluidide applied at the vegetative growth stage in 1980.

Mefluidide	Genotype				
	315-434	229-692	Ky 31	Ent.111	292-851
— kg/ha —	— grams —				
0	169.440	156.780	122.280	197.440	190.300
0+DMSO 10%	163.840	153.960	125.980	199.260	195.100
0.28+DMSO 10%	145.220	120.200	120.200	164.640	153.540
0.28	139.260	116.980	114.280	160.640	149.700
0.56	101.140	76.600	71.360	150.280	75.960
0.84	75.900	54.200	42.200	138.780	77.400
LSD .05	16.811				
LSD .01	22.219				

Chemical Analysis

Mean squares from the analysis of variance for ADF, CP, and WSC content are shown in Appendix Table 2. F-tests at the 1% probability level reveal that genotype, mefluidide, as well as the genotype x mefluidide interaction, were highly significant. This means that neither genotypes nor mefluidide are independent of each other. Genotypes interacted with mefluidide in determining ADF, CP, and WSC content.

ADF. Acid detergent fiber (ADF) content for each genotype at each rate of mefluidide is presented in Table 2. DMSO AT 10% did not affect ADF content in any genotype. This compound also did not enhance nor inhibit the activity of mefluidide. Note that the rate of 0.84 kg/ha mefluidide was required to reduce ADF content significantly (P, 0.01) in Entry 111 and 292-851. In contrast, 0.28 kg/ha was sufficient to reduce ADF in entries 315-434 and 229-692, while 0.56 kg/ha was required for reducing ADF in Ky 31.

CP (Crude Protein). The content of CP in various genotypes is shown in Table 3. Except for Ky 31, CP increased significantly (P, 0.01) from that of control plant at 0.28 kg/ha of mefluidide. Ky 31 required 0.56 kg/ha rate of mefluidide to show an increase in CP (P, 0.01). Addition of DMSO at 10% did not give consistent results and benefit from its addition is questionable.

WSC (Water Soluble Carbohydrate). Table 4 shows the percentage of WSC for each genotype at each level of mefluidide rate. In

Table 2. Acid detergent fiber (ADF) content as a percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the vegetative growth stage in 1980.

Mefluidide	Genotype				
	315-434	229-692	Ky. 31	Ent.111	292-85T
— kg/ha —	%				
0	30.51	28.81	30.04	29.94	28.42
0+DMSO 10%	31.11	27.59	29.73	30.37	27.39
0.28+DMSO 10%	23.31	26.92	29.22	28.87	27.28
0.28	24.23	25.99	29.16	29.16	26.37
0.56	22.76	24.31	27.27	28.33	26.74
0.84	22.41	22.85	23.25	26.58	24.40
LSD .05	1.76				
LSD .01	2.36				

Table 3. Crude protein (CP) content as a percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the vegetative growth stage in 1980.

Mefluidide	Genotype				
	315-434	229-692	Ky 31	Ent.111	292-851
— kg/ha —	%				
0	8.18	7.58	10.23	8.74	9.27
0+DMSO 10%	7.42	8.84	10.07	9.52	10.01
0.28+DMSO 10%	9.45	9.68	10.47	10.48	10.33
0.28	9.73	10.06	10.06	10.58	10.67
0.56	8.90	10.32	11.62	10.91	11.18
0.84	9.19	11.63	11.08	12.29	11.71
LSD .05		0.42			
LSD .01		0.56			

Table 4. Water soluble carbohydrate (WSC) content as a percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the vegetative growth stage in 1980.

Mefluidide	Genotype				
	315-434	229-692	Ky 31	Ent.111	292-851
— kg/ha —	%				
0	15.33	14.09	10.85	14.82	12.72
0+DMSO 10%	15.33	16.66	11.01	12.69	9.05
0.28+DMSO 10%	12.76	10.44	8.01	12.91	9.66
0.28	13.14	12.32	7.72	12.69	11.23
0.56	13.64	7.30	10.67	12.60	8.30
0.84	12.59	10.45	10.53	12.01	9.17
LSD .05		0.91			
LSD .01		1.23			

general, mefluidide reduced WSC content significantly ($P, 0.01$) at the rate of 0.28 kg/ha or higher. The reduction of WSC content caused by mefluidide was not consistent for genotype 229-692, Ky 31, and genotype 292-851. Again, DMSO did not perform consistently.

Experiment 2

Dry Matter Yield

The mean square from the analysis of variance for dry matter yield per individual clone on 5 selected genotypes as affected by mefluidide applied at the booting growth stage of regrowth in 1981 is presented in Appendix Table 3. The effect of genotype, mefluidide, and the genotype x mefluidide interaction are again highly significant ($P, 0.01$). Therefore, genotype and mefluidide are not independent of each other in determining dry matter yield.

Table 5 presents dry matter yield per individual clone of each genotype which was treated at the booting growth stage of regrowth. Mefluidide at the rate of 0.28 kg/ha reduced dry matter production significantly ($P, 0.01$) from that of untreated plants. This effect was reflected by all genotypes at rates higher than 0.28 kg/ha. However, Entry 111 produced the highest dry matter yield at all levels of mefluidide application but also showed a steady reduction in dry matter yield beyond 0.28 kg/ha of mefluidide. The loss of dry matter caused by 0.28 kg/ha of mefluidide relative to its respective untreated control plant were 13%, 42%, 21%, 44%, and 42% for genotype 315-434, 229-692, Ky 31, Entry 111, and 292-851, respectively.

Table 5. Dry matter yield of individual clones of 5 genotypes of tall fescue as affected by mefluidide applied at the booting growth stage in 1981.

Mefluidide	Genotype				
	315-434	229-692	Ky 31	Ent.111	292-851
kg ai/ha	grams				
0	351.620	489.520	418.820	596.400	450.980
0+DMSO 10%	348.000	485.880	401.580	603.420	438.580
0.28+DMSO 10%	320.160	258.360	339.340	357,560	254.020
0.28	306.780	253.200	331.620	332.500	260.240
0.56	267.840	209.900	297.900	329.600	223.060
0.84	204.280	200.800	294.200	324.620	201.360
LSD _{.05}	20.260				
LSD _{.01}	24.772				

DMSO at 10% did not affect dry matter yield significantly and did not inhibit nor enhance the activity of mefluidide. Mefluidide at higher rates showed variable effects on dry matter yield.

Chemical Analysis

The F-test for the effect of genotype and mefluidide on the content of ADF (acid detergent fiber), CP (crude protein), and WSC (water soluble carbohydrate) are shown in Appendix Table 4. The effects of genotype, mefluidide and genotype x mefluidide interaction are highly significant (P, 0.01), indicating that genotype and mefluidide are not independent of each other.

ADF. Acid detergent fiber (ADF) content for each genotype at each level of mefluidide is shown in Table 6. Mefluidide at the rate of 0.28 kg/ha reduced ADF content significantly at the 1% probability level on all tested genotypes, except for 292-851. This genotype required 0.56 kg/ha mefluidide to reduce its ADF content a significant amount (P, 0.01). By increasing mefluidide rates from 0.28 kg/ha to 0.56 kg/ha, ADF content was reduced significantly (P, 0.01) in Ky 31, Entry 111, and 292-851. Higher rates (0.84 kg/ha) had no further significant effect on ADF of Ky 31 or 292-851.

DMSO did not inhibit or enhance mefluidide activity at 0.28 kg/ha except on Entry 111. However, DMSO did not show a consistent effect.

CP (Crude Protein). Crude protein content for each genotype at

Table 6. Acid detergent fiber (ADF) content as a percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the booting growth stage of regrowth in 1981.

Mefluidide	Genotype				
	315-434	229-692	Ky 31	Ent.111	292-851
kg ai/ha	%				
0	37.50	39.90	36.70	41.30	38.20
0+DMSO 10%	38.00	39.60	36.30	39.95	39.50
0.28+DMSO 10%	38.00	28.20	31.50	36.45	38.65
0.28	37.90	27.60	31.70	33.00	37.90
0.56	36.10	27.55	28.50	30.40	29.70
0.84	33.00	25.80	28.50	27.50	29.30
LSD .05	1.38				
LSD .01	1.85				

each level of mefluidide is presented in Table 7. Genotype 229-692, Entry 111, and 292-851 exhibited higher CP content by the application of 0.28 kg/ha of mefluidide. For genotype 315-434, a rate of 0.84 kg/ha of mefluidide was required to increase protein content over that of the control (P, 0.01). Ky 31 did not show a significant response at any level of mefluidide application. DMSO did not enhance nor inhibit mefluidide activity except for Entry 111. However, results were not consistent.

WSC (Water Soluble Carbohydrate). WSC as a percentage of dry matter on all genotypes at each level of mefluidide rate is shown in Table 8. In general, WSC contents were increased on all genotypes by mefluidide. Rates of 0.28 kg/ha to 0.56 kg/ha appeared to give optimum WSC enhancement with 0.84 kg/ha superoptimum except for Ky 31 which showed a consistent increase in WSC with increasing rates of mefluidide.

