Res. Bull. Obihiro Univ., **7** (1971): 295~310.

The Promoting Effect and Utilization of Alcohol on Legume and Grass Forage Plants

I. The Growth and Production of Alfalfa and Orchard Grass

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Received May 31, 1971

草類に対するアルコール施用の効果

I. アルファルファならびにオーチャードグラスの生育と生産について

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WILDE (1964) was the first to report that allyl alcohol increased the growth of tree seedlings. Further studies by CHANG and CHUNG (1969) showed that in an experiment made at the College of Education, Seoul National University, silt loam soils, which are treated seven times with methyl and ethyl alcohol at rates ranging from 0.5 cc to 2.5 cc per pot, definitely increased the growth rate of radish and cabbage seedlings germinated from sterilized seeds. However, it was not until alcohol was used that the effect was seen throughout the entire growing season of plants were noted.

This paper describes the effects of methyl, ethyl, and buthyl alcohol on the growth, production, nodulation, and root development in a green-house of Obihiro Zootechnical University in Obihiro.

Experimental Methods

Green-house Experiments

Volcanic ash soils from the Attached Farm of the Obihiro Zootechnical

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University were filled in plastic Wagner pots and were used in green-house experiments. A basal application of fertilizer which supplied the equivalent of 4 kg of N, 12 kg of P, 10 kg of K and 2 kg of Ca $(H_2PO_4)_2H_2O$ per are was made at sowing, and soil moisture was maintained during the experiment, between field capacity and 50% available moisture. Inoculative plots of alfalfa (Du Puits) were inoculated with a suitable strain of rhizobium after sowing.

Methyl, ethyl, and buthyl alcohols were applied factorially to alfalfa, 7 days after germinating and to orchard grass (Colby's), 10 days after transplanting. The levels employed were 0, 0.1, 0.15, 0.2, 0.25, 0.5, 1.25, 2.5, 5.0, 7.5, 10, and 12.5 liter of methyl, ethyl, and buthyl alcohol per are but the plots of orchard grass were applied to 0, 0.25, 2.5 and 5.0 liters. The treatments were carried out at intervals of 7 or 10 days.

The experiment was run in a green-house maintained between 18°C and 28°C from November 13, 1970 until February 13, 1971. All the plots were harvested twice and the plant tops and roots were dried at 60°C and ground in a stainless steel mill for chemical analyses.

Chemical Analyses

The determination of total nitrogen was carried out by the micro Kjeldahl method and the amount of crude protein was calculated by multiplying N by 6.25. Crude fat was estimated by the usual method. Chemical analyses of lignin and cellulose were determinded by the methods of CRAMPTON *et al.* (1938), with other analyses following procedures recommended by the Association of Official Agricultural Chemists in Japan.

Results and Discussion

1. Growth-promoting Effects

In the first week, non-alcohol-treated alfalfa and orchard grass grew up slowly. During this period no differences were evident between the treated and non-treated, nor were there any morphological signs which were observed previously in alfalfa and orchard grass grown on the Attached Farm of Obihiro Zootechnical University.

The promoting effects developed in all treated plots of alfalfa and orchard grass where alcohol was applied for more than three times. Figure 1 shows the effect of alcohol on the height of alfalfa after 51 days of treatment.

Growth-promoting effects of each treatment of the alfalfa plot during 28 days after the first cutting were given in Fig. 2. The plant height of alfalfa in the first harvest (Fig. 1) was lower than in the second harvest (Fig. 2).

Effects of alcohol on the growth of orchard grass in the first and second harvest were as shown in Figs. 3 and 4 respectively.

As shown in Figs. 1, 2, 3 and 4, the plant height of alcohol-treated alfalfa and orchard grass was higher than that of control which was non-alcohol-treated.

Fig. 1. Effects of methyl, ethyl and buthyl alcohol on the growth of alfalfa at various levels after 51 days of treatment

