Forage Quality Responses of Selected Grasses to Tebuthiuron

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Abstract

Tebuthiuron pellets (20% active ingredient [a.i.]) applied at 0.6, 1.1 or 2.2 kg/ha (a.i.) to native stands of little bluestem and at 1.1 or 2.2 kg/ha to 1-year-old seeded stands of Bahiagrass, Bell rhodesgrass, green sprangletop, and little bluestem did not significantly alter the in vitro digestible organic matter concentrations of grass leaves. Leaf water concentrations of Bahiagrass, green sprangletop, and little bluestem were not consistently altered by application of tebuthiuron. However, application of 1.1 or 2.2 kg/ha of tebuthiuron pellets to seeded stands or to native little bluestem increased foliar crude protein concentrations. Application of 0.125, 0.188, or 0.25 ppm of tebuthiuron in aqueous solutions to pots containing grasses in the greenhouse significantly increased foliar crude protein concentrations, compared to that of untreated plants. Crude protein concentrations were increased only during the growing season of application in the native stand of little bluestem. These results suggest application of tebuthiuron for brush control may enhance rangeland forage crude protein concentrations while not affecting in vitro digestible organic matter.

Research on herbicides for brush management traditionally has emphasized efficacy as related to dosage and time of application with lesser emphasis on forage responses. Most studies of range forage responses to herbicide application have evaluated changes in standing crop and botanical composition of herbaceous stands as attributed to selective removal or suppression of associated woody species. However, sublethal applications of some herbicides may significantly alter various physiological processes within some forage species (Reis 1976). Nutritional constituents, such as crude protein, have increased following applications of simazine [2-chloro-4,y-bis(ethylamino)-*s*-triazine] (Allinson and Peters 1970) or atrazine [2-chloro-4-(ethylamino)-6-(isopropylamino)-*s*-triazine] (Baker et al. 1980).

The physiological mode of action of tebuthiuron [N-(5-(1,1-dimethylethyl)-1,3,4-thiadiazol-2--yl)-N,N'-dimethylurea] is similar to that of *s*-triazines (Klingman and Ashton 1975). Tebuthiuron effectively controls a number of woody species on rangeland, several of which are not controlled with conventional herbicide sprays (Scifres et al. 1979, Scifres et al. 1981a). However, selectivity of tebuthiuron is dosage dependent. Applications of 5.6 kg/ha active ingredient (a.i.) or more tebuthiuron may result in total vegetation control (Ford et al. 1974), while broadcast applications of 1.1 and 2.2 kg/ha (a.i.) of the pellet formulation for brush control may significantly increase production of desirable grasses the following growing season (Scifres and Mutz 1978, Scifres et al. 1981b). Selective application rates of tebuthiuron may also increase the proportion of species of good-to-excellent grazing value in the forage stands (Scifres and Mutz 1978, Scifres et al. 1981b). For example, presence and production of the genera *Chloris, Paspalum* and *Schizachryium* may increase considerably following tebuthiuron applications (Scifres and Mutz 1978, Scifres et al. 1981b). Green sprangletop (*Leptochloa dubia* (H.B.K.) Nees) and little bluestem (*Schizachryium scoparium* (Michx.) Nash) are native range species while Bahiagrass (*Paspalum notatum* Flugge) and Bell rhodesgrass (*Chloris gayana* Kunth) are introduced grass species used in south Texas.

Daily steer gains and total steer gains and grazing days/ha were increased, compared to that on adjacent untreated brush pastures, the growing season following application of 2.2 kg/ha of tebuthiuron to mixed hardwoods in east central Texas (Scifres et al. 1981b). Seasonal steer diets selected from tebuthiuron-treated pastures were higher in crude protein and in vitro digestible organic matter concentrations compared to diets of steers grazing untreated pastures (Kirby and Stuth 1982). Moreover, Scifres et al. (1983) reported that cattle apparently selected tebuthiuron-treated plots for grazing in preference to untreated areas. This grazing preference usually occurred only during the growing season of tebuthiuron application.

