STUDIES ON THE EFFECT OF SOME ERADICANTS ON MYCORRHIZAL DEVELOPMENT IN FOREST NURSERIES

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SELOSTUS:

KASVINSUOJELUAINEIDEN VAIKUTUS MYKORITSAIN KEHITYKSEEN METSÄTAIMITARHOISSA

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Contents

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Introduction

The importance of artificial reforestation is rapidly increasing in forestry today. At the same time proper management of forest nurseries is gaining more attention in forestry practice and research. Forest nurseries aim at production of high-grade seedlings for forest plantations at low cost. A high-grade seedling means a seedling which after transplanting survives and starts growing without stagnation. For this capacity a balanced ratio must prevail between top and roots and the roots should be able to become adapted to the entirely different environment. In this respect the importance of mycorrhizae has been stressed. Mycorrhizal association is a charateristic feature of the trees of the northern coniferous forests. As has been especially stressed by BJÖRKMAN (1944, 1953, 1956, 1962) and MOSER (1956, 1962, 1963), stagnation and poor survival of seedlings, and even complete failure of young plantations, may be due to absence or inadequate development of mycorrhizae in seedlings when they are transplanted from the nursery into natural forest soil. Therefore, a type of nursery management should be practised which guarantees optimum development of mycorrhizae, which, in turn, depends on adequate fertilization, and sometimes even artificial inoculation of mycorrhizal fungi may be necessary (MOSER 1959).

On the other hand, efforts to produce healthy seedlings and to avoid losses have given rise to large scale application of fungicides and insecticides in forest nurseries. In order to reduce the cost of weeding herbicides are also commonly used. Thus, in modern nursery practice several kinds of chemicals are requlary added to the soil. Naturally then the question arises of what kind of harmful effect these more or less poisonous chemicals may exert directly on the seedlings, on beneficial soil microbes, or on some other soil properties. The effect of fungicides on mycorrhiza formation in particular is an important question, for substances which are expecially intended to destroy fungi are likely to harm mycorrhizal fungi as well and, as was stated above, presence and healthy growth of mycorrhizal fungi is indispensable for the production of usable seedlings.

Of course, such an important problem has been the object of a number of investigations. Especially in America, where different kinds of eradicants have been used on the largest scale, several papers have been published concerning their effect on mycorrhiza formation (e.g. WILDE & PERSIDSKY 1954, 1956; PERSIDSKY & WILDE 1955, 1960; HACSKAYLO & PALMER 1957; PALMER & HACSKAYLO 1958; WALLIS 1959; HACSKAYLO 1961); likewise the effect of eradicants on

other soil microbes has been studied in several countries (e.g. WACRUP 1951; MOLLISON 1953; BOLLEN 1961). Since, however, the chemical industry is continuously inventing new biocidal compounds, the effect of which on mycorrhizal fungi is dependent on various soil factors, further research is necessary.

The purpose of the present study has been to determine what influence some fungicides and herbicides which are in common use in Finnish nurseries may exert on the commencement and early development of mycorrhizal infection in pine and spruce seedlings. Insecticides proper were not included in this study because they are used less frequently in Finnish nursery practice. The results are based mainly on field experiments in nurseries, which were supplemented with some pure culture experiments in the laboratory.

Before the effect of biocides on the commencement of mycorrhizal infection can be studied it is necessary to know the sequence of root development and fungal infection under normal conditions when no chemicals are applied. This subject has been dealt with in several studies (e.g. BURBRIDGE 1936; LADEFOGED 1939; WERLICH & LYR 1957) which, however, have been conducted under different climatic conditions. Therefore a detailed study of the normal rhythm of the growth of seedlings and of mycorrhizae in particular proved necessary, and the main results are presented in the first part of this paper.[1]

This paper constitutes part of a series of studies which are being conducted at the Department of Silviculture, University of Helsinki, with a grant under United States Public Law No. 480, 83rd Congress.

Experimental Conditions

All the field studies on the normal rhythm of seedling growth and mycorrhizal infection, as well as the bulk of the field work with biocides were conducted at the Forestry Field Station of the University of Helsinki (Hyytiälä). Experiments were there carried out in two nurseries which in the following are called Hyytiälä old and new nurseries. The old nursery had been under coniferous stock for seven years. The new nursery was an old farmland which in 1960 was converted to nursery use. As will be seen later, there were some interesting differences in mycorrhizal structure between the old and the new nursery.

Some additional material was collected from Punkaharju nursery in East Finland, where the Forest Research Institute was conducting experiments on the chemical control of weeds and parasitic fungi. Furthermore, root samples were taken from two commercial nurseries of the Foundation of Forest Tree Breeding (Pieksämäki and Vanaja), where herbicides and fungicides were applied regularly.

[1] A more detailed report is preserved at the Department of Silviculture, University of Helsinki (LAIHO 1963).

All these nurseries are located in Central Finland, representing the principal forest area of the country. Some soil properties of the nurseries are given in Table 1. In regard to mechanical composition the nurseries are rather similar, while in chemical properties there are noteworthy differences. The Hyytiälä nurseries, where the experiments were mainly conducted, had a considerably lower nutrient content than Finnish nurseries in general (MIKOLA 1957) and than has been recommended by WILDE (1938), while in Punkaharju nursery, which had been used as a vegetable garden some 10 years ago, both pH and nutrient contents were exceptionally high.

Table 1. Soil properties in the experimental nurseries.

	Hyytiälä old	Hyytiälä new	Punkaharju	Pieksämäki	Average in Finnish nurseries (MIKOLA 1957)
Mechanical composition, %					
Ø 20—2 mm	1.3	5.0	3.3	16.5	6.0
2.0—0.2 »	11.8	28.4	24.4	21.6	31.0
0.2—0.02 »	73.2	53.4	59.4	46.5	45.5
0.02—0.002 »	10.3	9.2	7.0	10.6	10.5
0.002 > »	3.4	4.0	5.9	4.8	7.0
pH	5.5	5.7	6.6	5.3	5.8—5.9
Total N, %	0.18	0.22	0.31	0.25	0.23
Org. matter,[1] %	5.2	6.7	9.3	6.8	6.0
Exch. nutrients,[2] mg/100 g					
K_2O	5.7	11.5	19.5	19.2	14.0
CaO	85	114	429	109	147
P_2O_5	1.3	1.2	13.1	5.0	3.3

[1] Loss on ignition.
[2] From fraction <2 mm. Extracted with 1 N acid NH_4 acetate, pH 4.65. Soil analyses were made at the Soils Department of the Finnish Forest Research Institute.

The nursery experiments were conducted in the summers of 1960 and 1961, and some supplementary experiments in 1962. The weather conditions of these seasons are presented in Fig. 1. As is seen, early June was wery warm in both 1960 and 1961; thereafter the temperature was near the average until the end of September in both years. October was exceptionally cold in 1960 and warm in 1961. The summer of 1962 was one of the coldest growing seasons ever recorded.

In regard to precipitation there are some divergences too. The main growing season of 1961 was exceptionally rainy, while September was drier than average in both 1960 and 1961.

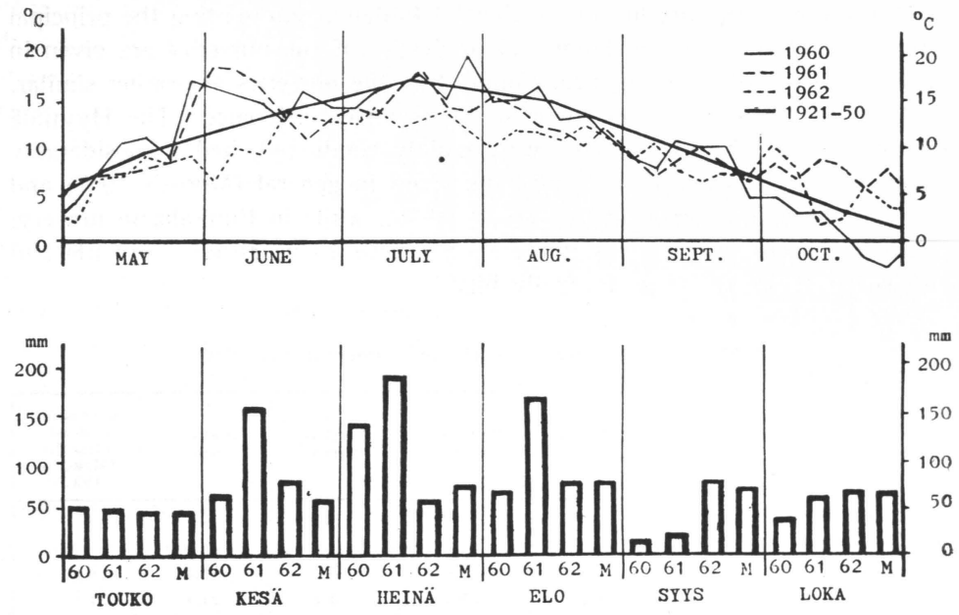

Fig. 1. Temperature (mean of 5 days) and monthly rainfall during the growing seasons of 1960—1962 and the means (M) of 1921—1950.

Commencement and Normal Development of Mycorrhizal Infection

Methods and Material

The growth rhythm of seedlings and the beginning and further development of mycorrhizae were observed by taking samples from nursery beds at short intervals. Subsequent samples were always taken from the same bed by choosing an average spot and removing a bunch of 30—50 seedlings.

