The effect of osmotic stress on growth characteristics of two spring wheat varieties
by Glenn Richardson

A THESIS Submitted to the Graduate Faculty in partial fulfillment of the requirements for the degree of Master of Science in Soils
Montana State University

Abstract:
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Master of Science in Soils

at

Montana State College

Approved:

Head, Major Department

Chairman, Examining Committee

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Bozeman, Montana
August, 1961

ACKNOWLEDGEMENT

The author wishes to express his sincere appreciation to his major professor, Dr. J. C. Hide, for his guidance and assistance in conducting the research and preparation of the thesis.

Thanks are also due Mr. William Richardson, Jr., who aided in carrying out a portion of the experiment, and to the members of the soils staff at Montana State College who aided in carrying out this study.

The research conducted in connection with this study was partially supported by funds from Regional Project W67.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Two spring wheat crops were grown on drip cultures involving two varieties and two levels of osmotic stress. Growth characteristics and moisture use were determined for four growth periods for each crop. The first crop was grown in the spring and summer so that day lengths were normal for the crop at the location.

An osmotic stress of approximately seven atmospheres reduced both growth and water use for both varieties below that obtained when the stress approximated three quarters of an atmosphere. This was true whether the stress was imposed throughout the entire growth period or when stress was not imposed until tillering stage. The two varieties behaved differently at different stages of growth. The behavior of the two varieties was not always comparable between the two crops which were grown under widely different day lengths and temperatures.

INTRODUCTION

Production of small grain is a major source of income from dryland areas in the western states. Drought frequently limits the yield of these crops. To obtain a maximum crop yield from a limited moisture supply all stages of crop growth should be adjusted to make their maximum contributions to a final yield on a minimum of water. To obtain this objective, it is necessary to know how widely different varieties of a crop differ in their responses to water tensions at each stage of growth. From such information, it should be possible to adjust fertility level to the water level prevailing at seeding time and expected during the growing season. Information on how widely different genotypes of a crop respond to variable conditions at their different stages of growth should assist the plant breeder to develop crop varieties that will make more efficient growth on a limited moisture supply. Since moisture tension is a variable factor influencing plant growth, emphasis was placed on this variable during the study.

REVIEW OF LITERATURE

Water is a major material in which absorption by plant must exceed the amount retained as a plant constituent. Several hundred pounds of water are used for each pound of dry plant material produced. While the dry matter usually makes up less than 30% of the weight of green plants, the water contained in the plants is only a small proportion of that used for transpiration. Water deficiency is the most common factor that limits plant growth. The importance of water deficiency in plant growth has stimulated numerous research workers to study the relationship between water tensions and plant growth. Yet, satisfactory methods have not been devised whereby a specific soil moisture tension can be maintained.

Osmotic solutions have frequently been used to study plasmolysis of cells or to induce tensions which restrict water entry into plants. Numerous studies with saline soils demonstrate that an important influence of salt is to restrict water movement into the plant. The crops grown on soils with excessive salt concentrations demonstrate many symptoms of drought.

In an attempt to evaluate the role of water transmissions as a factor affecting root growth, Gingrich and Russell (1957) compared soil moisture tensions to osmotic stresses. They found root growth to be significantly greater for those roots grown with osmotic stresses.

Water diffuses to the root surface from the soil solution due to moisture gradients within the small volume of soil between roots. Thus, the mean tension exerted by a mass of soil is lower than the stress to which the roots are subjected. The use of osmotic stresses and plant culture equipment should keep the plant root at a fairly constant osmotic stress in order to study plant responses to stresses associated with drought.

The use of various media in solution for the desired osmotic stress has been shown to exhibit side effects to plant growth which are not clearly understood. Harris (1924) has shown the toxicity to be of a chemical nature. Taking into account the side effects of salts or other media, this is the most feasible method of inducing the desired drought effects. Pearson and Bernstein (1959) have particularly noted rice to show characteristics associated with drought when induced stresses were caused by salts in solution. Hayward and Spurr (1941) found that sulfate salts were more inhibitory to flax growth than chloride salts, and were equal in the ability to produce drought effects. Alfalfa was noted by Brown and Hayward (1956) to exhibit droughty appearances when submitted to salinized conditions. Ayers, et al, (1952) showed the yield of wheat was not reduced nearly as much when the drought conditions occurred at a late growth stage as when they occurred at an early growth stage.