DMSO did not exhibit a consistent effect. In some genotypes, the addition of DMSO resulted in an increase in WSC, while in others a reduction occurred. Increasing rates of mefluidide (0.56 kg/ha and 0.84 kg/ha) were not effective in increasing WSC content in genotypes 229-692 and 292-851.

Experiment 3

(Perennial Ryegrass)

Dry Matter Yield, Leaf/Stem Ratio and LAI

Mean squares from the analysis of variance for dry matter yield

Table 7. Crude protein (CP) content as a percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the booting growth stage of regrowth in 1981.

Mefluidide	Genotype				
	315-434	229-692	Ky 31	Ent.111	292-851
kg ai/ha	%				
0	8.45	6.38	9.14	5.46	6.28
0+DMSO 10%	8.01	7.00	9.12	6.37	6.79
0.28+DMSO 10%	8.15	9.65	9.44	6.96	7.14
0.28	8.76	9.15	8.98	7.98	7.43
0.56	8.84	9.51	9.13	7.52	7.93
0.84	9.57	10.49	8.57	8.39	8.31
LSD .05	0.85				
LSD .01	1.15				

Table 8. Water soluble carbohydrate (WSC) content as a percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the booting growth stage of regrowth in 1981.

Mefluidide	Genotype				
	315-434	229-692	Ky 31	Ent.111	292-851
kg ai/ha	%				
0	4.60	9.11	8.52	5.91	5.74
0+DMSO 10%	8.88	7.89	7.56	4.53	6.65
0.28+DMSO 10%	4.03	14.31	10.63	8.35	6.83
0.28	9.77	10.31	15.71	7.44	18.79
0.56	9.36	13.61	18.87	14.38	16.17
0.84	11.74	12.38	19.32	10.95	14.28
LSD .05	0.82				
LSD .01	1.11				

are shown in Appendix Table 5. Leaf/stem ratio and leaf area index (LAI) mean squares are shown in Appendix Table 6. The effect of mefluidide on total dry matter yield and LAI of perennial ryegrass cv. Linn (Table 9) was not significant. But leaf/stem ratios were affected significantly at the 1% probability level. Mefluidide affected stem dry matter yield significantly (P, 0.05) but did not affect leaf dry matter.

The 0.28 kg/ha or 0.56 kg/ha of mefluidide rates were not significantly different in their effect. Reduction in stem dry matter was followed by increased leaf/stem ratio as expected.

Plant Height and Fertile Tillers

The effect of mefluidide on plant height and percentage of tillers producing inflorescences (heads) were highly significant at the 1% probability level (Appendix Table 7). Table 10 presents the average plant height and percentage of tillers producing head. Plant height and fertile tillers were significantly reduced (P, 0.01) by 0.28 and 0.56 kg/ha rates of mefluidide. By increasing mefluidide rates from 0.28 to 0.56 kg/ha, plant height was reduced non-significantly, but the percentage of fertile tillers was decreased significantly (P, 0.01). The onset of inflorescences emergence was also inhibited by mefluidide at rates of 0.28 kg/ha or 0.56 kg/ha applied at the floral initiation growth stage.

Table 9. Leaf and stem dry matter yield, leaf/stem ratio, and LAI of perennial ryegrass cv. Linn as affected by mefluidide applied at the floral initiation growth stage.

Mefluidide kg ai/ha	Dry matter yield/m ²		Leaf/Stem	LAI
	Leaf	Stem		
0	338.0	998.0	0.33	0.56
0.28	274.3	493.7	0.56	0.46
0.56	363.7	523.0	0.72	0.59
LSD _{.05}	ns	376.2	0.13	ns
LSD _{.01}		609.0	0.22	

Table 10. Plant height and percentage of fertile tillers as affected by mefluidide applied at the floral initiation growth stage in perennial ryegrass cv. Linn.

Mefluidide (kg/ha)	Plant height (cm)	Percentage of tiller producing head (\sqrt{x})
0	48.8	4.48 (20.10) ¹
0.28	25.6	2.71 (7.77)
0.56	22.2	1.44 (2.13)
LSD _{.05}	7.5	0.75
LSD _{.01}	12.5	1.25

¹Actual percentage is in parenthesis.

Chemical Analysis

F-test results showing the effect of mefluidide on the content of ADF, CP, and WSC are presented in Appendix Table 8.

The average ADF, CP, and WSC content on leaf and stem tissues are shown in Table 11. ADF and CP of leaf tissue were not significantly affected by mefluidide. However, stem ADF was significantly (P, 0.05) decreased from that of control by mefluidide treatment at the rate of 0.28 kg/ha, with a highly significant effect (P, 0.01) by 0.56 kg/ha of mefluidide. There was no significant difference in stem ADF resulting from 0.28 kg/ha or 0.56 kg/ha of mefluidide. Stem CP was increased significantly (P, 0.05) by 0.56 kg/ha of mefluidide, but not at the 0.28 kg/ha rate. The rate of 0.56 kg/ha of mefluidide also increased WSC content of leaf and stem tissues. At the high rate, leaf WSC was increased 116%, while stem WSC was increased 29% relative to the respective control.

B. Growth Chamber Experiments

Experiment 1

Dry Matter Yield per Tiller

The effect of different rates of mefluidide applied at pre-floral initiation or at floral initiation in 4 varieties of tall fescue under 50/60°F and 70-80°F night/day temperature regime are shown in Appendix Table 9. The F-test (1% probability level) shows a significant effect of variet x mefluidide x growth stage interaction on dry

Table 11. ADF, CP, and WSC content as a percentage of dry matter of perennial ryegrass cv. Linn as affected by mefluidide applied at the floral initiation growth stage.

Mefluidide (kg/ha)	Plant parts	ADF (%)	CP (%)	WSC (%)
0	leaf	26.64	11.52	7.96
	stem	30.22	5.26	21.38
0.28	leaf	24.51	12.86	10.80
	stem	25.56	6.40	26.46
0.56	leaf	23.69	12.95	17.18
	stem	22.72	7.58	27.50
LSD _{.05}	leaf	ns	ns	2.10
LSD _{.01}				3.49
LSD _{.05}	stem	4.15	1.49	1.74
LSD _{.01}		6.88	2.47	2.89

matter yield per tiller. This infers that variety, mefluidide and growth stage of plant at which mefluidide was applied are not independent of each other.

The average dry matter per tiller under both temperature regime is shown in Table 12. In all cases, except the untreated variety S-170, dry matter yield per tiller under 70/80°F temperature was higher than that under a 50/60°F night/day temperature regime. Mefluidide at the rate of 0.28 or 0.56 kg/ha, applied pre-floral initiation or at the floral initiation growth stage resulted in significantly (P, 0.01) lower dry matter yield. By delaying the application of 0.28 kg/ha of mefluidide from pre-floral initiation to the floral initiation growth stage, dry matter production per tiller would increase significantly (P, 0.01) over that from early applications on Alta and S-170 under 70/80°F night/day temperature regime. This effect was also noted for 0.56 kg/ha of mefluidide application on Fortune and Alta under 50/60°F and on Fawn under 70/80°F night/day temperature regime. If mefluidide was applied at the rate of 0.28 kg/ha at pre-floral initiation growth stage, dry matter yield reduction would be 59% for Fortune, 79% for Alta, 72% for S-170, and 71% for Fawn under the 50/60°F night/day temperature regime and would be 49%, 55%, 33%, and 28%, respectively for Fortune, Alta, S-170, and Fawn under a 70/80°F night/day temperature regime.

Under 70/80°F night/day temperature regime, dry matter yield was not only higher than that under 50/60°F night/day temperature regime, but also resulted in less loss of dry matter yield caused by mefluidide treatment. Consistent effects of mefluidide were not evident

Table 12. Dry matter yield per tiller of 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide (kg/ha)	Growth stage	Dry matter yield/tiller	
			50/60°F	70/80°F
			mg	
Fortune	0	Control	144.3	173.0
	0.28	P1 ¹	59.3	89.0
		1 ²	61.3	103.3
	0.56	P1	36.0	81.0
		1	72.0	87.7
	Alta	0	Control	221.0
0.28		P1	46.7	109.0
		1	60.0	156.0
0.56		P1	32.3	92.7
		1	50.0	80.0
S-170		0	Control	199.0
	0.28	P1	56.3	114.0
		1	93.3	124.0
	0.56	P1	40.3	118.0
		1	43.7	79.3
	Fawn	0	Control	175.0
0.28		P1	50.3	128.0
		1	63.7	136.0
0.56		P1	50.0	87.0
		1	52.0	102.0
LSD .05			4.6	9.4
LSD .01			6.1	12.6

¹P1 = Pre-floral initiation growth stage.

² 1 = Floral initiation growth stage.

under both temperature regime for each variety. Dry matter yields relative to each control for varieties are shown in Table 13.

Leaf/Stem Ratio

Mean squares from the analysis of variance for leaf/stem ratio are presented in Appendix Table 10. F-test values show significant (P, 0.01) effects of variety x mefluidide x growth stage interaction. This infers that none of these variables is independent.

