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Title: THE INFLUENCE OF PRE-HARVEST KILLING OF WESTERN HEMLOCK ON SUBSEQUENT STUMP INVASION BY Fomes annosus

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This study concerns the effect of pre-harvest killing of commercial sized, forty year old western hemlock (Tsuga heterophylla (Raf.) Sarg.) on subsequent stump infection by Fomes annosus (Fr.) Karst. Further evaluations were made on tree drying, loosening of bark, wood deterioration and rate of crown and cambial death. Findings on the susceptibility of injection axe cuts to Fomes annosus are included.

Two herbicides MSMA and Tordon 101 were applied during four seasons; incubation periods of six, nine and twelve months were allowed to lapse between treatment and felling.

Epidemiology of Fomes annosus in commercial thinnings of western hemlock was subject to deliberate prophylactic treatment by pre-harvest killing of trees with MSMA. At the same time the

trees dried faster, and bark was more easily removed. Wood deterioration and breakage increased, but at levels tolerable to pulp operations. The degree of stump infection reduction, drying and bark removal was directly related to the amount of cambial kill by the herbicide.

Tordon produced variable results, with stump infection sometimes exceeding that of the control. Furthermore, Tordon injection axe cuts were found to be highly susceptible to Fomes annosus infection.

The behavior of Fomes annosus is thought to be responsive to cambial condition. The two herbicides produced different cambial responses, hence different responses of the pathogen.

The Influence of Pre-harvest Killing of Western Hemlock on Subsequent Stump Invasion by *Fomes annosus*

by

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INTRODUCTION

Root and butt rot (*Fomes annosus* (Fr.) Karst) of western hemlock (*Tsuga heterophylla* (Raf.) Sarg.) is widespread in coastal forests of the Pacific Northwest. The disease is most familiar in the northwest as butt decay of old growth, but recent reports by Hunt and Krueger (1962) and Driver and Wood (1969) indicate a high potential for occurrence in young growth western hemlock stands.

Fomes annosus is known to be a virulent pathogen of many tree species in temperate Europe and Eastern North America where there has been a definite increase in incidence of disease with intensified forest management, especially thinning. Northwest forest managers are concerned that *F. annosus* may endanger thinning programs in western hemlock, which responds well to management and is economically attractive. The silvicultural merits of hemlock and the projected demand for wood, provide incentive for the development of control.

It is generally agreed that *F. annosus* flourishes in logging wounds and stumps (Hodges, 1969), whose freshly exposed living tissues are highly susceptible to *F. annosus* infection. Spread to adjacent trees occurs via root grafts or contacts. These close

associations of roots, abundant in second growth hemlock stands, further increase the risk.

Throughout the world control of stump infection has been obtained by chemical treatment of fresh stumps (Sinclair, 1964). Several workers have reported only variable success with these methods on hemlock stumps in the northwest (Edmonds, Driver and Russell, 1969; Weir, 1969).

The primary objective of this study was to test the effect of pre-harvest killing of western hemlock by herbicidal injection on subsequent stump invasion by F. annosus. Further objectives were to evaluate effects of the treatments on tree drying, loosening of bark, wood deterioration and rate of crown and cambial death. Findngs on the susceptibility of the injection axe cuts to F. annosus are also included.

LITERATURE REVIEW

General

Cultural characteristics of Fomes annosus (Fr.) Karst, family Polyporaceae of the class Basidiomycetes including colony morphology, temperature phenomena, nutrient requirements and growth rates have been described by Roll-Hansen (1940), Miller (1943), Rennerfeldt (1944, 1953), Risbeth (1951a), Etheridge (1955), Cowling and Kelman (1964) and Gooding, Hodges and Ross (1966).

The destruction of coniferous forests by F. annosus was first reported by Robert Hartig in 1874 (Sinclair, 1964). He noted both red-rot of the butts of older trees, and gaps in young stands via direct killing. He reported that conifers, in general, were susceptible. Since then reports from temperate zone areas throughout Europe and North America have revealed the wide distribution of damage caused by the pathogen.

The nature and chronological increase in the rate of damage in Great Britain is well documented by Hiley (1919), Risbeth (1950, 1951a, 1951b, and 1967), Low and Gladman (1960), Gladman and Low (1963) and Gladman and Greig (1965) among others. Descriptions of damage from the European continent are reviewed by Peace (1962) and Sinclair (1964). The pathogen, based on the number of reports

of damage, apparently has been much more serious in Germany, Scandinavian coutries and the Netherlands than in southern Europe.

Damage in Europe, although present in natural stands, has correlated with intensive management, especially in thinned plantations on previously non-forested land. Economic losses have warranted efforts to develop control programs.

Fomes annosus was not considered to be a primary pathogen in the United States prior to the early 1940's, however, the ubiquitous distribution of the fungus was well known in the southeast (Von Schrenk, 1898 and 1900), in California (Meinecke, 1914), in New York (Sumstine, 1917), in Michigan (Kaufman, 1917), in the northern Rocky Mountains (Weir, 1917 and Weir and Hubert, 1919), in North Carolina (Haasis, 1923), in Vermont (Spaulding, 1930), in Colorado (Shope, 1931), in Washington and Oregon (Boyce, 1932), in Connecticut (Stoddard, McDonnell and Hickock, 1939) and in the Great Lakes Region (Baxter, 1941). The lack of serious damage was, no doubt, correlated with lack of intensive management. Reports of damage have proliferated since 1940. Severe damage has been reported in the southeast in eastern white pine (Pinus strobus L.), (Hepting and Downs, 1944) and Boyce (1962), eastern red cedar (Juniperus virginiana L.) (Miller, 1943) and in southern yellow pine stands (Campbell and Hepting, 1954), (Powers and Boyce, 1961), (Driver and Dell, 1961a, b) and Powers and Vorrall (1962). These reports

indicate that damage is much more severe in thinned plantations than in natural stands. Damage in pulpwood stands of loblolly (Pinus taeda L.) and slash pine (Pinus elliotti var. ellioti Engelm.) has become so serious as to warrant clearcutting prior to rotation age (Southern Forest Pest Reporter, 1969).

Experience in pine stands of the northeast parallels that of southeast (Welch and Stone, 1953; Miller, 1960; Mook and Eno, 1960; and Stambaugh et al., 1962). Mook and Eno (1960) emphasize that the disease is particularly damaging in 20- to 30-year-old thinned plantations on formerly cultivated land.

Fomes annosus has caused widespread mortality and butt rot in stands of ponderosa (Pinus ponderosa Laws.) and Jeffrey pine (Pinus jeffreyi Grev. and Balf.) in California (Olson, 1941; Wagener and Cave, 1946). Bega (1962) found the fungus attacking 26 species of pine in a genetics arboretum at Placerville, California. Recent papers (Bega, 1965) and Cobb and Barber (1968) have emphasized the present and potential damage in California, and observe that greatest losses are associated with stumps left from thinning. Bega (1962) postulates that increased forest management is contributing to a higher incidence of the pathogen in present day stands.

Fomes annosus is apparently non-specific in host attack. Sinclair (1964) published a host list that includes over 50 species of angiosperms and 80 species of gymnosperms. He notes that,

although many hardwood and brush species are hosts, reports of severe damage is limited to conifers. However, the importance of hardwoods and brush in disease epidemiology should not be overlooked.

British workers, Risbeth (1957) and Greig (1962), have discussed the epidemiology of the disease on former hardwood sites. Risbeth found that pines were sometimes killed by infection from stumps of birch (Betula spp.), gorse (Ulex europseus L.) and broom (Sarothamnus acoparius Wimmer). In California Smith, Bega and Tarry (1966), have found the fungus on several species of manzanita (Arctostophylos spp.), which frequently precedes pine in the vegetative succession. The spectrum of host species is apparently broad enough to discriminate against crop rotation as a means of disease control.

Signs and symptoms of the diseases are excellently described and illustrated by Hartig (1894), Hiley (1919), Wagener and Cave (1946), Risbeth (1951a, 1957) Mook and Eno (1961) and Gladman and Greig (1965). The perennial sporophores are often observed fruiting from stumps, on downed logs and on roots, root crotches and butts of living trees. Englerth (1942) noted that sporophores of the fungus were frequently found in the root crotches of the fluted butts of old growth hemlock in coastal regions of the Pacific Northwest.

Symptoms of the disease vary with species and the age of the host. External symptoms are not characteristic only of F. annosus

and may be confused with diseases caused by other organisms, site deficiencies or suppression. Above ground symptoms of the disease are reduced growth, crown discoloration, tufting of needles at branch ends, and tree death. Trees may be severely decayed without external evidence of the disease.

The work of Hiley (1919) is considered a classic descriptive work on decay of F. annosus. Color photographs of decay in several conifers, excluding hemlock, are presented in the work of Gladman and Greig (1965). Englerth (1942), Buckland, Foster and Nordin (1949) and Wright and Issac (1956) present descriptions and black and white photographs of decay in western hemlock specifically.

The historical development of an understanding of establishment and spread of F. annosus in forest stands is quite interesting and involves considerable controversy; development of only the most accepted and current concepts is discussed here.

Robert Hartig, in 1874, revealed the ability of the fungus to infect healthy trees via contact between the roots of diseased and healthy trees (Sinclair, 1964). This method of spread has been substantitated by Olson (1941), Hepting and Downs (1944), Wagener and Cave (1946), Day (1948), Risbeth (1950), and Towers (1964), among others.

Infection centers may persist through successive generations of trees on a given site, or may arise during the development of the

stand. Certain stands have, no doubt, contained F. annosus infection centers for centuries, the disease being transmitted from generation to generation by root contact. Even when stands are clear cut the residual stumps may harbor the fungus well into the next rotation. According to Low and Gladman (1959) Fomes annosus may remain active for decades on old stumps, the oldest recorded being a European larch (Larix decidua Mill.) stump 63 years old. Once the fungus is established in an area it is capable of remaining for long periods of time.

Means of establishment in stands not previously infected has been cause for considerable discussion. Risbeth (1950 and 1951c) showed that infection occurs only through living roots. He noted the fungus to be incapable of competing with saprophytes in dead tissue and of establishing itself and growing in unsterilized soil. His work refuted the previously held theory that F. annosus entered the boles of trees through dead roots.

Hepting and Downs (1944) were the first to associate the disease with thinnings, an observation that has since been confirmed (Risbeth, 1950, 1951a, b, c; Gladman and Low, 1959; Powers and Verral, 1962; Powers and Boyce, 1963; Sinclair, 1964; and Yde Andersen, 1968). While Highley (1919) showed the role of infected stumps in infection of remaining trees by root contact, it remained for Risbeth (1950, 1951a) to prove that newly cut stumps could be colonized by air borne

spores of F. annosus and that the mycelium from these spores could penetrate stump root systems and subsequently, by root contact or graft, enter roots of neighboring healthy trees. Stump infection and subsequent spread of the fungus through entangled root systems is now recognized as the major means of establishment of F. annosus in previously un-infected stands. Other possible methods of establishment are known to exist.

Jorgensen (1961), working with red pine (Pinus resinosa Ait.) in Canada, found that spores deposited on seedling roots during transport could infect the seedlings with subsequent wounding while planting.

Rhodes and Wright (1946) demonstrated that logging scars on western hemlock were highly susceptible to F. annosus infection. Low and Gladman (1960) reported that F. annosus infection may also occur through pruning wounds, logging wounds to bole and roots, wounds from removal of double leader in Christmas trees and animal caused wounds. Witcher and Beach (1962) also report pruning wounds of slash pine to be infected by F. annosus.

The possibility of infection of roots by spores filtering through soil has been demonstrated (Risbeth, 1951a; Hendrix and Kuhlman, 1964). When a suitable substrate (living tissue) was present F. annosus spores filtering through the soil were able to germinate and compete successfully with soil saprophytes. Work of Risbeth (1951a)

and Kuhlman (1969) has shown that spores of the pathogen exist and can remain viable in the soil at the root level for periods up to ten months.

Risbeth (1951b) and Hodges (1968) have shown that stump roots may be infected by spores filtering through the soil. Hodges found that five to ten percent of the infection of stumps may arise in this manner. However, infection of living tree roots in this way is questionable. Wallis (1961) found spore inoculum consistently failed to infect uninjured Scotch pine (Pinus sylvestris L.) roots. Wallis also noted considerable difficulty obtaining infection of healthy roots using various other inoculation techniques.

While other possible methods of establishment of F. annosus have been demonstrated, workers concur that stump infection is the main avenue of entrance and spread in forest stands. Only in western hemlock stands, where considerable infection of logging wounds also occurs does an apparent exception exist.

By root contacts or grafts the disease spreads radially from the infected stumps of trees to produce somewhat circular infection centers in the stand. Rate of radial spread appears variable. Risbeth (1949) has estimated an average of six feet per year in pine plantations on land formerly under agricultural cultivation in Great Britain. Driver (1968) reports that slash pine may show crown symptoms one year after thinning, with mortality occurring two years after thinning.

On the other hand, in Great Britain, Low and Gladman (1960), working with species susceptible to butt rot but not mortality, report that from six to ten years may elapse before stain appears in a tree adjacent to a stump infected after thinning.

No experimental evidence exists on differential of susceptibility of various conifers to the pathogen. Observations on plantations in England by Risbeth (1951b), Low and Gladman (1960), Gladman and Greig (1965) and Gibbs (1968) generally agree that pines are more resistant to attack than other conifers, but that under certain conditions damage in pines can become extremely severe. They also concur that species of pine, when attacked, are more susceptible to killing than other genera, where the disease is most frequently expressed as a butt end root rot. However, Gibbs (1968) reports that when other conifers are grown on typical pine sites they too may suffer mortality. These workers agree that in general western hemlock is the most susceptible tree to attack in Europe and that damage is generally expressed as a root or butt rot rather than mortality. However, many of these observations on susceptibility were confined to plantations on exotic sites, and may or may not be indicative of susceptibility of hemlocks in its native habitat.

Weather influences spore production, spore dispersal, stump colonization by F. annosus and its fungal competitors. Effects of temperature on sporophore and basidiospore production are presented

in papers by Risbeth (1951a), Meredith (1959), Yde Andersen (1962), Stambaugh et al. (1962), Reynolds and Wallis (1966), Wood (1966), Kallio (1968), and Driver and Ginns (1969). These authors agree that spore production by F. annosus ceases only during freezing temperatures and is otherwise present throughout the year. Inoculum loads, of course, may vary with seasonal and even local changes in the environment.

Effects of temperature and moisture on stump surface colonization by F. annosus have been examined by Yde Andersen (1962), Driver and Ginns (1964, 1969) and Gooding et al. (1966). They have found that the effects of temperature and moisture by F. annosus on stump surface colonization are confounded with the effect of competing fungi (Driver and Ginn, 1969). However, freezing weather and stump surface temperatures over 95 °F are considered directly inhibitory to surface colonization by F. annosus.