Data for comparing wheat yields under dry land conditions where drought is a major factor in production reflect the yields of varieties but does not reflect desirable or undesirable growth characteristics for the interim growth periods. This study was designed to determine if differences occur between varieties at the various stages of growth.

METHODS

The study involved growing two widely different genotypes of spring wheat in nutrient media at two osmotic stresses. Plant growth was evaluated by periodic sampling and water use was determined by the difference between inflow and outflow on sand cultures.

The plants were grown on a coarse sand base in quart oil cans which had been coated with asphalt paint. The nutrients were provided by using a continuous drip culture technique. A stock bottle was used as the source of the appropriate solution for each pot. The solution was continuously syphoned from each stock bottle to the appropriate pot at a rate that exceeded evaporation and transpiration. A drainage hole was provided in each pot around which a small piece of copper tubing had been soldered and drainage was collected from each pot. The difference between inflow and outflow from the pot between sampling periods provided data on evapotranspiration. The pots were wetted with nutrient solution before the start of the experiment so that soil moisture storage was not a variable.

In addition to the planted pots, there were two pots left in each replication without plants growing in them. One pot had plant nutrient solution at high stress dripped on it at a rate in excess of evaporation and the other pot was similarly treated with low stress solution. The amount added to the stock bottle minus the amount of drainage was used to evaluate evaporation during growth periods. The loss attributed to evaporation was subtracted from the total use to give transpiration.

Hoagland's nutrient solution, as described by Eaton (1936), was used as a base solution. The base solution used for low osmotic stress developed an osmotic pressure equivalent to seventy-five hundredths atmospheres. High stress was obtained by adding calcium chloride and sodium chloride to the base solution. Each salt added three and one-quarter atmospheres of osmotic stress; thus, the solution used had a total stress of seven and one-quarter atmospheres of which seventy-five one hundredths atmosphere was developed from the nutrient solution. The proper amount of each salt used for this osmotic stress was determined from Agricultural Handbook 60, Diagnosis and Improvement of Saline and Alkalie Soils (1954).

The varieties used for this study were Thatcher and a short-strawed selection from a cross of (Norin 10 x Brevor 14) X Centana. Throughout the study this variety will be referred to as Japanese Dwarf. Both varieties are spring wheats.

A randomized block type of design was used for the study. This design allowed the planting of sixteen pots per replication plus two blank pots for measurement of evaporation and two pots for any side study. This randomized block was replicated three times. The individual pot location in each replication was selected by use of randomized tables. The pots were numbered one through twenty for each replication. Numbers one through eight were allocated the Thatcher variety; pots nine through sixteen grew the Japanese Dwarf variety. Pots seventeen and eighteen held Japanese Dwarf and Thatcher wheats respectively for side experiments. The last four pots of each of the two main series were used for high osmotic stresses for the two varieties. Pots nineteen and twenty were not planted and were used to measure evaporation.

Each pot was planted with ten seeds of the desired variety to a depth of one-half inch. After germination and emergence the seedlings were thinned back to five plants per pot. This procedure allowed for the uniform spacing and stand per pot.

Sampling dates were selected upon the basis of readily distinguished major plant growth stages. These stages were at tillering, between heading and blossoming, soft dough stage, and maturity. The pots sampled at each sampling date were picked according to a table of randomized numbers for each variety, treatment, and replication. The entire plant population on the appropriate pot was harvested on each sampling date.

Plant measurements on the sampling dates were taken from the harvested pots. These measurements were dry plant material weight, number of heads, number of tillers, and plant height. Also at each sampling date transpiration was determined as explained above.

The method of analysis for the collected data is the one employed by Snedecor (1946) on complex experiments. The shortest significant range mean separation, as used by Duncan (1955), was used to test differences between three or more means.