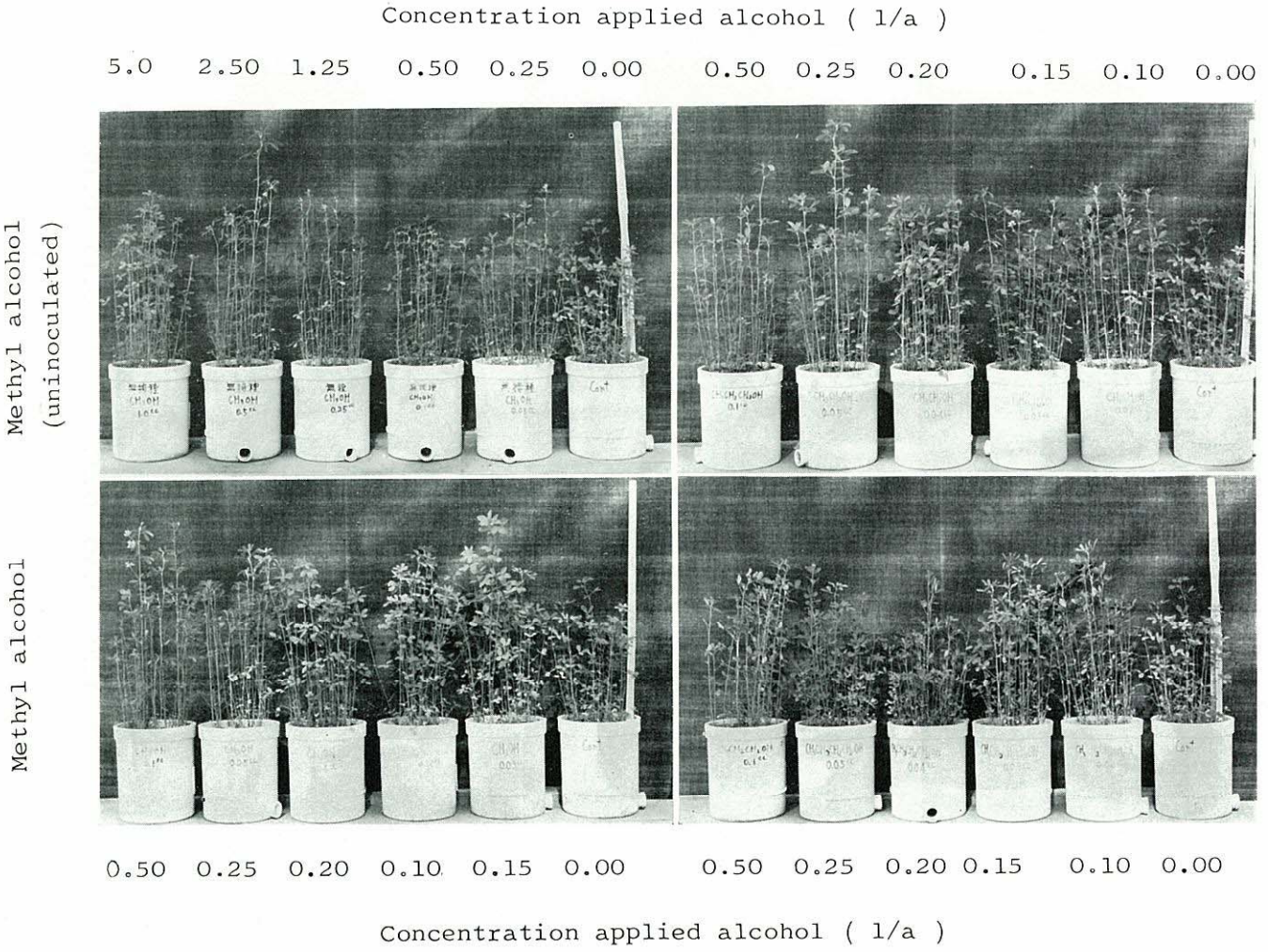

Fig. 2. Effects of methyl, ethyl and buthyl alcohol on the growth of alfalfa at various levels in the second harvest