Sinclair (1964), Froelich et al. (1966) and Towers and Stambaugh (1968) report that in general soil conditions unfavorable either for the host or competing microorganisms favor the disease. High acidity, sand or clay content, low organic matter and extremes in soil moisture regimes have been found to favor the disease.

The effect of soil microorganisms on F. annosus epidemiology have been widely investigated. Gibbs (1968) and Hyppel (1968) have stressed the effect of soil factors and competing microorganisms.

In areas where populations of antagonistic soil organisms are low; as in depleted agricultural land with low soil organic matter and high pH, growth of F. annosus is epiphytic and damage is most severe. On the other hand, where endophytic growth is enforced, as in soild with abundant antagonistic microflora, spread of the fungus is much slower (Risbeth, 1950, 1951c). When the pathogen is restricted to slow growth in the stump root system the possibility is enhanced that stump roots will senesce enough to become invaded by competing soil saprophytes preventing F. annosus spread to points of contact with roots of living trees. Risbeth (1951c) notes that stands with high stump infection rates after thinning may suffer little damage if antagonistic soil and root microflora restrict the pathogen to the stumps.

Control

Many methods of control have been recommended, mostly prior to knowledge of the effect of stumps on the epidemiology was known (Risbeth, 1952, and Sinclair, 1964). Risbeth (1951a, 1952) first presented methods aimed at reducing disease incidence by preventing stump infection. Risbeth (1951a) and Meredith (1959) tested coinciding cutting operations with conditions adverse to production and survival of spores. This method, impractical in Great Britain, has shown some promise elsewhere (Yde Andersen, 1962; Stambaugh et al., 1962; Driver, 1963; Sinclair, 1964, Driver and Ginns, 1964;

1968, 1969; Gooding et al., 1966; Cobb and Barber, 1968).

Nearly two decades ago Risbeth (1952) suggested the use of chemical protectants and fungal competitors to exclude F. annosus from stumps. Since then Risbeth and many others have tested these methods of controlling stump infection. Some of these procedures have reached local operational development but none has gained universal acceptance.

Risbeth has developed both biological (Risbeth, 1963, 1968) and chemical (Risbeth, 1959a, 1959b, 1967) stump control methods into operational measures. Driver and Ginns (1968) have listed a multitude of papers by other workers following Risbeth's principles. These methods have not proven adequate in the Pacific Northwest (Driver and Ginns, 1968) and Edmonds et al., 1968).

Risbeth (1959) pointed out that chemical stump protectants may have different modes of action against F. annosus. Preservative chemicals to protect stumps from all fungal invaders, high nitrogen compounds encourage stump colonization by microorganisms competitive or antagonistic to F. annosus; herbicides to kill stump tissues, thereby removing selectivity for parasites and allowing colonization by saprophytes.

Driver and Ginns (1968) have classified chemicals currently being used as stump protectants as to their mode of action. Borax and creosote act directly against F. annosus. Urea, ammonium

bifluoride and sodium nitrite assist in biological control by promoting growth of competing fungi.

Other reports of control attempts involve reduction of substrate by clear felling around infection centers (Linnard, 1965) and habitat poisoning by soil fumigation (Houston and Eno, 1969).

The Disease West of the Cascades in the Pacific Northwest

Early papers of Boyce (1929, 1932) indicated that F. annosus was of little significance in the coastal regions of the Pacific Northwest. He reported the fungus was rarely found on the Olympic peninsula of Washington except on down logs and stumps, and caused only a small percentage of the decay in old growth western hemlock and Douglas fir. From the results of a decay study in Douglas fir Boyce (1932) went as far as to say "The conifer root fungus F. annosus, which frequently kills young Douglas firs in Europe, has not been found as a parasite in this region, although it occurs occasionally on old stumps and down trees."

Englerth (1942), in a study that encompassed the area west of the Cascades in Washington and Oregon, reported Fomes annosus to be responsible for 21 percent of the total decay in old growth hemlock. Rot columns were observed extending 40 feet up the tree, with an average of 17 feet. Sixty percent of the infections were reported to have entered through roots, 35 percent via scars from falling

trees, with the remainder attributed to fire scars and unknown causes. The fungus was found as a root rot in younger age classes, but no infections were found in trees less than 60 years of age. He postulated the age of infection (based on Meinke's (1916) terminology) to be between 60 and 85 years.

Englerth found Fomes pini (Thore) Karst. (19.2%) and Fomes applanatus (Pers.) Gill. (17.1%) causing appreciable decay. F. pini was the most important agent in Cascade Mountain plots while F. annosus was most damaging on the coast. Armellaria mellea (Vahl.) Quel., Poria subacida (Pk.) Sacc., Poria colorea Engl. and Polyporus schweinitzii Fr., also were found primarily in butts and roots. In all, 16 species were found causing decay in western hemlock.

The amount of decay caused by a fungal species varies considerably from one area to another. Buckland Foster and Nordin (1949), found that Fomes pinicola (Sw.) Cke. (40.8%), F. pini (12.8%) and P. subacida (10.2%) to cause more decay than F. annosus (7.3%) on Vancouver Island. Foster, Craig and Wallis (1954) found 26 species of decay fungi attacking hemlock in the Upper Columbia region of British Columbia. They found, as did Weir (1918), that Echinodontium tinctorium E. and E. was the principal agent of decay in Inland Empire stands of western hemlock. Fomes annosus was responsible for only 0.2% of the total decay in this area. Foster, Browne, and Foster (1954) found Fomes pini to be responsible for nearly 50 percent

of the total hemlock decay in northwest British Columbia. Fomes annosus ranked sixth with 1.1% of the total decay.

Partial cutting in mature stands increases the incidence of Fomes annosus in the residual trees (Englerth and Issac, 1947; Rhoads and Wright, 1946; Wright, Rhoads and Issac, 1947; Wright and Issac, 1956; and Shea, 1960). Infection entry is through logging wounds on boles and roots. Wright and Issac (1956) reported that 63 percent of the logging scars, broken tops, and sunscald lesions following harvest to be infected by Fomes annosus in coastal areas, forty percent in Cascade Mountain stands.

Information on damage in young stands is deficient, although some findings have been published recently. In the Puget Sound area, Hunt and Krueger (1962) found that 61 percent of the wounds on young hemlock contained decay, and that F. annosus was responsible for 80 percent of the decay volume. Decay increased with the proximity of the wound to the ground and the size of the wound. They estimated that 5.5 percent of the net periodic annual increment after thinning was lost due to decay of wounds incurred during thinning. Short rotations were suggested to reduce loss.

Driver and Wood (1968) examined both thinned and unthinned stands at 20- to 60-year-old hemlock. Their results indicate that substantial infection may occur through stumps. They found up to 90 percent probability of stump infection after thinning. Work

by Edmonds, Driver and Russell (1969) have substantiated their findings.

Information available to date seems to indicate that increased infection of hemlock stands can be expected to accompany with increased members of stumps and bole wounds associated with intensive management.

Edmonds, Driver and Russell (1969) and Weir (1969) have tested different formulations of borax to control stump infection in compounds to control stump infection in western hemlock. They report that borax-glycerol solutions and borax-glycol were generally more effective than dry borax, but still permitted up to ten percent infection. Dry borax, highly effective in southeastern United States was ineffective, probably as a result of removal of the chemical from the surface by rain.

Substrate Colonization

Fomes annosus is most frequently thought of as a root parasite, although bole entry via wounds is well documented. The fungus has demonstrated a certain capacity for saprophytism. It has been observed persisting in dead organic material, such as dead roots and stumps, for long periods. However, initial colonization of dead material, unless under sterile conditions, is extremely rare if not non-existent. Hartig (1894) considered F. annosus a saprophyte

in roots and stumps only if it it had entered the host while still alive. Although several workers have since referred to the fungus as saprophyte, the work cited below convincingly supports the view of Hartig.

Risbeth (1950) has presented points of evidence that demonstates the poor competitive ability of the fungus on saprophytic substrates. First, no worker has been able to grow the fungus on forest humus or soil, unless these substrates were previously sterilized. Second, the fungus does not colonize forest litter, for it has never been isolated from needles or twigs that have been embedded in sporophores for some time. Third, in stands heavily infected with F. annosus, root systems of pines killed by suppression were colonized to a much less degree by the pathogen than non-suppressed trees. Fourth, basidiospores will germinate readily and produce mycelium on surface sterilized roots, but not on unsterilized roots. Infection can be obtained on unsterilized roots only if fresh wounds are made shortly before inoculation.

Work dealing with fungal colonization of stumps has further revealed the poor competititive nature of F. annosus. European workers, Risbeth (1951), Meredith (1960), Yde Andersen (1962), have shown that pine and spruce stumps are not likely to be colonized by F. annosus spores that land on the surface ten or more days after felling. Meredith (1960) reports freshly exposed pine stumps are colonized by a limited number of species, considering that the species

represented among air spora number in the thousands. He reasoned that stump tissues are still alive for some time after felling, and initially possess the normal resistance of living tissue in the intact tree. This resistance deteriorates in time, usually within one or two weeks, and the stump surface becomes increasingly susceptible to saprophytic invasion. Stumps thus lose the capacity to serve as selective substrates for the few specialized fungi that are capable of colonizing freshly exposed wood surfaces.

Risbeth (1959c) has found the initial selectivity of freshly exposed pine wood to be very useful in sampling the air spora for the presence of F. annosus. The technique involves brief exposure of freshly cut pine disks to air spora and counting the number of colonies of F. annosus on the disk after a period of incubation. He points out that when the pine disks are autoclaved before exposure the selectivity for F. annosus is lost and the disks are rapidly overrun with molds. Thus, he concluded that F. annosus colonization is favored by the initial resistance of the freshly exposed wood to saprophytic organisms. Subsequently, he employed this principle to control stump colonization by F. annosus. He reasoned that the sooner a stump and its root system were dead the more they would be colonized by saprophytes to the exclusion of the pathogen. In support of this, he applied di-sodium octaborate, ammonium sulfamate and urea to kill stump surface tissues at the time of felling. The result was a

rapid colonization of molds rather than primary wound colonizers including F. annosus.

Cobb and Schmidt (1964), Ross (1968) and Cobb and Barber (1968), have found in the United States that stump susceptibility decreases with time after felling in white pine, loblolly pine, ponderosa pine, redwood (Sequoia sempervirens (D. Don) Endl.) and Douglas fir. Cobb and Barber (1968) however, observed that redwood and ponderosa pine stumps may increase in susceptibility to F. annosus the first week after cutting, before reverting to a decreasing trend. The above workers generally agree that the changes in susceptibility of stumps to F. annosus is due to alterations in tissue resistance to invading fungi. They also point out that seasonal effects may influence both changes in substrate and abundance of competing fungi as well as the condition of tissue at the precise time of felling. The point of emphasis here, however, is that stumps become less susceptible to F. annosus with increasing time after felling and that this phenomena is due primarily to the loss of the stump's resistance to saprophytic invaders; these saprophytes, moreover, act negatively on F. annosus colonization through mechanisms of competition and antagonism.

PROCEDURES

The Study Approach

As seen from the literature F. annosus has a competitive advantage over saprophytic fungi on freshly exposed wood surfaces of living trees. When trees are felled the tissues in the remaining stump still possess some of the resistance mechanisms to invasion by microorganisms that were present when the tree was intact. These resistance mechanisms exclude saprophytic organisms, but not the more virulent parasites, such as F. annosus. As the stump tissue dies this residual resistance is lost and the stump surface becomes increasingly susceptible to colonization by saprophytes, and is no longer a selective substrate for the small group of fungi capable of colonizing freshly exposed wood.

The specific hypotheses under test in this paper are: 1) destruction of tissue selectivity of living trees by killing the tree prior to cutting will render stumps non-selective to F. annosus, and receptive to saprophytes. And 2) that the competitive development of saprophytes will exclude F. annosus, from the stumps.

Although these hypotheses have never been tested in temperate zones, work in tropical areas, with Armellaria mellea offers some support. Leach (1937) reported control by girdling of jungle trees before felling in order to encourage saprophytic invasion of the root

system. Napper (1939) found that killing of stumps of rubber tree (Ficus sp.) with sodium arsenite resulted in rapid invasion of saprophytes to the exclusion of parasites of rubber trees. Fox (1965) notes that rubber trees girdled and poisoned before felling have the added advantage that logs left in the woods, as well as stumps, are rendered unsuitable for colonization by primary parasites.

The primary specific objectives of this work was to determine if colonization of western hemlock stumps by F. annosus can be prevented by killing the tree with herbicides prior to felling and to describe the causative mechanism. The effect of the herbicides used on the rate of tree death, on drying, on debarking and deterioration between treatment and felling, and on breakage at the time of felling also were examined. Data was also taken on the effect of the herbicide treatment on stump senescence, and the amount of F. annosus infection of the axe cuts used to inject the herbicide into the tree.

Location of Study

The study was conducted in a 40 year old stand on the Crown Zellerbach Clatsop tree farm eight miles east of Seaside, Oregon. The stand was composed of predominantly hemlock (75 to 80% hemlock) interspersed with Sitka spruce, Douglas fir, red alder (Alnus rubra Bong.) and a very few western red cedar. The stand on an east slope was generally in good vigor. A few of the trees exhibited

top and bole damage, as described by Shea (1961), due to a cold wave of 1955. These and trees with damaged butts were excluded from the study.

Experimental Design

A completely randomized design was used to evaluate the effect of the following treatments on invasion of stumps by F. annosus:

1. Herbicide injected
   a. Ansar 170 (MSMA)
   b. Tordon 101
   c. Control (no treatment)
2. Seasons of herbicide injection
   a. Fall
   b. Winter
   c. Spring
   d. Summer
3. Interval between injection and felling
   a. Six months
   b. Nine months
   c. Twelve months
4. Date of Felling
   a. March 1968
   b. June 1968

c. September 1968

d. December 1968

e. March 1969

f. June 1969

The general scheme of operations permitting hierarchical classification of the data for analysis is summarized in Table 1. The 26 resultant stumps of each of the treatments were divided into two groups of 13 each of which ten were selected at random for observations. This allowed for surplus trees and provided supernumeraries for unforeseen contingencies. One group was inoculated with a spore suspension of F. annosus conida, while the other was exposed to the natural air spora only. The latter measured the amount of naturally occurring infection.

A chronological arrangement of the detailed procedures of treating the trees and collecting data follows.

Field Methods

Selection of Trees

All trees selected were free from visible bole defects and had normal healthy crown. Suppressed trees were excluded. Tree diameter at breast height ranged from eight to 18 inches.