The average leaf/stem ratios for all 4 varieties as affected by mefluidide applied pre-floral initiation or at floral initiation growth stage under 50/60°F and under 70/80°F are shown in Table 14. Comparing the leaf/stem ratio to untreated plants of each variety, mefluidide at the rate of 0.28 kg/ha or 0.56 kg/ha applied at pre-floral initiation growth stage, under 50/60°F or under 70/80°F night/day temperature regime, would increase the leaf/stem ratio significantly at the 1% probability level, except for selection S-170 under 70/80°F night/day temperature regime. Again, these effects were not consistent when mefluidide was applied at floral initiation growth stage across temperature regime. Application of mefluidide at the rate of 0.56 kg/ha at the pre-floral initiation growth stage resulted in a significantly (P, 0.01) higher leaf/stem ratio with the exception of Alta under the 70/80°F night/day temperature regime. This effect would change if mefluidide was applied at the floral initiation growth stage.

A greater difference in leaf/stem ratios resulted from early mefluidide application than that of late mefluidide application.

Table 13. Dry matter yield per tiller relative to that of untreated plants of 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regime in growth chambers.

Variety	Mefluidide (kg/ha)	Growth stage	Dry matter yield/tiller	
			50/60°F	70-80°F
			% —————	
Fortune	0	Control	100	100
	0.28	P1 ¹	41	51
		1 ²	42	60
	0.56	P1	25	47
		1	50	51
Alta	0	Control	100	100
	0.28	P1	21	45
		1	27	64
	0.56	P1	15	38
		1	23	33
S-170	0	Control	100	100
	0.28	P1	28	67
		1	47	73
	0.56	P1	20	69
		1	22	47
Fawn	0	Control	100	100
	0.28	P1	29	72
		1	36	77
	0.56	P1	29	49
		1	30	58

¹Pre-floral initiation growth stage.

²Floral initiation growth stage.

Table 14. Leaf/stem ratio of 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F temperature regimes in growth chambers.

Variety	Mefluidide (kg/ha)	Growth stage	Leaf/stem ratio	
			50/60°F	70-80°F
Fortune	0	Control	1.9	3.0
	0.28	P1 ¹	3.2	4.6
		1 ²	2.1	4.4
	0.56	P1	3.3	5.4
		1	2.9	5.0
	Alta	0	Control	3.0
0.28		P1	3.5	5.2
		1	2.8	4.9
0.56		P1	4.3	4.8
		1	4.1	4.6
S-170		0	Control	3.4
	0.28	P1	4.6	5.0
		1	4.0	4.1
	0.56	P1	5.5	5.9
		1	4.7	4.9
	Fawn	0	Control	2.1
0.28		P1	3.8	4.7
		1	2.3	3.7
0.56		P1	5.0	5.3
		1	3.4	4.0
LSD .05				0.3
LSD .01			0.5	0.9

¹Pre-floral initiation growth stage.

²Floral initiation growth stage.

The difference became less under higher temperatures. In general, later applications of mefluidide resulted in lower leaf/stem ratios. In all cases, leaf/stem ratios were higher under 70-80°F night/day temperature regimes than under a 50-60°F night/day temperature regime.

ADF (Acid Detergent Fiber)

Appendix Table 11 shows mean squares from the analysis of variance for ADF. The F-test shows that the variety x mefluidide x growth stage interaction is highly significant (P, 0.01). Therefore, these variables were interdependent.

The average ADF content, which was expressed as percentage of dry matter on leaf and stem tissues, is shown in Table 15. In all cases, ADF content of the stem is higher than that of leaves under either a 50-60°F or a 70-80°F night/day temperature regime. Consistent reduction in ADF contents by mefluidide was not observed on leaf or stem tissues under either temperature regime. Stem and leaf ADF of Fortune was not reduced by mefluidide under a 50/60°F regime, but was significantly reduced (P, 0.01) under the 70-80°F night/day temperature regime for the 0.56 kg/ha rate at both growth stages. A significant reduction (P, 0.01) in stem ADF for Alta resulted from the application of 0.28 kg/ha or 0.56 kg/ha of mefluidide applied pre-floral initiation under 50-60°F night/day temperature regime. This effect was also observed for leaf ADF of Alta under the 70/80°F night/day temperature regime.

Mefluidide at 0.56 kg/ha was required to reduce leaf and stem ADF of S-170 under 50/60°F night/day temperature regime. This rate

Table 15. ADF content of leaf and stem as a percentage of its respective dry matter on 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide kg/ha	Growth stage	ADF				
			50/60°F		70/80°F		
			leaf	stem	leaf	stem	
			— % —	— % —	— % —	— % —	
Fortune	0	Control	26.4	28.3	27.7	29.9	
	0.28	P1 ¹	25.1	29.6	26.5	27.1	
		1 ²	26.9	28.6	25.3	29.5	
	0.56	P1	26.1	30.3	24.6	20.1	
		1	26.0	29.8	24.8	20.9	
	Alta	0	Control	26.0	28.7	27.2	31.6
0.28		P1	26.1	26.2	23.5	30.2	
		1	26.5	28.2	27.7	28.4	
0.56		P1	25.5	26.3	23.4	27.4	
S-170		0	Control	24.2	28.0	26.6	28.7
		0.28	P1	23.8	29.1	25.3	27.3
	1		26.9	29.1	19.3	27.3	
	0.56	P1	21.0	22.2	16.2	22.3	
		1	22.7	24.4	23.0	27.8	
	Fawn	0	Control	25.4	27.9	26.3	30.3
0.28		P1	23.8	26.2	24.7	30.3	
		1	26.4	31.6	23.5	29.2	
0.56		P1	20.2	30.3	21.1	27.4	
		1	24.8	29.1	20.1	28.2	
LSD _{.05}				1.3	1.0	1.8	1.5
LSD _{.01}			1.8	1.4	2.5	2.0	

¹Pre-floral initiation growth stage.

²Floral initiation growth stage.

also resulted in the reduction of stem ADF under a 70/80°F night/day temperature regime. A significant reduction (P, 0.01) of leaf and stem ADF of Fawn resulted from the application of 0.56 kg/ha of mefluidide at the pre-floral initiation or floral initiation growth stage under a 70/80°F night/day temperature regime. Under a 50/60°F night/day temperature regime, a significant reduction (P, 0.01) of leaf ADF only occurred with the 0.56 kg/ha rate.

Crude Protein (CP)

The analysis of variance for leaf CP under 50/60°F reveals that variety x mefluidide and mefluidide x growth stage interactions are highly significant (P, 0.01). The variety x mefluidide x growth stage interaction is also significant for stem CP under the 50/60°F night/day temperature regime and for leaf and stem CP under a 70/80°F night/day temperature regime (Appendix Table 12).

Table 16 shows the CP content of leaf and stem tissues grown under 50/60°F and 70/80°F night/day temperature regimes. A consistent effect was not shown for CP content, but there was the indication that CP content increased significantly (P, 0.01) from that of control by mefluidide applied at the rate of 0.28 kg/ha or 0.56 kg/ha in plant stems grown under 50/60°F and in leaves and stems grown under 70/80°F night/day temperature regimes.

The effect of variety and mefluidide on leaf CP under the 50/60°F night/day temperature regime is shown in Table 17. For Fortune, the rate of 0.56 kg/ha of mefluidide decreased CP

Table 16. Crude protein (CP) content of leaf and stem as a percentage of its respective dry matter on 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide kg/ha	Growth stage	CP			
			50/60°F		70/80°F	
			Leaf	stem	Leaf	stem
			—— % ——	—— % ——		
Fortune	0	Control	9.4	5.2	12.5	5.8
	0.28	P1 ¹	16.5	12.9	16.2	9.2
		1 ²	18.5	13.1	14.5	9.3
	0.56	P1	12.3	12.5	18.4	10.4
		1	12.3	11.3	17.0	10.1
	Alta	0	Control	11.4	8.2	15.5
0.28		P1	18.4	11.6	19.1	10.8
		1	21.6	17.2	17.3	9.6
0.56		P1	21.6	16.7	16.6	9.6
		1	20.4	18.4	18.0	12.8
S-170		0	Control	12.0	7.2	16.0
	0.28	P1	15.6	14.5	15.7	7.8
		1	18.5	12.4	18.7	9.7
	0.56	P1	20.5	18.9	18.9	11.1
		1	20.8	17.7	16.6	10.5
	Fawn	0	Control	13.3	7.9	14.5
0.28		P1	16.8	9.6	17.7	8.1
		1	16.8	13.4	17.5	8.0
0.56		P1	21.1	17.2	16.5	7.4
		1	19.1	14.0	22.3	13.1
LSD .05				ns	1.4	2.0
LSD .01				1.8	2.7	1.5

¹Pre-floral initiation growth stage.

²Floral initiation growth stage.

Table 17. Mean of CP content of leaf as a percentage of its dry matter on 4 varieties of tall fescue as affected by three levels of mefluidide under a 50/60°F night/day temperature regime in growth chambers.