The study was conducted in two phases. The first phase was performed in a temperature-controlled greenhouse. The major portion of this phase compared growth characteristics between the varieties upon salinized and unsalinized cultures from germination to maturity. This phase was conducted from May 9, 1960, through August 12, 1960. A side experiment was also conducted during this phase, between varieties, when salinization occurred at the tillering stage. This was done in order to determine if the seven atmosphere stress was sufficient to cause drought effects when the varieties were salinized at a later date.

Since there were only enough pots for one sampling date on this side study (Treatment III), sampling occurred at maturity. However, water use measurements could be determined without plant sampling for the growth periods which occurred after tillering and prior to maturity. The data for plant height, final number of tillers, total heads, grain harvested, kernels per spikelet, and average kernal weight were compared with other treatments at maturity. Plant weight for this treatment is not compared with other treatments as there were no data for the interim periods.

The second phase of the study compared the growth characteristics when salinization occurred at tillering. The randomized block design with three replications was used as in the first phase of the study. This crop was grown between August 20, 1960, and January 8, 1961. From August 20 till late September the greenhouse evaporative cooler was in operation. From late September through January the greenhouse was heated. During the last month of the experiment the day length was lengthened to sixteen hours by the use of 250 watt incandescent lamps.

The sampling techniques and sampling stages were the same as those used in the earlier phase. Data analyses were also the same.

Table I - Treatments

Treatment I	Low osmotic stress from germination through harvest.
Treatment II	High osmotic stress from germination through harvest. 200 millequivalents NaCl and $CaCl_2$ on a combined electrical conductivity of 18 millimhos /cm.
Treatment III	Low osmotic stress prior to tillering time and high stress from tillering to maturity.

RESULTS AND DISCUSSION

This experiment consisted of growing two successive crops in the greenhouse on nutrient solutions as listed under methods and outlined in Table I. Since the first crop was grown under summer conditions with prevailing day lengths, and the second crop under fall and early winter conditions, the two phases of the study are discussed separately. The data for each crop are discussed by growth periods.

Comparative measurements of conductivities of the stock nutrient solutions and the drainage from the pots indicated a slight increase in conductivity as the solution passed through the pot. However, this increase did not reach statistically significant levels. Pearson and Bernstein (1959) found that the conductivity of nutrient solutions increased as water was used by caloro rice for evapotranspiration.

Data were analyzed by periods as the study progressed to guide the course of the experiment. Since the data were already available and give the statistical interpretation by periods it was believed to have sufficient value to remain a part of the thesis. In summarizing the data, statistical treatments were used which aided the interpretation of the data for the several periods.

First Crop

<u>First Growth Period</u> (Germination to tillering)

Data obtained during the first growth period for dry weight of plant material and water use are presented in Tables II and III. Preliminary experiments indicated that wheat emergence was delayed about one day

Table II -Gain in plant weight in gram during the first growth period (germination to tillering).

	Treatment I Replications				Treatment II Replications				
Varieties	I	II	III	Total	I	II	III	Total	G. Total
Thatcher	1.137	1.502	1.450	4.089	.181	.220	.127	.528	4.617
Japanese Dwarf	1.552	.800	1.198	3.550	.142	.240	.150	.532	4.082
Total	2.689	2.302	2.648	7.639	.323	.460	.277	1.060	8.699

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	.0040	.066
Varieties	1	.0238	.393
Treatments	1	3.6069	59.52***
Treatments x varieties	1	.0246	.466
Error	6	.0606	
Total	11	4.0266	

***Significant at 0.5%

Table III - Amounts of water used in milliliters through the first growth period.

Varieties	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	G. Total
Thatcher	440	787	654	1881	43	227	110	380	2261
Japanese Dwarf	453	799	321	1573	224	326	158	708	2281
Total	893	1586	975	3454	267	553	268	1088	4542

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	82,633	10.00**
Varieties	1	33	.0039
Treatments	1	466,496	54.46***
Treatments x varieties	1	33,709	4.079*
Error	6	8,263	
Total	11		

***Significant at 0.5%
**Significant at 2.5%
*Significant at 10 % level

under high stress. Consequently, Treatment II was planted a day earlier than Treatment I.