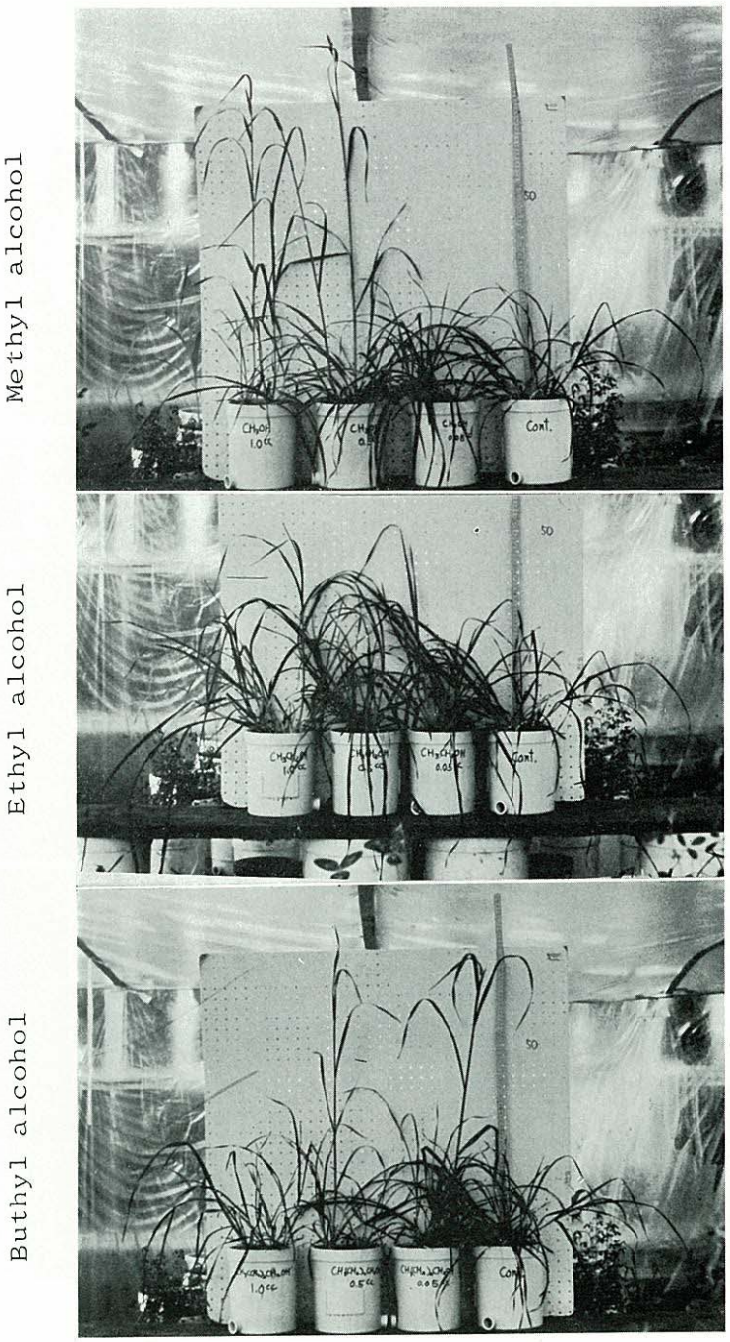

Fig. 3. Effects of methyl, ethyl and buthyl alcohol on the growth of orchard grass at various levels in the first harvest