Herbicide Treatment of Trees

Chemical treatments consisted of injecting four parts Tordon 101, one part water or full strength Ansar 170 into axe cuts spaced equally around the tree about four feet from the ground. Trees up to ten inches in diameter received one axe cut for each inch in diameter at breast height. Those over ten inches D. B. H. received one additional cut for each inch over ten, e. g. 15-inch trees would receive 20 injections. The additional cuts for the larger trees were selected arbitrarily to compensate for the large crowns of the big trees. Approximately 1 1/2 cc. of chemical was injected into each axe cut with a polyethylene wash bottle, immediately after each individual cut was made. Dates of herbicide treatment were selected to represent the "four seasons" of the year (Table 1).

The active ingredients of Tordon 101 (Dow Chemical) are 10. 2% 4-amino, 3, 5, 6,-trichloropicolinic acid and 39. 6% 2, 4-dicloro-phenoxy acetic acid, both as the tri-isopropanoalamine salts. Acid equivalents are 5. 7 (. 44 lbs. per gal) and 21. 2 (1. 6 lbs. per gal) percent respectively, in the dilution used. The active ingredient of Ansar 170 (Ansul Company) is 51% (6. 6 lbs. per gal) monosodium acid methanearsonate hereafter referred to as MSMA.

At the time the study was initiated, research by Crown Zellerbach Research Division had shown that Tordon 101 applied with the axe

Table 1. Schedule of treatments of the study trees

Dates of felling	Dates of treatment				Total[a]
	Fall Sept. 67	Winter Dec. 67	Spring March 68	Summer June 68	
March 68	26 MSMA[b] 26 Tordon				78
June 68	26 MSMA 26 Tordon	26 MSMA 26 Tordon			130
Sept. 68	26 MSMA 26 Tordon	26 MSMA 26 Tordon	26 MSMA 26 Tordon		182
Dec. 68		26 MSMA 26 Tordon	26 MSMA 26 Tordon	26 MSMA 26 Tordon	182
March 69			26 MSMA 26 Tordon	26 MSMA 26 Tordon	130
June 69				26 MSMA[c] 26 Tordon	78
Total	78 MSMA 78 Tordon	78 MSMA 78 Tordon	78 MSMA 78 Tordon	78 MSMA 78 Tordon	

[a]Total date for felling includes 26 live untreated control trees for each felling date.

[b]Herbicide formulations discussed in next section.

[c]Adequate data was collected by March 1969 so that the June felling was not deemed necessary.

injection method was effective for thinning in pre-commercial-sized hemlock, while research at Oregon State University demonstrated that MSMA was highly effective with pine and Douglas fir (Newton, 1967).

Evaluation of Herbicide Effect on Trees

The effect of the herbicide on the trees was rated using the following six point scale:

1. No observable effect.
2. Appearance of abnormal foliage color.
3. Top kill, but lower crown still living.
4. Two thirds or more of crown dead.
5. Crown is essentially dead, only few branches at base of crown still retaining green needles.
6. Crown completely dead, no living foliage present.

The crowns were rated three, six, and nine months after the time of treatment. Six and nine-month evaluations were made only if mortality was not nearly complete at the time of the previous rating period (i.e. three and six months).

In order to evaluate flashback, i.e. herbicide damage on untreated trees adjacent to trees that have been treated, records were kept on all trees over eight inches D.B.H. that were observed to have suffered flashback damage. Ten of the trees that died due to

flashback were examined for root grafts with adjacent treated trees.

Sampling of Trees for *Fomes annosus* and Associated Microorganisms at Time of Felling

The trees were felled six, nine, or 12 months after the date of herbicide treatment by logges employed by Crown Zellerbach. In order to evaluate the tree interior at stump level for presence of microorganisms cross sectional wafers were cut from stump surfaces at the time of felling, numbered with a number corresponding to that of the felled tree, and placed in polyethylene poultry bags. The bags were immediately sealed to prevent water, vapor or liquid, exchanges between the air and the disk. These disks were later sampled for *F. annosus* in order to obtain an estimate of the incidence of the pathogen in the stand prior to felling. Care was taken to avoid exposure of disks to direct sunlight subsequent handling. Also at the time of felling, the stumps were numbered on the surface to correspond with the tree number.

Except for the March 1968 felling, the disks were stored at 70° F. seven to ten days before examination. The disks from the March 1968 felling were stored in a cooler at 34° F. for three weeks prior to examination. The disks were stored vertically, in the manner of phonograph records in a rack, so as not to press the polyethylene bag against the cross sectional surface of the disk. The samples were

examined with 10-30X magnification of a dissecting microscope for colonies of F. annosus. The conidial heads of the imperfect stage of the fungus were easily detected with this procedure.

Examination of the disks revealed that areas infected while the tree was living could often be delineated from areas infected after the tree was killed. Infections present in the tree while living, whether Fomes annosus or any of many other organisms are always outlined by a reaction zone line. A F. annosus infection present in a poisoned tree before it was killed is portrayed in Figure 1. The purple stain inside the zone line (outer deep purple line around the stain) represents the infection in the tree prior to killing. A purple coloration is usual for many F. annosus infections, but is also caused by other organisms in western hemlock. The yellow brown staining beyond the reaction zone represents the advance of the fungus prior to felling after the host had died. The color of the stain inward from the reaction zone is not always so purple. The inner stain may vary from shades of deep purple to pink to shades of yellowish brown. These differences in color may be due to the stage of development of the pathogen, to differences in species of associated microorganisms, or to differences in the host reaction to the infection at some instant in time. However, infections in living trees were always surrounded by a reaction zone.

The above differences between wood invaded by F. annosus

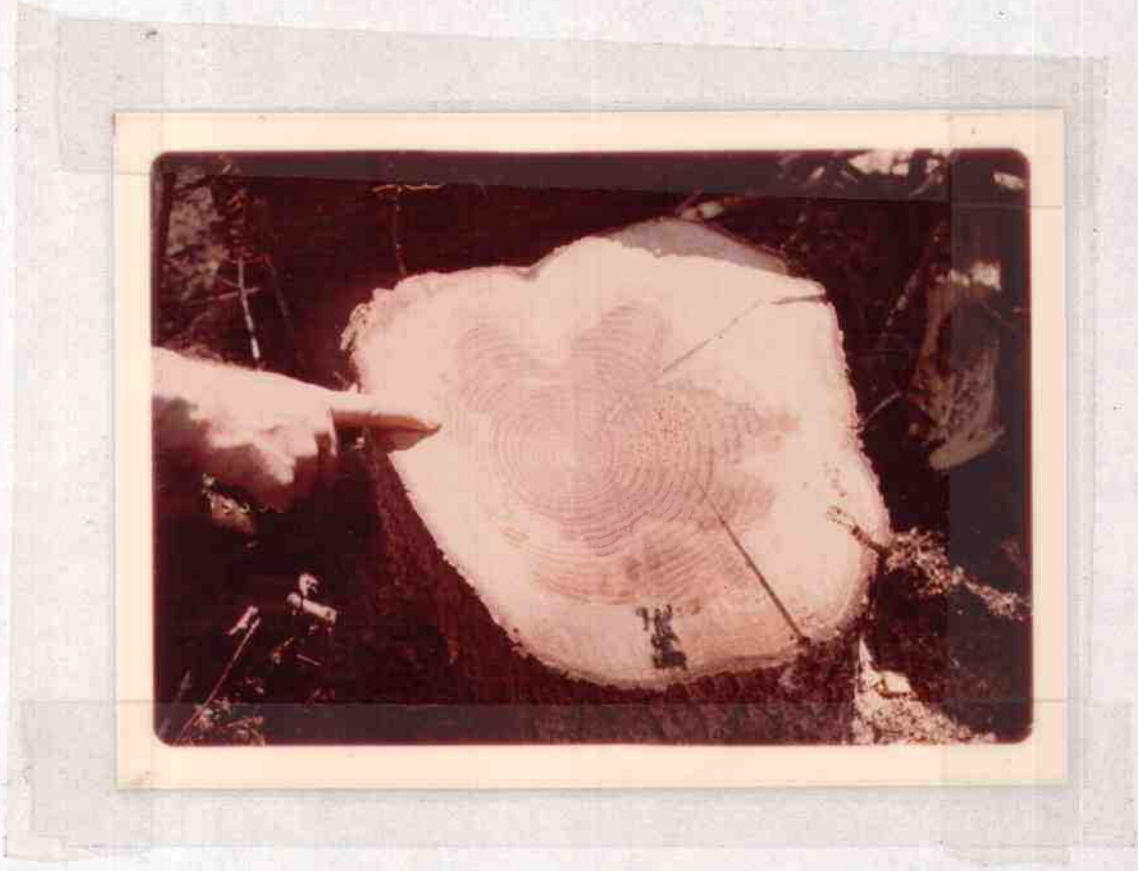

Figure 1. Section of a stump. A Fomes annosus infection that was present in a poisoned tree before it was killed, photographed nine months after treatment. The purple stain inside the zone line (center deep purple line around the stain) represents infection in the tree prior to killing. The brown to yellow areas represent fungal invasion after death of the tree but prior to felling.

before and after tree death (i.e. purple discoloration in general and reaction zone specifically in wood invaded before death and the brown discoloration with no reaction zone of wood infected after death) allowed distinction between infections before death due to herbicidal treatment and those that had progressed down into the bole after death from axe cut injection wounds.

Five trees infected as indicated by brown staining and lack of reaction zones in the sapwood of the cross sectional disks, were dissected in field and the infection was traced from the axe cut to the stump. Records were kept of all trees showing axe cut infections for purposes of correlating them with herbicide treatments.

The cross-sections taken from all of the trees were observed to evaluate stain and decay present in the stand before treatment and the amount of fungal colonization of bole tissue at stump level at the time of felling. The number of trees exhibiting stain and decay were recorded; the reaction zone mentioned above was used to determine whether the stain was present in the treated trees before treatment.

The percentage area of disk cross section colonized at the time of felling was determined to evaluate the amount of fungal colonization of the bole tissue, at stump level, at the time. This is of primary importance, since tissue already colonized by some organism at the time of felling has an extremely low probability of being infected by F. annosus.

Percentage area colonized was determined by first drawing three parallel lines across the surface of the disk, so as to divide

the disk into four approximately equal areas. Thirty sample points along these lines were selected by randomly stopping while scanning these lines with 30X magnification. Each sample point (a circle approximately one cm. in diameter) was designated as colonized if fungal mycelium was observed on the surface or if the wood was stained or excessively moist (wetwood) within any portion of the sample point. The number of sample points colonized divided by 30 was used as an estimate of the percent area inhabited by microorganisms.

Stump Inoculation

One-half of the stumps were exposed to only the natural inoculum in the air. The others were inoculated artificially within 24 hours after felling, except for the stumps of the December felling which required modified procedures because of lack of viable inoculum. The stumps were re-inoculated three weeks later using fresh inoculum. A three inch disk was sawn off of the stump before re-inoculation and ten new control trees were felled and inoculated as checks on the effect of the delayed inoculation.

All inoculum was prepared and applied using the following procedure. A fresh isolate of F. annosus was obtained from an infected bolt (three feet by 12 inches in diameter) stored in a 34° F. cooler by extracting wood chips and placing them on Kuhlman-Hendrix agar (Kuhlman and Hendrix, 1962) in petri dishes. Isolates were then transferred to petri dishes containing 2.5% malt agar. Colonies growing on these plates after seven to ten days were used as inoculum for the stumps. By this time the colonies had completely

covered the plates. Several plates of each batch were observed for production of conidial head production; in every case conidial production was abundant.

Inoculum was applied adding distilled water, brushing the surface of the agar with a 1 1/2 inch nylon paint brush and then painting the resultant spore suspension over the stump surface. In general, one petri dish produced enough suspension to cover an entire stump. Two dishes were required for some of the larger stumps. Each stump surface was inoculated with an exceedingly heavy dose of inoculum, far heavier than would ever be expected in nature. The method proved so effective that over 90% of the surface area of stumps 18 inches in diameter was infected under certain conditions.

Sampling of Incubated Stumpes for Fomes annosus, Associated Microorganisms, Assessment of Cambial Condition and Ambrosia Beetle Infestation

The stumps were harvested after the following incubation periods; March 68 felling, 16 weeks; June 68, 16 weeks; Sept. 68, 25 weeks; December 68, 23 weeks; March 69, 16 weeks. Longer incubation periods were used during winter months to compensate for the slower growth of the fungus during cool weather. Length of incubation had little effect on lateral spread of F. annosus. Lateral development of the fungus in wood is slow, due to cell arrangement, but even more limiting here is the fact that the colonies of the pathogen were surrounded in a horizontal direction by other organisms that had colonized the stump surface.

Stumps from trees which at the time of felling were found to be

infected with F. annosus or were found to contain decay by disk sampling were replaced randomly using a table of random numbers from a group of supernumerary trees treated in a manner identical to that of the trees they replaced. As it turned out, since the original number of replicates was reduced from 13 to ten, it was seldom necessary to use supernumeraries.

At the time of sampling the stumps were severed approximately six inches below the stump surface with a chain saw and placed in a polyethylene bag. At the same time, three to five lateral roots were sampled at the root collar by scraping the bark back to the cambium to determine the condition of the cambium.

Originally, five categories described the condition of the cambium in the roots.

1. alive, as indicated by a glistening white moist cambium.
2. alive and dormant or first stage of senescence, as indicated by a white dry cambium.
3. yellow to tan
4. brown
5. brown plus evidence of fungal activity, i.e., mycelial mats, fans or fungal staining.

In the final analysis, however, the data were grouped into two classes: "one," including categories one and two, and "two," the remaining categories. Cambial condition and patterns of ambrosia beetle infestations of the stump were examined to determine the relationship of these phenomena with the colonization of the stump by Fomes annosus.

The stumps were transported to the laboratory and stored at 34° F. until they were examined for Fomes annosus infection. All of the stumps were examined within seven to ten days after they were cut.

Stumps were examined for Fomes annosus colonization as follows. First, all of the bark was scraped off with a hatchet. The width of living cambial streaks was determined to the nearest inch, in order to determine the percent of living cambium for each stump. From a 6" x 6" sample area selected at random on the cambial surface, the number of ambrosia beetle holes per square foot was determined. The stumps were then cut in half longitudinally with a power band saw. A two inch disk was then sawn off the top of each stump half and placed in a polyethylene bag, which was sealed immediately. These disks were used later to determine the percent area of colonization by the pathogen at the two inch level.

In addition, three inoculated stumps from each treatment, and all of the stumps of the March 68 harvest, were examined for vertical distribution of F. annosus. Observations were also made on the comparative vertical distribution of associated fungi. In these stumps, cross sectons representing the two to four and four to six inch levels were placed in polyethylene bags and incubated.