This study was designed to determine if tebuthiuron applied at rates used for brush control would affect forage quality of selected grass species by altering leaf water, crude protein and in vitro digestible organic matter concentrations; and the affects of varying concentrations of the herbicide on grass seedling growth and leaf crude protein concentrations. Plant crude protein and in vitro

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Authors appreciate the assistance of G.A. Rasmussen in the conduct of the research, and the efforts of J.R. Scifres in manuscript preparation and typing.

Manuscript received September 30, 1982.

digestible organic matter concentrations are recognized as indicators of forage quality (Dietz 1970). Leaf water concentration as a measure of plant succulence may be an indicator of palatibility (Archibald et al. 1943).

Materials and Methods

Field Experiments

Field experiments were conducted on a brush-free site on the Texas A&M University Range Research Area near College Station. The site is a Tabor sandy loam (Udertic Paleustalf) with 50% sand and 11% clay and 1.6% organic matter in the surface 8 cm (Mutz et al. 1979).

Native Stand

A 0.63-ha, little bluestem-dominated pasture was divided into 36, 11 by 16-m plots for application of treatments in a latin square design. Treatments included tebuthiuron pellets (20% active ingredient) at 0, 0.6, 1.1, and 2.2 kg/ha (a.i.) applied with a hand-operated fertilizer spreader on April 4, 1979. In addition, picloram (4-amino-3,5,6-trichloropicolinic acid) and 2,4-D [(2,4-dichlorophenoxy) acetic acid] as broadcast sprays were applied at 1.1 kg/ha with a hand-operated sprayer on April 7, 1979. All plots were mowed to a 12-cm stubble height on July 31, 1979, and May 27, 1980, to produce uniform vegetation height across the experimental area.

Randomly located little bluestem plants were harvested to a 2.5-cm stubble height from each plot 44, 63, 110, 182, 202, 405, and 435 days after application of tebuthiuron. Only previously unclipped plants were harvested at each sampling date. Approximately a 100-g subsample of green leaf tissue was taken from each herbage sample and fresh weight recorded. The subsample was then oven dried at 105°C for 1 hr, at 60°C for 22 hr, then at 105°C for 1 hr and dry weight recorded. This drying scheme was employed to avoid nitrogen losses attributable to excessive heat associated with a 24-hr, 105°C drying cycle. Leaf water concentration was calculated as a percentage of fresh weight.

Dried samples were ground to pass a 1-mm screen. Leaf samples were analyzed for percent crude protein based on the micro-kjeldahl procedure for nitrogen determination (A.O.A.C. 1970). Digestible organic matter concentration was determined on an ash-free basis utilizing the fermentation stage (Tilley and Terry 1963) followed by extraction in a neutral detergent (Van Soest and Wine 1967). A standard forage, buffelgrass (*Cenchrus ciliaris* L.), of known in vitro digestibility was included in each digestion analysis to correct in vitro digestible organic matter to apparent digestibility. Soil samples were taken at 0–8, 8–15 and 15–30 cm increments from each herbage collection plot and percent soil water was determined using the gravimetric technique.

Seeded Stands

A 0.36-ha area immediately adjacent to the native grass stand was plowed in May and disced to a smooth seedbed in June 1979. The study area was divided into 16, 11 by 17-m plots separated by 2.4-m wide strips. Bahiagrass, Bell rhodesgrass, green sprangletop, and little bluestem were each planted at 5 kg/ha of pure live seed to each of 4 plots in randomized complete block design arranged as a split plot. Grass species constituted the main plot effect and herbicide the subplot effect. The plots were sprinkler irrigated during the summer and early fall of 1979 and 1980 to ensure establishment and maintenance of the stands.

Vegetation on the study site was mowed to a 12-cm stubble height and raked from the plots on February 29, 1980. All plots were hand weeded from April 20 to May 2, 1980, leaving pure stands of the seeded species. Tebuthiuron pellets (20% a.i.) were applied at 0, 1.1 and 2.2 kg/ha (a.i.) with a hand-operated spreader on May 3, 1980, to a third of each of the plots.