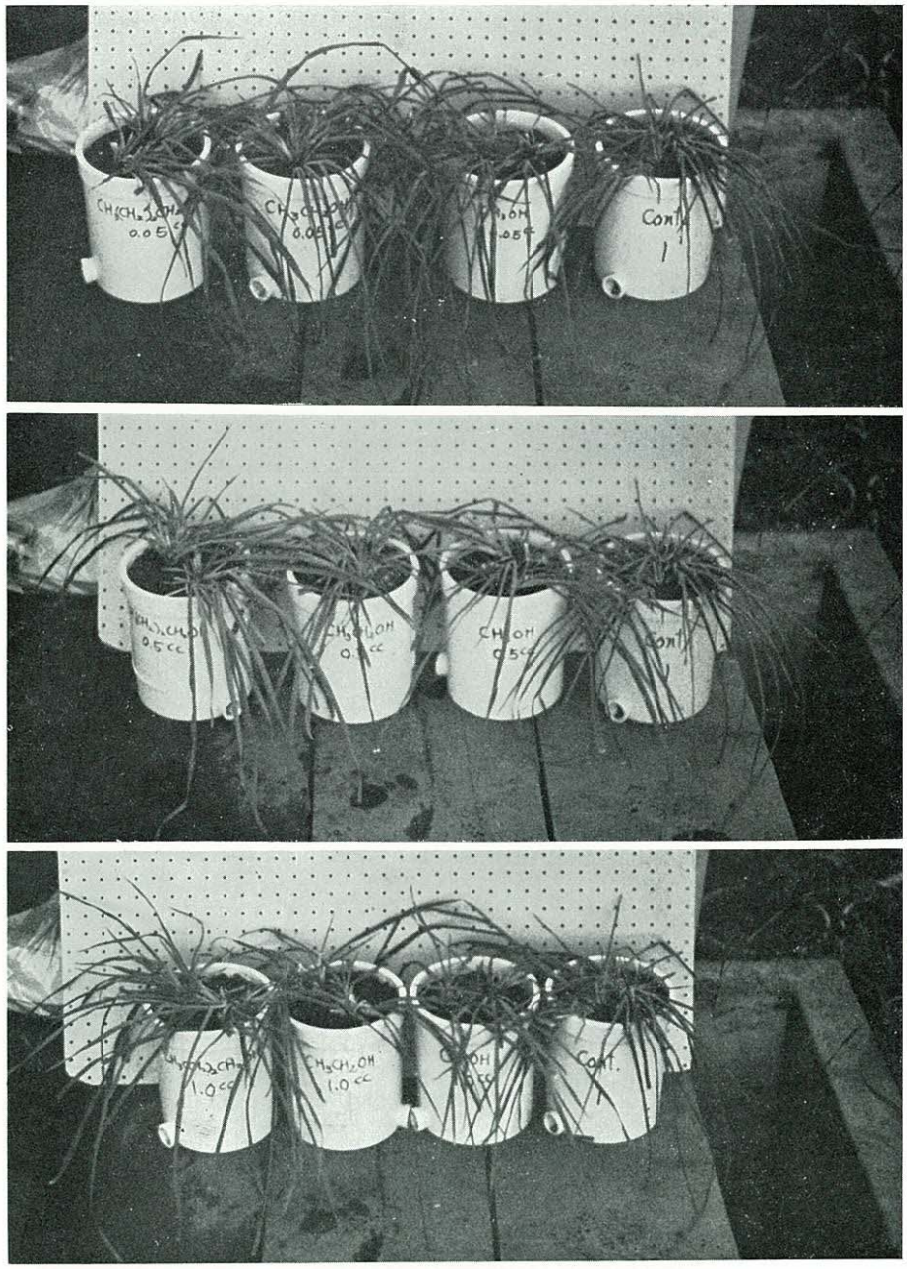

Fig. 4. Effects of methyl, ethyl and buthyl alcohol on the growth of orchard grass at various levels in the second harvest

The estimates of the plant height in each treatment of the plots are shown in Tables 1 and 2. The differences between each treatment and control were highly significant at more than 0.01 levels. Comparing the plant height among the treatments of each plot, there were no significant differences but the treatments of 7.50, 10.00 and 12.50 liter per are of buthyl alcohol were depressed. As indicated in Table 2, the plant height of alcohol-treated orchard grass in the second harvest were lower than that of control or the same. However, the treatments of 0.25 liter per are of methyl, ethyl and buthyl alcohols were higher than in the control.

Table 1. Plant height of alfalfa in the first and second harvest (cm, average)

Concentration applied alcohol (l/a)	Methyl alcohol (uninoculated)	Methyl alcohol	Ethyl alcohol	Buthyl alcohol
The first harvest				
0.00	24.0			
0.10		29.9	33.2	32.5
0.15		32.8	33.8	32.9
0.20		33.3	40.2	32.0
0.25	28.5	32.4	32.5	29.8
0.50	27.8	33.9	36.6	29.7
1.25	34.4	33.2	33.1	34.0
2.50	33.3	34.5	31.8	30.1
5.00	32.9	33.8	31.7	30.7
7.50	32.6	33.2	30.8	29.9
10.00	31.4	33.4	31.9	23.5
12.50	34.3	32.8	34.8	19.6
The second harvest				
0.00	27.0			
0.10		40.4	40.0	35.7
0.15		39.7	38.2	37.4
0.20		38.9	39.3	35.0
0.25	34.5	37.4	44.2	34.0
0.50	30.7	40.2	39.0	36.5
1.25	37.6	41.2	40.4	35.0
2.50	38.2	41.0	37.6	37.2
5.00	36.0	36.0	40.2	36.2
7.50	39.5	33.7	33.8	36.3
10.00	33.2	35.7	35.0	32.0
12.50	30.0	39.6	31.6	24.5

Table 2. Plant height of orchard grass in the first and second harvest (cm, average)

Concentration applied alcohol (l/a)	Methyl alcohol	Ethyl alcohol	Buthyl alcohol
The first harvest			
0.00	53.7		
0.25	60.0	57.8	74.4
2.50	78.4	61.2	61.3
5.00	67.6	57.4	48.2
The second harvest			
0.00	43.8		
0.25	57.0	49.0	45.5
2.50	44.0	37.6	41.7
5.00	42.3	33.0	40.3

Comparing the color of the leaves between the treatments of alcohol and control, the former had a yellowish-green color and the latter had a blue-green color.

In the case of orchard grass, treatment with 2.50 and 5.00 liter per are of methyl alcohol and 0.25 and 2.50 liter per are of buthyl alcohol brought about earing but no effects of the treatment with ethyl alcohol was observed. The results observed in those treatments suggest that methyl and buthyl alcohols promote earing and flowering of grasses. The flower promoting effect in the plot of alfalfa could not observed in this experiment (Figs. 1, 2, 3 and 4).

Analyses of chlorophyll, carotene, organic carbon and total N in comparative studies between alcohol treatment and non-alcohol treatment are carried out in the next paper.

2. Production-promoting Effects

Figure 5 is a diagram of green yields in the first and second cutting obtained from this experiment. The relationship appears a curve and shows a marked increase in low concentration applied methyl, ethyl and buthyl alcohol. As an indication of the difference between this relationship for different concentrations, each plateau of the curves of the green weights may be distinguished from the available data (Fig. 5).