Variety	Mefluidide kg/ha	Crude protein (CP)
		———— % ————
Fortune	0	9.3
	0.28	17.5
	0.56	12.3
Alta	0	11.4
	0.28	20.0
	0.56	21.0
S-170	0	12.0
	0.28	16.8
	0.56	20.7
Fawn	0	13.3
	0.28	17.6
	0.56	20.1
LSD .05		1.2
LSD .01		1.6

Table 18. Mean of CP content of leaf as a percentage of its dry matter as affected by three levels of mefluidide applied at pre-floral initiation or at floral initiation under a 50/60°F night/day temperature regime in growth chambers.

Mefluidide kg/ha	Growth stage	Crude protein
		———— % ————
0	Control	11.5
0.28	P1	16.8
	1	19.2
0.56	P1	18.9
	1	18.2
LSD .05		0.8
LSD .01		1.1

significantly (P, 0.01) compared to 0.28 kg/ha of mefluidide. However, for S-170 and Fawn this effect was not true.

Table 18 shows mefluidide and growth stage responses for CP content under 50/60°F night/day temperature regimes. The rate of 0.28 kg/ha of mefluidide applied at floral initiation increased CP content significantly (P, 0.01) over that of the early application (at pre-floral initiation). Mefluidide at the rate of 0.28 kg/ha or 0.56 kg/ha, applied at pre-floral initiation or at floral initiation, would increase leaf CP content significantly (P, 0.01) over the untreated control.

WSC (Water Soluble Carbohydrate)

The F-test for the analysis of variance for leaf and stem WSC concentration is shown in Appendix Table 13, for both temperature regimes of 50/60°F and 70/80°F night/day. Under both of these temperature regimes, the variety x mefluidide x growth stage interaction is highly significant (P, 0.01), indicating that variety, mefluidide and growth stage are not independent of each other.

The average leaf and stem WSC for each variety and growth stage as affected by three levels of mefluidide under the two temperature regimes is presented in Table 19. A consistent effect was not observed. However there was an indication that mefluidide at the rate of 0.28 kg/ha or 0.56 kg/ha applied at pre-floral initiation or at floral initiation growth stage reduced leaf and stem WSC content from that of each control for both temperature regimes. WSC concentration

Table 19. WSC content of leaf and stem as a percentage of its respective dry matter on 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation (P1) or at floral initiation (1) under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide	Growth stage	WSC			
			50/60°F		70/80°F	
			leaf	stem	leaf	stem
	kg/ha		%			
Fortune	0	Control	19.7	23.8	7.7	21.2
	0.28	P1	6.1	6.0	4.8	12.2
		1	5.7	8.4	4.4	8.5
	0.56	P1	3.6	4.3	3.6	14.3
		1	7.1	16.7	3.9	9.0
	Alta	0	Control	20.8	19.2	6.7
0.28		P1	5.1	9.3	3.3	13.9
		1	6.5	4.7	4.6	7.9
0.56		P1	3.8	5.1	3.7	11.4
		1	4.9	7.1	4.6	13.6
S-170		0	Control	16.8	23.9	8.4
	0.28	P1	17.7	8.4	4.9	9.2
		1	8.7	8.2	2.8	5.7
	0.56	P1	12.6	5.8	5.3	9.6
		1	7.9	4.5	3.7	11.2
	Fawn	0	Control	10.4	16.1	4.0
0.28		P1	16.3	10.4	3/0	12.2
		1	3.3	6.1	5.4	13.5
0.56		P1	6.2	9.4	4.6	16.5
		1	2.1	4.0	2.8	4.6
LSD .05				0.8	1.0	0.9
LSD .01			1.0	1.3	1.2	1.4

under the 50/60°F night/day temperature regime was higher than that under 70/80°F night/day temperature regime. Stem WSC was higher than leaf WSC which was more pronounced and consistent under the 70/80°F night/day temperature regime.

Experiment 2

Dry Matter Yield per Tiller

The mean squares from the analysis of variance for dry matter yield per tiller of 4 varieties of tall fescue as affected by three levels of mefluidide applied at pre-booting or booting growth stage under night/day temperature regimes of 50/60°F and 70/80°F are shown in Appendix Table 14. Under the low temperature regime, the variety x mefluidide x growth stage interaction was not significant. But the variety x mefluidide, variety x growth stage, and mefluidide x growth stage interactions were highly significant (P, 0.01).

Under the 70/80°F night/day temperature regime, variety x mefluidide x growth stage interaction was highly significant (P, 0.01). This means that neither variety nor mefluidide nor growth stage act independently.

The mean dry matter yield per tiller under the 50/60°F night/day temperature regime is presented in Table 20. Regardless of growth stage, mefluidide rates of 0.28 kg/ha or 0.56 kg/ha reduced dry matter yield significantly (P, 0.01) for all varieties. The variety Fortune did not, however, show differences between these two mefluidide rates. Mefluidide applied at booting growth stage produced significantly

Table 20. Dry matter yield per tiller of 4 varieties of tall fescue as affected by three levels of mefluidide applied at pre-booting (PB) or at booting (B) under 50/60°F night/day temperature regimes in growth chambers.

Mefluidide (M)	Growth stage (G)	Variety (V)				Mean M x G
		Fortune (V ₁)	Alta (V ₂)	S-170 (V ₃)	Fawn (V ₄)	
kg/ha		mg				
0	PB	120.0	172.0	228.0	172.3	173.1
	B	121.7	173.3	228.3	173.3	174.2
0.28	PB	91.7	55.0	176.7	65.0	97.1
	B	97.0	67.0	200.0	72.7	109.2
0.56	PB	92.0	34.0	92.0	58.0	69.0
	B	96.0	48.0	115.3	72.0	82.8
Mean: V x M						
0		120.8	172.7	228.2	172.8	
0.28		94.3	61.0	188.3	68.8	
0.56		94.0	41.0	103.7	65.0	
V x G	PB	101.2	87.0	165.6	98.4	
	B	104.9	96.1	181.2	106.0	
V x M	LSD _{.05} = 5.8	V x G	LSD _{.05} = 4.8	M x G	LSD _{.05} = 4.1	
	LSD _{.01} = 7.8		LSD _{.01} = 6.4		LSD _{.01} = 5.5	

(P, 0.01) higher dry matter yield than pre-booting growth stage application in all varieties except Fortune. This was reflected in the variety x growth stage interaction, where dry matter yield at booting stage was significantly (P, 0.01) higher than that at pre-booting growth stage.

Under the 70/80°F night/day temperature regime (Table 21), all varieties except S-170, produced higher dry matter yield per tiller than under the lower temperature regime. At the 70/80°F night/day temperature regime, mefluidide at rates of 0.28 kg/ha or 0.56 kg/ha, resulted in a significant (P, 0.01) reduction of dry matter yield, either at pre-booting or booting growth stage. However, the application of 0.56 kg/ha mefluidide at the booting growth stage produced a significantly (P, 0.01) higher dry matter yield than at the pre-booting growth stage for varieties Fawn and S-170, but not for Fortune or Alta. This difference among varieties was not exhibited at the 0.28 kg/ha mefluidide rate. Generally, the higher rate of mefluidide produced significantly (P, 0.01) lower dry matter yield for all varieties. This was not as consistent at the pre-booting growth stage, particularly on varieties Fortune and Alta.

Leaf/Stem Ratio

The F-test for the analysis of variance on leaf/stem ratio reveals that the variety x mefluidide x growth stage interaction is highly significant (P, 0.01) as shown in Appendix Table 15.

The average leaf/stem ratio is presented in Table 22. By delaying mefluidide application from pre-booting to the booting

Table 21. Dry matter yield per tiller of 4 varieties of tall fescue as affected by three levels of mefluidide applied at pre-booting (PB) or at booting (B) under a 70/80°F night/day temperature regime in growth chambers.

Variety	Mefluidide kg/ha	Growth stage	Dry matter yield per tiller
			mg
Fortune	0	Control	155.0
	0.28	PB	107.0
		B	129.3
	0.56	PB	104.0
		B	106.3
	Alta	0	Control
0.28		PB	112.3
		B	172.7
0.56		PB	119.7
		B	102.3
S-170		0	Control
	0.28	PB	97.7
		B	101.0
	0.56	PB	66.7
		B	89.0
	Fawn	0	Control
0.28		PB	103.3
		B	110.3
0.56		PB	80.7
		B	101.3
LSD .05			
LSD .01			7.5

Table 22. Leaf/stem ratio of 4 varieties of tall fescue as affected by three levels of mefluidide applied at pre-booting (PB) or at booting (B) under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide kg/ha	Growth stage	Leaf/stem ratio	
			50/60°F	70/80°F
Fortune	0	Control	2.4	2.6
		PB	3.2	3.2
	0.28	B	2.4	2.9
		PB	2.9	4.6
	0.56	B	2.8	3.8
Alta	0	Control	3.2	2.4
		PB	4.5	3.6
	0.28	B	2.1	2.6
		PB	2.7	4.3
	0.56	B	2.5	3.6
S-170	0	Control	2.8	3.1
		PB	3.2	4.2
	0.28	B	2.3	3.2
		PB	4.3	4.2
	0.56	B	2.5	3.4
Fawn	0	Control	2.4	3.4
		PB	3.4	4.8
	0.28	B	1.9	4.3
		PB	2.8	4.2
	0.56	B	2.0	4.1
LSD .05			0.3	0.6
LSD .01			0.4	0.8

growth stage at the low rate (0.28 kg/ha), a significantly ($P, 0.01$) lower leaf/stem ratio resulted. The higher rate of mefluidide, 0.56 kg/ha, did not show this reduction on varieties Fortune and Alta under the 50/60°F night/day temperature regime.