This part of the study was concentrated in Hyytiälä new nursery. Seeding was done on June 10, 1960, and germination took place round June 20. The first samples were taken on June 29 and subsequent ones at weekly intervals. During the following summer the same benches were sampled at two-week intervals, starting on May 4 and continuing until Oct. 24.

In order to check the results of 1960, sampling was repeated from beds sown on June 5, 1961. Germination took place round June 15; sampling was started on June 29 and repeated at two-week intervals until October.

Roots were examined immediately after sampling under a binocular microscope (magnification 5—20 ×). Special emphasis was placed on the short roots, on their probable mycorrhizal infection, and in pine on their dichotomous branching. The following determinations of the samples were made:

Average height,
Length of the main root,
Number of long roots,
Number of short root tips, and separately:
the number of mycorrhizal short root tips,
the number of tips of dichotomous short roots.
Dry weight (dried at room temperature),
shoot and roots separately.

Further, parts of root systems were fixed for later microscopic examination.

With microscopic examination the macroscopic classification of short roots and mycorrhizae was checked and the anatomic structure of different mycorrhizal types was studied. Young stages of mycorrhizal infection were especially traced, and the later development and senescene of mycorrhizae were observed. Altogether 700 short roots were sectioned with a paraffin microtome. The thickness of the sections was 7 microns. Double staining with safranin and fast green was used, a method which was developed especially to detect early stages of mycorrhizal infection (Mikola & Persidsky 1951).

Normal Rhythm of Seedling Growth

The weight increase of the shoots and roots of the seedlings in the Hyytiälä experiments are shown in Figs. 2 and 3. During the first growing season the development of shoots and of roots is similar; both increase in weight evenly until September. In the second growing season there are distinct differences. The shoot weight increases sharply about midsummer, although 2-year-old seedlings still have a longer growth period than older trees. The roots have two distinct growth periods, the first in early summer (between May and June, which may be partly due to the marked warm period at that time in 1961), and the second, more pronounced, growth period in late summer. In the second summer root growth commenced earlier than shoot growth and continued longer in the autumn. The same type of periodicity of root growth, with a maximum rate in the late summer, has also been noticed elsewhere (Ladefoged 1939; Heikinheimo 1940; cf. Kramer & Kozlowski 1959, pp. 52—53).

The sharp increase of root weight in the late summer is mainly due to the active branching of the roots and the emergence of new short roots. The growth rate of the long roots, however, is at its maximum at midsummer. Hence in the midsummer the growing long roots had tips without branches for several centimeters, while in the fall branching extended near the long root tips.

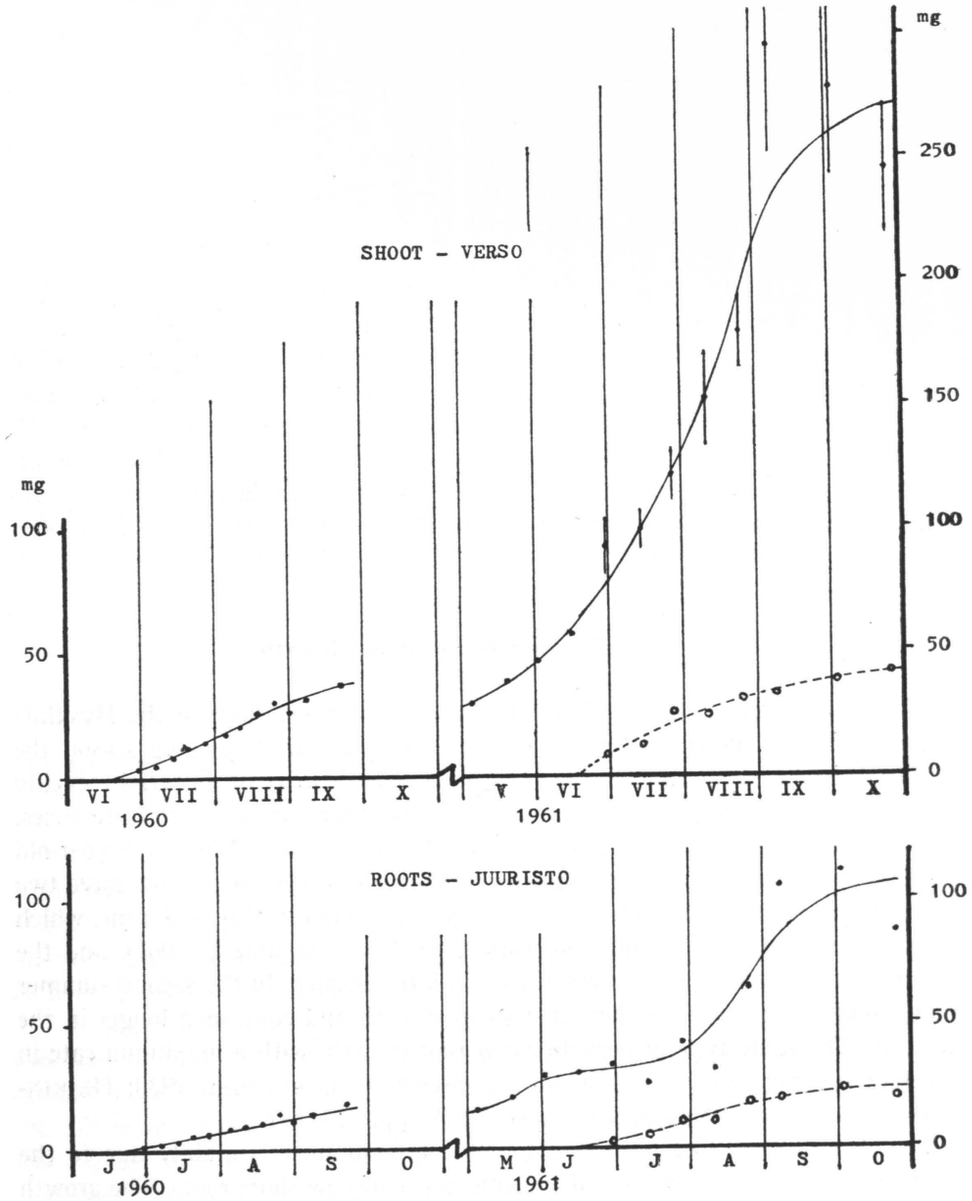

Fig. 2. Weight increase of shoot and roots of pine in Hyytiälä nursery. Solid line: Seeding of June 10, 1960. Broken line: Seeding of June 5, 1961.

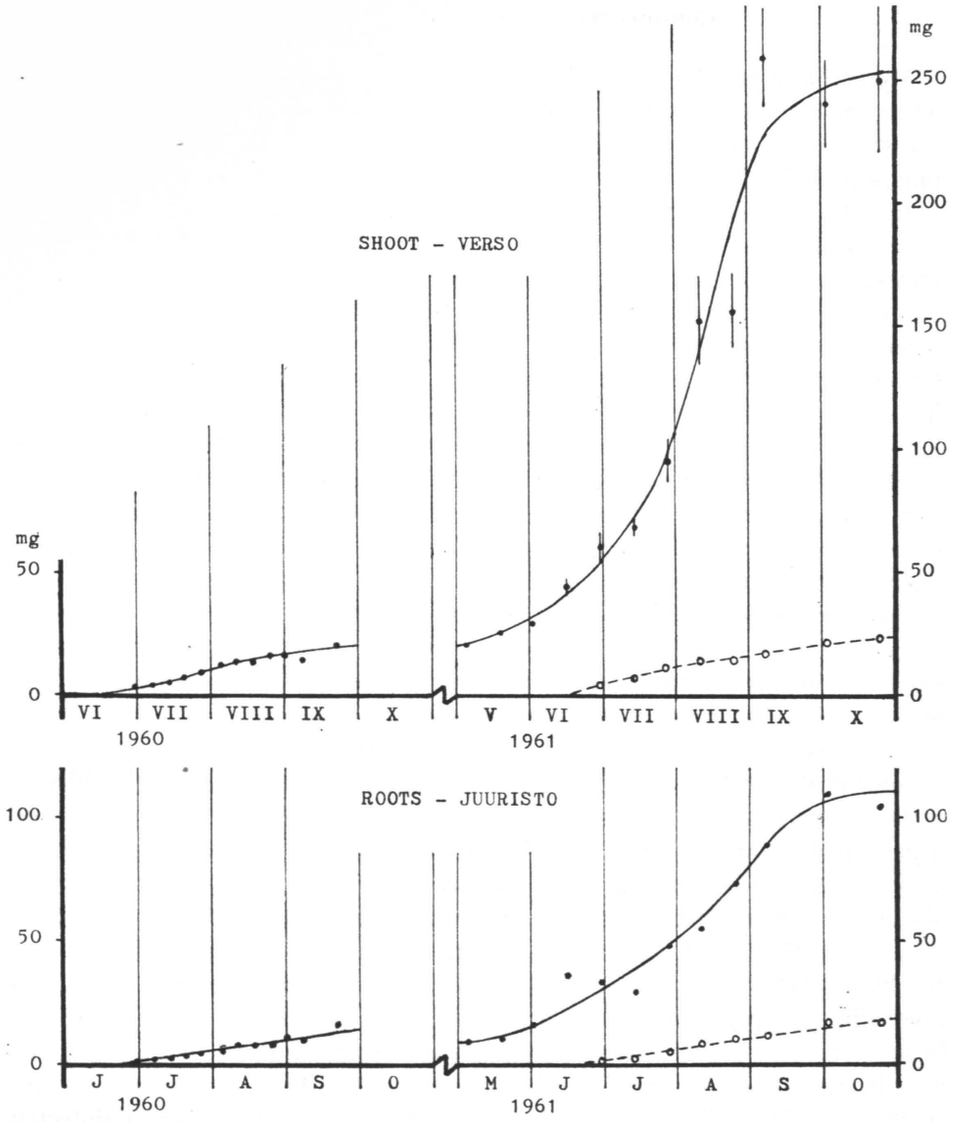

Fig. 3. Weight increase of shoot and roots of spruce in Hyytiälä nursery. Solid line: Seeding of June 10. 1960. Broken line: Seeding of June 5. 1961.