As shown in Fig. 5, the green yields of the inoculative treatments of methyl alcohol of alfalfa were higher than those of the uninoculative treatments. There was a highly significant relationship between these two treatments. The results suggest that the yields in plots of alfalfa were affected by rhizobium inoculation. Moreover, those of the uninoculative treatments of methyl alcohol were higher than those of the control uninoculated and non-alcohol-treated plots. Therefore,

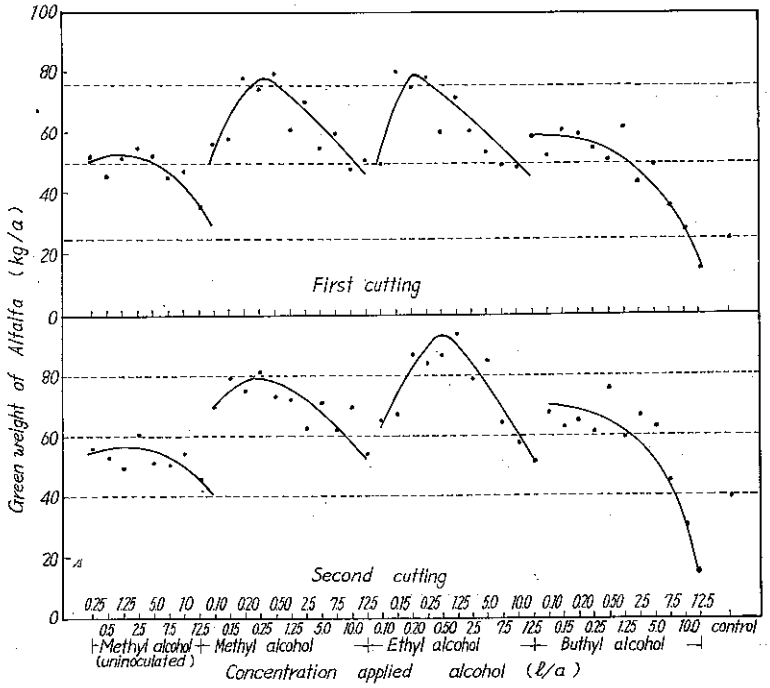

Fig. 5. Green yields of alfalfa alcohol-treated in the first and second cutting

it may be stated that alcohol promotes the growth of alfalfa and rhizobium.

The green yields of the inoculative treatments of methyl and ethyl alcohol were significantly increased as compared with the uninoculative treatments of methyl alcohol and the inoculative treatment of buthyl alcohol.

Generally speaking, as the concentration of alcohol rises to 0.25 liter per are, the green yield increases remarkably and shows a maximum value at 0.25 liter per are. The green yields of the inoculative treatments from 0.1 to 10.0 liter per are of methyl and ethyl alcohol showed values more than twice of those of the control in the first harvest and more than one and a halffold in the second harvest. The highest yield of all the treatments were three fold of the control in the first harvest and two fold in the second harvest. The yields of all treatments except for 10.0 and 12.5 liter per are of buthyl alcohol were higher than those of the control and showed a highly significant difference at 0.01 level and over.

The water content and dry weight of the green yield of alfalfa were given in Table 3 and Fig. 6 respectively.

The water content of alfalfa in all alcohol treatments was higher than that in the control in the first and second cutting. According to the result of "t" test, the water content showed a significant difference between the alcohol-treated plots and the control.

Table 3. The water content of the green yield of alfalfa

Concentration applied alcohol (l/a)	Methyl alcohol (uninoculated) (%)	Methyl alcohol (%)	Ethyl alcohol (%)	Buthyl alcohol (%)
The first harvest				
0.00	75.8			
0.10		81.4	79.3	81.7
0.15		82.8	81.1	80.2
0.20		82.2	81.6	78.6
0.25	79.0	82.3	82.0	77.4
0.50	76.4	81.0	84.2	83.9
1.25	82.5	81.4	83.9	78.3
2.50	82.6	82.6	82.5	80.0
5.00	82.4	74.1	79.1	80.4
7.50	81.4	77.4	82.4	79.2
10.00	81.3	77.5	81.4	80.9
12.50	79.3	89.7	80.5	80.4
The second harvest				
0.00	71.1			
0.10		77.3	74.7	72.1
0.15		79.2	76.2	73.7
0.20		74.8	75.4	73.2
0.25	75.5	77.5	73.0	73.3
0.50	76.3	76.2	75.3	76.8
1.25	76.4	75.3	78.2	76.9
2.50	78.9	78.8	78.0	75.4
5.00	79.1	81.9	81.1	75.7
7.50	75.4	71.6	75.3	78.9
10.00	78.1	77.4	73.9	78.5
12.50	74.2	77.3	73.3	78.7

The effect of methyl, ethyl and buthyl alcohol on the dry matter yield of alfalfa in the first cutting (Fig. 6) was highly significant. An application of 0.15 to 5.00 liter per are of methyl and ethyl alcohol in the inoculative treatments increased the yield by 50% and over, while the 0.15 to 1.25 liter per are increased the yield by 100% and over respectively. Where rhizobium was not inoculated, methyl alcohol from 0.25 to 5.0 liter per are promoted the yield by 30% and over. An application of 0.15 to 5.00 liter per are of buthyl alcohol increased the yield by 40% and over but 10.00 and 12.50 liter per are depressed alfalfa yield by less than 10%. In the second cutting, the effect of alcohol on the dry matter yield showed the same tendency as in the first cutting. The