Under a 70/80°F night/day temperature regime, a lower leaf/stem ratio was exhibited by varieties Fortune and S-170 at 0.56 kg/ha and by varieties Alta and S-170 at the 0.28 kg/ha mefluidide rate.

Apparently, increasing the leaf/stem ratio by mefluidide treatment was not consistent under the two temperature regimes for all varieties. However, results reported here indicate that leaf/stem ratio could be increased by mefluidide application, especially when applied at the pre-booting growth stage rather than the booting growth stage.

ADF (Acid Detergent Fiber)

Appendix Table 16 shows the results of the F-test for the effect of treatments upon ADF content of leaf and stem tissues under 50/60°F and 70/80°F night/day temperature regimes. Obviously, variety, mefluidide, and growth stage are not independent of each other (second order interaction).

The average ADF content of leaf and stem are presented in Table 23. In terms of ADF content reduction, mefluidide rate of 0.56 kg/ha was superior when applied at the pre-booting growth stage. However, the high rate of mefluidide (0.56 kg/ha) was required to reduce ADF content significantly ($P, 0.01$) if mefluidide

Table 23. ADF content of leaf and stem as a percentage of its respective dry matter on 4 varieties of tall fescue as affected by three levels of mefluidide applied at pre-booting (PB) or at booting (B) under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide kg/ha	Growth stage	ADF			
			50/60°F		70/80°F	
			leaf	stem	leaf	stem
			%			
Fortune	0	Control	30.8	32.2	26.0	31.4
	0.28	PB	28.3	29.9	25.8	29.2
		B	27.1	29.3	27.1	29.3
	0.56	PB	26.3	29.9	25.8	28.5
		B	27.1	28.3	26.0	29.4
	Alta	0	Control	30.4	33.2	32.1
0.28		PB	28.5	31.2	31.2	34.2
		B	29.5	30.4	31.2	34.1
0.56		PB	28.9	29.7	29.3	30.6
		B	25.9	27.6	28.5	29.4
S-170		0	Control	31.9	35.5	28.7
	0.28	PB	31.5	35.4	26.2	29.9
		B	29.1	30.3	27.3	31.6
	0.56	PB	27.6	30.3	25.9	28.2
		B	27.3	29.1	25.0	29.9
	Fawn	0	Control	30.2	32.4	29.1
0.28		PB	29.3	28.4	27.4	30.6
		B	30.1	32.9	28.5	30.1
0.56		PB	25.4	28.4	26.9	29.8
		B	26.2	27.9	28.8	30.6
LSD _{.05}				1.6	1.6	1.0
LSD _{.01}			2.2	2.1	1.3	1.3

was applied at the booting growth stage. Consistent reduction in ADF content of leaf and stem tissues was not reflected by all varieties. Varieties responded differently under the different temperature regimes.

A significant reduction in ADF content resulted from 0.28 kg/ha of mefluidide application at pre-booting growth stage in Fawn (stem ADF under 50/60°F and leaf ADF under 70/80°F); Alta (leaf ADF under 50/60°F) and Fortune (leaf and stem ADF under 50/60°F and stem ADF under 70/80°F). The results indicate an inconsistent and unexplainable effect of mefluidide on ADF content in different tissues under different temperature regimes.

CP (Crude Protein)

An F-test on the effect of treatments reveals that variety, mefluidide, and growth stage are not independent of each other in determining CP content of leaf and stem tissues under 50/60°F and 70/80°F night/day temperature regimes (Appendix Table 17).

The average CP content of leaf and stem tissues in each variety is shown in Table 24. No increase in CP content of Fawn leaf tissue and stem tissue of S-170 by mefluidide application was shown under the 50/60°F night/day temperature regime. Leaf and stem CP changes were not consistent for each variety or growth stage. Generally, these results indicate that mefluidide application can increase CP content in both leaf and stem tissues under 50/60°F and 70/80°F night/day temperature regimes.

Table 24. Crude protein (CP) of leaf and stem as a percentage of its respective dry matter on 4 varieties of tall fescue as affected by three levels of mefluidide application at pre-booting (PB) or booting (B) under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide kg/ha	Growth stage	CP			
			50/60°F		70/80°F	
			Leaf	stem	Leaf	stem
			%			
Fortune	0	Control	16.3	10.0	17.2	12.9
	0.28	PB	21.7	18.4	19.3	14.7
		B	16.4	13.4	16.4	13.4
	0.56	PB	18.2	17.5	19.2	14.1
		B	17.8	13.3	17.4	12.2
	Alta	0	Control	14.6	9.9	18.7
0.28		PB	21.0	17.6	20.2	15.8
		B	16.6	15.9	20.1	16.0
0.56		PB	18.3	15.2	22.3	16.4
		B	18.4	17.6	19.9	13.3
S-170		0	Control	15.5	10.6	14.7
	0.28	PB	16.2	10.6	19.1	14.3
		B	17.4	10.6	17.7	13.6
	0.56	PB	18.8	10.9	20.9	15.6
		B	16.1	15.0	18.8	15.1
	Fawn	0	Control	16.3	10.3	16.3
0.28		PB	16.4	14.5	19.8	13.7
		B	15.9	12.0	18.3	12.5
0.56		PB	16.2	15.4	21.3	14.9
		B	17.5	18.3	21.3	15.2
LSD _{.05}				1.5	1.3	1.1
LSD _{.01}			2.0	1.7	1.4	1.1

WSC (Water Soluble Carbohydrate)

Mean squares from the analysis of variance for WSC content are shown in Appendix Table 18. The F-test reveals that variety x mefluidide x growth stage interaction is highly significant (P, 0.01) in determining WSC content on stem tissue under 50/60°F and on leaf WSC under 70/80°F night/day temperature regimes.

The average leaf and stem WSC under both temperature regimes are presented in Table 25. In general, WSC concentration of leaves or stems were higher under 50/60°F than under 70/80°F night/day temperature regime. Treatments (variety, mefluidide and growth stage) did not affect WSC content of leaves under the 50/60°F night/day temperature regime. Under the 70/80°F night/day temperature regime, stem WSC of S-170 is higher than for the other varieties. There was significant effect (P, 0.01) of variety only on WSC content of stem tissue under the 70/80°F night/day temperature regime.

The results shown in this table indicate that mefluidide reduced WSC content of leaves under 70/80°F and stem WSC under 50/60°F night/day temperature regime. However, stem WSC of S-170 increased significantly (P, 0.01) under 50/60°F night/day temperature regime at 0.56 kg/ha of mefluidide applied at booting growth stage. Again, the results reported here show inconsistent and unexplainable effects of mefluidide.

Table 25. WSC content of leaf and stem as a percentage of its respective dry matter on 4 varieties of tall fescue as affected by three levels of mefluidide applied at pre-booting (PB) and booting (B) under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Variety	Mefluidide kg/ha	Growth stage	WSC			
			50/60°F		70/80°F	
			leaf	stem	leaf	stem
			%			
Fortune	0	Control	6.4	12.6	2.7	3.6
	0.28	PB	3.1	3.4	2.2	3.4
		B	3.0	3.4	3.0	3.4
	0.56	PB	5.0	6.9	3.4	4.3
		B	5.3	5.7	2.6	3.2
	Alta	0	Control	8.2	11.6	3.4
0.28		PB	2.4	2.4	2.2	3.8
		B	1.9	4.4	2.2	3.9
0.56		PB	4.3	5.4	2.4	2.0
		B	4.5	6.5	2.2	2.4
S-170		0	Control	6.7	9.6	9.4
	0.28	PB	6.7	10.0	4.3	9.4
		B	7.1	10.0	4.8	9.1
	0.56	PB	7.0	10.7	4.4	7.4
		B	8.8	12.1	3.6	8.6
	Fawn	0	Control	4.2	11.8	4.8
0.28		PB	6.8	7.6	2.1	3.4
		B	5.5	9.1	2.0	3.5
0.56		PB	5.7	6.6	3.4	5.4
		B	3.3	5.2	4.1	5.4
LSD .05				ns	0.8	0.2
LSD .01				1.1	0.3	
Mean:	Fortune					3.6
	Alta					3.9
	S-170					10.5
	Fawn					3.6
LSD .05						1.29
LSD .01						1.75

DISCUSSION

A. Field Trials

Experiments 1 and 2

In the first experiment, plants were treated with mefluidide at the vegetative growth stage, while in the second experiment, it was applied at the booting growth stage. Entry 111 produced the highest dry matter yield and also exhibited steady reduction of dry matter yield in both experiments. Experimental results reported here suggested that delaying mefluidide application from the vegetative growth stage to the booting growth stage, dry matter yield obtained would be higher (Table 1 vs Table 5). However, as plants in Experiment 2 were more mature (at the booting growth stage) than in Experiment 1 (at the vegetative growth stage), ADF content increased (Table 2 vs Table 6) and CP decreased (Table 3 vs Table 7). The ADF presented in Table 2 and Table 6 indicate that suppression of ADF in the more mature plants required a high rate of mefluidide.