Commencement of Mycorrhizal Infection

Mycorrhizae are short roots which have been infected with fungi. Therefore, when the commencement of mycorrhizal infection and its further development are studied the emergence of short roots, their increase in number, and in the pine especially their dichotomous branching must be observed (Fig. 4).

In both summers the first short roots of pine and spruce formed 25—28 days after sowing or 16—20 days after germination. Thereafter their number increased steadily, reaching levels of 300—400 and round 200 per seedling of pine and spruce respectively by the end of the first summer. At the beginning of the second growing season the number of short roots is somewhat smaller because the ground frost breaks many long roots and some non-mycorrhizal short roots die out during the winter. In the second summer the number of pine root tips increases most sharply in August and September because of intensive dichotomous branching. No corresponding phenomenon occurs in spruce, because at the end of the first growing season all spruce short roots are still unbranched and even during the second summer branching is infrequent.

Mycorrhizae are often hard to distinguish from uninfected short roots either macroscopically or with 5—20 times magnification. The earliest stages of infection can not be detected without stained sections. Therefore, microscopic examination was the principal method used in this study to determine the time of first infection, to check the macroscopic classification, and to observe the further development and senescence of the mycorrhizae.

Some of the mycorrhizae could be distinguished easily; they were classified as »good» mycorrhizae. They were thicker than the uninfected short roots and usually had a smooth surface and light color. Mycorrhizae with a slender basal part or »stalk» and swollen tip (B mycorrhizae of MELIN, 1927) were also common. Microscopic examination of »good» mycorrhizae always revealed a hypertrophied cortex with Hartig net and on pine mycorrhizae usually a mantle. The ectendotrophic mycorrhiza of pine, which was a common and in some nurseries even a predominant type, however, had a very thin (5—10 μ) mantle, if any. Mycorrhizae of nursery-grown spruce seedlings were usually unbranched and had no mantle, and since the cortical cells were only slightly hypertrophic the mycorrhizae could hardly be distinguished from pseudomycorrhizae and uninfected short roots. Dichotomous short roots of pine were usually »good» mycorrhizae at the same time, *i.e.* thick and light-colored. There were also, however, slender dichotomous short roots, the mycorrhizal nature of which could be identified by microscopic examination only. The dichotomous short roots of pine invariably proved mycorrhizal, thus the occurrence of dichotomous short roots could be considered a reliable indication of the presence of mycorrhizal association and their number as an index of the degree of infection. True, dichotomy without mycorrhizal association is know under experimental conditions (HATCH & DOAK

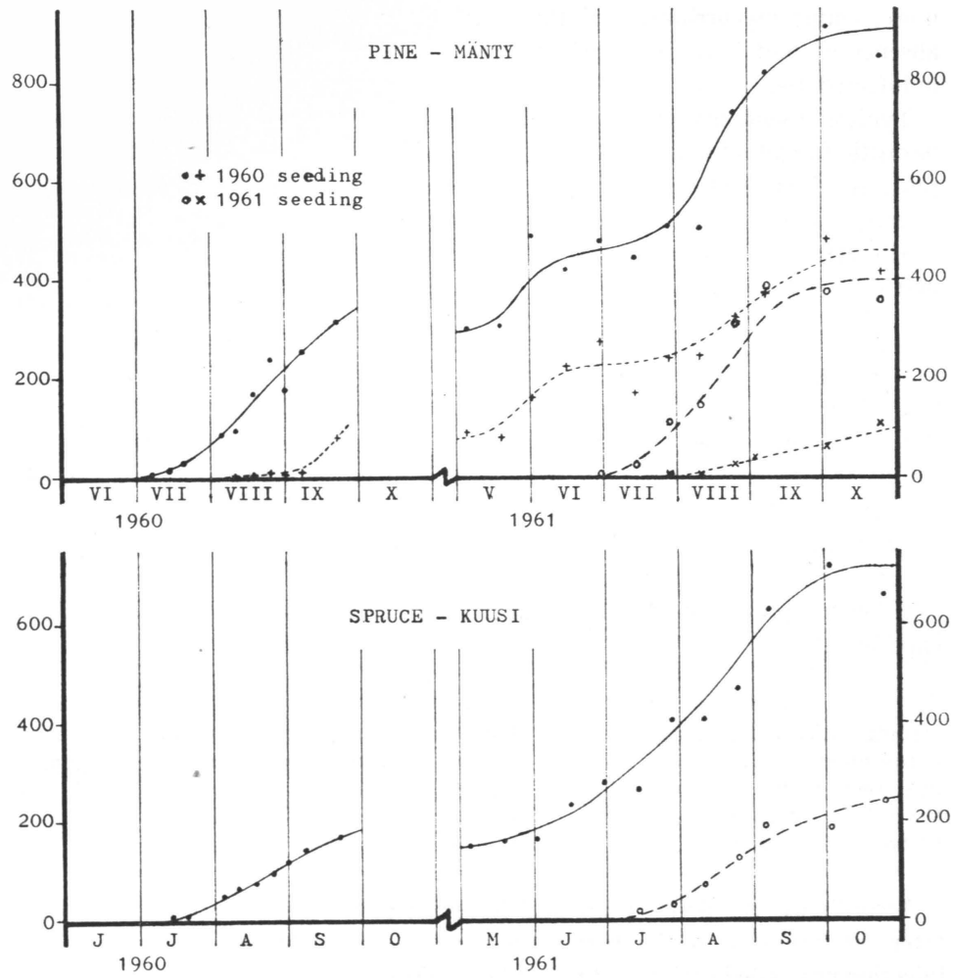

Fig. 4. Increase of the numbers of short root tips and tips of dichotomous short roots (dotted lines, pine only) of pine and spruce seedlings in Hyytiälä nursery. Seedings: June 10, 1960 (solid line) and June 5, 1961 (broken line).

1933; SLANKIS 1948), in Hyytiälä nursery, however, dichotomous non-mycorrhizal short roots were not found (cf. RICHARDS & WILSON 1963).

In microscopic examination mycorrhizal structure, *i.e.* the Harting net, was generally also detected in the unbranched slender short roots which in no way differed from uninfected short roots. The uninfected short roots often had root hairs, but short roots without either root hairs or fungal infection were common too. Some hyphae grew sporadically on the root surface and in the epidermis; such short roots, however, can hardly be called pseudomycorrhizae. Further-

more, young mycorrhizae sometimes still had root hairs, thus the presence or absence of root hairs is no reliable basis for distinguishing mycorrhizae from uninfected roots.

Typical pseudomycorrhizae, *i.e.* short roots with intracellular, probably parasitic fungal infection alone, were rare in the nursery. On the contrary, short roots, in which even careful microscopic examination could not reveal any fungal infection, were common in seedlings during their first summer.

The results of the microscopic examination of 700 short roots can be summarized as follows:

1. All the short roots which were macroscopically classified as »good» mycorrhizae were in fact mycorrhizal.
2. Dichotomous short roots of pine were always mycorrhizal.
3. Slender unbranched short roots which were classified as uninfected or pseudomycorrhizae on macroscopic examination included mycorrhizae too, particularly in older seedlings. This finding led to a correction of the percentage of mycorrhizae. Thus, in the second summer practically all short roots were mycorrhizal, although only 40—50 % were classified as »good» mycorrhizae.

Below are presented some essential dates of seedling development in Hyytiälä experiments.

	Pine		Spruce	
	1960	1961	1960	1961
Seeding	Jun. 10	Jun. 5	Jun. 10	Jun. 5
Germination	Jun. 18	Jun. 13	Jun. 18	Jun. 18
First short roots	Jul. 6	Jun. 29	Jul. 6	Jun. 29
Mycorrhizal infection	Jul. 25	Jul. 27	Aug. 4	Jul. 27
Dichotomous short roots	Aug. 4	Jul. 27		

Accordingly, the first mycorrhizal infection took place as late as 3—4 weeks after the formation of the first short roots. Once established, the mycorrhizal infection proceeded rapidly. At the end of the first growing season the precentages of mycorrhizal short root tips were as follows:

	1960	1961
Pine	60	89
Spruce	59	94

The difference between the years 1960 and 1961 is probably due to the widely different weather conditions of the respective fall seasons (cf. Fig. 1).

