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THE EFFECT OF DD ON GRASS AND CLOVER IN STERILIZED SOIL[1]

G. KOOISTRA[2]

Institute of Phytopathological Research (I.P.O.), Wageningen[3]

A greenhouse experiment in steam-sterilized potting soil and thus without eelworms, proved that DD can protect clover from disappearing in a perennial ryegrass and white clover culture. At the normal dosage or at half the normal dosage, DD produces clover yields 2.1 and 2.6 times as large as the yield of the blank pots. These results suggest that DD enhances the ability of the white clover to compete with perennial ryegrass even when there are no parasites.

INTRODUCTION

For several years research has been going on into the reason why, especially on sandy soil, there is a decline in white clover growth in grassland and leys. When a grass-clover mixture of 30% clover and 70% grass is sown, this may in the course of a few years result in a growth of almost exclusively grass with only a small percentage of clover. The question of whether parasites were responsible was soon raised. Because of the beneficial effect that pre-sowing treatment of the soil with DD[4] and other nematicides has on the growth of grass and clover, OOSTENBRINK (1954) concluded that poor grass and clover growth is due to nematodes such as *Pratylenchus crenatus* Loof, *Tylenchorhynchus dubius* Bütschli and *Heterodera trifolii* Goffart. According to ENNIK *et al* (1962), after disinfection of the soil growth was better than on untreated soil, the improvement being short-lived in the case of grass but very longterm in the case of white clover. This effect of DD 4 might be due to the killing of harmful nematodes. It also occurred, however, where there were only a few nematodes in the soil and certainly not enough to cause damage. In this case it is quite possible that the few nematodes create points of entry for parasitic soil fungi.

In order to study the effect of a DD treatment of soil that does not contain nematodes on the growth of grass and white clover, a pot experiment was carried out with sterilized soil.

MATERIALS AND METHOD

Potting soil consisting of a mixture of two parts sandy soil, one part leaf mould and one part old farmyard manure was steam-sterilized for $\frac{3}{4}$ hour at 105°C and $1\frac{1}{2}$ atm. pressure. Twenty 6-litre Mitscherlich pots were filled with this sterilized soil. In groups of four the pots were then treated with respectively 3 ml, 1.5 ml, 0.7 ml and 0.35 ml DD per pot ($2\times$, $1\times$, $\frac{1}{2}\times$ and $\frac{1}{4}\times$ the normal dosage of 50 ml DD per m^2 = per 200 kg of soil). Four pots were not treated. The DD was injected into the soil immediately above the bottom of the pots.

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[2] Seconded from Woodstock Agricultural Research Centre, "Shell" Research Ltd.

[3] Present adress: Laboratory for Flowerbulb Research, Lisse.

[4] Shell-DD, a mixture of dichloropropane and dichloropropene.

For three weeks the soil was then kept as far as possible out of contact with the atmosphere by a water seal. Next, at the end of February 1962, 5 young white clover plants and 5 perennial ryegrass plants, all about 3 weeks old, were planted in each Mitscherlich pot.

The plants were trimmed to about 1 cm above the soil 24, 52, 76 and 100 days after planting and the cuttings dried. The dry weights of the grass and clover were each determined separately and per pot.

RESULTS

Table 1 presents the yields per treatment in grams of dry material taking the average of four replicates of grass and white clover.

Table 2 gives the quotients of (a) total yields of grass and white clover up to a given date and (b) yields at the first cutting. With white clover, there is in each case little difference on the same crop dates between the quotients for different treatments. With the ryegrass, the differences are somewhat greater.

This relatively good correlation in crop development after 24, 52, 76 and 100 days prompted calculations, from both crops given in Table 1, of the interrelationship between the blank and the DD treatments. These ratios for grass and clover are presented in the last columns of Table 2.

Fig. 1 shows the position 71 days after the start of the test; it clearly illustrates the effect of the different dosages.

DISCUSSION

The tables show that application of the normal dosage and of half the normal dosage of DD result in a considerable improvement in the amount of clover (by comparison with the blank); half the normal dosage is even slightly better than the full (normal) dosage. This is presumably the result of greenhouse conditions: the constant temperature and humidity of the atmosphere make it possible to give a somewhat smaller dosage than in the open. According to Table 2, the beneficial effect of DD with half the normal dosage produces 2.6 times the yield and with the normal dosage 2.1 times the yield of the blank. Pots treated with DD dosages of twice and of a quarter of the normal quantity were almost the same as the blank. In the case of the double dosage this result is obviously caused by phytotoxicity; in the case of the quarter dosage, by the fact that the amount of DD injected was in this case too small to have any effect.