The cross sectional halves were incubated in the polyethylene bags in which they were placed at the time of sawing. Wet paper

towels were inserted and the bag resealed by twisting and tying with vegetable ties. Plastic surveyor's ribbon was used to keep the wet towels from touching the wood by tying the tape around the neck of the bag between the towels and the disk. This formed a constriction that permitted air passage, but prevented the towels from touching the wood. Preliminary tests had shown that F. annosus would not fruit if liquid water existed on the wood. At the other end of the bag a three inch section of half-inch plastic tubing was punched through the bag, and tied in place. These tubes served as air vents between the humid interior and the external environment. The air vent was deemed necessary from the results of Risbeth (1951a) that indicated that F. annosus will not produce conidia under high oxygen tensions, and from Cobb, Krstic and Zavarin (1968) that indicate that volatile compounds given off from wood in saturated conditions were inhibitory to F. annosus.

The disks were incubated in the polyethylene incubation chambers for seven days before examination for the conidial heads of the fungus. The incubation chambers were carefully stacked so that the bag was not pressed against the wood surface. Wooden blocks, about the size of dominoes, were used as separators between chambers so that the chambers could be stacked one upon the other, touching only on the wood blocks inserted between them.

At the end of the incubation period, each disk from the two-inch

level was examined for F. annosus. The entire surface of the disk was scanned for colonies of the pathogen in a systematic manner with 10-30X magnification. As colonies were encountered they were outlined with a red lead pencil. The red lines were then traced with a felt marker so that the boundaries of the colonies could be observed through tracing paper. After the infected area and the disk cross section were traced on paper, the area infected was determined by placing the traced areas over engineering cross section paper. The traced areas were expressed in numbers of squares; the percent area infected was the number of squares of infected area divided by the number of total squares of cross sectional area of the disk surface.

In stumps from which three disks were incubated, vertical penetration of the fungus was determined to the nearest cm. according to which disk surfaces the colonies were detected on and the depth of stain from the last disk on which the fungus was detected. Vertical patterns of penetration could be traced by matching colonies with infections of the disk below, above or both.

The amount of fungal colonization of the disks by associated fungi was estimated by eye as either less than 25%, 25% to 50%, 50% to 75% and 75% to 100%. Colonized wood was easily distinguished from noncolonized wood, by differences in coloration. Wood colonized previous to felling was included in the estimation. Differences between fungal colonization in the various stump treatments were so great that

more precise limits of estimation were not necessary.

Determination of the Effect of the Herbicide Treatment on Drying, Deterioration and De-barking

The effect of the chemical treatments on rates of timber deterioration, drying and de-barking were evaluated only on the September 68, December 68, and March 69 treatments that were felled in September 1969. At this time trees of each treatment had been on the stump for twelve, nine and six months respectively.

Deterioration of the trees was evaluated on the day of felling by removing squares of bark at least 6" x 6" at the butt, at mid-bole and at the top (6" top). From these sampling points the number of ambrosia beetles per sq. ft., presence or absence of inner bark boring beetles and condition of the cambium was noted. Cambium was noted as dead or streaked. Dead cambium is not white but varies from light yellow to brown. Cambium designated as streaked is characterized by vertical streaks of dead cambium (apparently along conduits of herbicide translocation), running through the live cambium. Fungal radial penetration was evaluated by measurement of the depth of stain at the top and butt sampling points.

Moisture content of trees was determined by weighing one-inch-thick cross sections cut and placed in a polyethylene bag at the time of felling. The samples were carried to the truck and weighed to the

nearest tenth of a gram on a portable field balance (Figure 2). After weighing, the disks were immediately placed back in the bag and transported to the laboratory, where they were dried for 14 days at 105 ° C. Moisture content of the wood was expressed as percentage of the oven dry weight of the wood (Panshin and DeZeeuw, 1964).

The effect of the chemicals on bark removal was determined by evaluating the ease of removal of the bark section at mid-bole. The following classification system is a modification of that used by Wort (1954):

1. Inner bark dry and loose, separates easily from dry wood.
2. Bark easily removed in large pieces (6"x6" or larger) by forcing axe blade between bark and wood.
3. Same as above except that considerable force is required to push axe between bark and wood.
4. Some of the bark can be removed only by chopping, although pieces (usually narrow strips) may be removed easily.
5. All bark is tightly fastened to the wood and some wood is removed while chopping the pieces of bark off.

While this method is rather crude, it was considered adequate to detect differences in bark removal between the treatments.

Breakage during felling was noted only if damage occurred below the 6" top. Nearly all of tops of treated trees were brittle, and breakage frequently occurred above the 6" top. Breakage was merely

Figure 2. Field balance used to determine moisture content of disks.

indicated as present or absent. Some breakage probably also occurred during yarding operations, but this was not assessed.

FINDINGS

Most of the results presented in following sections can be correlated with the amount of cambial kill produced by the herbicide treatment. MSMA produced mortality more completely and quickly than Tordon, and within the MSMA treatments, the December application produced a superior mortality. Results for the December MSMA treatments should be noted particularly, if this is done the interrelationships of the different responses to the herbicide treatments becomes readily apparent.

Herbicide Treatment of Trees

Mortality resulting from herbicidal treatments is described in Table 2. The axe cut injection method used in this study provided almost total mortality response in commercial-sized hemlock. A reduction in dosage might have provided acceptable results, and deserves testing.

The response of the tree to the herbicide was highly correlated with seasonal biological activity. It took nine months through the winter season to reach the levels of response that were obtained in three months following spring and summer applications. No significant effect was noted in March for trees treated in December, but these trees as well as those treated in March showed a high degree

Table 2. Mortality response of commercial sized, western hemlock to injections of MSMA and Tordon 101 at four seasons of the year.

Date of Treatment	Degree of kill on dates of rating							
	Dec. 67		March 68		June 68		Sept. 68	
	MSMA	Tordon	MSMA	Tordon	MSMA	Tordon	MSMA	Tordon
	Mean rating of 6 point scale							
Sept. 67	4.82	5.10	5.60	5.35	6.00	5.90	--	--
Dec. 67	--	--	no significant effect		5.98	5.69	6.00	6.00
March 68	--	--	--	--	5.98	5.52	6.00	5.98
June 68	--	--	--	--	--	--	5.97	5.98

*6 point rating scale, where 1 is no effect and 6 is completely dead. Minimum sample size equals 90 trees.

of mortality when rated in June (MSMA giving almost complete kill). Much of this mortality was evident by late April, and if the trees had been rated at this time rates of kill would have approached those of June ratings, i.e., most of the defoliation occurred with the onset of substantial physiological activity in spring.

Flashback that was observed in trees treated with Tordon is summarized in Table 3. These results include only trees over eight inches in D. B. H.; many smaller trees were probably similarly affected.

The stand was last evaluated for flashback in July of 1969. It is probable that additional flashback will occur from the March and June 1968 treatments, since new tree mortality was observed in connection with these treatments following the previous evlau ation in April 1969.

Root grafts were found between the treated and flashback tree in each of the ten cases examined. These results do not preclude the possibility that some damage occurs to the residual stand due to release of Tordon 101 from the root systems, but at least the major damage occurs from direct root graft.

No observable crown kill was detected as a result of flashback from MSMA-treated trees. However, dead streaks of cambium one to three inches wide were found running nearly 20 feet up the bole from the base in two felled control trees that had been adjacent to

Table 3. Total amount of flashback (chemical damage to untreated adjacent trees) resulting from Tordon 101 treatments through June of 1969.

Number and percent of trees inciting flashback mortality by date of treatment									
Sept. 67		Dec. 67		March 68		June 68		Total	
#	%	#	%	#	%	#	%	#	%
8	10.0	8	10.0	10	12.5	6	7.5	32	10.0

MSMA-treated trees. The total damage in the stand due to this phenomena was not estimated. However, these dead streaks may be very detrimental to the stand as entry courts to decay fungi.

Fomes annosus Invasion of Stumps

At the initiation of the experiment it was expected that a comparable effect of the herbicides on tree death would result in a similar effect on F. annosus invasion of the stump. It soon became apparent that, despite similar ratings of response of the crowns to the herbicides, their effect on the internal physiology of the tree was radically different. It was further found that development of F. annosus as well as many other biological phenomena responded differentially to these differences. The mean percentage of stump area colonized two inches below the stump surface by F. annosus for each treatment is presented in Tables 4 and 5. Table 4 includes stumps exposed to natural inoculum only whereas Table 5 illustrates natural plus artificial inoculum. The data used to calculate these percentages is presented in the Appendix in Tables 1-3.

The infection rates in stumps exposed to natural inoculum only was lower than expected. When the data are espressed in terms of numbers of stumps infected, however, percentage infection in some treatments is as high as 70 percent (Table 1, Appendix). Data expressed merely in percent of individual stumps infected may be a

Table 4. Effect of pre-killing of trees with MSMA and Tordon 101 on stump invasion after felling by Fomes annosus. Stumps exposed to natural inoculum only.

Date of herbicide treatment or control	Mean percent stump cross sectional area colonized two inches below surface at time of sampling for respective dates of felling Ten stumps per treatment Date of harvest														
	March 1968			June 1968			Sept. 1968			Dec. 1968			March 1969		
	MSMA	Tordon	Control	MSMA	Tordon	Control	MSMA	Tordon	Control	MSMA	Tordon	Control	MSMA	Tordon	Control
Control			2.5			4.4			3.3			1.8			10.9
Sept. 1967	0.1	6.2		1.0	1.7		0.0	3.7							
Dec. 1967				0.1	0.9		0.3	2.9		0.0	3.3				
March 1968							0.0	1.5		0.0	3.5		0.0	3.0	
June 1968										0.0	0.0		0.0	1.8	
Grand Means				MSMA	0.1	Tordon	2.4			Control	4.3				

Basic data presented in Table 1 of Appendix

Table 5. Effect of pre-killing of trees with MSMA and Tordon 101 on stump invasions after felling by Fomes annosus. Stumps exposed to both natural and artificial inoculum.

Date of herbicide treatment or control	Mean percent stump cross sectional area colonized two inches below surface at time of sampling for respective dates of felling. Ten stumps per treatment. Date of Harvest: March 1968			June 1968			Sept. 1968			Dec. 1968			March 1969		
	MSMA	Tordon	Control	MSMA	Tordon	Control	MSMA	Tordon	Control	MSMA	Tordon	Control	MSMA	Tordon	Control
Control			15.8			42.7			24.4			3.0			81.4
Sept. 1967	5.4	57.0		0.7	26.3		6.5	30.7							
Dec. 1967				0.0	31.7		0.6	13.4		0.2	3.2				
March 1968							4.8	31.2		0.3	1.6		1.2	24.4	
June 1968										0.6	1.2		6.5	12.9	
Grand Mean			MSMA 2.7			Tordon 18.4			Control 33.5						

Basic data presented in Table 2 or Appendix

poor indicator of the amount of actual stump infection that has occurred. For example March 1969 control stumps have four times the surface area colonized by F. annosus as the March 1968 control, yet both treatments yield a 70 percent value for number of stumps infected per treatment.

Tables 4 and 5 indicate that stumps of trees treated with MSMA are unfavorable substrate for F. annosus colonization. The grand means presented in Tables 4, 5 and 6 show for artificial inoculation that MSMA stumps are colonized to a much lesser extent than stumps of trees treated with Tordon or control stumps. Colonization of MSMA stumps exposed to natural inoculum only is insignificant. These tables illustrate that the reduction of stump infection in Tordon stumps, as compared to control, is not only less than the reduction in MSMA stumps, but that it also is very inconsistent. Indeed stump colonization in Tordon stumps has at times exceeded that in control stumps.

The results indicate that environment at the time of felling affects stump colonization by F. annosus. Colonization was minimal in December. Although the delayed stump inoculation (discussed in Procedures section) may have had some effect, the primary reason for low infection appears to have been environmental. Low rates of infection are similar for both inoculated and non-inoculated stumps. In addition, the ten additional fresh felled controls at the time of the

Table 6. Grand means of (percent of surface area colonized) Fomes annosus invasion in stumps treated with MSMA, Tordon 101 and control stumps.

Treatment	Inoculated stumps	Stumps subjected to natural inoculum only
MSMA	2.7	0.1
Tordon 101	18.4	2.4
Control	33.5	4.3

second inoculation were also found to contain extremely low levels of infection (Table 7).

Table 7. Fomes annosus infection of individual stumps of control trees felled at the time of the delayed December inoculation.

Percent area colonized 2" below stump surface			
Inoculated		Non-inoculated	
1.2		0.0	
0.0		0.0	
0.0		0.0	
0.0		0.9	
0.0		0.0	
1.2		0.9	
Mean	0.2	Mean	0.2

The two March fellings of the control inoculated stumps offer another example of the effect of environment at the time of felling. The March 1969 felling produced stumps with the highest levels of infection observed (81.4%) while March infection of 1968 stumps (15.8%) exceeded only that of December (Table 5). Severe winter conditions at the time of felling and for several weeks after are believed to be responsible for low infection rates of stumps from trees felled in December. Freezing temperatures predominated and snow blanketed the ground and stumps for over two weeks after inoculation. Temperatures in the area dropped to below freezing on the night of inoculation (U. S. Weather Bureau, 1968). Yde Andersen (1962) reports such conditions prevent colonization by F. annosus.

The differences between March treatments indicate that daily fluctuations in environment may play a definite role. Although, precise weather data were not recorded, it was noted that the March 1969 felling was the only felling when some rainy weather was not encountered.

The effect of the chemicals and the time of felling is obviously significant. A statistical analysis helped to verify the significance of these effects, and to test less obvious effects. The standard deviation of the percent area colonized was determined for the inoculated stumps (Table 2, Appendix). The infection levels in the non-inoculated stumps was in general so low that the standard deviation would have been meaningless and was not computed.

Standard deviations obtained for the control and Tordon-inoculated stumps were quite high. This appears to have resulted from 1) tree to tree variations in resistance to F. annosus, 2) prior colonization of stump sections by microorganisms in the living tree before felling and 3) variations in surface environment of the stumps, mainly due to differences in exposure to the sun.

Analysis of variance of the data used to compute means, in Tables 4 and 5 and Appendix Tables 1 and 2 showed significant differences between herbicides, and herbicides and the control. All differed significantly in their effects on F. annosus invasion of both the inoculated and non-inoculated stumps (Table 8). Time of felling was significant only in the inoculated stumps. This may indicate that time of

Table 8. Analysis of variance of the data presented in Tables 4 and 5 on the effect of pre-killing of trees with MSMA and Tordon 101 on stump invasion after felling by Fomes annosus. Basic data presented in Tables 1 and 2 of Appendix.

Source of variation	d.f.	Inoculated Stumps Sums of Squares	Mean Square	F-values
Total	269	218,549		
Treatments				
Chemical, Control	2	38,423	19,212	46.6**
Time of Felling	4	20,745	5,187	12.6**
Error		159,480	606	
Orthogonal Contrasts				
Chemical vs. Control	1	19,409	19,409	47.2**
MSMA vs. Tordon 101	1	48,892	48,892	118.7**
		Non-inoculated Stumps		
Total	269	7,085		
Treatments	26			
Chemical, Control	2	657	329	15.4**
Time of Felling	4	141	35	1.6
Error	263	6,286	239	
Orthogonal Contrasts				
Chemical vs. Control	1	278	278	12.8**
MSMA vs. Tordon 101	1	292	292	13.4**

**1% level
*5% level

felling is critical to the inoculum applied to the stump at that time but not to the natural inoculum falling on the stump surface for a period of weeks after felling. However, infection from natural inoculum may have been so low that differences with time of felling were statistically undetectable. It is for this reason that only inoculated stump data was selected for the more critical examination that follows.