Approximately 100 g of herbage was sampled from near the center of each subplot by harvesting several plants to a 2.5-cm stubble height 31, 57, 93, and 120 days following herbicide application. Grass leaves were analyzed for water crude protein, and in vitro digestible organic matter concentrations as previously described. Percent soil water from each plot at depths of 0–8, 8–15, and 15–30 cm was determined using the gravimetric method.

Greenhouse Experiment

The greenhouse experiment was designed as a randomized complete block with 5 replications arranged as a split plot. Two trials of the experiment were conducted during the period, May 27 to September 27, 1980. Species used in the field experiment were seeded in 946-ml pots containing 1,140 g of air dried aeolian sand. With each trial, 100 pots were seeded with each of the 4 species. Half of the pots were then treated with tebuthiuron when the grasses had developed 2 fully expanded leaves. A suspension of tebuthiuron wettable powder (80% a.i.) in distilled water was pipetted onto the sand surface to achieve dosages of 0, 0.063, 0.125, 0.188, and 0.25 ppm (wt/wt), such that all pots within a species and seedling growth stage were treated with herbicide at the same time. The remaining pots received herbicide treatments when grasses reached the four-leaf growth stage. Grass stands were thinned to 10 plants per pot at time of herbicide application. Lee and Ishizuka (1976) found the two- and four-leaf growth stages to be appropriate for herbicide application when evaluating susceptibility of barley (*Hordeum vulgare* L.) and wheat (*Triticum aestivum* L.) to thiadiazolyl urea herbicides.

Plants and soil were removed from the pots 30 days after herbicide application. Sand was soaked from the roots and numbers of live and dead plants from each pot were recorded. Plants were separated into roots and shoots, ovendried at 60°C for 24 hr, and

Table 1. Average concentrations of water and crude protein of little bluestem leaves at various times after application of tebuthiuron pellets on April 4, 1979 near College Station, Texas.[1]

Tebuthiuron rate (kg/ha)	Time (days) after tebuthiuron application[2]						
	44	63	110	182	202	405	435
	Water concentration (%)						
0	69 b	62 a	66 a	66 a	58 a	71 a	70 a
0.6	66 a	63 a	67 a	66 a	57 a	71 a	70 a
1.1	68 b	65 ab	69 a	68 a	61 a	71 a	70 a
2.2	70 b	68 b	68 a	67 a	58 a	72 a	70 a
	Crude protein concentration (%)						
0	6.1 a	5.7 a	6.5 a	6.9 a	6.5 a	7.0 a	10.3 a
0.6	6.3 ab	5.7 a	7.7 ab	7.3 ab	7.1 a	7.3 a	9.8 a
1.1	7.1 bc	7.1 b	8.4 b	8.6 b	7.4 a	7.6 a	10.0 a
2.2	8.3 c	6.2 ab	8.0 b	8.0 ab	7.1 a	7.4 a	9.9 a

[1]Crude protein concentrations are reported on dry weight basis.
[2]Means within time after application and an attribute followed by the same letter are not significantly different ($\alpha = 0.05$) according to Tukey's *w*-procedure.

weighed. Shoots were separated into leaves and stems, and leaves ground in a Wiley mill to pass a 1-mm mesh screen. Leaf samples were analyzed for percent crude protein based on the micro-kjeldahl procedure for nitrogen determination (A.O.A.C. 1970).

Statistical Analysis

Analysis of variance was conducted on each variable from the native stand with sampling time as the main plot effect and tebuthiuron application rate as the subplot effect. Hierarchical order of sources of variation for analysis of measurements from seeded stands in descending order were species, herbicide rate and sampling time. Hierarchical analysis of variance was applied to each variable from the greenhouse study with sources of variation in descending order as grass species, trial, grass growth stage and herbicide treatment.

Results and Discussion

Regardless of variable evaluated in the greenhouse study, the effect of grass seedling growth stage when treated with tebuthiuron and growth stage by herbicide rate interactions were not significant (α=0.05). Therefore, only grass species by herbicide treatment for all experiments, and sampling time by grass species by tebuthiuron treatment means for the native and seeded stand experiments are presented.