The grass yield with half the normal dosage was only better than the other treatments on 23rd March and 20th April. On subsequent crop dates this effect has disappeared. It can be clearly seen that after 100 days the grass yield is already on the decline while that of the clover is then still on the increase. In the case of grass, the development of nitrogen deficiency might be responsible for this; clover makes its own provision for its nitrogen requirements by its root-knots.

The above results had become more or less definitive by the end of 24 days (23rd March); they were found to be significant in the variance analysis[5], especially on 20th April and 14th May. On 7th June they were rendered less

[5] Carried out by Mr. C. A. VAN DEN ANKER, I.P.O., Wageningen.

TABLE 1. Treatment of grass-clover culture in sterile potting soil with DD in four different dosages. Dry weight in grams of grass and clover. Each figure is the average weight of four pots.

	Grass								Clover							
Data at which the dry weights were determined	Per treatment				Cumulative				Per treatment				Cumulative			
	23/3	20/4	14/5	7/6	23/3	20/4	14/5	7/6	23/3	20/4	14/5	7/6	23/3	20/4	14/5	7/6
Intervals in days	24	28	24	24	24	52	76	100	24	28	24	24	24	52	76	100
Blank	0.41	1.76	1.59	0.83	0.41	2.17	3.76	4.59	0.14	0.49	1.03	1.50	0.14	0.63	1.66	3.16
$\frac{1}{4}\times$ DD practical dosage	0.36	1.46	1.29	1.05	0.36	1.82	3.11	4.16	0.15	0.56	0.96	1.66	0.15	0.71	1.67	3.33
$\frac{1}{2}\times$ DD practical dosage	0.55	2.31	0.99	0.63	0.55	2.86	3.85	4.48	0.37	1.55	2.22	3.36	0.37	1.92	4.14	7.50
$1\times$ DD practical dosage	0.48	2.15	1.30	0.70	0.48	2.63	3.93	4.63	0.29	1.45	2.21	2.49	0.29	1.74	3.95	6.44
$2\times$ DD practical dosage	0.37	1.30	0.77	1.12	0.37	1.67	2.44	3.56	0.17	0.74	1.02	1.67	0.17	0.91	1.93	3.60

TABLE 2. Ratios for the cumulative part of Table 1 and further interrelationships between the blank and the different DD dosages.

	Grass				Interrelationship between the blank and the DD dosage	Clover				Interrelationship between the blank and the DD dosage
Number of days since the experiment started	24	52	76	100		24	52	76	100	
Blank	1.0	5.3	9.2	11.1	1.0	1.0	4.5	12.0	22.5	1.0
$\frac{1}{4}\times$ DD practical dosage	1.0	5.1	8.7	11.6	0.9	1.0	4.7	11.1	22.2	1.1
$\frac{1}{2}\times$ DD practical dosage	1.0	5.2	7.0	8.2	1.3	1.0	5.2	11.2	20.2	2.6
$1\times$ DD practical dosage	1.0	5.5	8.2	9.7	1.2	1.0	6.0	13.3	22.2	2.1
$2\times$ DD practical dosage	1.0	4.5	6.6	9.7	0.9	1.0	5.4	11.4	21.2	1.2
Average	1.0	5.1	7.9	10.1		1.0	5.2	11.8	21.7	

reliable by the great differences among them, which presumably arose because the different dosages had produced varying yields of grass and clover per Mitscherlich pot. As a result, the nutritive condition of the potting soil varied and manure should have been added. This, however, was not done. The experiment was discontinued as it had clearly shown that in sterilized soil, that is free from parasites, DD can have the effect of promoting good clover growth. Examination in June confirmed this finding; practically no nematodes were found, still fewer soil insects, such as Collembola and Diptera larvae, and also no mites such as Oribatidae.

A possible explanation of this DD effect is that DD can check the disappearance of the clover and that this process has already begun in the untreated soil and the quarter-dosage treatment. As there is no question of parasitic attack, it must be attributed to competition between the grass and the clover. The perennial ryegrass used in these experiments is more aggressive than most other grasses towards white clover (ENNIK, 1959).

The reason for the effect of the DD on white clover is not known. It can, however, be concluded from the results of this experiment that outside, too, when white clover growing in grassland does not disappear following DD treatment of the soil, whereas it does disappear in untreated soil, this may be the result of an improvement in the competitive position of white clover versus grass, rather than of the killing of parasites.

In addition to investigation into a possible parasitic cause as explanation of the disappearance of the white clover, experiments on competitive aspects should certainly also be made.