Data from the inoculated stumps had revealed that the choice of herbicide and the time of felling had definite significant effects on F. annosus stump invasion. At this point it was felt desirable to determine if season of treatment of the herbicide and the length of time between treatment and felling significantly effected invasion of the stumps by the pathogen. In order to do this the confounding of the effects of time of felling had to be minimized.

In an attempt to minimize the effects of the local environmental conditions at the time of felling and stump inoculation, the data for the inoculated individual MSMA and Tordon 101 stumps (expressed as the percent area of surface area infected) were expressed as a percent of the mean area infected of inoculated control (non treated) trees felled on same date. Thus, if an MSMA stump showed 10 percent area infected and the mean of ten inoculated control stumps on the same date of felling showed a 50 percent area infected then the percent of control mean area infected was 10/50 or 20 percent. The mean

percent of control infection for each treatment is expressed in Table 9. The basic data used to calculate these means appears in Table 4 of the appendix. The data in Table 4 was arranged in a hierarchical system of classification (Steel and Torrie, 1960). Analysis of variance of this data (Table 10) revealed significant differences for season of herbicide treatment, length of time between treatment and felling and length of time between treatment and felling within the same season of treatment.

Grand means of infection-expressed as percent of infection occurring in control stumps- of MSMA stumps by length of incubation time and season of treatment are presented in Tables 11 and 12. Nine and 12 month incubation periods result in an obvious reduction in infection as compared to the six month incubation.

Of the means presented in Table 12, December exhibits, by far, the greatest reduction in stump infection. Analysis of variance reveals the December contrast with the other treatments to be highly significant, as is the contrast of March and June treatments with the September treatment.

Similar grand means for Tordon treated trees appear in Tables 13 and 14. Length of incubation period produced some baffling results. Here the nine month wait produced a greater reduction than the 12 month period. Possible reasons for this and the general inconsistency of the Tordon results will be discussed later. Note that June

Table 9. Mean Fomes annosus infection of treated tree residual stumps expressed as a percent of the mean surface area infected per treated stump over the mean percent area infected control stumps felled on same date.

Herbicide	Date of treatment	Date of felling	Length of incubation	Area infected
			Months	Percent
MSMA	Sept. 1967	March 1968	6	32.2
		June 1968	9	2.1
		Sept. 1968	12	26.9
MSMA	Dec. 1967	June 1968	6	0.2
		Sept. 1968	9	2.4
		Dec. 1968	12	0.7
MSMA	March 1968	Sept. 1968	6	20.5
		Dec. 1968	9	11.7
		March 1969	12	1.4
MSMA	June 1968	Dec. 1968	6	18.9
		March 1969	9	8.0
Tordon	Sept. 1967	March 1968	6	360.6
		June 1968	9	61.7
		Sept. 1968	12	125.8
Tordon	Dec. 1967	June 1968	6	74.4
		Sept. 1968	9	81.9
		Dec. 1968	12	106.0
Tordon	March 1968	Sept. 1968	6	117.8
		Dec. 1968	9	53.3
		March 1969	12	49.8
Tordon	June 1968	Dec. 1968	6	41.7
		March 1969	9	24.8

Basic data appears in Table 4 of Appendix.

Table 10. Analysis of variance for the various sources of variation in stump infection from Table 9 and data Table 4 of appendix.

Source of variation	df	SS	MS	F
Among incubation times	21	1,302,079	62,004	10.5**
Among dates of treatment	7	737,189	105,313	17.8**
Among incubation times within dates of treatment	14	564,890	39,064	6.62**
Among stumps within incubation times = sampling error	198	1,168,846	5903	
Total	219	2,470,925		

Table 11. Grand means of stump infection of MSMA treated trees by length of time between treatment and felling date, expressed as a percent of the mean area infected in control stumps of the same felling date. (Basic data Tables 9 and Table 4 of appendix)

	Extent of infection		
	Percent of control infection by length of time between treatment and felling date of each of three incubation periods		
	Six months	Nine months	Twelve months
	Percent		
Grand Mean	17.9	6.0	9.7

Table 12. Grand means of stump infection of MSMA treated trees by season of treatment expressed as percent of mean area infected in control stumps of the same felling date. (Basic data Table 9 and Table 4 of appendix)

	Extent of control infection by season of treatment			
	September	December	March	June
	Percent			
Grand Mean	20.4	1.1	11.2	13.4

Table 13. Grand means of stump infection of Tordon 101 trees by length of time between and felling date as expressed as percent of mean area infected in control stumps of the same felling date (Basic data Table 9 and Table 4 of appendix)

	Extent of control infection by length of time between treatment and felling date		
	Six months	Nine months	Twelve months
	---------- Percent ----------		
Grand mean	148.6	55.4	110.4

Table 14. Grand means of stump infection of Tordon 101 treated trees by season of treatment as expressed as percent mean area infected in control stumps of the same felling date. (Basic data from Table 9 and Table 4 of appendix)

	Extent of control infection by season of treatment			
	September	December	March	June
	---------- Percent ----------			
Grand mean	182.7	87.4	73.6	33.2

was the season of the best treatments with Tordon. The minimum for Tordon, 33% of the control infection still exceeds the highest value for MSMA (20% in September).

In summary, MSMA treatments applied in the winter are clearly superior to both Tordon and no treatment. After treatment with MSMA the nine- and 12-month incubation periods before felling reduced infection significantly below those with six month delay.

All ten of the inoculated stumps of the MSMA, Tordon and control felled trees in March were examined for vertical penetration of F. annosus. On the basis of findings in the first examination, only three stumps of the ten stump samples were examined for vertical penetration in the treatments felled at later dates. The values in Table 15 are a group of all MSMA and Tordon treatments felled at the particular date, i.e., Tordon trees treated in September and December of 1967 and March of 1968 and felled in September of 1968 were grouped together. The data is not differentiated as to separate dates of treatment or length of tree seasoning times in this table, the stumps are merely grouped by the herbicide treatment for each particular date of felling.

It is noteworthy that penetration is reduced during cold stump incubation periods (incubation after September and December felling). Control stumps offered some resistance to F. annosus penetration as compared to stumps of the herbicide treated trees. Horizontal

Table 15. Mean vertical penetration of Fomes annosus into inoculated stumps by felling dates.

	Date of Felling				
	March 68	June 68	Sept. 68	Dec. 68	March 69
			Centimeters		
Control	100	150+	95	40	120
MSMA	150+	150+	145	45	150+
Tordon	150+	150+	145	45	150+
Length of stump incubation	16	16	25	22	16
Numbers of stumps sampled					
Control	10	3	3	3	3
Each herbicide	10	6	9	9	6

distribution of the fungus on successive disks taken from a given stump differed very little. Apparently competition from surrounding colonies prevented lateral expansion of F. annosus.

Stump Colonization by Fungi other than F. annosus

Data pertaining to stump colonization by associated fungi is presented in Table 16 and in Table 5 of the appendix.

Table 16. Colonization of stumps by fungi other than F. annosus.

Depth of sample disk	Mean colonization disk class* by treatment		
	MSMA	Tordon	Control
	----------Percent----------		
2"	4.0	3.0	3.4
4"	3.7	2.6	2.1
6"	3.6	2.3	1.4
replicates	40	40	20

*Percent area colonization disk classes, 1.0-25%; 2.25-50%; 3.50-75%, 4.75-100%. Absolute data presented in Appendix Table 5.

MSMA stumps appear to be more heavily colonized by associated fungi than Tordon and Control stumps. Hyphomycetes were conspicuously present on the MSMA stumps. Species of Graphium, Leptographium, Diplosporium, Penicillium, Cladosporium, Hormodendron and Cephalosporium predominated; whereas, non-sporulating fungi, mainly resembling Basidiomycetes, were present

along with species of Trichoderma, Graphium and Leptographium on the disks from Tordon and control stumps. Much of the fungal colonization of the MSMA stumps appeared to have penetrated centripitally while penetration of control stumps was vertically downward from the stump surface. Much of the colonization of the MSMA stumps occurred prior to felling. Colonization behavior in Tordon stumps was variable.

Fungal Colonization of the Lower Bole Prior to the Time of Felling

Data on fungal colonization of disks collected at time of felling from stump surfaces is presented in Table 17. Colonization of the Control trees was meager and represented the amount of decay or stain presented in the living tree.

The MSMA trees have been colonized much more extensively than the Tordon trees in the interval between treatment and felling. As expected, colonization increases with the time the tree remains on the stump after poisoning. Fungal species colonizing these disks were similar to those reported to colonize stumps.

Axe Cut Infection by Fomes annosus

One of the most important findings of this study was that axe cuts used to insert Tordon into the tree were highly susceptible to

Table 17. Fungal colonization of the lower bole at the time of felling.

Treatment		Number of stumps in each percentage range of colonization 0-20%	21-40%	41-60%	61-80%	81-100%	Means*
September felling (1968)**							
	Treated						
	September 67	0	0	0	4	21	86.8
MSMA	December 67	0	0	0	1	24	89.2
	March 68	0	0	0	8	17	83.6
	September 67	1	2	8	10	4	61.2
Tordon	December 67	6	5	8	6	0	39.2
	March 68	8	7	6	4	0	34.8
Control		10	0	0	0	0	10.0
December felling (1968)							
	Treated						
	December 67	0	0	0	0	25	90.0
MSMA	March 68	0	0	0	0	25	90.0
	December 67	7	7	6	6	0	37.2
Tordon	March 68	1	7	9	4	4	43.6
	June 68	6	6	7	6	0	40.4
Control		10	0	0	0	0	10.0
March felling (1969)							
	March 68	0	0	0	1	24	89.2
MSMA	June 68	0	4	6	5	10	71.2
	March 68	0	3	3	15	4	58.0
Tordon	June 68	2	3	9	4	7	58.8
Control		10	0	0	0	0	10.0
Tordon Mean 46.6							
MSMA Mean 83.4							

*Means calculated using class midpoints.

**Data was not taken for March and June 1968 fellings.

F. annosus infection while none of the MSMA axe cuts were infected (Figures 3 and 4, Table 18). It should be pointed out that the fungus had to progress from the axe cuts to the point of cutting before the tree was felled in order to be detected in the disks collected at the time of felling. Low infection rates in some treatments may be a result from failure of the fungus to reach the level of cutting. The main factors correlated with this vertical penetration were time of incubation between treatment and felling (i.e., six, nine and 12 months) and the season that this incubation period occurred.

From the 12 month incubation period data the 48, 36, and 44 percent infection levels indicate that substantial infection may be through these axe cuts (Table 18). In Figure 3 note the high infection rate of axe cuts a disk taken from the bole at injection level. In most cases only one or two cuts were infected by the pathogen in each tree.

Growth rates of the pathogen in the bole after entrance via the axe cut were determined by measuring decay columns (Figure 4) from the five trees that were dissected to trace the fungus from the infection into the stump. The maximum rate of growth that occurred was 24" in nine months from March to December. The mean of the five trees, all treated in March and felled in December, was 20.4" and the minimum 17.5". In Figure 8 the arrow points to the end of the 24" infection column. This column would no doubt reach the lateral roots by the following summer.

Figure 3. Axe cut infection frequency in a Tordon killed tree by Fomes annosus. In most cases only one or two cuts per tree are infected.

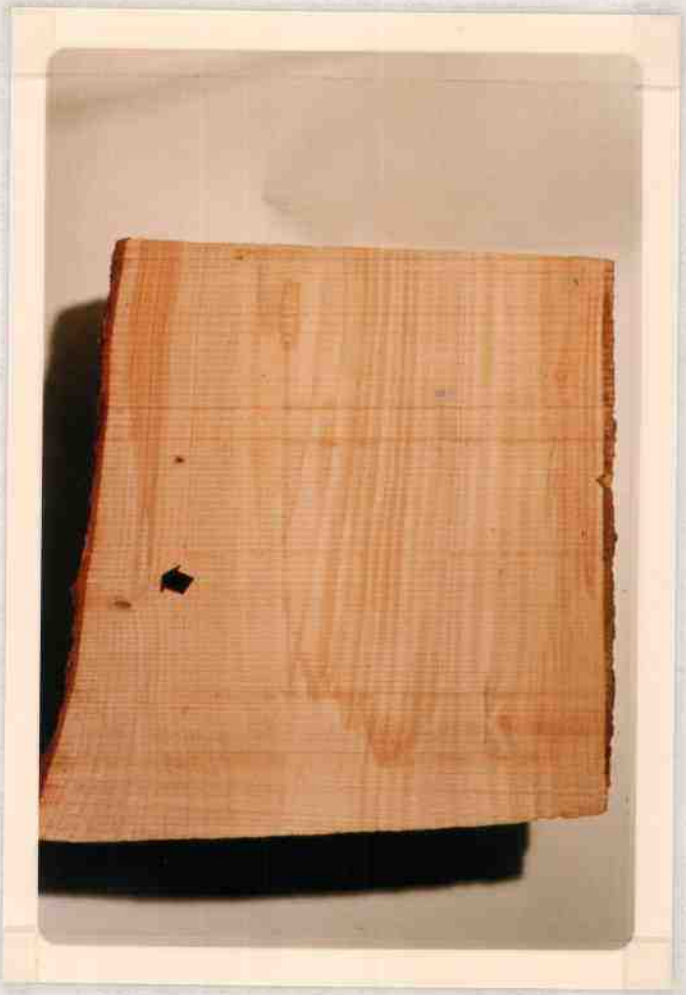

Figure 4. An infection column of Fomes annosus in the lower stump of a tree that was infected through an axe cut.

Table 18. Number and percent of Tordon-treated stumps infected by Fomes annosus entering through axe cuts.

Date of Treatment	Date of Felling									
	March 68		June 68		Sept. 68		Dec. 68		March 69	
	Number	Percent	Number	Percent	Number	Percent	Number	Percent	Number	Percent
Sept. 67	0	0	0	0	12	48	-	-	-	-
Dec. 67	-	-	0	0	6	24	9	36	-	-
March 68	-	-	-	-	5	20	9	36	11	44
June 68	-	-	-	-	-	-	1	4	1	4

Condition of the Stump Cambium and Lateral Roots at the Time of Stump Harvest

Stumps of injected and control trees were examined for cambial death within 24 hours after felling. Control stumps showed no signs of death. MSMA stumps were more senescent than the Tordon stumps. Differences between Tordon and MSMA stumps (Table 19) were tested by the Student's "t-test for condition of both the cambium and the lateral roots. Differences between effects of the two herbicides were highly significant for both cambium and roots.