Native Stand

Rainfall during the period 60 days prior to tebuthiuron application, about 25 cm, was sufficient to promote rapid vegetative growth of little bluestem. Only 3 cm of rainfall were received during June, 19 cm were received in July, and 8 cm in August. Soil water concentration of the surface 8 cm varied from 29% in March to 11% in July. However, soil water concentration at 15–30 cm deep was always greater than 15%.

Standing crop data are reported in a study of grazing preference of cows for tebuthiuron-treated areas (Scifres et al. 1983). Tebuthiuron in that study did not decrease standing crop of little bluestem, regardless of application rate. Moreover, standing crop of little bluestem was increased in the fall after application of 2.2 kg/ha of tebuthiuron in the spring.

Average crude protein concentration of little bluestem leaves was not increased by 0.6 kg/ha of tebuthiuron compared to that of tissues from untreated plots (Table 1). However, crude protein concentrations of little bluestem from plots treated with 1.1 kg/ha of tebuthiuron were increased in samples recovered at 63, 110 and 182 days after herbicide application. Crude protein concentrations were increased in little bluestem leaves 44 and 110 days following application of 2.2 kg/ha of tebuthiuron. In addition, there was a trend for an increase in little bluestem crude protein concentrations 63 and 182 days after application of 2.2 kg/ha of the herbicide. Average crude protein concentrations were unaffected the growing season after herbicide application. With exception of plants sampled 63 days after herbicide application, leaf water concentrations were unchanged, regardless of rate of tebuthiuron applied or sampling date (Table 1). Digestible organic matter concentrations of little bluestem leaves were significantly changed by herbicide treatment regardless of sampling date. The average digestible organic matter concentration of little bluestem leaves was 49.9% ± 2.04.

Seeded Stands

Plots in seeded stands were not of adequate size to allow sampling for standing crop and also retain enough tissue for laboratory analysis. However, based on periodic visual inspections during the study, there were no apparent yield reductions attributable to herbicide application.

Tebuthiuron at 1.1 or 2.2 kg/ha increased crude protein concentrations of leaves of all grass species at 31 days after application of the herbicide, and increased the crude protein concentrations of all species except little bluestem 93 and 120 days after application (Table 2). Both herbicide rates increased the crude protein concentrations of Bahiagrass leaves 57 days after tebuthiuron application.

Table 2. Average crude protein concentration (%) of Bahiagrass, Bell rhodesgrass, green sprangletop and little bluestem leaves at various times after application of tebuthiuron pellets on May 3, 1980 near College Station, Texas.

Tebuthiuron rate (kg/ha)	Time (days) after tebuthiuron application[1]			
	31	57	93	120
		Bahiagrass		
0	8.9 a	6.4 a	8.7 a	8.5 a
1.1	12.6 b	8.0 b	10.7 b	10.9 b
2.2	13.8 b	9.3 b	11.2 b	11.7 b
		Bell rhodesgrass		
0	6.2 a	5.1 a	6.0 a	7.4 a
1.1	7.9 b	5.6 a	7.4 b	8.8 b
2.2	9.0 b	5.9 a	8.4 b	9.7 b
		Green sprangletop		
0	9.9 a	7.8 a	12.1 a	12.4 a
1.1	14.4 b	9.0 ab	14.3 b	16.0 b
2.2	16.6 c	9.5 a	16.0 c	17.4 c
		Little bluestem		
0	7.1 a	5.8 a	8.2 a	7.4 a
1.1	8.6 b	6.9 a	9.7 a	8.1 ab
2.2	9.7 b	7.1 a	9.4 a	9.1 b

[1]Means within time after application and species followed by the same letter are not significantly different (α=0.05) according to Tukey's *w*-procedure.