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FIG. 1. Part of an experiment in which 5 grass- and 5 white clover plants were grown in Mitscherlich pots with steam-sterilized potting soil treated with DD. From left to right: blank, $\frac{1}{4}\times$, $\frac{1}{2}\times$, $1\times$ and $2\times$ the DD dosage used in practice. Notice the excellent clover growth at the $\frac{1}{2}$ and $1\times$ DD dosage pots. Photograph taken on 9th May, 71 days after the beginning of the experiment.

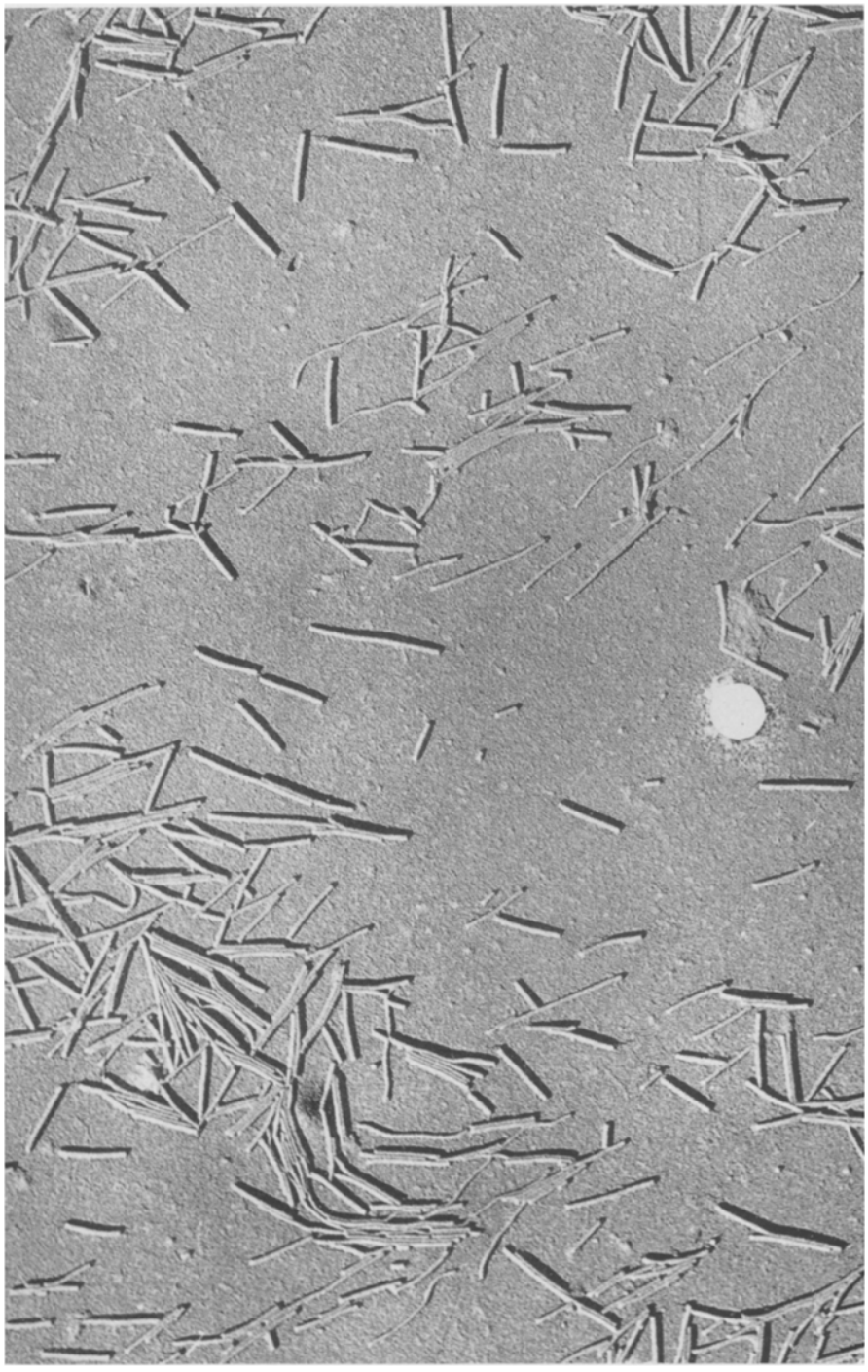

FIG. 1. Electron micrograph of tobacco mosaic virus isolated by chromatography on cellulose using polyethylene glycol containing solutions as solvent. Particles dissolved in 0.1 M NH_4-acetate solution (pH 7).