In the second summer (seeding of 1960 in 1961), mycorrhizal infection took place immediately as fast as new short roots emerged. Thus from June on the percentage of mycorrhizae was nearly 100.

Accordingly, the early development of seedlings in the nursery can be divided into three phases:

1. Non-mycorrhizal phase: from germination to late July (approximately 5 weeks).
2. The phase of mycorrhizal infection, until early June of the second year.
3. Mycorrhizal phase, from this time on, practically all short roots being mycorrhizal.

The dichotomous branching of pine short roots began immediately after fungal infection had taken place and then proceeded rapidly. Thus, in the seeding of 1961 the first dichotomous roots with 4 tips were found on Aug. 1, roots with 8 tips on Aug. 7, and with 16 tips in the middle of September. Also in the second growing season forking of short roots took place mainly in the late summer (cf. the different course of the number of pine and spruce short root tips, Fig. 4).

The delayed formation of mycorrhizae in first-year seedlings was first noticed by MELIN (1917, p. 367), and there are several later references too (e.g. HUBERMAN 1940; WILSON, cit. HARLEY 1959, p. 39). The reason for the slow infection may be the fact that the photosynthetic capacity of the cotyledons is too weak to bring the concentration of soluble carbohydrates in the roots to the level necessary for mycorrhiza formation (BJÖRKMAN 1949). This hypothesis is supported by the observation that the mycorrhizal infection coincided with the formation of the first juvenile needles, which means a considerable increase of photosynthetic capacity. This has also been noticed by MOSER (1962), who recommendes that the inoculation of *Pinus cembra* seedlings should be postponed until transplanting, because younger seedlings are not able to produce sufficient carbohydrates for the fungi.

The mode of infection is also different. The first short roots contract the infection from the surrounding soil. Hyphae penetrate between the cortical cells and therefore a Hartig net is usually the first sign of mycorrhizal infection (Fig. 5, A—B). Later on, short roots are probably infected mainly by hyphae growing along the long roots, either on their surface or in the cortex. Hence the short root tip is generally covered by a fungal mantle as soon as it has penetrated the cortex of the long root (Fig. 5, C). Ectendotrophic infection, which is very common on pine in many Finnish nurseries, also takes place along with the formation of short roots; because ectendotrophic mycorrhizae often lack a mantle, however, the infection begins with a Hartig net (Fig. 5, D) and intracellular infection follows a little later. In seedlings with ectendotrophic mycorrhizae intracellular hyphae of the same fungus are usually very numerous in the cortex of long roots too.

Later Development of Mycorrhizae

As was stated previously, the fungal infection of short roots may commence in different ways, either immediately at the formation of the short root or later. Likewise the advance of infection in the roots and the later development of

mycorrhizae may be different, depending on the fungal species, environmental conditions, ect. Fig. 5 shows schematically some different models of the development of mycorrhizae.

A. (1) The short root has grown without infection for about three weeks. Root hairs are common at this stage but may also be lacking. (2) Infection has taken place; a Hartig net is forming near the root tip, behind the meristem. (3) At the end of the first growing season the mycorrhizal cortex is covered by a fungal mantle. Infection has not advanced to the basal part of the short root, where sporadic intracellular (pseudomycorrhizal) hyphae are growing. Root hairs are disappearing. (4) In the second growing season the growth of the mycorrhiza continues. In the basal part the cortex degenerates and gradually disappears. (5) Growth still continues in the third season. In the oldest part the cortex has disappeared and a thin suberized tissue protects the stelar tissue. This kind of development is typical of mycorrhizae which form on pine roots in the first growing season. Repeated dichotomous branching is common from stage (2) on.

B. (1)—(2) As in case A infection begins when the short root is about 4 weeks old. (3) By the end of the first growing season the Hartig net has advanced from the zone of the first infection to the older part of the short root up to its base. The cortical cells are not hypertrophied, however, and no mantle has formed; hence the short root is not thickened. Root hairs are disappearing. (4) Growth continues in the second growing season and (5) in the third, when the cortex of the oldest part has died and disappeared. This is the common mode of infection of spruce seedlings.

C. (1) After penetration of the cortex of the long root, the root is covered by a fungal mantle. (2) A few days later the Hartig net has formed. (3) At the end of the growing season the short root is a typical A mycorrhiza. (4) In the following growing season growth continues. The cortex of the older part is also still alive and active. (5) In the third summer growth still continues; the cortex of the oldest part has died. This type of development is characteristic of ectotrophic pine mycorrhizae which form in the second year or later.

D. (1) When penetrating the long root cortex the short root is still uninfected. (2) A few days later the Hartig net is already present but there is no mantle so far. (3) At the end of the summer the well-developed mycorrhiza may be covered by a thin mantle. (4)—(5) Development continues as in C. This was the commonest type of development of spruce mycorrhizae in the nursery after the first year. Likewise the ectendotrophic mycorrhiza of pine developed by a corresponding pattern.

E. (1) A young short root with root hairs is infected intracellularly. The pseudomycorrhizal cortex is fairly shortlived. (2) In the late summer the cortex is dead in the basal part; the tip is still growing and pseudomycorrhizal infection follows. The short root often dies at this stage. (3) Mycorrhizal infection (Hartig

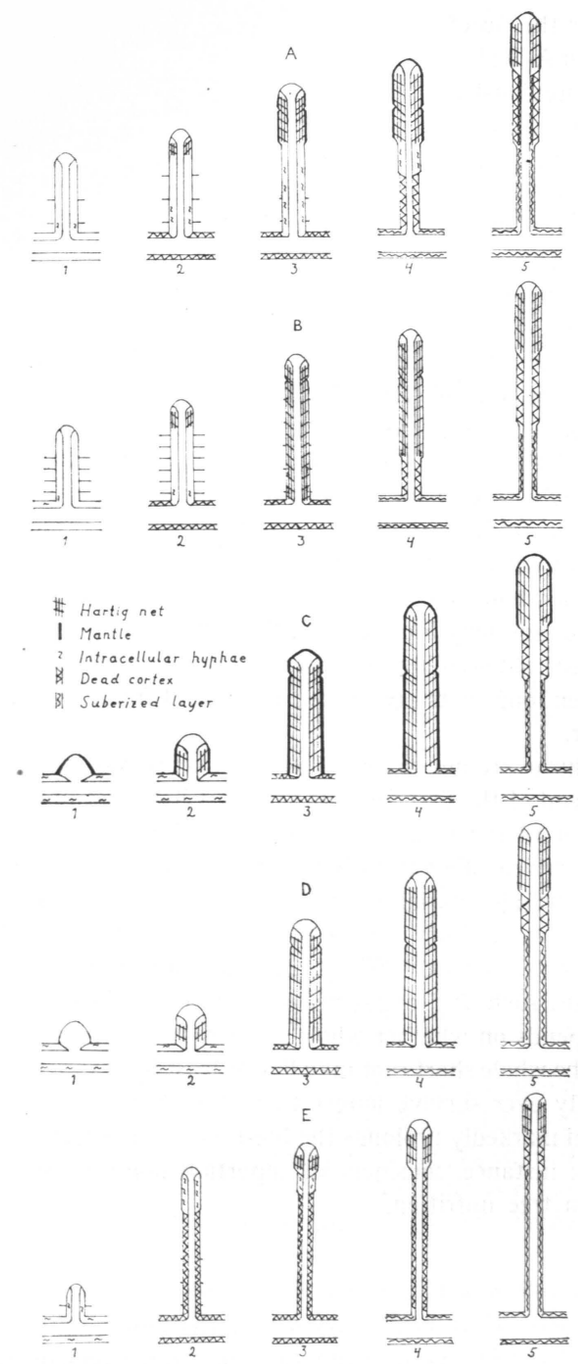

Fig. 5. Patterns of mycorrhiza development. Explanation in the text.

net) appears near the tip of the pseudomycorrhiza. (4) Growth continues in the next summer as in A-D above.

As was stated previously, the percentage of mycorrhizae approaches 100 in the beginning of the second growing season, and thereafter the number of short roots also increases rapidly. For example, a 2-year-old pine seedling with a shoot dry weight of 0.25 g (Fig. 2) has more than 800 mycorrhizal short root tips (Fig. 4). Thus the mycorrhizal short roots constitute an extensive and effective nutrient-absorbing system for the young seedling.

The formation and branching of short roots continues in the third summer. The question that now arises is how long the first mycorrhizae remain alive and active. In the present study the development of seedlings and especially their mycorrhizal relations were observed for two growing seasons and some samples were still taken in the third season from the same beds. The models of Fig. 5 show examples of the development of mycorrhizae in the second and third seasons. Accordingly, mycorrhizae live and grow at least 2 or 3 years. The mycorrhizal cortex of short roots is short-lived, however, individual cells remaining alive and active 1 or 2 seasons only.

The mortality of whole mycorrhizae was small during the study period. Thus, in 2-year-old seedlings only 5 per cent of the total number of mycorrhizae were dead, and in 3-year old seedlings 10—15 per cent. In addition to that, mycorrhizae die out when long roots are broken by ground frost, as generally happens during the winter.

Pseudomycorrhizae are not so long-lived. If a short root has not become mycorrhizal by the end of the first growing season it has most probably contracted pseudomycorrhizal infection, *i.e.* semiparasitic fungi have grown into the cortical cells. Pseudomycorrhizae die out during or even before the second year.