In general, WSC content of mefluidide treated plants in the second experiment (booting growth stage of mefluidide application) were higher than those in the first experiment, where mefluidide was applied at the vegetative growth stage (Table 4 vs Table 8). Low WSC content of untreated plants in the second experiment was probably due to the accumulation of carbohydrates at the stem base (13, 19) and this stem base portion was not included in the harvested forage. Another factor which may contribute to inconsistent WSC obtained was

the evidence that WSC concentration within the shoot increases from the top to bottom internode but the gradient within the stem is not clearly consistent among plants (19) and during sampling, the lower leaf blade which contains higher WSC than upper leaf blade, might not have been included in the sample.

In the first experiment, mefluidide decreased WSC content, but in the second experiment, WSC content was increased by mefluidide (Table 4 vs Table 8). This occurred in all tested genotypes. This suggests that mefluidide should not be applied at vegetative growth stage.

In both experiments, CP content was increased by mefluidide. WSC concentration in the first experiment was negatively related to increasing CP content. This is in agreement with Minson (13). Higher CP content of mefluidide treated plants was partly attributed to leafiness of treated plants, and leaf CP content is higher than stem CP content (20). This probably resulted in the reduction of WSC concentration of treated plants in the first experiment, because untreated plants were more stemmy than treated plants and grass culm contains higher WSC concentration than leaves. This is the reverse of CP.

ADF content shown in Table 6 for the second experiment and in Table 2 for the first experiment suggested that if mefluidide has to be applied on the more mature plant, a higher rate of mefluidide will be required to reduce ADF content. This was reflected by treated plants with 0.28 kg/ha of mefluidide in the first experiment,

compared to treated plants with 0.84 kg/ha of mefluidide in the second experiment of genotype 229-692 and Ky 31.

Apparently, the application of mefluidide at booting stage could eliminate the loss of dry matter caused by earlier application. This advantage was followed by an increased CP and WSC and reduced ADF content. Further study is required to determine if those benefits also are followed by increasing digestibility of subsequent regrowth of treated forage grasses.

Experiment 3 (Perennial Ryegrass)

A reduction of dry matter occurred on mefluidide treated plants but was followed by an increase in protein and WSC and a reduction in ADF. Reduction of dry matter yields were more pronounced in the stem than the leaf (Table 9). This was consistent with the effect of mefluidide on LAI (Table 9) and on plant height (Table 10). As the plant became shorter, and LAI did not change, stem dry matter yield would be decreased by mefluidide. The disproportionate retardation of stem versus leaf tissue resulted in high leaf/stem ratio for treated plants (Table 9).

The reduction of ADF content and the increase of CP content caused by mefluidide were more pronounced in stem than leaf tissues (Table 11). The inhibition of inflorescence (Table 10) improved forage quality in terms of ADF, CP and WSC content. This study also requires further study to examine the digestibility of treated plants.

B. Growth Chamber Experiments

Experiments 1 and 2

Under both temperature regimes, 50/60°F and 70/80°F night/day, mefluidide reduced dry matter yield substantially when it was applied at the pre-floral initiation, initiation, pre-booting or at booting growth stage (Tables 12, 20, 21). The reduction of dry matter yield caused by mefluidide was more pronounced in the early application of mefluidide (at pre-floral initiation or at floral initiation) than that in the late application (pre-booting or at booting). This reduction effect was less obvious under high temperature regimes (70/80°F night/day). Unproportional retardation by mefluidide on stem versus leaf part was reflected in high leaf/stem ratio of treated plants (Tables 15 and 22).

Leaf/stem ratios in the first experiment were higher than those in the second experiment because plants in the second experiment were more mature, with more culms than those in the first experiment at the time of mefluidide application. By delaying mefluidide application from pre-floral initiation or floral initiation to pre-booting or booting growth stage, leaf and stem ADF would increase (Tables 15 and 23). This is reasonable, since the plants in the second experiment were more mature than those in the first experiment at the time of mefluidide application. But CP and WSC of leaf and stem tissues performed inconsistently, though in both experiments mefluidide resulted in the increase of CP and the reduction of WSC. CP of the

plants in the first experiment (treated with mefluidide in the early growth) in some cases were lower than that which resulted from treated plants in the second experiment (treated at pre-booting or at booting growth stage). But in other cases, CP content of leaf and stem tissues were higher in the first experiment than those in the second experiment. This indicates inconsistent effect of mefluidide.

Temperature regimes of 50/60°F and 70/80°F night/day did not show a conspicuous effect on ADF and CP content of leaf and stem tissues, but WSC concentration of plants under the low temperature regime was higher than those under the high temperature regime. This result agrees with Smith, 1973 (19).

Inconsistent results obtained in these growth chamber experiments may be attributed to several factors. Plants which were grown for these experiments were removed from the field and transplanted into pots in growth chambers. This method would allow greater variation in plant material and stage of development and this would vary with variety. All tillers might not be in the same growth stage at the time of mefluidide application. Improper selection of the appropriate growth stage when mefluidide was applied could also lead to inconsistent results.

However, there were some indications that mefluidide, when applied at the booting growth stage, could eliminate some dry matter reductions, especially under warm temperatures and accompanied by reduction in ADF and increase in CP content.

SUMMARY AND CONCLUSION

The application of mefluidide on tall fescue at vegetative growth stage under field conditions resulted in substantial loss in dry matter yield. The same was true under controlled environmental conditions. Delaying mefluidide application resulted in reduced loss of dry matter yield and some forage quality parameters improvement (ADF, CP and WSC). Warmer temperatures are preferable in order to reduce dry matter lost.

In perennial ryegrass, reduction of ADF and the enhancement of CP are more pronounced in stem parts rather than in leaves. Leaf area was not affected significantly but plant height and the onset of inflorescence were inhibited by mefluidide.

Mefluidide can be used in manipulating growth performances in order to get a balance between quantity and quality factors of forage grass production. As far as ADF, CP, WSC and dry matter yield are concerned, mefluidide application at the booting growth stage is not too late under field conditions or in growth chambers. Higher rates of mefluidide will be required if it has to be applied to older plants. Further study is essential for testing the digestibility of dry matter resulting from mefluidide application on forage grass regrowth.

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APPENDIX LITERATURE REVIEW

Forage production comprises quantity and quality aspects. Quantity is reflected in dry matter yield and quality by nutritive value of the forage produced. Dry matter yield improvement has been successfully accomplished through breeding work and many attempts have been devoted to genetic improvement of forage quality (7, 25, 32). With further plant improvement and improved processing, forage quality is likely to be increased.

Nutritive value of forage is determined by its chemical composition, digestibility and the nature of digested products (24). Chemical composition is a factor associated with the plant and the environment under which it is grown. Natural factors that have a bearing on the chemical composition and correspondingly on the nutritive value are the kind of plant, climatic conditions, soil fertility, the weather preceding and during harvest, the age of the plant at harvest time, and the season of the year (6, 35).

As a forage becomes mature, it also becomes less digestible (19, 22, 26, 28). Apparent digestibility of protein, organic matter and fiber are highest in the early growth stages as exhibited by tall fescue (27) and perennial ryegrass (20, 22). Digestibility of forage grasses starts to decline soon after the onset of reproductive growth (20, 22, 31). As the grass plant matures, its fiber content increases and becomes less digestible (6, 23, 29, 30, 38), partly as a consequence of lignification of the stem (1, 15, 19).

Crude protein (CP) content is positively correlated with

digestibility (15, 26, 34) and its content decreases along with the advance of plant maturity (15, 26, 27, 37). Changes in dry matter digestibility are inversely related to the lignin content of the forage (1, 13, 15, 19, 38), but lignin content expressed as a proportion of crude fiber content decreases with increasing plant age (15). In contrast, carbohydrates (monosaccharides, disaccharides, fructosans and starch) in a wide range of feeds are completely digestible by ruminants (10). An increase in protein content is often offset by a depression in soluble carbohydrate level (2). Peaks of water soluble carbohydrate (WSC) in tall fescue occurred in early spring and irrigation during the summer resulted in higher concentration of WSC in leaf tissue. However, WSC accumulated in stem bases of water-stressed plants (17).

In vitro dry matter digestibility (IVDMD) of the leaf blade of tall fescue, as well as perennial ryegrass, is higher than that of stems and position of that leaf also influences the digestibility (15). As the season progresses, digestibility of the stem declines more rapidly than that of the leaf but young stem tissue is as digestible and sometimes more digestible than leaf tissue (36).