It was obvious throughout the growth period that the high osmotic stress seriously reduced growth. (See Figure 13) At the tillering stage the plants on Treatment I had produced eight to twenty times more growth than Treatment II. This difference was significant at the 0.5% level. There was no significant difference between replications or varieties and no interactions were found.

The total water use was considerably greater for Treatment I than for Treatment II and this difference was significant at the 0.5% level. The water use per gram of dry matter produced was much higher under Treatment II than for Treatment I (Table XX). The same table shows the water requirement to be similar for the two varieties under Treatment I, but Japanese Dwarf used much more water to produce a unit of dry matter than did Thatcher for Treatment II.

<u>Second Growth Period</u> (Tillering to soft dough)

The data for this period of growth are presented in Tables IV and V. The analysis of variance (Table IV) shows the replications to be different at the 2.5% level of significance, while the interaction, varieties, and treatments differed at the 0.5% level.

Averaging the two varieties, plants on Treatment I produced about ten times more plant material than plants on Treatment II. Thatcher produced about five and one-half times as much plant material on Treatment I as on Treatment II. A similar comparison for Japanese Dwarf produced a ratio of about sixteen to one, with Treatment I producing the most plant

Table IV - Gain in plant weight in grams during second growth period (tillering to heading).

Varieties	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	G. Total
Thatcher	.862	.297	1.469	2.628	.141	.169	.154	.464	3.092
Japanese Dwarf	1.735	2.057	2.601	6.393	.066	.050	.261	.377	6.770
Total	2.597	2.354	4.070	9.021	.207	.219	.415	.831	9.862

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	.40	8.0**
Varieties	1	1.13	22.60***
Treatments	1	5.58	116.00***
Treatments x varieties	1	1.23	24.60***
Error	6	.05	
Total	11		

**Significant at 2.5%
***Significant at 0.5% level

Table V – Amounts of water used in milliliters during second growth period.

Varieties	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	Treatment III Replications I	II	III	Total	G. Total
Thatcher	411	437	352	1200	45	54	63	162	215	277	373	865	2227
Japanese Dwarf	642	563	677	1882	13	77	64	154	285	218	337	840	2876
Total	1053	1000	1029	3082	58	131	127	316	500	495	710	1705	5103

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	3,142	1.10
Varieties	1	24,305	8.54**
Treatments	2	318,783	111.9***
Treatments x varieties	2	26,705	9.38**
Error	10	2,847	
Total	17		

***Significant at 0.5%
**Significant at 2.5% level

MSR Mean separation showed a significant difference between all treatment means.

material. Japanese Dwarf produced about three times more plant material than Thatcher on Treatment I. On Treatment II, Thatcher produced about one-fourth more plant material than Japanese Dwarf. The plant yield for Treatment III is not available because this treatment was a minor study, as discussed under methods, and yields were obtained only at maturity.

For Treatment I, water use by Thatcher (Table V) was about eight times more than that for Treatment II, and about a fourth more water was used by Thatcher for Treatment I than Treatment III. Japanese Dwarf for Treatment II used about one-twelfth of the water used by Treatment I. When comparing varieties, Japanese Dwarf used about a third more water on Treatment I than II. The two varieties used about the same amount of water on each of Treatments II and III.

The analysis of variance illustrates the treatment differences as discussed above are significant at the 0.5% level, varieties at the 2.5% level, and the interaction at the 2.5% level of significance.

Third Growth Period (Heading to soft dough)

The gain in weight for this period is presented in Table VI. On Treatment I, Thatcher produced about three times more dry matter than Japanese Dwarf. Thatcher produced about one-fifth more plant material than Japanese Dwarf for Treatment II. Thatcher produced eleven times more dry plant matter on Treatment I than on Treatment II. Japanese Dwarf produced five times more plant material for Treatment I than for Treatment II.

The analysis of variance indicates these differences to be significant for treatments at the 0.5% level, for varieties at the 2.5% level,

Table VI - Gain in plant weight in grams during third growth period (heading to soft dough).