The literature includes variable data on the life-time of individual mycorrhizae, estimates varying from 1 to 4 years (Melin 1923, p. 110; Burbridge 1936, p. 38; Werlich & Lyr 1957, p. 22; Lobanov 1960, p. 186). Probably climatic conditions, such as drought periods, exert a great influence. The result obtained also depends on whether whole short roots or annual growth portions are considered. The whole short root may live at least 3—4 years, while individual cortex cells hardly ever survive longer than 2 growing seasons. Anyway, mycorrhizal infection markedly prolongs the life-time of the short root, a fact which Hatch (1937), for instance, considers an important point when judging the role of mycorrhizae in tree nutrition.

Experiments with Eradicants

The Chemicals Used

The commonest eradicants in Finnish nursery practice are volatile or unstable poisons which are applied to the soil before seeding to kill damping-off fungi and weed seeds. For decades formaldehyde has been used for such partial sterilization of soil; recently several new substances have been introduced for the same purpose, *e.g.* methyl bromide and allyl alcohol. Since such soil sterilants are likely to be detrimental to mycorrhizal fungi the main emphasis was put on them in this study. In addition, some modern herbicides which are in common use today were included, as well as some fungicides which are used as sprays or dusts on growing seedlings and then come into the soil too. Application of insecticides is not a regular practice in Finnish nurseries; therefore they were not considered in this study.

In the experiments the application of the chemicals varied from the rates recommended by the manufacturers up to amounts several times greater. The following chemicals were included:

A. Soil disinfectants

1. Methyl bromide, which has proved very effective for weed eradication. Because of its gaseous state and highly poisonous nature its application is cumbersome; it is allowed to act under a sheet of plastic for 1—2 days; the rate recommended is 1—2.5 kg/100 m²; the highest rate in the experiments was 15 kg/100 m².

2. SMDC (Vapam, manufactured by Roehm & Haas Co., Philadelphia, U.S.A., containing 32.7 % Na methyl dithiocarbamate) is a liquid which, because ot its lower price and more convenient handling, has lagerly taken the place of methyl bromide. The recommended application is 5—10 liters of Vapam + 250—500 liters of water per 100 m². A rate even five times as high was used in the experiments. A sharp rise of pH (up to 9) results from SMDC application. Seeding can be done 2—3 weeks after soil treatment.

3. Formalin (35 % formaldehyde) is a rather weak disinfectant which has been in use for a long time. It is recommended especially against damping-off fungi and it also kills weed seeds; the normal application is 10—20 liters of formalin (corresponding to 3—6 kg of formaldehyde) per 100 m², diluted with a 50-fold amount of water. The highest rate used in this study was 200 liters of formalin per 100 m².

4. Allyl alcohol is more effective than formalin and therefore has recently largely taken its place especially as a weed eradicant. The normal application is 1 kg/100 m², diluted with a 500-fold amount of water. The highest application in the experiments was 25 kg/100 m².

B. Weed killers

5. Simazine (50 % 2-chlor-4,6-bisethyl aminotriazine, J. R. Geigy A.G., Basel, Switzerland), very slightly soluble in water, is used as a water suspension between seedling rows. The normal rate of application in nurseries is 2—4 kg/hectare; in the experiments as much as 50 kg/hectare was applied.

6. Dalapon (2,2-dichloropropionic acid, Pan Britannica Industries, Waltham, England) is recommended especially against *Agropyron repens* and other graminaceous weeds, used at the rate of 20—50 kg/ha; this was exceeded by 20-fold in the experiments. Soil treatment is done at least 3 weeks before seeding.

C. Fungicides

7. PCNB (20 % pentachoronitrobenzene, Brassicol of Farbwerke Hoechst A. G., Frankfurt, Germany) is mainly used as a dust against needle cast and snow blight fungi and can also be mixed into the soil to control damping-off. The recommended rates are 20—300 kg/ha, which was exceeded by 20-fold in the experiments.

8. Cu oxychloride (87.5 %, Soltosan of Plant Protection Ltd., London, England) is used in 0.5 % solution as a spray against needle cast fungi at the rate of 5—8 kg/ha.

9. Zineb (65 % Zn ethylene bisdithiocarbamate, Dithane Z—78 of Roehm & Haas Co., Philadelphia, U.S.A.) is also used as a sprays against needle cast fungi at the rate of 0.6—1.2 kg/ha. In the nursery experiment both Soltosan and Dithane were applied to the soil before seeding at a rate exceeding twice that recommended.

The Effect of Eradicants of Mycorrhizae in the Nursery

The effect of eradicants on the formation of mycorrhizae was studied with field experiments, which were conducted in the old and new sections of Hyytiälä nursery in 1960—62. The main experiments were as follows:

1 A. Hyytiälä new nursery in 1961. All the above chemicals were used, most of them in two different concentrations and in two replicates. The size of the plots was 2 m². Different treatments were separated with untreated controls. Treatments were performed on June 9—14 and both pine and spruce were seeded on June 19. Samples were taken in two seasons (1961 and 1962).

1 B. Another similar experiment was started in the same nursery in the spring of 1962. The same chemicals were used, but in considerably larger doses. Because of the exceptionally cold summer the growth of seedlings was slow. Samples were taken in the fall of 1962 only.

2. Hyytiälä old nursery in 1961. The experimental conditions were the same as in Exp. 1 A and the same treatments were included. Some previous studies had revealed that the ectendotrophic mycorrhiza was the prevalent type in the old nursery and lacking in the new one; therefore parallel experiments were conducted in both nurseries to show whether there are any differences in the sensitivity of the two types of fungi to different chemicals. A heavy application of Vapam was further included in a fertilizer experiment in 1962 (2 B).

In addition to these main experiments, supplementary material was obtained from the following sources:

3. An experiment with methyl bromide in Hyytiälä new nursery. The experiment was started in 1960 and sampled in 1960 and 1961.

4. Punkaharju nursery. Experiments with methyl bromide and Simazine were started in the spring of 1959, and samples were taken in 1960.

5. Punkaharju nursery. Experiments with five different chemicals were started in the spring of 1960; samples were taken in 1960 and 1961.

6. Punkaharju nursery. Experiments with Vapam, allyl alcohol, and Simazine in 1962.

7. Vanaja nursery. Samples were taken in the spring of 1961 from beds treated with methyl bromide one and two years earlier.

8. Pieksämäki nursery. Samples were taken in the fall of 1961 from beds where Simazine had been applied two months earlier.

The results of the methyl bromide experiments are summarized in Table 2. Although there is great variation in the rate of mycorrhiza formation a general trend can be noticed, viz. retardation of mycorrhizal infection by methyl bromide treatment. In the untreated controls initial infection was usually present in early August, as was stated previously (p. 12). By the end of the first season the percentage of mycorrhizae usually amounts to 50—60 and by the middle of the second summer to 100. Methyl bromide treatment usually delayed the commencement of infection by a few weeks. In general, however, infection took place during the first summer, and the difference between treated plots and controls disappeared by the end of the second summer. The stronger the application, the more the infection was delayed. The retarding effect of the normal application, 1—3 kg/100 m², was rather insignificant.

The delay of mycorrhizal infection was not harmful to the growth of the seedlings. On the contrary, in the treated plots the seedlings were, on the average, 30 % bigger and the number of short roots was also greater than in the controls, probably owing to the effects of methyl bromide on the soil.

As regards the structure of the mycorrhizae, the methyl bromide plots did not differ from the controls; this was to be expected since the poisonous gas probably had disappeared from the soil long before the commencement of infection.

Methyl bromide treatment probably kills the hyphae and spores of mycorrhizal fungi in the surface layer of the soil. This layer is relatively thin, however, and the hyphae deeper in the soil remain alive. In fact, in the treated plots the first mycorrhizae usually appeared at a depth of around 5 cm, while in the

Table 2. Percentage of mycorrhizal short root tips in experiments with methyl bromide.

Experiment Date of seeding	Date of sampling	Pine						Spruce					
		Methyl bromide, kg/100 m²											
		0	1	2	3	4–7	10–15	0	1	2	3	4–7	10–15
1 A	Aug. 28, 1961	2			0	0	0	0					0
Jun. 19, 1961	Oct. 24, 1961	11			12	0	1	21			0		0
	Jul. 2, 1962	100			100		60						
	Oct. 12, 1962	100					100	100					100
1 B													
Jun. 21, 1962	Oct. 10, 1962	11			9	0	0	30			0		0
2	Aug. 16, 1961	7			0	17							
Jun. 19, 1961	Sept. 9, 1961	43				8							
	Oct. 2, 1961	41				21							
	Oct. 24, 1961	64			22	55							
	Jul. 2, 1962	100			100	100							
3	Sept. 3, 1960	17		7				6		4			
Jun. 10, 1960	Sept. 15, 1961	100		100				100		100			
4	May 27, 1960	29	26		14	23		8	4		2	2	
Jun. 12, 1959	Sept. 23, 1960	100	100		100	100		100	100		100	100	
5	Sept. 23, 1960	46			46	0		58			16	8	
Jun. 13, 1960	Sept. 30, 1961	100			100	100		100			100	100	

controls infection always was first observed nearer the surface. Later on in the fall, some sporadic mycorrhizae also appeared in the treated plots quite near the surface, suggesting air-borne infection through spores.