Figures 5 and 6 show that the treatment that gave the best reduction in F. annosus invasion of stumps, December MSMA, also produced the most rapid killing of the stump cambium and the lateral roots.

Ambrosia Beetle Infestations of Stumps

Species of Trypodendron and Gnathotrichus were collected from trees and stumps. Ambrosia beetle infestations of the stumps of the various treatments are summarized in Table 20. The data refer to the number of ambrosia holes per stump without reference to which species bored the hole.

MSMA stumps were attacked more frequently and more intensively than were the control and Tordon stumps. The attack in MSMA

Table 19. Condition of the stump cambium and lateral roots at time of stump harvest.

Treatments		Percent of cambium alive	Percent of lateral roots still living
MSMA			
Treated	Felled		
Sept. 67	March 68	17.4	21.0
	June 68	15.8	33.0
	Sept. 68	4.4	11.0
Dec. 67	June 68	3.2	11.0
	Sept. 68	0.1	0.0
	Dec. 68	0.0	0.0
March 68	Sept. 68	39.2	61.0
	Dec. 68	7.7	48.0
	March 69	5.4	0.0
June 68	Sept. 68	6.5	39.0
	March 69	1.0	0.0
Tordon			
Treated	Felled		
Sept. 67	March 68	23.2	73.0
	June 68	25.6	46.0
	Sept. 68	4.0	19.0
Dec. 67	June 68	64.1	91.0
	Sept. 68	11.9	47.0
	Dec. 69	9.0	47.0
March 68	Sept. 68	19.0	49.0
	Dec. 68	12.5	65.0
	March 69	14.4	11.0
June 68	Sept. 68	9.1	51.0
	March 69	2.0	2.0

Student T-test $t_{.01} = 2.898$
t_{01} for cambium = 2.905**
t_{01} for roots = 6.777**

Length of stump incubation prior to harvest by felling date; March 68, June 68 and March 69 - 16 weeks, Sept. 68 - 25 weeks, Dec. 68 - 22 weeks.

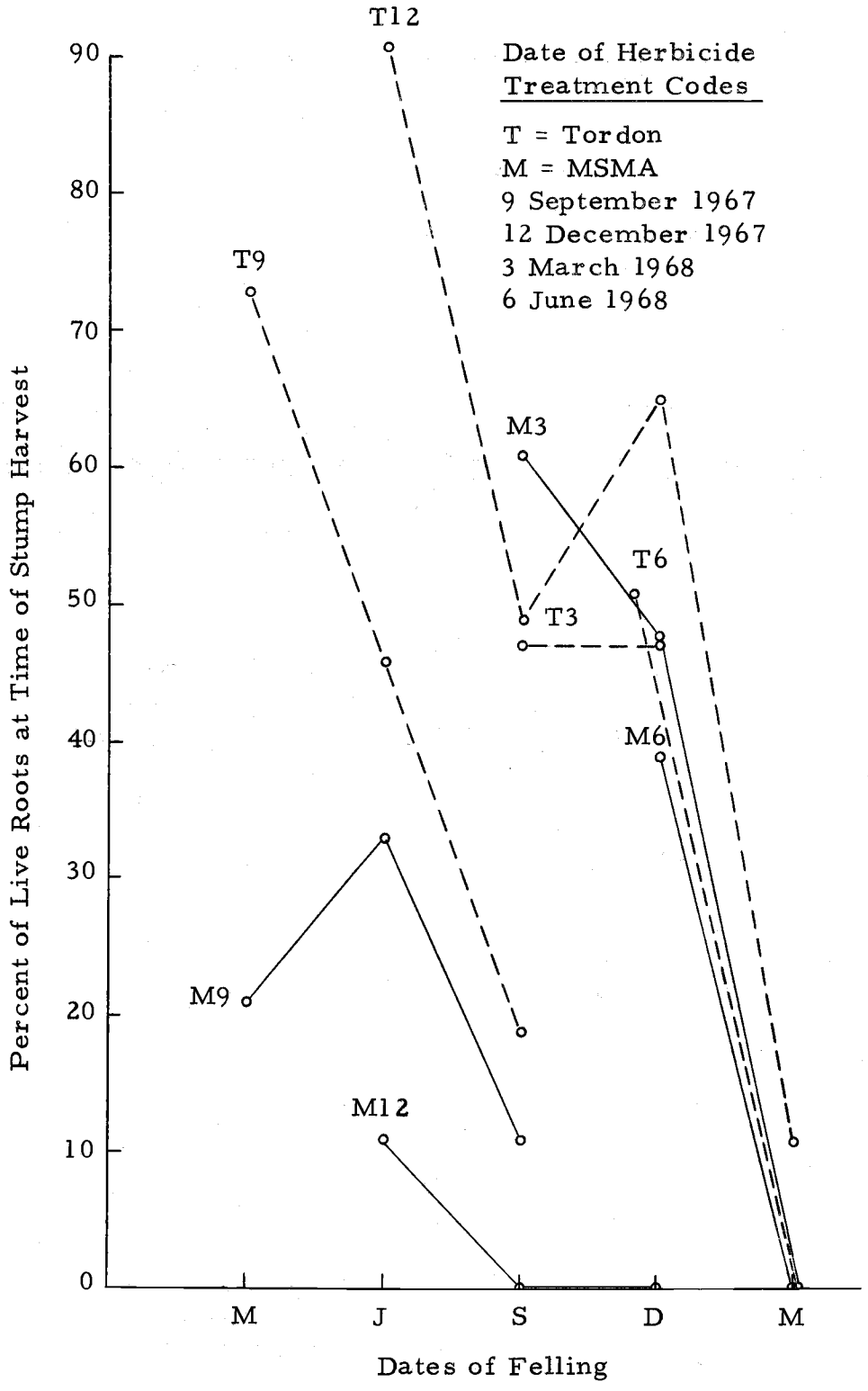

Figure 5. Percent of lateral roots alive at time of stump harvest.

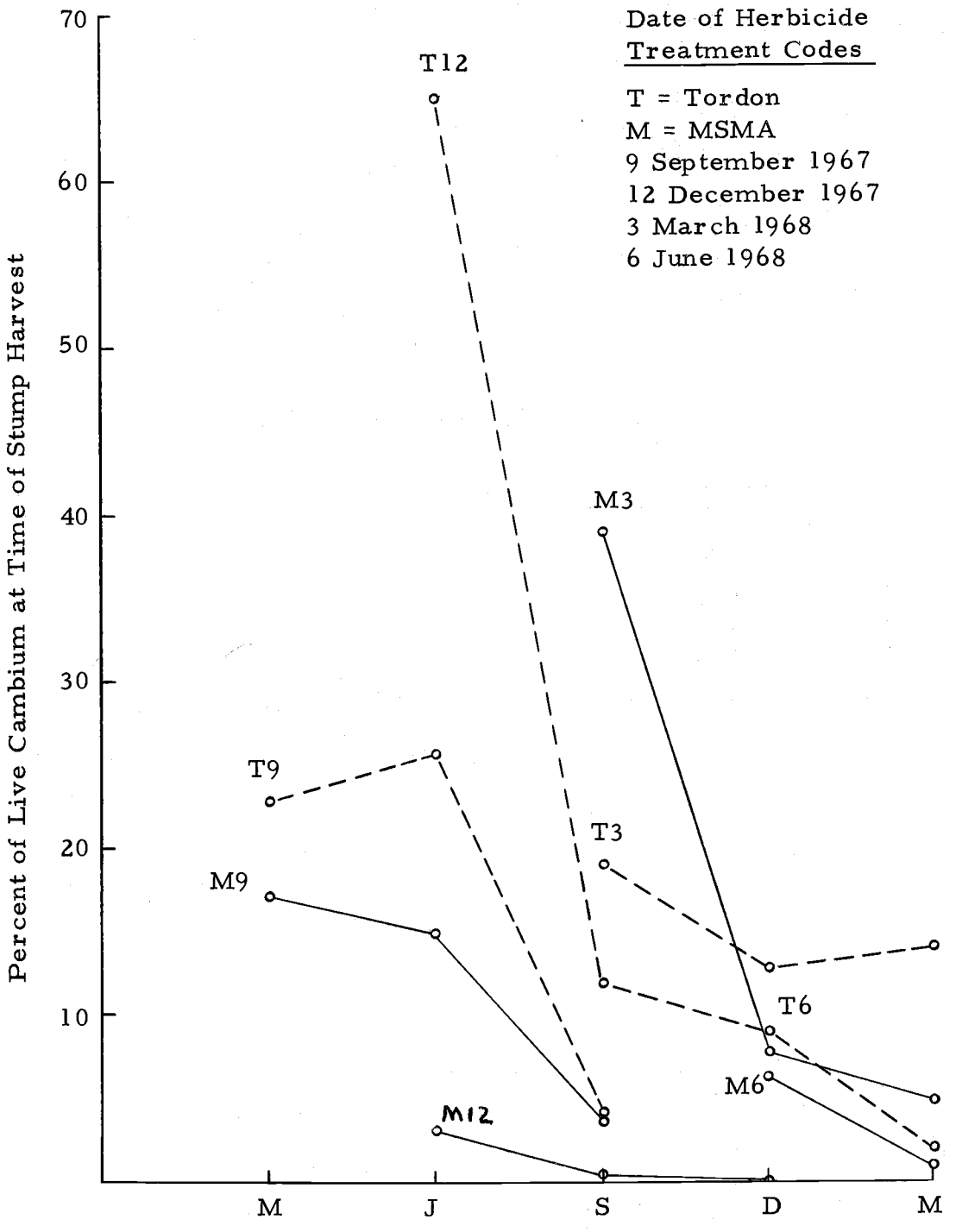

Figure 6. Percent of stump cambium alive at time of harvest.

Table 20. Ambrosia beetle infestation of stumps based on 20 trees for each herbicide, date of treatment and month of felling.

Treatment	Month treated	Month felled	Mean no. of holes per sq. ft.	No. of trees attacked	Maximum attack per tree; holes per sq. ft.
MSMA	Sept. 67	March 68	14.5	14	79
		June 68	16.6	15	108
		Sept. 68	14.5	19	38
Tordon	Sept. 67	March 68	0.4	2	5
		June 68	1.1	3	14
		Sept. 68	1.9	7	20
MSMA	Dec. 67	June 68	22.9	19	90
		Sept. 68	19.2	19	32
		Dec. 68	17.4	18	102
Tordon	Dec. 67	June 68	0.7	1	14
		Sept. 68	2.6	2	30
		Dec. 68	3.2	12	12
MSMA	March 68	Sept. 68	16.5	18	36
		Dec. 68	18.6	18	32
		March 69	19.2	12	63
Tordon	March 68	Sept. 68	2.0	3	24
		Dec. 68	4.1	8	32
		March 69	5.9	8	72
MSMA	June 68	Dec. 68	9.8	17	28
		March 69	11.2	12	36
Tordon	June 68	Dec. 68	3.4	8	20
		March 69	3.9	10	24
Control	--	March 68	9.5	6	73
	--	June 68	0.0	0	0
	--	Sept. 68	0.0	0	0
	--	Dec. 68	6.3	6	36
	--	March 69	0.0	0	0

trees was higher than that reported by Prebble and Graham (1958) for western hemlock logs on the ground.

During sampling of the stumps for F. annosus in the laboratory it was observed that ambrosia larval cells were not found in the MSMA or Tordon stumps nor were many dead adults found in the tunnels. The insects were apparently unable to reproduce in these stumps. Since the number of adults found in the galleries was extremely low, it is probable that the beetles had left and these trees may not be functioning as trap trees for ambrosia beetles.

The ambrosia beetle galleries open the stump interior to rapid colonization by fungal competitors of F. annosus. In no case was a F. annosus infection observed to originate from these galleries.

Pathological Condition of the Stand Prior to Establishment of Treatments

The pathological condition of the stand at the time of felling as determined from the disks collected at that time are presented in Table 21.

It is possible that the percent of total decay attributed to F. annosus should be higher since some of the decay not yielding isolates of the pathogen may have been initiated by it, only to have been replaced by secondary organisms later.

Table 21. Pathological condition of the stand at the time prior to treatment. Based on disks collected from the stump surface at the time of felling.

Condition	Percent
Trees containing stain	77.5
Trees containing decay	7.0
Trees containing Fomes annosus (stain and decay)	6.3
Trees containing decay from which F. annosus was successfully isolated	1.4
Percent of the total decay attributed to F. annosus	19.5
Trees less than 12" D.B.H containing decay	7.8
Trees over 12" D.B.H. containing decay	3.9
Trees less than 12" D.B.H. containing F. annosus	4.1
Trees over 12" D.B.H containing F. annosus	3.2

All trees with visible scars were omitted from study

Drying, Deterioration, De-barking and Breakage

Results in this section were obtained from data collected during the September felling of 1968 of trees treated in September and December of 1967 and March of 1968. Ambrosia beetle attack, radial fungal penetration, and cambial condition were the parameters used as measures of deterioration.

Ambrosia beetle attacks on the upper, middle and lower bole at the time of felling are presented in Table 22. Apparently the

Table 22. Ambrosia beetle attack of trees treated in Sept. and Dec. of 1967 and March of 1968 at time of felling in Sept. 1968.

Treatment	Average attack per sq. ft. for 30 trees		
Date treated	butt	mid-bole	top
	Number		
MSMA			
September 1967	5.2	2.9	1.5
December 1967	2.9	6.9	2.1
March 1968	7.3	11.8	5.3
Tordon			
September 1967	0.4	0.0	0.0
December 1967	1.1	0.0	0.0
March 1968	0.6	0.0	0.0

Table 23. Cambial condition of trees treated in Sept. and Dec. 1967 and March 1968 at time of felling in Sept. 1968.

Treatment	Percent of trees with dead cambium* (30 trees)		
Date treated	butt	mid-bole	top
MSMA			
September 1967	66.7	100.0	100.0
December 1967	93.3	100.0	100.0
March 1968	80.0	100.0	100.0
Tordon			
September 1967	20.0	66.7	93.3
December 1967	6.7	53.3	100.0
March 1968	6.7	26.7	100.0

*equals the number of 6x6 patches that were dead divided by total number examined (30).

MSMA trees treated in March, during the height of beetle activity, were more attractive to the insects than other treatments. This agrees with Wort's (1954b) findings in chemical treated hemlock. Attacks on Tordon trees were generally very light, and occurred only at the butt. MSMA-treated trees are highly subject to degrade (loss of economic value) due to pin holes from the beetle attack.

The condition of the cambium at the three sampling points is presented in Table 23. The cambial sections that were streaked with dead tissue were grouped with the unstreaked sections because of presence of live tissue. Streaked sections were very few and were observed only in the butts of the Tordon trees.