Green sprangletop leaves from plots treated with 2.2 kg/ha of tebuthiuron contained more crude protein than did samples from untreated plots, regardless of sampling time, and contained more crude protein at 31, 93, and 120 days after application than leaves from plots treated with 1.1 kg/ha of the herbicide. Different responses to tebuthiuron among grass species cannot be readily explained. Ashton and Crafts (1973) indicate that because effects of herbicides on RNA and protein synthesis are complex, interpretation of results is difficult. The ability of Hill reaction inhibitors (i.e. *s*-triazines and tebuthiuron) to increase plant protein synthesis is generally accepted. However, the mechanism by which these plant enhancements occur is not well understood (Reis 1976).

Regardless of date of sampling or application rate, tebuthiuron did not cause significant changes in the digestible organic matter concentrations of the 4 grass species. The following are mean digestible organic matter concentrations (%) for each species: Bahiagrass (60.2 ± 1.1), Bell rhodesgrass (57.7 ± 1.3), green sprangletop (63.5 ± 1.3), and little bluestem (56.5 ± 1.6). Other researchers (Biondini and Pettit 1980) have reported that the 24-hr IVDMD values of little bluestem were increased by 20 to 25% after application of 1 kg/ha. However, the significant source of variation in our experiment was attributed to grass species, primarily the result of greater digestible organic matter concentrations of green sprangletop leaves compared to those of other species.

Variation in water concentrations of Bahiagrass leaves did not follow a consistent pattern relative to herbicide treatment. Water concentrations of Bahiagrass leaves were increased by both herbicide rates at 31 days after application, by the higher rate 93 days after application, by the intermediate rate 120 days after application, but were unaffected at 57 days after treatment (Table 3). Green sprangletop leaf water concentrations were increased by 1.1 kg/ha of tebuthiuron 31 days after application and by 2.2 kg/ha rate 31 and 120 days after herbicide application. Increases in little bluestem leaf water concentration occurred only 31 days after application of 2.2 kg/ha. Water concentrations of Bell rhodesgrass leaves were unaffected, regardless of sampling time or herbicide treatment. Reis and Wert (1972) found uptake of water and nitrate to be highly correlated with simazine-induced increases in total

Table 3. Average waer concentration (%) of Bahiagrass, Bell rhodesgrass, green sprangletop and little bluestem leaves at various times after application of tebuthiuron pellets on May 3, 1980 near College Station, Texas.

Tebuthiuron rate (kg/ha)	Time (days) after tebuthiuron application[1] 31	57	93	120
	Bahiagrass			
0	71 a	59 a	67 a	67 a
1.1	75 b	60 a	70 ab	71 b
2.2	76 b	63 a	71 b	70 ab
	Bell rhodesgrass			
0	72 a	66 a	69 a	69 a
1.1	74 a	66 a	71 a	70 a
2.2	75 a	67 a	72 a	70 a
	Green sprangletop			
0	67 a	58 a	67 a	63 a
1.1	71 b	61 a	69 a	66 ab
2.2	73 b	61 a	69 a	68 b
	Little bluestem			
0	65 a	54 a	63 a	62 a
1.1	68 ab	57 a	63 a	59 a
2.2	69 b	57 a	65 a	61 a

[1]Means within time after application and species followed by the same letter are not significantly different (α=0.05) according to Tukey's w-procedure.

plant nitrogen concentration. Therefore, observed increases in Bahiagrass, green sprangletop, and little bluestem leaf water concentrations may be linked to increases in leaf crude protein concentrations caused by tebuthiuron application.

Although soil water concentrations were relatively low in July 1980, water applications by sprinkler irrigation ensured the soil water concentrations (14% at 0–8 cm, 11% at 15–30 cm) were adequate for growth of grasses in August and September 1980.