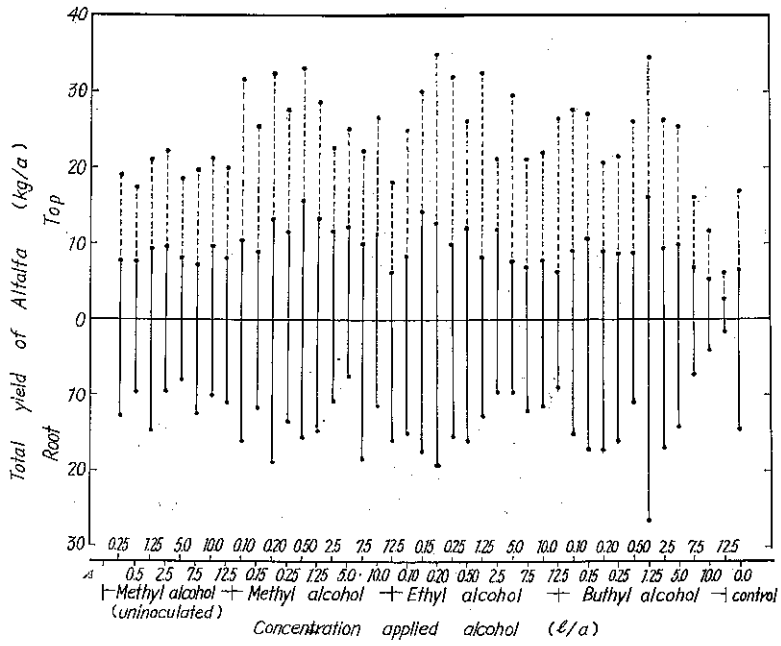

Fig. 6. The effect of alcohol on the dry matter yield of top and root of alfalfa in the first and second cutting

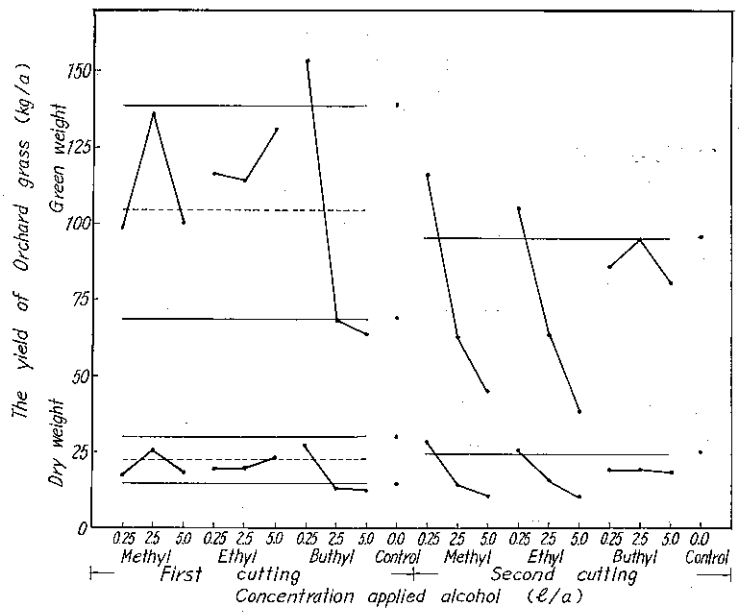

Fig. 7. The green and dry matter yields of alcohol-treated orchard grass in the first and second cutting

total alfalfa yield was calculated by the total of the first and second harvests. Therefore, the promoting effect of alcohol on the total yield of alfalfa also showed the same tendency as in the first and second cuttiug (Fig. 6). It is certain that one part of the amount of an increase according to the promoting effect on the green yield is water which alfalfa contains and other part is dry matter of alfalfa. Thus, the effect of alcohol on some chemical compositions in the two experimental plants was discussed in the next paper.

The yield of orchard grass was affected by methyl, ethyl and buthyl alcohol likewise. The data of green and dry matter yields of alcohol-treated orchard grass in the first and second cutting are indicated in Fig. 7.

In the first cutting, an application of 0.25 to 5.00 liter per are of methyl and ethyl alcohol increased the green yield of orchard grass by more than 40%, while 2.50 liter per are of methyl, 5.00 liter per are of ethyl and 0.05 liter per are of buthyl alcohol increased the green yield by 80% and over respectively. However, 2.5 and 5.0 liter per are of buthyl alcohol depressed orchard grass yield as compared with the control (Fig. 7).