Varieties	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	G. Total
Thatcher	4.181	8.408	3.611	16.200	.227	.538	.811	1.576	17.776
Japanese Dwarf	2.189	.681	2.654	5.524	.553	.289	.339	1.181	6.705
Total	6.370	9.089	6.265	21.724	.780	.827	1.150	2.747	24.481

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	3.09	1.87+
Varieties	1	15.22	9.22**
Treatments	1	28.71	17.40***
Treatments x varieties	1	5.07	3.07+
Error	6	1.65	
Total	11		

+Significant at the 25% level
**Significant at the 2.5% level
***Significant at the 0.5% level

Table VII - Amounts of water used in milliliters during third growth period.

Varieties	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	Treatment III Replications I	II	III	Total	G. Total
Thatcher	1032	1992	1783	4807	70	71	209	350	521	978	744	1691	6848
Japanese Dwarf	808	2145	1759	4712	18	98	205	321	631	383	677	2243	7276
Total	1840	4137	3542	9519	88	169	414	671	1152	1361	1421	3934	14124

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	334,802	3.44*
Treatments	2	3,336,847	34.33***
Varieties	1	10,176	.10
Treatments x varieties	2	21,126	.22
Error	10	97,200	
Total	17		

***Significant at 0.5%
*Significant at 10% level

SSR Mean Separation[1]

Treatment I Treatment III Treatment II

[1]Underline shows nonsignificant difference between treatments

and for both interaction and replications to be significant at the 25% level.

The amounts of water used during this period are given in Table VII. Water use for Treatment III is also shown. Thatcher, on the low stress treatment, used almost fourteen times more water than on Treatment II, and about three times more water than on Treatment III. Japanese Dwarf used about fourteen times more water for Treatment I than for Treatment II and about two times more than on Treatment III. There was no difference between the varieties in water uptake during this period. The analysis of variance shows these differences between treatments and replications to be significant at the 0.5% and 2.5% levels respectively. The SSR mean separation shows the difference for water uptake between Treatments II and III was not significant at the 1% level. Table XX indicates Thatcher was the most efficient user of water for this period, requiring about one-third as much water on Treatment I as Japanese Dwarf to produce a gram of dry matter. For Treatment II about the same amounts of water were required to produce a gram of dry matter for both varieties. Plant height for the period is shown in Table VIII. For Treatment I Thatcher and Japanese Dwarf produced twice the height as for Treatment II. At this time Thatcher was a fifth taller than Japanese Dwarf on both treatments. The analysis of variance showed varieties to be significantly different at the 10% level of significance and the difference in plant height for treatments to be significantly different at the 0.5% level.

Also at this time Thatcher produced about a fifth more tillers than Japanese Dwarf (Table IX) for Treatment I. For Treatment II there was

Table VIII – Plant height at end of third growth period.

Varieties	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	G. Total
Thatcher	27	30	27	84	16	14	12	42	126
Japanese Dwarf	23	21	24	68	13	10	11	34	102
Total	50	51	51	152	29	24	23	76	228

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	2.0	.016
Varieties	1	48.0	3.84*
Treatments	1	481.0	38.48***
Treatments x varieties	1	8.0	.64
Error	6	12.5	
Total	11		

*Significant at 10%
***Significant at 0.5% level

Table IX - Number of tillers at end of third growth period.

Varieties	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	G. Total
Thatcher	20	28	17	65	6	5	3	14	79
Japanese Dwarf	17	17	18	52	4	4	5	13	65
Total	37	45	35	117	10	9	8	29	144

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	8	.92
Varieties	1	16	1.84*
Treatments	1	675	77.59***
Treatments x Varieties	1	15	1.72
Error	6	8.7	
Total	11		

*Significant at 10%
***Significant at 0.5% level

no difference between varieties. On Treatment II both varieties produced only about a fourth as many tillers as on Treatment I. The analysis of variance shows varieties and treatments to differ at the 10% and 0.5% levels of significance respectively.

Table X shows there is very little difference between the number of heads produced at this time between varieties at either salt level. Treatment II at this stage of growth reduced the number of heads produced to about a third as compared to Treatment I, and the difference is significant at the 0.5% level.