The effect of SMDC (Table 3) was very much the same as that of methyl bromide. Strong application distincly retarded mycorrhizal infection in the first summer, while the difference between treated and control plots disappeared during the second summer. The fungus of the ectendotrophic mycorrhiza which was present in Hyytiälä old and Punkaharju nurseries (Expts. 2 and 6) was perhaps somewhat more resistant than the ordinary ectotrophic fungi. No apparent influence on the growth of seedlings could be observed, the size of the seedlings being the same in both plots.

The above results with methyl bromide and SMDC agree well with those obtained by HACSKAYLO and PALMER (1957; PALMER & HACSKAYLO 1958).

Formaldehyde and allyl alcohol were fairly similar in their effect on the development of seedlings and mycorrhizae (Tables 4 and 5). A moderate application did not retard mycorrhizal infection, neither could any influence on

Table 3. Percentage of mycorrhizal short root tips in experiments with SMDC.

Experiment Date of seeding	Date of sampling	Pine			Spruce		
		SMDC, kg/100 m²					
		0	3	10	0	3	10
1 A	Aug. 28, 1961	2	0	1	0		
Jun. 19, 1961	Oct. 24, 1961	11	14	0	21	0	0
	Jul. 2, 1962	100	100	100	100	60	
	Oct. 12, 1962	100	100	100	100	100	100
1 B							
Jun. 21, 1962	Oct. 10, 1962	11	7	2	30	0	0
2.	Aug. 16, 1961	7	12	0			
Jun. 19, 1961	Aug. 28, 1961	42		32			
	Oct. 24, 1961	64	25	25			
	Jul. 2, 1962	100		100			
	Oct. 12, 1962	100	100	100			
2 B							
Jun. 11, 1962	Oct. 10, 1962	70		0			
6							
Jun. 15, 1961	Sept. 30, 1961	64	33		33	8	

Table 4. Percentage of mycorrhizal short root tips in experiments with formaldehyde.

Experiment Date of seeding	Date of sampling	Pine				Spruce			
		Formaldehyde, kg/100 m²							
		0	3	10	20–70	0	3	10	20–70
1 A									
Jun. 19, 1961	Aug. 28, 1961	2		7		0			
	Oct. 24, 1961	11	18	23		21	1	20	
	Oct. 12, 1962	100		100		100	100	100	
1 B									
Jun. 21, 1962	Oct. 10, 1962	11	3		6	30	0		0
2									
Jun. 19, 1961	Aug. 16, 1961	7			12				
	Oct. 24, 1961	64			50				
	Oct. 12, 1962	100			100				
5									
Jun. 10, 1960	Sept. 23, 1960	56	27	42		31	40	24	
	Sept. 30, 1961	100	100	100		100	100	100	

Table 5. Percentage of mycorrhizal short root tips in experiments with allyl alcohol.

Experiment Date of seeding	Date of sampling	Pine					Spruce				
		Allyl alcohol, kg/100 m²									
		0	1	3	5—7	10—20	0	1	3	5—7	10—20
1 A											
Jun. 19, 1961	Aug. 28, 1961	2	3		8		0				
	Oct. 24, 1961	11	32	19	16		21	17	10	3	
1 B											
Jun. 21, 1962	Oct. 10, 1962	11	6		0	0	30	5		1	0
2	Aug. 16, 1961	7	13	25	11						
Jun. 19, 1961	Sept. 9, 1961	43			45						
	Oct. 24, 1961	64	51	60	74						
6											
Jun. 5, 1961	Sept. 30, 1961	64	83				33	43			

Table 6. The effect of formaldehyde treatment on growth of seedlings during their first growing season. Experiment 5.

Formaldehyde kg/100 m²	Pine		Spruce	
	Shoot weight mg	No. of short roots per seedling	Shoot weight mg	No. of short roots per seedling
0	20	90	12	80
3	37	149	26	160
10	43	213	35	173

the structure or relative number of mycorrhizae be observed. In fact, the absolute number of mycorrhizae per seedling was greater in the treated plots than in the controls, for the size of seedlings and the number of short roots were much greater in the treated plots (Table 6).

The promoting effects of formaldehyde and allyl alcohol on seedling growth have been noticed in other studies too. Furthermore, in some American experiments both substances also retarded the fungal infection and caused abnormalities in the structure of the mycorrhizae (WILDE & PERSIDSKY 1954, 1956; PERSIDSKY & WILDE 1955). Strong application exerted a retarding effect in the present study too (Expt. 1 B).

The herbicides Simazine and Dalapon were included in Experiments 1 A, 1 B, 2, and 5, and Simazine further in Experiments 3, 6, and 8. A strong application was definitely harmful to the seedlings and hence, of course, conditions

for mycorrhiza formation were also worsened. On the other hand, an application that was harmless to the seedlings did not retard mycorrhizal infection either. Evidently tree seedlings are more sensitive to the above herbicides than their mycorrhizal fungi, as PERSIDSKY and WILDE (1960) have also found.

The three fungicides (PCNB, Cu oxychloride, and Zineb, Expts. 1 A, 1 B, and 2) which are used as sprays or dusts in ordinary nursery practice, had no retarding or disturbing effect on mycorrhizal development when mixed with the soil before seeding.

The Effect of Eradicants on Other Soil Microbes

The effect of eradicants on soil microbes in general was studied in Hyytiälä new nursery in connection with Experiments 1 A and 1 B. Soil samples were taken from the same plots where mycorrhizal development was studied, and the numbers of bacteria, molds, and actinomycetes were determined with the ordinary dilution plate method. The first samples were taken immediately after the treatments, the second ones one week later, and sampling was then continued at two-week intervals. The plate substrates were the Hagem agar which is generally used for mycorrhizal and other fungi (MODESS 1941, p. 16) and soil extract agar for bacteria and actinomycetes. At each sampling the numbers of microbes were compared with the simultaneous counts of the controls. In untreated soil the numbers of bacteria amount to around 1—2 million per g of dry soil, the corresponding figures for molds and actinomycetes being 100 000—400 000. The results of the two summers were fairly consistent.

The results of the plate count studies are summarized as follows:

Immediately after methyl bromide, SMDC, and formaldehyde treatments there were hardly any microbes in the surface soil. The number of bacteria, however, increased rapidly; after one week it was at the same level as in the controls, and after two more weeks as much as 10 times higher. Thereafter the number of bacteria decreased again, reaching the level of the controls in the late summer. Such an effect of partial soil sterilization on bacteria has been generally found. The number of molds increased more slowly approaching the level of the controls after 1—2 months. The return of actinomycetes was still slower. After methyl bromide and SMDC treatments their number only started to increase after 2 months and had not attained the control level by the end of the season. After formaldehyde treatment actinomycetes were not found at all in the same summer.

The effect of allyl alcohol was similar but considerably weaker. The treatment did not destroy the microbes so completely, and correspondingly, the number of bacteria did not rise much above the control level.

The other chemicals had little influence on the soil microbes. After Simazine

treatment the numbers of all microbes remained somewhat below the control level through the summer. The highest application of fungicides probably slightly reduced the number of microbes.

The most significant finding in these experiments was the remarkable influence of formaldehyde and allyl alcohol on the composition of the mold population. Both of the treatments promoted *Trichoderma viride* tremendously. *Trichoderma viride*, which only occurred sporadically in the controls and other treatments, became the overwhelmingly dominant mold after formaldehyde or allyl alcohol treatment (Table 7; the results of the 1962 experiment were consistent). The predominance of *Trichoderma* continued even through the second growing season.

The strong increase of *Trichoderma viride* after formaldehyde and allyl alcohol treatments is known from other studies too (WARCUP 1951; MOLLISON 1955; EVANS 1955; YATAZAWA, PERSIDSKY & WILDE 1960). In this connection it should especially be pointed out that the same treatments also strongly promote the growth of coniferous seedlings but have little effect on the formation of mycorrhizae.

Table 7. The number of *Trichoderma* (1000/g) in nursery soil after treatment with different chemicals (Treatment on June 11, 1961).

Treatment	Date of sampling						
	Jun. 15	Jul. 2	Jul. 18	Aug. 1	Aug. 17	Oct. 1	Oct. 24
Control	0	4	0	0	0	0	0
Methyl bromide ..	0	0	0	0	11	0	47
SMDC	0	0	0	0	0	0	0
Formaldehyde	0	10	42	110	11	39	210
Allyl alcohol	30	21	61	79	83	72	80

The Effect of Eradicants on Fungi in vitro

Pure culture experiments were conducted to show whether mycorrhizal fungi are more sensitive to nursery chemicals than other soil fungi, or *vice versa*. The fungi were grown in petri dishes on Hagem agar to which biocides had been added in different concentrations. The same chemicals were used as in nursery experiments, except methyl bromide, which, being a gas, had proved difficult to dissolve in agar. Simazine and other difficultly soluble powders were suspended in melted agar. The relative growth of fungi was determined by measuring the diameters of colonies and comparing them with the growth on control plates. Measurements were made when the diameter of the controls was around 2 cm; accordingly the duration of the experiment was different for different fungi, depending on their rate of growth.