Dry matter yields of alcohol-treated orchard grass were also increased by more than 20%. An application of 2.5 liter per are of methyl, 5.0 liter per are of ethyl and 0.25 liter per are of buthyl alcohol promoted dry matter yields of orchard grass by more than 50% respectively.

Table 4 is a summary of the water content of alcohol-treated orchard grass. As shown in Table 4, the water content of all the treatments with methyl, ethyl and buthyl alcohol were higher than those of the controls.

The above results show that alcohol treated in the plot of orchard grass promotes absorption of water and dry matter yield respectively.

Table 4. The water content of the green yield of orchard grass

Concentration applied alcohol (l/a)	Methyl alcohol (%)	Ethyl alcohol (%)	Buthyl alcohol (%)
The first harvest			
0.00	79.12		
0.25	82.31	82.35	78.58
2.50	81.30	82.50	76.69
5.00	82.05	82.38	80.38
The second harvest			
0.00	75.13		
0.25	75.92	76.02	77.57
2.50	78.08	75.67	79.54
5.00	76.29	72.77	76.78

In the second cutting, the green and dry matter yield of alcohol-treated orchard grass was far less than that of the control. This result suggests that in much the same manner as in the non-application of nitrogen fertilizer after the first cutting, the yield of orchard grass in the second cutting did not increase. Moreover, it is clear that in order to increase the green and dry matter yield of a plant, the application of alcohol and nitrogen must be controled.

The range of concentration of alcohol which promotes the growth and production of plants is in agreement with the results of CHANG and CHUNG (1969).

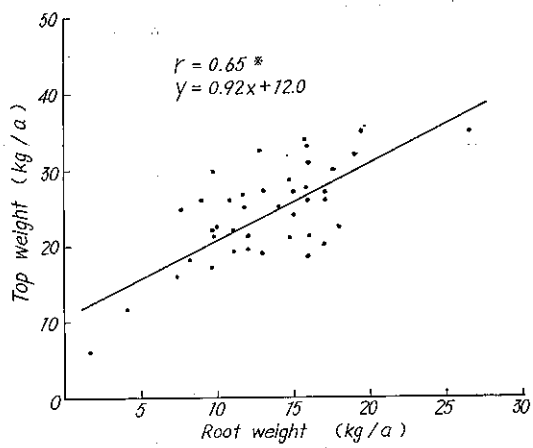

Fig. 8. Relationship between top and root weight of alcohol-treated alfalfa

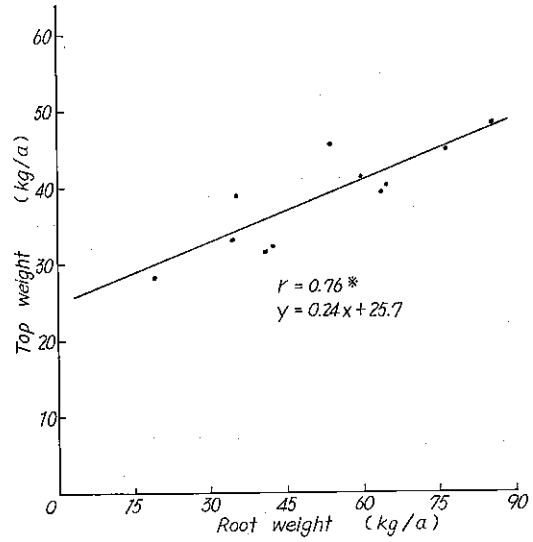

Fig. 9. Relationship between top and root weight of alcohol-treated orchard grass

3. Effects of alcohol on the root

The dry matter yields of roots, shown in Figs. 6, 8, and 9, generally increased with top weights. The top weight of alfalfa and orchard grass in the first and second cutting were correlated very highly and positively ($r=0.65$ and $r=0.76$) with the root weight.

However, the ratio of top weight of the first and second cutting, and root weight of alcohol-treated alfalfa and orchard grass was higher than that of the control respectively (Tables 5 and 6).

As indicated in Table 6, the higher the dry matter yield of top of alcohol-treated orchard grass, the higher the ratio of top and root weight is. This result seems to indicate that the top growth of a plant is affected principally by the application of alcohol. However, the alcohol treatment at the high level of root weight has a higher yield than the alcohol treatment at low level (Figs. 8 and 9).