Fourth Growth Period (Soft dough to maturity)

Differences between the two varieties in dry matter production did not reach significant levels on any of the treatments in this growth period (Table XI). The difference between treatments shows Thatcher produced about twenty-five times more plant material for Treatment I than for Treatment II. Treatment III produced intermediate yields although the data are not presented. Japanese Dwarf produced forty-six times more plant material on Treatment I than on Treatment II, and again Treatment III yields were intermediate.

The analysis of variance indicates the treatments were significantly different at the 0.5% level. An SSR mean separation indicates these differences to be significant between each treatment at the 1% level.

Water uptake could not be measured during this period for Treatment II. A salty crust appeared on the cropped pots, preventing normal evaporation. Table XII includes only data gathered for Treatments I and

Table X - Number of heads at end of third growth period.

	Treatment I				Treatment II				
	Replications				Replications				
Varieties	I	II	III	Total	I	II	III	Total	G. Total
Thatcher	9	17	11	37	4	3	3	10	47
Japanese Dwarf	7	10	13	30	4	4	4	12	42
Total	16	27	24	67	8	7	7	22	89

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	6.5	.97
Varieties	1	2.0	.30
Treatments	1	169.0	25.22***
Treatments x varieties	1	7.0	1.44
Error	6	6.7	
Total	11		

***Significant at 0.5% level

Table XI - Gain in plant weight in grams during fourth growth period (soft dough to maturity).

Variety	Treatment I Replications I	II	III	Total	Treatment II Replications I	II	III	Total	G. Total
Thatcher	10.120	7.893	9.678	27.691	-.080	.604	.626	1.150	28.841
Japanese Dwarf	5.324	11.562	11.547	28.433	.071	.097	.442	.610	29.043
Total	15.444	19.445	21.225	56.124	-.009	.701	1.068	1.760	57.884

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	3.079	1.02
Varieties	1	.003	.001
Treatments	1	246.287	81.33***
Treatments x varieties	1	3.867	1.28
Error	6	3.028	
Total	11		

***Significant at 0.5% level

Table XII - Amounts of water used in milliliters during the fourth growth period.

Varieties	Treatment I Replications I	II	III	Total	Treatment III Replications I	II	III	Total	G. Total
Thatcher	2623	2291	2860	7777	202	228	228	658	8435
Japanese Dwarf	1223	863	1608	3694	770	366	170	1306	5000
Total	3849	3154	4468	11471	972	594	398	1964	13435

Analysis of variance

Variation due to	d.f.	Mean Square	Cal. F
Replications	2	100,137	1.40
Varieties	1	983,269	13.73**
Treatments	1	7,531,921	113.19***
Treatments x varieties	1	1,865,169	26.05***
Error	6	71,602	
Total	11		

**Significant at 2.5%
***Significant at 0.5% level

III. During this period Thatcher under Treatment I required almost twice as much water as Japanese Dwarf, but on Treatment III it used only half as much as the dwarf variety. Thatcher under Treatment I needed twelve times the amount of water it used for Treatment III, while Japanese Dwarf needed about three times more water for Treatment I than for Treatment III. Table XX indicates Thatcher on Treatment I needed less water than Japanese Dwarf for a unit of dry matter production.

Treatments II and III reduced plant height of Thatcher for this period by two fifths and a sixth respectively over Treatment I. The Japanese Dwarf height was reduced by one half on Treatment II below those of Treatments I and III. The statistical differences between treatments were significant at the 10% level. There was no statistical difference between varieties. The SSR mean separation shows this difference to occur between all treatments. This data is shown in Table XIII.

Table XIV presents the total number of tillers and analysis of variance for tillers at harvest. Thatcher produced the most tillers on all treatments. For Treatment I, Thatcher produced about six times more tillers than Treatment II and about three times more than Treatment III. Japanese Dwarf produced about four-fifths as many tillers as Thatcher for Treatment I, a slight reduction on Treatment II, and about half as many tillers on Treatment III. The dwarf variety produced a sixth as many tillers on Treatment II and a third as many on Treatment III as on Treatment I. The analysis of variance indicates that treatment and variety differences on total tillers were significant at the 0.5% level.