MSMA trees have undergone considerably more senescence than the Tordon trees, especially below mid-bole. Within the MSMA treatments, the December application caused greater cambial mortality in the butt than did the September and March applications.

Radial fungal penetration of the bole, Table 24, as indicated by stain on the log ends, was greatest in MSMA. Apparently bark saprophytes find little resistance in their penetration of the inner bole after the living cells of the rays and cambium have succumbed due to MSMA toxicity. The heavy attacks of ambrosia beetles in MSMA trees also aided in introducing fungi into the bole.

The effects of the chemical treatments on bark removal are presented in Table 25. December MSMA and possibly the March MSMA

Table 24. Radial penetration of fungi in trees treated in Sept. and Dec. 1967 and March 1968 and evaluated at the time of felling in Sept. 1968.

Treatment	Mean radial fungal penetration for 30 trees	
	butt	top
	mm.	
MSMA		
September 1967	73	58
December 1967	73	44
March 1968	64	53
Tordon		
September 1967	40	44
December 1967	22	28
March 1968	18	28

Table 25. Effect of the herbicides applied in Sept. and Dec. of 1967 and March 1968 on bark removal at the time of felling in Sept. of 1968: Expressed as units of 1-5 rating scale.

Treatment	Mean rating* for 30 trees	Mean
	----------Scale Units----------	
MSMA		
Sept. 1967	3.0	2.0
Dec. 1967	1.1	
March 1968	1.8	
Tordon		
Sept. 1967	3.7	3.5
Dec. 1967	3.5	
March 1968	3.4	
Control	3.0	3.0

*based on bark removal rating scale described on page 39.

treatments show definite promise for use as bark removing agents. Superiority of the December treatment is no doubt related to the superior lateral spread of the herbicide resulting in more cambial kill. Other factors are involved, however; these will be discussed later.

These results agree with those of Wort (1954b) in that the arsenicals are effective agents for bark removal and that the growth regulator herbicides are not. Wort, however, did not test winter treatments and his methods of application were much more tedious than those applied here. He used the toxic inorganic arsenical sodium arsenite.

Rates of drying for treated trees are given in Tables 26 and 27. Drying of trees receiving the September and December MSMA treatments, on the stump twelve and nine months respectively, considerably exceeded the March MSMA treatment (on the stump six months).

Although drying has occurred in the MSMA treatments, on the average, it is questionable whether this drying took place in the potential "sinker trees" (trees that yield butt logs that sink during water transport). It seemed probable that the herbicide treatments would have little if any effect in reducing the number of sinkers, since this condition is caused by a pathological condition of the wood. In order to analyze drying more closely the data was grouped into nine wood

Table 26. Mean moisture content of the butts and tops of trees treated in Sept. and Dec. of 1967 and in March 1968 and compared at the time of felling in September 1968 with freshly cut controls.

Treatment	Moisture content				Replicates
	Butts		Tops		
	Mean	Range	Mean	Range	
MSMA	Percent				
Sept. 1967	105.6	73-152	108.0	64-146	24
Dec. 1967	108.4	71-142	93.4	64-103	21
March 1968	116.7	95-149	121.1	69-115	22
Tordon					
Sept. 1967	108.8	74-155	135.2	94-154	23
Dec. 1967	112.7	86-138	133.2	106-172	22
March 1968	118.9	85-145	143.5	107-160	23
Control	118.2	75-158	136.9	74-176	74

Table 27. Arrangement of butt moisture content results for the September and December 1968 MSMA treatments and controls felled in September 1968, into moisture content classes.

Percent moisture content* class	Number of trees falling into class					
	Control	MSMA				
			September 1968		December 1968	
	No.	Percent	No.	Percent	No.	Percent
71-80	3	4	3	13	1	4
81-90	5	7	2	8	1	4
91-100	7	9	7	29	5	24
101-110	13	18	4	16	4	16
111-120	17	23	1	4	5	24
121-130	17	23	2	8	3	12
131-140	7	9	2	8	1	4
141-150	1	1	2	8	1	4
151-160	1	1	1	4	0	0

*Moisture content is expressed as a percentage of the oven dry weight of the wood.

moisture content classes, with a range of ten percent, ranging from 71.0 to 160.0. The data appear in Table 27 and Figure 7.

The MSMA treatments have more trees in the drier classes; they also have more in the wetter classes (141-160) which are likely to be sinkers.

These results on drying were obtained from trees that were exposed to one of the wettest summers on record in this region. Monthly rainfall and the deviation from the norms are as follows: June 6.33 + 3.21; July 0.97, -.26; August 4.75, + 3.22; September 3.99, + 1.05. The wet summer, no doubt influenced the drying effects of the treatments in a negative manner.

Results on breakage for the three treatments sampled in September of 1968 are expressed in Table 28. The December MSMA treatment conspicuously increased breakage. However, in only two of the 32 treated trees that breakage occurred was the bole shattered to the point that the tree was left in the woods.

It was noted that 30 of the 36 trees (83%) that were broken were felled across logging trails, also 30 of the 96 (32%) trees felled across trails were broken, while only six of the 136 (47%) not felled across trails were broken. This indicates that the breakage was due to a loss of the cushioning effect of the live crowns of the treated trees, and the felling across the logging trails.

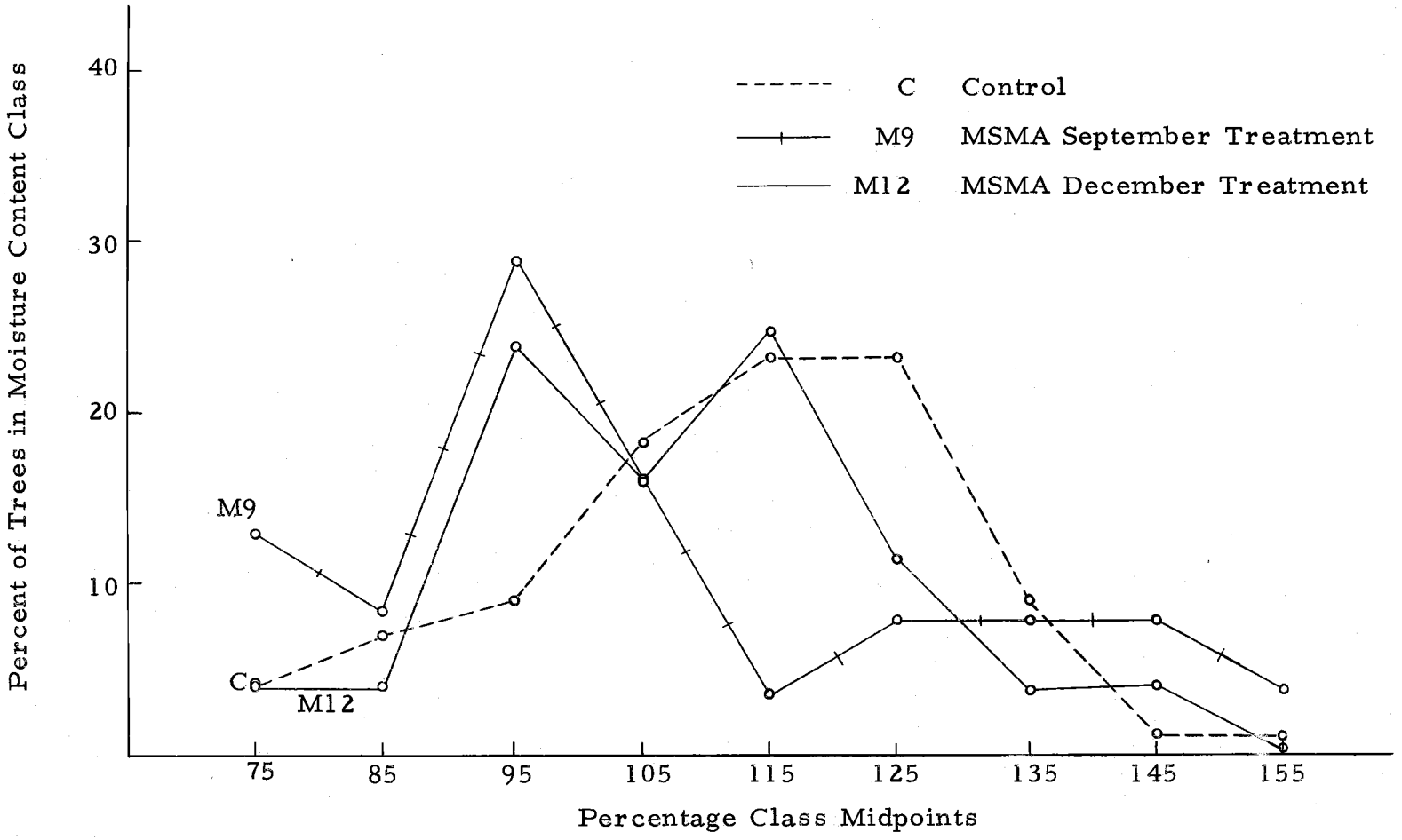

Figure 7. Moisture content distribution among September and December 1967 MSMA treated trees and controls felled in September of 1968.

Table 28. Breakage of trees treated in September and December of 1967 and March 1968 when felled in September of 1968.

Treatment	Trees	Broken trees		Damaged in more than one log
		No.	Percent	No.
MSMA				
September 1967	30	4	13	1
December 1967	30	10	33	0
March 1968	30	8	27	1
Tordon				
September 1967	30	6	20	0
December 1967	30	2	7	0
March 1967	30	2	7	0
Control	52	4	8	0

DISCUSSION AND CONCLUSIONS

Effect of Herbicide on Rates of Tree Mortality

Both herbicides did an acceptable job of killing commercial sized (eight to 18" D. B. H.) western hemlock when applied using the axe cut and injection method. The response of the tree to the herbicide varied with the seasonal biological activity that succeeded treatment. It took nine months through the winter season to reach the same degree of defoliation that was obtained in less than three months during the growing season (Table 2). Reasons for differences in tree mortality rates will be discussed later under mechanism of herbicidal action.

Although both chemicals did an adequate job of killing trees from a silvicultural standpoint, it soon became obvious that extreme differences existed in their modes of action within the tree. Tordon, for example, was found to cause considerable flashback among residual trees adjacent to treated trees (Table 3). Damage in residual trees adjacent to MSMA trees was practically undetectable. It was also observed, that the effect of the two chemicals on the cambium of the bole and roots was radically different (Table 19). MSMA caused an initial rapid kill, followed by a slow senescence. Cambial death resulting from Tordon was slow from

the beginning. These differences in cambial kill were found to have a profound effect on other processes and their results in this study.

Effect on Colonization of Stumps by Fomes annosus

There was a striking reduction in F. annosus infection of stumps treated with MSMA. Tordon, on the other hand, was inconsistent with infections exceeding the controls. It is also obvious that factors other than the herbicide treatments have influenced F. annosus invasion of stumps. Environmental conditions at the time of felling had a major influence on stump infection. The December felling was probably influenced primarily by severe weather conditions but the fellings at other times were affected by a combination of weather and biological factors.

The key to most of the results reported in the thesis lies in the effect of the herbicide on cambial tissues (Tables 19 and 23). MSMA treatments have produced a much better cambial kill than Tordon and within the MSMA treatments the December application resulted in the superior cambial killings. The failures of F. annosus to colonize the stump and axe cuts on the bole, and success of saprophytes in entering these wounds is directly related to amount of cambial kill. Increases in ease of de-barking drying, ambrosia attack, breakage during felling and general deterioration are all greater in MSMA treated trees than Tordon and are greatest in the MSMA December

treatment and are presumably consequences of the same phenomenon; while the same general pattern exists with the Tordon treatments it is not as consistent. The major discrepancy is that F. annosus infection of stumps does not necessarily decrease with increased Tordon induced cambial mortality. This discrepancy [discussed later in detail] is probably due to confounding of the stump infection data by infection of axe cuts on the bole shortly after treatment.

The data in Tables 19 and 23 show that only the cambial area in the tops of the Tordon trees was killed extensively. Most of the cambium of the Tordon tree remained fresh, is moist, glistening white and alive for a considerable time after the crown died. It then senescences slowly, changing from white to yellow, then tan and finally brown. It retains its moisture even in the brown stage for a period of time, the interval preceding drying, depending on environmental moisture. On the other hand the cambium of the MSMA treatments in general, and the December treatment in particular died very rapidly and was soon observed to be dark brown and either dry or wet, depending on the external environment at the time of felling. This data does not distinguish between the stages of senescence of the Tordon killed cambium. All discolored cambium was regarded as dead. Also the data in Table 23 expresses the number of trees with dead cambium at a certain level on the bole while data in Table 19 represents the percent of live cambium in the stump at

time of stump harvest. Both parameters only serve as approximations of the percent of cambial death at stump level at the time of felling.

The senescence of these cambial area cells is of prime importance. As long as they remain alive, the physiological functions of resistance of the tree continue. Water and food transport in the xylem, phloem, cambium and wood rays apparently does not cease immediately with the loss of the crown. Cessation of physiological function of these cells is apparently due to starvation. While alive these cells are still capable of producing phenolic compounds, phenol oxidizing systems and other systems important in disease resistance. The effectiveness of the MSMA treatment in preventing stump colonization by F. annosus appears directly related to its ability to kill the cambial area cells quickly. Death of the living cells of the rays probably occurs shortly thereafter since the tree interior is open to rapid invasion of saprophytes, after the cambium is killed.

Once the cambium of the treated standing trees has lost its resistance, rapid invasion of the food rich cells of the outer bole and rays commences by fungi that were previously inhabitants of the bark. The results in Table 16 indicate that the colonization of the bole at the time of felling in MSMA trees is substantially greater than in Tordon trees. This is of prime importance to later colonization of stumps by F. annosus, since the pathogen can invade fresh,

uncolonized wood only.

Once the tree is felled and the stump exposed, F. annosus enters into competition for stump substrates. This competition can occur only on the uncolonized areas of the stump surface. The uncolonized areas of the MSMA stumps are not only proportionately smaller than those of Tordon and control stumps, but differ substantially in ability to act as a selective substrate for F. annosus (or other primary wood invading organisms).

The killing of the cambium, during the incubation period between treatment and felling in the MSMA trees, results in destruction of the wood rays. If they are not killed directly by the chemical, they will starve after the death of the cambial area cells. Once these cells are dead, living resistant physiological barriers to invading organisms cease to be effective. All that remains as a barrier to fungal invasion is the residual toxic substances stored in heartwood i.e. resins and tannins. Western hemlock wood, however, has a notoriously low content of resinous materials content (Panshin et al., 1964). The interior gaseous environment in the MSMA trees was radically altered by the elimination of the living hollow cylinder that separates the tree interior from the external atmosphere, and by the tunneling of ambrosia beetles. The work of Cobb et al. (1968) and the reviews of Wagener and Davidison (1954), Hillis (1962) and Shigo (1965) leave little doubt that the host mechanisms of resistance to

fungal attack will be lost by disruption of the polyphenolic producing systems and the internal gaseous nature of living trees and that such disfavor will promote development of secondary organisms over primary invaders.