Table 5. The ratio of top weight in the first and second cutting, and root weight of alcohol-treated alfalfa

Concentration applied alcohol (1/a)	Methyl alcohol (uninoculated)	Methyl alcohol	Ethyl alcohol	Buthyl alcohol
0.00	1.18			
0.10		1.92	1.65	1.84
0.15		2.15	1.71	1.60
0.20		1.69	1.81	1.19
0.25	1.47	2.04	2.19	1.32
0.50	1.77	2.08	1.64	2.42
1.25	1.43	1.95	2.55	1.30
2.50	2.29	2.13	2.23	1.55
5.00	2.26	3.28	3.06	1.99
7.50	1.56	1.18	1.74	2.21
10.00	2.04	2.29	1.96	2.95
12.50	1.76	1.14	2.95	3.75

Table 6. The ratio of top weight in the first and second cutting, and root weight of alcohol-treated orchard grass

Concentration applied alcohol (1/a)	Methyl alcohol	Ethyl alcohol	Buthyl alcohol
0.00	0.66		
0.25	0.56	0.52	0.86
2.50	1.10	0.62	0.73
5.00	1.32	0.95	0.72

4. Effects of alcohol on nodulation

The effects of the various treatments of alcohol on the nodulation of alfalfa were highly significant. The mean nodule scores and weight are shown in Table 7.

Two types of nodules were observed namely, pink nodules and white ones. It was thought that the small white nodules were formed by wild rhizobium. Alfalfa from the uninoculated treatments of methyl alcohol had small nodules on the low parts of root only but the inoculated treatments of methyl, ethyl and buthyl alcohol showed nodules on the upper and low parts of the root respectively. Only healthy nodules were scored.

The nodule scores of the uninoculated treatments of methyl alcohol were higher than those of the uninoculated control and non-alcohol-treated. This suggests that an application of alcohol promotes the nodule scores and increases the forage production significantly.

Table 7. Mean nodule scores and weight of alcohol-treated alfalfa

Concentration applied alcohol (l/a)	Methyl alcohol (uninoculated) (g/pot)	Methyl alcohol (g/pot)	Ethyl alcohol (g/pot)	Buthyl alcohol (g/pot)
0.00	0.028 (127)			
0.15		0.234 (960)	0.206 (362)	0.209 (181)
0.50	0.082 (161)	0.221 (450)	0.210 (170)	0.326 (450)
5.00	0.081 (154)	0.188 (440)	0.177 (194)	0.163 (280)
10.00	0.147 (181)	0.463 (275)	0.436 (375)	0.177 (4)

* ( ); Mean nodule scores per pot

Summary

The effects of methyl, ethyl and buthyl alcohol on the growth and yield of alfalfa and orchard grass were investigated. The experimental results are summarized as follows.

1. The plant height of alcohol-treated alfalfa and orchard grass was higher than that of the uninoculated control and non-alcohol-treated. There was no significant difference among the alcohol treatments.

2. Comparing the color of leaves of plants between the alcohol treatments and control, the former had a yellow-green color and the latter had a blue-green color.

3. In the first and second cutting, application of 0.15 to 1.25 liter per are of methyl and ethyl alcohol increased the yield of alfalfa by about 100% as compared with that of control, while 0.10 to 5.00 liter per are of buthyl alcohol increased the yield by about 50%. However, application of 7.50, 10.00 and 12.50 liter per are of buthyl alcohol decreased the yield.

4. In the case of orchard grass, administration of 2.5 liter per are of methyl alcohol, 5.0 liter per are of ethyl alcohol and 0.25 liter per are of buthyl alcohol promoted yields by more than 50% respectively.

5. Treatment with 2.50 and 5.00 liter per are of methyl alcohol and 0.25 and 2.50 liter per are of buthyl alcohol in a plot of orchard grass brought about earing but no effects of the treatment with ethyl alcohol was observed.

6. The ratio of top weight of the first and second cutting, and root weight of alcohol-treated alfalfa and orchard grass was higher than that of control.

7. Application of alcohol in an alfalfa plot promoted both the nodule scores and weight.

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摘　　要

アルファルファならびにオーチャードグラスの生育と生産に対するメチル，エチルおよび，ブチルの各アルコール施用の効果について検討したが，この結果を要約するとつぎのごとくである。

1. 各アルコール施用区は対照区に比較して草丈は高くなるが，施用量間には差が認められない。

2. 各アルコール施用により葉色の緑度が減退する。

3. アルファルファの草量に対して，メチルおよびエチルアルコール濃度が1アールあたり0.15～1.25 ℓ 施用では100% の増収がみられ，ブチルアルコールでは1アールあたり0.1～5.0 ℓ で約50% 増収したが，1アールあたり7.5～12.5 ℓ 施用では収量が減少した。

4. オーチャードグラスでは，1アールあたりメチルアルコール2.5 ℓ，エチルアルコール5 ℓ，ブチルアルコール0.25 ℓ 施用で，対照区に比較して50% 以上の増収が認められる。

5. メチルアルコールの1アールあたり5 ℓ，ブチルアルコールの1アールあたり0.25，2.5 ℓ 施用で出穂が促進される。

6. 各アルコール施用により草類の T/R 率が増加される。

7. アルファルファでは根粒菌の着生がアルコール施用の場合良好で，数量と重量の増加が認められる。