Obviously, the leaf blade is lower in fiber and higher in protein content than that of stem (20). Because these fractions have been assumed to be closely correlated with digestibility, herbage breeding programs have emphasized breeding for leafiness. However, there is frequently only a small correlation between herbage digestibility and leaf/stem ratio, and leafy cuts of different herbage

species may also differ markedly in digestibility. Perennial ryegrass has consistently been more digestible than cocksfoot at the same growth stage (8).

Among grass species, at the flowering stage, late maturing grasses maintain a higher level of digestibility than the early maturing grasses. However, early maturing grasses have a higher IVDMD at the flowering stage than later maturing grasses (28). It was noted that change in digestibility from an almost consistent high level to lower level was closely associated with the first emergence of flowering heads in *Dactylis glomerata* and *Lolium perenne* as well as in *Festuca arundinacea* (28, 30).

Apparently, there are large differences in digestibility between different plant parts (15). Pritchard et al. (28) found that the upper segments of the stem tended to have a lower IVDMD than the basal segments. This phenomenon might be associated with the presence of higher concentrations of total nonstructural carbohydrates (TNC) in the basal segments. TNC concentration usually increases from the top to bottom internodes on the stem, especially as the shoot advances in maturity (23, 33).

Poor forage quality of tall fescue and perennial ryegrass is noted after the onset of reproductive growth (23, 28, 30). Delaying or prolonging the vegetative stage, or inhibiting maturation could also delay the time of forage quality decline and effective grazing duration could be improved. Mechanical means which result in maintaining high forage quality such as frequent cutting, are cumbersome and laborious.

Mefluidide {N-[2,4-dimethyl-5-[[trifluoromethyl)-sulfonyl]amino]phenyl]acetamide}, previously known as MBR 12325, is a plant growth regulator that has been found to inhibit seedhead production, decrease cellulose content, increase N content, increase digestibility of dry matter, decrease ADF (acid detergent fiber) and NDF (neutral detergent fiber), and reduce dry matter production (11). This chemical reduces leaf blade length, seedhead density, and plant height in bermudagrass turf (16). It severely inhibits plant growth, especially top growth (9) and the common responses of many plant species to mefluidide is death of the apical meristem and axillary buds (9).

Uniformly ring-labelled ^{14}C -mefluidide was applied to the uppermost fully expanded leaf of two-week old corn, only 3% of the radioactivity remained in the treated leaf after 72 hours and this was found to be associated with the nucleus, chloroplast, mitochondria and ribosomes. The remaining 90% was in the supernatant, indicating the association of the radioactivity with the cytoplasm and soluble enzymes. At low rates (0.001 μM) mefluidide stimulates elongation, but at concentrations of 0.1 μM , it will reduce coleoptile elongation. Mefluidide at rates from 0.1 to 1.0 μM stimulates incorporation of leucine into protein so that mefluidide acts as an auxin-like substance (14).

Other works showed that absorption of mefluidide was greater through the foliage than the roots, and foliar-applied mefluidide moved in the phloem along with the assimilate stream mainly to the

areas of high metabolic activity (5). The reduction of top growth following mefluidide application might be attributed to its action within the root of the plant, since most of the absorbed mefluidide is translocated to the roots (12).

Temperature and relative humidity (RH) influence the translocation of mefluidide. At a constant level of RH, 40% or 100%, an increase in air temperature from 22 to 32°C resulted in a drastic increase in absorption and translocation of mefluidide in soybeans. At a constant temperature, 22 or 32°C, an increase of RH from 40% to 100% resulted in less than a twofold increase of mefluidide absorption and translocation. The effect of RH was more pronounced in soybean, while temperature was more pronounced in johnsongrass and common cocksbur (21).

Mefluidide is rapidly absorbed and excreted by the cow or sheep, eliminated almost totally through the urine in the form of unmetabolized mefluidide. This chemical also does not result in toxicological hazards to the exposed animals nor contamination of meat or milk used for human consumption (18).

Decrease in dry matter yield of tall fescue by mefluidide application has been reported by many investigators, though it is followed by an increase in protein and carbohydrate content (11), decrease in cellulose content and delaying of flowering head production (11, 13, 16). Mefluidide should be applied at a specific time (growth stage) so that a yield reduction by mefluidide will be compensated for high forage quality.

Forage quality is often related to CP content (15, 26, 27, 37). The problem becomes more complex if forage quality is measured by parameters other than protein, such as carbohydrate content and fiber. As plants mature, crude fiber increases, crude protein decreases, and carbohydrate may increase, while the proportion of stem also increases, which in turn decreases dry matter digestibility. Further work is needed to utilize the use of mefluidide for use in pasture improvement in the diverse climate areas within the United States and elsewhere.

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A P P E N D I X

Appendix Table 1. Mean square from the analysis of variance for dry matter yield of individual clones of 5 genotypes of tall fescue as affected by mefluidide applied at the vegetative growth stage in 1980.

Source of variation	df	MS
Treatments	29	56526.368
Genotypes (G)	4	21087.600**
Mefluidide (M)	5	34561.500**
G x M	20	877.268**
Error	120	180.218
Total	149	

**Significant at the 1% probability level.

Appendix Table 2. Mean square from the analysis of variance for ADF, CP and WSC as percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the vegetative growth stage in 1980.

Source of variation	df	MS		
		ADF	CP	WSC
Treatments	29	70.6246	16.4432	58.3204
Genotypes (G)	4	21.6885**	7.8236**	37.3067**
Mefluidide (M)	5	44.5884**	7.8634**	15.9860**
G x M	20	4.3478**	0.7562**	5.0277**
Error	30	0.7384	0.2498	0.1991
Total	59			

**Significant at the 1% probability level.

Appendix Table 3. Mean square from the analysis of variance for dry matter yield of individual clones of 5 genotypes of tall fescue as affected by mefluidide applied at the booting growth stage in 1981.

Source of variation	df	MS
Treatments	29	317999.500
Genotypes (G)	4	77656.400**
Mefluidide (M)	5	228612.000**
G x M	20	11731.100**
Error	120	1569.770
Total	149	

**Significant at the 1% probability level.

Appendix Table 4. Mean square from the analysis of variance for ADF, CP and WSC as percentage of dry matter on 5 genotypes of tall fescue as affected by mefluidide applied at the booting growth stage of regrowth in 1981.

Source of variation	df	MS		
		ADF	CP	WSC
Treatments	29	241.7796	15.7604	220.9046
Genotypes (G)	4	60.6422**	9.4230**	70.5986**
Mefluidide (M)	5	167.7250**	5.2691**	134.8950**
G x M	20	13.4124**	1.0683**	15.4110**
Error	30	0.4547	0.1748	0.1619
Total	59			

**Significant at the 1% probability level.

Appendix Table 5. Mean square from the analysis of variance for dry matter yield per m² of perennial ryegrass cv. Linn as affected by mefluidide applied at the floral initiation growth stage.

Source of variation	df	MS		
		Leaf	Stem	Total
Mefluidide	2	6346.33 ^{ns}	240419.00*	234336.45 ^{ns}
Replication	2	3436.33	42800.10	55947.11
Error	4	8412.67	26243.60	78166.95
Total	8			

ns = non significant ($P \leq 0.05$)

*Significant at the 5% probability level.

Appendix Table 6. Mean square from the analysis of variance for LAI and leaf/stem ratio of perennial ryegrass cv. Linn as affected by mefluidide applied at the floral initiation growth stage.

Source of variation	df	MS	
		LAI	Leaf/stem ratio
Mefluidide	2	0.0144 ^{ns}	0.1132**
Replication	2	0.0495	0.0137
Error	4	0.0549	0.0034
Total	8		

ns = non-significant at the 5% probability level.

**Significant at the 1% probability level.

Appendix Table 7. Mean square from the analysis of variance for plant height and percentage of fertile tillers as affected by mefluidide applied at the floral initiation growth stage of perennial ryegrass cv. Linn.

Source of variation	df	MS	
		Plant height	Fertile tiller ¹
Mefluidide	2	629.3065**	7.0098**
Replication	2	4.3303	0.5762
Error	4	11.0176	0.1106
Total	8		

**Significant at the 1% probability level.

¹Transformed to $\sqrt{x\%}$

Appendix Table 8. Mean square from the analysis of variance for ADF, CP and WSC as percentage of dry matter of perennial ryegrass cv. Linn as affected by mefluidide applied at the floral initiation growth stage.

Source of variation	df	MS					
		ADF		CP		WSC	
		Leaf	Stem	Leaf	Stem	Leaf	Stem
Mefluidide	2	6.9728 ^{ns}	42.9406*	1.9428 ^{ns}	4.0489*	66.8372**	32.1712**
Replication	2	0.9559	0.2083	2.2321	0.3734	0.0280	1.8981
Error	4	3.0167	3.3541	1.7607	0.4304	0.8620	0.5909
Total	8						

ns = non-significant at the 5% probability level.

*Significant at the 5% probability level.