The following 10 fungi served as test objects:

1. *Boletus variegatus* } mycorrhizal fungi; isolated
2. *Boletus luteus* } from sporophore tissues.
3. *Cenococcum graniforme*, a mycorrhizal fungus; isolated from a sclerotium.
4. *Corticium bicolor* (?) a mycorrhizal fungus; isolated from a bright yellow mycorrhiza of spruce (MIKOLA 1962).
5. *Collybia dryophila*, a saprophytic basidiomycete of forest humus; isolated from sporophore tissue.
6. *Mycelium radicis atrovirens*, a ubiquitous root associate, pseudomycorrhizal and probably weakly parasitic; isolated from a spruce mycorrhiza.
7. *Rhizoctonia solani*, a parasitic damping-off fungus; obtained from the Department of Plant Pathology, the Finnish Agricultural Research Institute.
8. *Trichoderma viride* } common soil molds; isolated from Hyytiälä new
9. *Verticillium terrestre* } nursery; identification is based on GILMAN's
10. *Penicillium spinulosum* } (1944) manual.

The results of the pure culture experiments are summarized in Figs. 6—13. The growth of fungi at different concentrations of eradicants are expressed relatively by taking the growth of the control as 100.

As is seen from Figs. 6—13, the sensitivity of different fungi to biocides varies greatly. Mycorrhizal fungi are the most sensitive and soil molds the most tolerant. Common soil molds generally tolerate concentrations 100—1000 times higher than mycorrhizal fungi. According to these experiments, *Cenococcum graniforme* is the most sensitive species; this is rather surprising since the drought resistance of *Cenococcum* has drawn attention in several connections (e.g. WORLEY & HACSKAYLO 1959; TRAPPE 1963). Pseudomycorrhizal *Mycelium radicis atrovirens* was approximately as tolerant as the common molds. Regarding sensitivity, the saprophytic basidiomycete *Collybia dryophila* was between mycorrhizal fungi and soil molds, as well as the damping-off parasite *Rhizoctonia solani*. These results agree well with those of RENNERFELT (1954), who showed that mycorrhizal fungi are more sensitive to selective brush-killers than saprophytic and blue-staining fungi.

The relative sensitivity of different fungi to chemicals was not always the same. This was well illustrated by the otherwise tolerant *Mycelium radicis atrovirens*, which was very sensitive to Zineb, and by *Rhizoctonia solani*, which tolerated PCNB surprisingly well.

Trichoderma was tolerant in general but especially to formaldehyde and allyl alcohol. This is in good agreement with the results of the field experiments, where these two chemicals strongly promoted *Trichoderma*. Likewise WARCUP (1952) found that on agar *Trichoderma* tolerates formaldehyde better than other molds or damping-off fungi. Furthermore, *Trichoderma* has proved to be able to utilize allyl alcohol as a nutrient (JENSEN 1959).

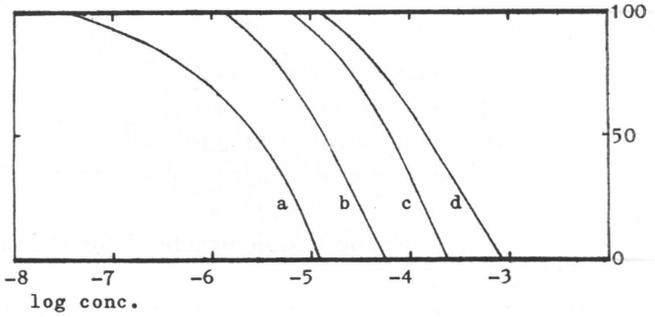

Fig. 6. Relative growth of fungi on agar at different concentrations of formaldehyde

a. *Boletus luteus, B. variegatus, Cenococcum graniforme, Corticium bicolor, Collybia dryophila*
b. *Rhizoctonia solani*
c. *M.r. atrovirens, Penicillium spinulosum, Verticillium terrestre*
d. *Trichoderma viride*

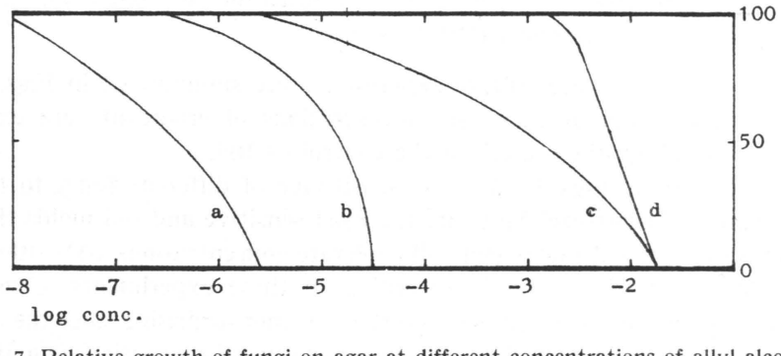

Fig. 7. Relative growth of fungi on agar at different concentrations of allyl alcohol

a. *Boletus luteus, Cenococcum graniforme*
b. *Boletus variegatus, Corticium bicolor, Rhizoctonia solani*
c. *Collybia dryophila, M.r. atrovirens, Penicillium spinulosum, Verticillium terrestre*
d. *Trichoderma viride*

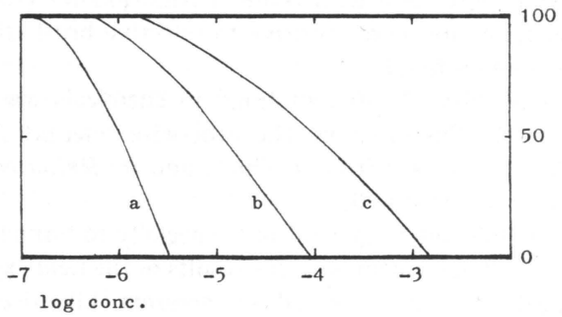

Fig. 8. Relative growth of fungi on agar at different concentrations of SMDC (Vapam)

a. *Cenococcum graniforme*
b. *Boletus luteus, B. variegatus, Corticium bicolor, Collybia dryophila, Rhizoctonia solani*
c. *M.r. atrovirens, Penicillium spinulosum, Verticillium terrestre, Trichoderma viride*

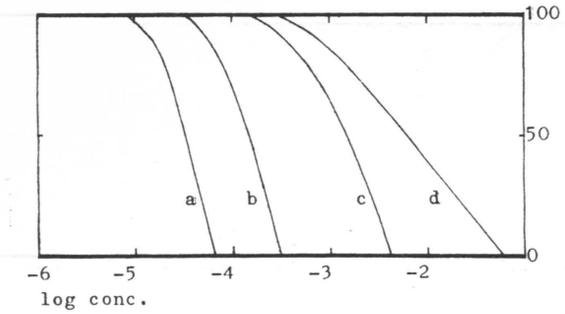

Fig. 9. Relative growth of fungi on agar at different concentrations of Simazine

a. *Cenococcum graniforme*
b. *Corticium bicolor*
c. *Boletus luteus, B. variegatus, Collybia dryophila, Rhizoctonia solani*
d. *M.r. atrovirens, Penicillium spinulosum, Verticillium terrestre, Trichoderma viride*

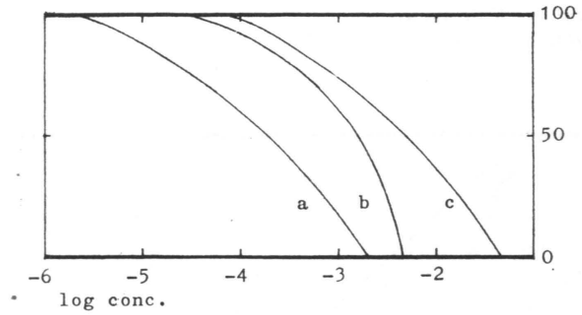

Fig. 10. Relative growth of fungi on agar at different concentrations of Dalapon

a. *Cenococcum graniforme*
b. *Boletus luteus, B. variegatus, Corticium bicolor, Collybia dryophila*
c. *Rhizoctonia solani, M.r. atrovirens, Penicillium spinulosum, Verticillium terrestre, Trichoderma viride*

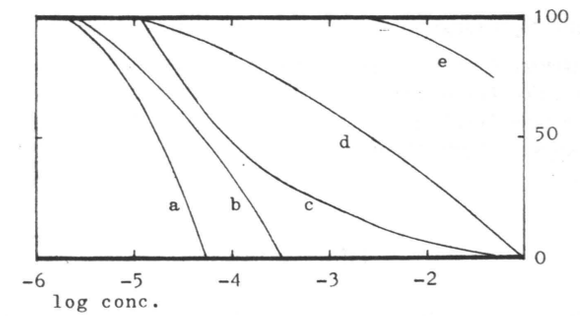

Fig. 11. Relative growth of fungi on agar at different concentrations or PCNB (Brassicol)

a. *Cenococcum graniforme, Corticium bicolor, Collybia dryophila*
b. *Boletus luteus, B. variegatus*
c. *Penicillium spinulosum*
d. *M.r. atrovirens, Verticillium terrestre, Trichoderma viride*
e. *Rhizoctonia solani*