Greenhouse Experiment

Based on average mortality at 30 days after application of tebuthiuron at 0.188 and 0.25 ppm, Bell rhodesgrass and green sprangletop seedlings were less susceptible to the herbicide than were seedlings of Bahiagrass or little bluestem (Table 4). This differential susceptibility was also indicated by differences in dry shoot and root weights among surviving plants, especially with those seedlings treated with the higher tebuthiuron rates. Shoot weights of Bahiagrass and little bluestem were significantly reduced when treated with 0.25 ppm of tebuthiuron. Bahiagrass and little bluestem shoot weights were decreased by 90 and 88%, respectively, compared to 11 and 30% reductions in Bell rhodesgrass and green sprangletop, respectively, after application of 0.25 ppm of the herbicide. Root weights of the 4 grasses were reduced, in comparison to untreated plants when treated with 0.125, 0.188, or 0.25 ppm of tebuthiuron. The 0.25-ppm rate of tebuthiuron caused a 97% reduction in Bahiagrass and little bluestem root weights and 70% decrease in root weights of Bell rhodesgrass and green sprangletop.

Regardless of grass species, application of 0.125 ppm or more tebuthiuron increased leaf crude protein concentrations compared to those of untreated plants (Table 4). These results are consistent with those of Biondini and Pettit (1979), who reported tebuthiuron-treated grass seedlings of several species contained an average of 42% more crude protein than did untreated species. However, they also noted that increased crude protein concentrations were accompanied by reductions in heights and weights of treated seedlings. Increased crude protein concentrations of leaves of Bahiagrass and Bell rhodesgrass occurred in our study with no reduction in shoot production when 0.125 ppm of the herbicide was applied. However, shoot production of all species except Bell rhodesgrass was decreased, compared to that of untreated seedlings, after 0.188 or 0.25 ppm of tebuthiuron was applied.

Table 4. Mortality, shoot and root weights, and leaf crude protein concentration of grass seedlings at 30 days after application of various tebuthiuron concentrations to pots in the greenhouse.[1]

Tebuthiuron rate (ppmw)	Species[2] Bahiagrass	Bell rhodesgrass	Green sprangletop	Little bluestem
	Mortality (%)			
0	1 a	1 a	4 ab	1 a
0.063	4 ab	1 a	3 ab	4 ab
0.125	21 bc	7 ab	9 ab	21 bc
0.188	67 de	18 abc	14 abc	54 d
0.25	94 f	32 c	30 c	86 ef
	Shoot wt (mg/plant)			
0	78 de	197 fg	319 i	95 e
0.063	79 de	187 fg	339 i	76 de
0.125	70 cde	176 f	276 h	50 bcd
0.188	34 abc	173 f	267 h	29 ab
0.25	8 a	175 f	221 g	11 ab
	Root wt (mg/plant)			
0	73 d	144 f	234 h	66 d
0.063	51 bcd	100 e	192 g	40 bc
0.125	36 b	63 cd	151 f	19 a
0.188	10 a	5bcd	111 e	7 a
0.25	2 a	43 bc	68 d	2 a
	Crude protein concentration (%)			
0	4.7 a	4.3 a	4.6 a	4.2 a
0.063	6.4 ab	6.7 ab	6.4 ab	6.5 ab
0.125	8.4 bc	9.5 cde	8.1 bc	9.2 de
0.188	11.4 de	11.4 de	9.9 cde	11.1 de
0.25	—[3]	10.3 cde	12.1 e	—

[1]Data represents average of two trials each where the herbicide was applied tc seedlings in the two- and four-leaf growth stage.
[2]Means within an attribute followed by the same letter are not significantly differen (α=0.05) according to Tukey's w-procedure.
[3]Insufficient amount of plant tissue for analysis.

Conclusions

Based on the experimental evidence, we can accept the hypothesis that tebuthiuron applications at rates normally used for brush control can increase crude protein concentrations in some grasses. Measurable differences in crude protein concentrations attributable to tebuthiuron treatment under field conditions occurred only during the growing season of application in the case of the native stand experiment. These tebuthiuron-induced differences in forage crude protein concentrations the year of application may explain the increased crude protein concentrations of cattle diets on tebuthiuron-treated rangeland (Kirby and Stuth 1982).

This research and other studies (Scifres and Mutz 1978, Scifres et al. 1981b) indicate that applications of tebuthiuron for brush control may improve rangeland herbage for grazing by increasing forage production, altering botanical composition, and producing short-term increases in crude protein of some grass species.

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