**Significant at the 1% probability level.

Appendix Table 9. Mean square from the analysis of variance for dry matter yield per tiller of 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS	
		50/60°F	70/80°F
Treatment	23	145455.22**	72159.36**
Variety (V)	3	1565.38**	4074.50**
Mefluidide (M)	2	138172.00**	63143.60**
Growth stage (G)	1	1995.01**	2560.52**
V x M	6	2868.43**	316.68**
V x G	3	75.90**	398.24**
M x G	2	464.22**	1186.85**
V x M x G	6	314.28**	478.96**
Error	48	7.83	32.93
Total	71		

**Significant at the 1% probability level.

Appendix Table 10. Mean square from the analysis of variance for leaf/stem ratio of 4 varieties of tall fescue as affected by mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS	
		50/60°F	70/80°F
Treatment	23	35.2149**	22.8117**
Variety (V)	3	8.7272**	2.6218**
Mefluidide (M)	2	15.8812**	14.0316**
Growth stage (G)	1	7.0688**	3.6136**
V x M	6	0.7162**	1.0332**
V x G	3	0.5941**	0.4221**
M x G	2	1.839**	0.9781**
V x M x G	6	0.3835**	0.1113**
Error	48	0.0431	0.1869
Total	71		

**Significant at the 1% probability level.

Appendix Table 11. Mean square from the analysis of variance for ADF content of leaf and stem as a percentage of its respective dry matter of 4 varieties of tall fescue as affected by 3 rates of mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS			
		50/60°F		70/80°F	
		Leaf	Stem	Leaf	Stem
Treatment	23	58.2523**	49.2489**	159.1543**	179.8473**
Variety (V)	3	14.7919**	16.5172**	34.1713**	35.6906**
Mefluidide (M)	2	17.5433**	5.1289**	76.7752**	91.2552**
Growth stage (G)	1	16.1008**	5.4675**	0.5419	5.2008**
V x M	6	2.7744**	13.3587**	6.1197**	13.1558**
V x G	3	3.9386**	1.5181**	0.5630	24.3139**
M x G	2	3.1033**	2.7269**	32.0744**	7.2477**
V x M x G	6	1.8678**	4.5316**	8.9088**	2.9833**
Error	24	0.4050	0.2379	0.7723	0.5154
Total	47				

**Significant at the 1% probability level.

Appendix Table 12. Mean square from the analysis of variance for crude protein (CP) of leaf and stem parts as a percentage of its respective dry matter of 4 varieties of tall fescue as affected by 3 rates of mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS			
		50/60°F		70/80°F	
		Leaf	Stem	Leaf	Stem
Treatment	23	329.8900**	379.9544**	74.3784**	73.0133**
Variety (V)	3	48.7847**	22.0647**	11.2641**	2.4435**
Mefluidide (M)	2	245.6540**	325.9765**	44.6800**	51.5258**
Growth stage (G)	1	3.5209**	0.1752	1.4352	5.6719**
V x M	6	19.3537**	6.4959**	4.1589**	2.2108**
V x G	3	0.9136	8.7880**	4.9569**	2.1524**
M x G	2	11.1889**	12.1690**	1.0508	4.3425**
V x M x G	6	0.4743	4.2851**	6.8325**	4.6664**
Error	24	0.6363	0.4323	0.9056	0.3056
Total	47				

**Significant at the 1% probability level.

Appendix Table 13. Mean square from the analysis of variance for leaf and stem WSC content as a percentage of its respective dry matter of 4 varieties of tall fescue as affected by 3 rates of mefluidide applied at pre-floral initiation or at floral initiation under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS			
		50/60°F		70/80°F	
		Leaf	Stem	Leaf	Stem
Treatment	23	765.5773**	1152.9969**	53.2937**	527.0447**
Variety (V)	3	57.0404**	25.9387**	5.9674**	13.8141**
Mefluidide (M)	2	519.5590**	1030.9000**	37.7162**	378.5400**
Growth stage (G)	1	51.2120**	2.2925**	0.0683	33.5922**
V x M	6	55.9343**	24.8688**	4.9428**	59.3853**
V x G	3	36.4829**	35.0390**	2.0402**	6.5278**
M x G	2	31.7260**	13.9988**	0.7693**	11.7790**
V x M x G	6	13.6227**	19.9591**	1.7895**	23.4063**
Error	24	0.1409	0.2163	0.1775	0.2607
Total	47				

**Significant at the 1% probability level.

Appendix Table 14. Mean square from the analysis of variance for dry matter yield per tiller of 4 varieties of tall fescue as affected by 3 levels of mefluidide applied at pre-booting or at booting under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS	
		50/60°F	70/80°F
Treatment	23	94128.6080**	64080.8304**
Variety (V)	3	25432.3000**	2461.2000**
Mefluidide (M)	2	61029.8000**	56112.1000**
Growth stage (G)	1	1458.0000**	1953.1300**
V x M	6	5770.7400**	1969.8400**
V x G	3	112.4810**	39.4954**
M x G	2	286.6250**	795.1250**
V x M x G	6	38.6620	749.9400**
Error	24	25.2917	11.7222
Total	47		

**Significant at the 1% probability level.

Appendix Table 15. Mean square from the analysis of variance for leaf/stem ratio of 4 varieties of tall fescue as affected by 3 rates of mefluidide applied at pre-booting or at booting under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS	
		50/60°F	70/80°F
Treatment	23	14.8297**	16.5997**
Variety (V)	3	1.0805**	2.6700**
Mefluidide (M)	2	0.1492**	8.1616**
Growth stage (G)	1	9.0029**	3.7310**
V x M	6	0.5790**	0.8141**
V x G	3	0.3722**	0.1455**
M x G	2	2.9567**	0.9837**
V x M x G	6	0.6892**	0.0938**
Error	48	0.0270	0.0136
Total	71		

**Significant at the 1% probability level.

Appendix Table 16. Mean square from the analysis of variance for ADF content of leaf and stem as a percentage of its respective dry matter of 4 varieties of tall fescue as affected by 3 levels of mefluidide applied at pre-booting or at booting under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS			
		50/60/°F		70/80°F	
		Leaf	Stem	Leaf	Stem
Treatment	23	77.9133**	109.1345**	58.1568**	38.4557**
Variety (V)	3	5.0747**	15.6163**	38.6374**	11.5341**
Mefluidide (M)	2	65.5565**	73.9952**	11.5544**	14.7058**
Growth stage (G)	1	0.7752	5.6719**	0.2269	0.0169
V x M	6	2.5126**	2.0105*	2.5558**	4.6939**
V x G	3	1.0891	4.9719**	2.6219**	3.9297**
M x G	2	0.4040	1.3856	1.8981**	2.7700**
V x M x G	6	2.5012**	5.4831**	0.6623*	0.8053**
Error	24	0.6202	0.5685	0.2135	0.2065
Total	47				

*Significant at the 5% probability level.

**Significant at the 1% probability level.

Appendix Table 17. Mean square from the analysis of variance for leaf and stem CP content as a percentage of its respective dry matter of 4 varieties of tall fescue as affected by 3 levels of mefluidide application at pre-booting or at booting under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS			
		50/60°F		70/80°F	
		Leaf	Stem	Leaf	Stem
Treatment	23	52.1102**	174.7185**	80.6632**	52.8843**
Variety (V)	3	4.5239**	20.2030**	19.1167**	12.5591**
Mefluidide (M)	2	21.0052**	113.9030**	38.9715**	23.4019**
Growth stage (G)	1	9.0133**	0.9633	8.0033**	2.4752**
V x M	6	3.9891**	11.5889**	5.7823**	6.5783**
V x G	3	2.8350**	10.6039**	1.3978**	0.9719**
M x G	2	5.8752**	13.5233**	6.0365**	4.4877**
V x M x G	6	4.8685**	3.8422**	1.3551**	2.4102**
Error	24	0.5188	0.3863	0.2633	0.1681
Total	47				

**Significant at the 1% probability level.

Appendix Table 18. Mean square from the analysis of variance for WSC content of leaf and stem as a percentage of its respective dry matter of 4 varieties of tall fescue as affected by 3 levels of mefluidide application at pre-booting or at booting under 50/60°F and 70/80°F night/day temperature regimes in growth chambers.

Source of variation	df	MS			
		50/60°F		70/80°F	
		Leaf	Stem	Leaf	Stem
Treatment	23	139.1709	169.2373**	86.1308**	35.9604
Variety (V)	3	27.1717	28.2826**	63.8790**	29.1516**
Mefluidide (M)	2	6.0753	110.5590**	13.6006**	1.4800
Growth stage (G)	1	20.1632	0.6256*	0.0154	0.3605
V x M	6	16.7295	26.1079**	7.8451**	3.4287
V x G	3	27.2715	1.3618**	0.0130	0.5482
M x G	2	19.2416	1.3708**	0.5973**	0.3824
V x M x G	6	22.5181	0.9296**	0.1804**	0.6090
Error	24	18.9855	0.1669	0.0087	2.3581
Total	47				

*Significant at the 5% probability level.

**Significant at the 1% probability level.