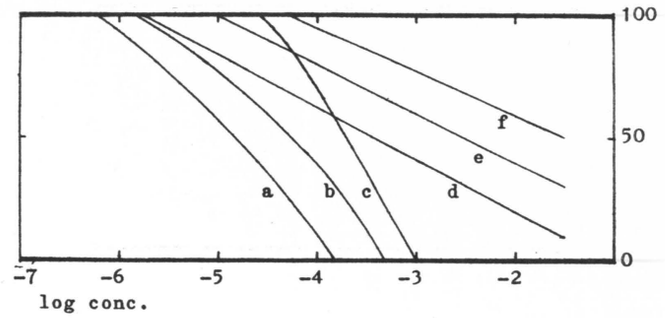

Fig. 12. Relative growth of fungi on agar at different concentrations of Cu oxychloride (Soltosan)

a. *Boletus luteus, Cenococcum graniforme*
b. *Boletus variegatus, Corticium bicolor*
c. *Collybia dryophila, Penicillium spinulosum*
d. *Rhizoctonia solani, Verticillium terrestre*
e. *Trichoderma viride*
f. *M.r. atrovirens*

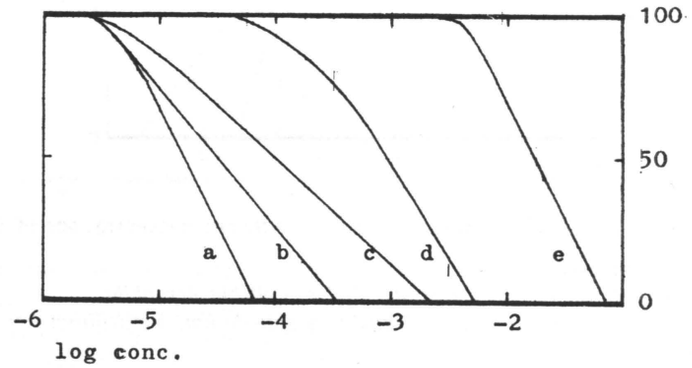

Fig. 13. Relative growth of fungi on agar at different concentrations of Zineb (Dithane Z-78)

a. *Cenococcum graniforme, Corticium bicolor, M.r. atrovirens*
b. *Boletus variegatus, Collybia dryophila*
c. *Boletus luteus, Rhizoctonia solani*
d. *Penicillium spinulosum, Verticillium terrestre*
e. *Trichoderma viride*

Discussion

In the first growing season mycorrhizal infection commences fairly late even under normal conditions, *i.e.* 6—7 weeks after seeding and 3—4 weeks after the formation of the first short roots. This must be taken into consideration when the effect of biocides on mycorrhiza formation is discussed.

Soil disinfectants are used before seeding and they are supposed to evaporate or disintegrate in a few days or 1—2 weeks, *i.e.* long before the time of mycorrhiza formation. Their effect on mycorrhizae depends, therefore, on two points, viz. how completely they can destroy the mycorrhizal fungi, and what chances the fungi have to reinvade the soil and infect the roots. As is shown by pure culture experiments, the soil disinfectants used are also very poisonous to mycorrhizal fungi; thus complete extermination of fungi is probable in the soil layer penetrated by the poisons. The depth of the influence of the chemicals depends on the strength of the treatment and also on the soil properties. The larger the doses of chemicals used, the deeper they penetrate and, correspondingly, the greater is the delay in mycorrhiza formation. Normal application hardly affects soil deeper than 5 cm; this penetration is sufficient to control weeds and parasitic organisms.

The delay of mycorrhizal infection does not seem to harm the seedlings in any way. Even in untreated soil the seedlings grow for a great part of their first growing season without mycorrhizal association; the mycorrhizal phase of the first summer is fairly short and, furthermore, covers the late part of the season when the seedlings have already largely completed their annual growth. In other experiments (unpublished) the authors have also observed that when mycorrhizal and nonmyccorrhizal seedlings are grown under similar conditions, hardly any difference in their size or vigor can be noticed at the end of the first season. Likewise, in the well-known nursery experiments in different parts of the world, where coniferous seedlings without mycorrhizal association suffered and died, the difference between mycorrhizal and non-mycorrhizal pine seedlings only became apparent at the time of the formation of the first fascicle needles (Hatch 1937; McComb 1943). Under Finnish conditions fascicle needles never grow before the second summer.

In the second summer, however, differences of mycorrhizal relations between treated and control plots disappeared. Accordingly, the influence of biocides on mycorrhizae, when applied in the customary concentrations, does not extend beyond the first growing season.

Different chemicals, however, differed in their effect on the mycorrhizae. Methyl bromide and SMDC retarded mycorrhiza formation distincly, while formaldehyde and allyl alcohol had no effect, except perhaps in very high concentration. The difference is hard to explain, for in pure culture experiments the critical concentration of formaldehyde and allyl alcohol was approximately the same as that of SMDC (ca. 0.001 %). Apart from not retarding mycorrhizae, formaldehyde and allyl alcohol promoted seedling growth and, furthermore, favored *Trichoderma viride* in the soil. What kind of relationship the increase of *Trichoderma* has to the other two phenomena is obscure. *Trichoderma* is known to be antagonistic to many other fungi, e.g. damping-off fungi (Bollen 1961) and the root-rot fungus (Rishbeth 1957). Melin (1934) showed that *Trichoderma*

is able to decompose some humus substances which are poisonous to mycorrhizal fungi. However, it is not known whether this happens in natural soils too. The relationship between *Trichoderma*, mycorrhizal development, and promotion of seedling growth deserves further investigation.

The herbicides and fungicides which were included in the nursery experiments had no harmful influence on mycorrhiza formation. It must be remembered, however, that the chemicals were applied before seeding, *i.e.* several weeks before the normal time of mycorrhiza formation. It is evident, however, that the herbicides and fungicides did not destroy the mycorrhizal fungi of the surface soil to such a degree as did methyl bromide and SMDC, for instance. In pure culture experiments mycorrhizal fungi proved several times more sensitive than parasitic and indifferent soil molds to herbicides and fungicides. Therefore, when new chemicals are introduced into nursery practice their possible harmful effects on useful soil fungi should be taken into consideration and such effects should be studied before new chemicals are recommended.

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SELOSTUS:

KASVINSUOJELUAINEIDEN VAIKUTUS MYKORITSAIN KEHITYKSEEN METSÄTAIMITARHOISSA

Nykyaikainen taimitarhatekniikka käyttää monenlaisia kemikaaleja rikkaruohojen sekä tuhosienien ja -hyönteisten torjuntaan. Varsinkin sienien tuhoamiseen tarkoitettujen aineiden kohdalla on pelättävissä, että ne vaikuttavat haitallisesti myös hyödyllisiin organismeihin, ennen muuta puiden kanssa symbioosissa eläviin mykoritsasieniin ja sitä kautta myös puun taimiin.

Tutkimuksen alkuosassa tarkastellaan puun taimien ja niiden mykoritsain kehitystä normaaliolosuhteissa, kun mitään kasvinsuojeluaineita ei ole käytetty. Männyn ja kuusen taimien ensimmäisenä kesänä mykoritsainfektio tapahtuu verraten myöhään eli heinä-elokuun vaihteessa, ts. 6—7 viikkoa kylvön ja 3—4 viikkoa ensimmäisten lyhytjuurten syntymisen jälkeen. Taimien ja niiden juuristojen kehitys on esitetty kuvissa 2—4.

Mykoritsainfektion tapahduttua se leviää nopeasti, ja jo toisena kesänä miltei kaikki lyhytjuuret kehittyvät mykoritsoiksi kohta syntyessään. Eräitä mykoritsain infektio- ja kehitysmalleja on esitetty kaavamaisesti kuvassa 5.

Kasvinsuojeluaineiden vaikutusta mykoritsain kehitykseen tutkittiin kenttäkokein taimitarhassa (ss. 18—23) sekä samojen aineiden vaikutusta itse sieniin puhdasviljelykokein laboratoriossa (ss. 24—28). Edelleen seurattiin taimitarhassa vaikutusta myös maan muuhun mikrobistoon (ss. 23—24).

Maan sterilointiaineet, kuten metylbromidi ja SMDC (Vapam), joilla maa käsitellään ennen kylvöä rikkaruohojen ja taimipoltesienien hävittämiseksi, vaikuttivat mykoritsain muodostumista viivästyttävästi (Taul. 2 ja 3). Viivästys oli kuitenkin niin vähäinen, että toisena kesänä ero käsittelemättömään maahan verrattuna hävisi. Formaliini ja allylalkoholi eivät hidastaneet mykoritsain muodostusta, paitsi suuria väkevyyksiä käytettäessä (Taul. 4 ja 5). Maan muuhun mikrobistoon formaliini ja allylalkoholi vaikuttivat siten, että *Trichoderma viride* lisääntyi voimakkaasti muiden homelajien kustannuksella (Taul. 7). Kokeilluilla rikkaruohomyrkyillä (Simatsiini ja Dalapon) samoin kuin ruiskutteina käytettävillä sienimyrkyillä ei ollut selvää haitallista vaikutusta mykoritsain kehitykseen.

Puhdasviljelykokeet osoittivat, että mykoritsasienet ovat kokeilluille myrkyille monin verroin herkempiä kuin yleiset parasiittiset tai saprofyyttiset maasienet (Kuv. 6—13). Uusia kasvinsuojeluaineita kehitettäessä on niiden mahdollinen haittavaikutus hyödyllisiin mikrobeihin siis otettava aina huomioon.