In summation, MSMA apparently kills the living cells of the cambial area shortly after contact. The result is a rapid destruction of the main phenolic producing cells and the living cylinder that isolates the inner tissues. At the same time or soon thereafter the cells of the wood rays die, further reducing phenol production. The result is that the mechanisms that may favor primary invading organisms over secondary invaders have been destroyed. Secondary organisms, moreover, are already inside the cork, giving an immediate advantage. The primary invaders, unable to compete with the secondary saprophytes, essentially never compete seriously for the substrate. Thus it is not surprising that the amount and speed of cambial kill is the key to stump infection by F. annosus.

The exception to this pattern relates to length of incubation of infection of Tordon stumps (Table 13). The primary reason for this is the axe cut infection of the Tordon treated trees by F. annosus (Table 18). At the time of felling, many of these infections had progressed down the bole and into the stump. The result was that these infections were confounded with infections occurring at the stump surface after felling. Thus, with longer periods of incubation (12

months over nine) more of the infection columns from the axe cuts reached the stump level, hence the higher infection rate after 12 months than after nine months.

Infections from axe cuts can also partially explain the high incidence of infection in Tordon stumps as compared to controls. However, the largest increase, 360% over control stumps (Table 9) for the September 67 treatment felled in March of 68 occurred before axe cut infection had reached stump level (Table 18). It is postulated here that, during certain stages of senescence of the Tordon killed trees, the cells actually became more susceptible to F. annosus than if exposed in a fresh living condition. At this state, it is hypothesized that the cells have lost their ability to resist F. annosus but not saprophytes. Freshly exposed healthy cells, on the other hand, are able to offer some resistance to all invaders, including F. annosus.

Failure of Fomes annosus to infect axe cuts of MSMA-trees is of primary interest. MSMA cuts supported a heavy growth of mold, while Tordon cuts did not. Two explanations are offered: 1) MSMA unlike Tordon kills the tissue and eliminating resistant mechanisms of the host, the wound is thus no longer selective to primary wound invaders, but is subject to saprophytic invaders; 2) MSMA is toxic to F. annosus. F. annosus was found to be completely inhibited on 2.5% malt agar containing 500 ppm. MSMA (Appendix, page 117).

Molds grew luxuriantly on agar containing this level of herbicide. Some growth occurred at 50,000 ppm. Chemical content of the wood near the axe cuts exceeds levels toxic to F. annosus.

MSMA is not believed to influence fungal successions of stump surfaces nor Fomes annosus present in trees prior to injection. Alston, Fox (1965), found arsenic concentrations in stumps of rubber trees poisoned with sodium arsenite to be too low to inhibit growth of fungal pathogens. Fomes annosus present in trees prior to treatment with MSMA was not affected as evidenced by the profuse growth and fruiting on incubated disks taken from such trees at the time of felling.

The finding that the axe cuts treated with Tordon are extremely susceptible to F. annosus is of importance to forest managers, since vast acreages of pre-commercial western hemlock are currently being thinned with this herbicide.

Axe cuts may pose a serious threat of F. annosus damage in the residual stand. Once the pathogen has gained entrance to the bole via the axe cut it finds little difficulty in advancing rapidly in the bole, and may reach the root systems before saprophytes penetrate the slowly dying cambium to check its spread. In the meantime, use of Tordon 101, or other herbicides that produce a slow cambial death, should be discontinued until the effects of these axe cut infections on the residual stand can be determined.

The effects of injection of MSMA are not limited to surface

colonization of stumps. The results in Table 16 demonstrate that vertical fungal colonization behavior is affected. Colonization of the four and six inch levels by associated fungi is more extensive than that of Tordon and control stumps. This is important, since the area colonized by other fungi is no longer a potential substrate for F. annosus. Cobb and Barber (1968) found that stain fungi and F. annosus to be mutually exclusive in stumps of ponderosa pine, Douglas fir and redwood. These stumps, however, were alive, and the Saprophytes did not enjoy the degree of competitive advantage observed here in the dead MSMA stumps.

It is probable that colonization of MSMA stumps by competing fungi will limit invasion by F. annosus to the area colonized at the time of felling. Colonies of the pathogen observed on disks taken from MSMA stumps were nearly always surrounded by competing fungi. Control and Tordon stumps, conversely retained considerable areas that were uncolonized. The F. annosus colonies not surrounded by saprophytes may expand into these uncolonized areas. Thus, low levels of infection in MSMA stumps will not increase, and may in fact disappear completely in a short time. Infections levels observed in the Tordon stumps, however, may or may not increase; they will almost certainly increase in control stumps.

The low percentage area infection rates obtained in stumps exposed to just natural inoculum are of interest (Table 4). Research

is needed to determine what percentage of a stump must be colonized before it can be considered an infection center hazard for the residual stand. It seems reasonable to assume that stumps with low percentage areas of colonization would be a minimum hazard. If this is so, it also seems logical that evaluation of stands for stump infection where the data is experssed in number of, or percentage, of stumps infected may be of little value.

Significance of Results on the Pathological Condition of the Stand Prior to the Study

The importance of the pathological data presented in Table 21 is limited in that it was taken from one stand, and that it represents only hidden decay. However, certain interesting points stand out. For example, the number of trees containing butt stain is very large. The number of trees containing stain is approximately ten times that containing decay (Table 21). Casual observations of stumps at logging operations throughout the Clatsop Tree Farm at Seaside, and in scattered areas throughout the northwest suggests that this condition is common.

Various fungi were isolated from many of the stains, while others contained only bacteria. Species Graphium spp. were most commonly associated with stain. Research is needed on the relationship between these stains and decay processes.

It is of significance to forest managers that trees under 12" D. B. H contained decay twice as often as larger trees (Table 21). Fomes annosus is only slightly more abundant in the smaller trees (4.1 percent versus 3.2 percent), respectively (Table 21).

It is noteworthy that Fomes annosus was responsible for only 6.3% of the stain and 19.5% of the decay (Table 21). Fomes annosus is only one member of many organisms that are causing an appreciable amount of trees with decay (7.0%) and a tremendous amount of staining (77.5%) in apparently sound hemlock of this stand. Observations of fruiting bodies on damanged and down trees is indicative that Fomes pinicola, Poria subacida, Fomes applanatus, and Armellaria mellea were present in the study area.

Drying, Deterioration, Breakage, and De-barking

The effect of cambial kill on drying and deterioration is not surprising; trees dry rapidly, once the cambium is dead. This may result in loosening of the bark, and in exposure of the inner wood to changes in the external environment. The treatments had no apparent effect on "sinker" trees (Table 27 and Figure 7), there even may have been an increase in sinkers. Since wet wood is a pathological condition (Boyce, 1961) it would not be expected to be appreciably affected by the herbicide treatment. The increase may be possibly explained by the fact that when the bark loosens moisture is allowed

to seep into the butts from the external environment, especially in trees growing in old decayed logs. Reasons for increased deterioration have already been discussed in the section of fungal invasion of the bole prior to felling.

Breakage increased (Table 28) with drying brought on by the MSMA treatments (Table 26). Much of this breakage occurred due to the felling of the trees across logging trails, and presumably could have been avoided to some degree.

Results on bark removal (Table 25) provide information of considerable practical use. December MSMA treatment gave excellent cambial kill, which apparently occurs at a time unfavorable for bark removal. The December treatment had little apparent effect until the trees became physiologically responsive to this compound. Apparently the trees initiated spring growth before the cambium dried. The bark was easily separated where the new wood had been initiated but killed before secondary wall thickenings occurred. In trees killed by MSMA when the bark was tight (September) little advantage in bark removal was obtained. The increased lateral spread of the herbicide in the December treatment was advantageous in that it eliminated hard-to-peel strips of live bark.

Herbicidal Action

Although no experiments were designed to test for the mechanism of herbicide action in the trees, it was clear that different mechanisms were operating. MSMA is known to be a contact herbicide (one that kills tissue on contact) whose main mechanism of action is as a respiration inhibitor, while picloram and 2, 4-D (the constituents of Tordon) are known to be growth regulator compounds. Thus it would be expected that MSMA would have a greater effect on a wide spectrum of tree tissues while Tordon would be more active in the most actively growing tissues. Both compounds produced most rapid crown deterioration when applied during seasons of active growth. However, MSMA applied in winter gave the best kill of stem tissues even though mortality does not occur until the advent of physiological activity of the tree in the spring.

Apparently when applied in the active season the chemical is rapidly transported to the crown of the transpiration stream, lateral distribution is less, resulting in living streaks of cambium between the dead areas adjacent to herbicide conduits. It is not surprising that the chemical is less active in the winter when the tree is physiologically inactive, since arsenicals are generally known to be respiration inhibitors and cells with low respiration would be much less affected than ones with high rates.

Summary and Conclusions

The results clearly show that little stump infection by F. annosus occurs in western hemlock killed by MSMA. At the same time the trees dry quickly on the stump, and bark is very easily removed. Disadvantageously wood deterioration and breakage increase. Breakage problems can be reduced by careful felling, but the deterioration cannot be so easily prevented. The type of deterioration encountered is tolerable in pulp wood operations, however, and in stands managed for pulp, treatments such as these could probably be used to economic as well as pathological benefit. The effects of each herbicide on various factors studied in relation to herbicide treatment is presented in Table 29.

Further research is needed before some of the questions arising from this study can be understood. For example, what degree of F. annosus stump infection poses a threat to the residual stand? Are these levels reached in western hemlock stumps?

The role of the vast complex of other organisms responsible for decay and stain in western hemlock must be investigated. Little is known about the potential of these organisms for damage in second growth hemlock, nor as their relationship to F. annosus. The stands under consideration are not old field plantations with limited wood decay organisms, but have existed for centuries in balance with a

Table 29. Summary of the effects of herbicide on factors under test as compared to controls.

Response	MSMA	Tordon
Stump infection by Fomes annosus	a practical control	variable
Saprophyte colonization of boles and stumps	rapid increase	slow at first rapid increase later
ambrosia beetle attack	increase	negligible to slight increase
foot graft damage	slight	heavy
breakage	large increase	slight increase
Debarking	excellent	poor
drying	increase	negligible
deterioration	increase	increase
bole infection via axe cuts	no effect	increase

host of wood decaying organisms. The immediate question is: do harvest and thinning operations upset this balance to favor F. annosus and possibly other disease causing organisms?

This thesis concludes that epidemiology of F. annosus in commercial thinnings of western hemlock is subject to deliberate prophylactic treatment by pre-harvest killing with MSMA. The utility or necessity for such treatment on an economic and long-term basis is beyond the scope of this paper, but appears encouraging.

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APPENDICES

Appendix Table 1. Effect of pre-killing of trees with MSMA and Tordon 101 on Fomes annosus invasion of stumps subjected to natural air spora only.

Percent area colonized two inches below each stump surface

C3*	C6	C9	C6	C9	C12	C9	C12	C15	C12	C15
M9-6	M9-9	M9-12	M12-6	M12-9	M12-12	M3-6	M3-9	M3-12	M6-6	M6-9
0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.3	0.0	9.0	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.0	2.2	0.0	0.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.0	5.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.9	0.8	0.0	0.0	2.7	0.0	0.0	0.0	0.0	0.0	0.0
0.2	1.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Σ1.5	9.9	0.1	1.0	2.7	0.0	0.0	0.0	0.0	0.0	0.0
%0	30	0	0	10	0	0	0	0	0	0
C3	C6	C9	C6	C9	C12	C9	C12	C15	C12	C15
T9-6	T9-9	T9-12	T12-6	T12-9	T12-12	T3-6	T3-9	T3-12	T6-6	T6-9
6.3	4.5	4.9	0.9	7.0	3.8	0.0	0.0	0.0	0.0	0.0
2.2	4.6	2.0	0.0	3.3	0.0	1.0	19.3	6.8	0.0	1.8
0.2	0.0	6.8	0.3	0.0	0.0	1.9	0.0	2.0	0.0	0.0
0.6	0.0	9.2	1.6	0.9	0.0	2.7	0.0	8.1	0.0	0.0
3.0	2.0	0.3	0.9	8.5	0.0	0.8	15.6	0.0	0.0	0.0
20.6	1.2	0.0	0.0	1.2	0.0	0.0	0.0	0.0	0.0	0.0
13.2	1.3	5.9	0.5	0.3	0.0	4.5	0.0	7.6	0.0	0.0
3.2	0.0	5.8	2.7	0.0	0.0	2.8	0.0	0.0	0.0	0.0
0.2	0.6	2.1	0.0	7.5	15.3	0.4	0.0	5.2	0.0	0.0
12.2	3.3	0.3	1.6	0.3	14.1	0.8	0.0	0.0	0.0	0.0
Σ61.7	17.5	37.3	8.5	29.0	33.2	14.9	34.9	29.7	0.0	1.8
% 100	70	70	30	50	30	50	20	50	0	10

C3	C6	C9	C12	C15
2.6	0.5	7.5	0.0	39.9
2.1	6.5	0.0	0.0	52.5
11.8	5.8	0.0	0.0	2.5
1.3	2.0	7.0	0.0	3.6
0.0	9.5	13.5	0.0	0.0
2.5	0.0	3.9	0.0	0.8
2.2	14.4	1.8	0.0	0.0
0.0	1.8	0.0	1.8	2.3
2.5	0.7	0.0	0.0	5.7
0.3	3.1	0.0	0.0	1.9
Σ25.3	44.3	33.7	1.8	109.2
% 70	70	50	10	70

*Treatment codes explained in Table 3

ΣTotals

% Percentage of stumps having percent area colonized larger than 1.0.

Appendix Table 2. Effect of pre-killing of trees with MSMA and Tordon 101 on Fomes annosus invasion of stumps subjected to both natural and artificial inoculation.

Percent area colonized two inches below each stump surface

C3* M9-6	C6 M9-12	C9 M9-12	C6 M12-6	C9 M12-9	C12 M12-12	C9 M3-6	C12 M3-9	C15 M3-12	C12 M6-6	C15 M6-9
3.2	5.5	5.9	0.2	0.3	0.0	1.8	0.0	3.2	2.1	8.6
0.5	0.0	6.5	0.0	0.0	0.0	2.3	0.0	0.0	0.0	9.4
1.4	0.2	3.8	0.0	0.5	2.3	0.0	0.0	1.2	1.9	7.0
3.5	0.2	0.5	0.3	0.9	0.0	3.4	0.0	1.1	0.0	9.2
6.4	0.0	7.8	0.0	0.0	0.0	5.2	0.0	2.0	0.0	17.5
4.0	0.0	1.7	0.0	0.0	0.0	10.0	0.0	0.0	0.0	5.6
10.9	0.0	1.2	0.0	0.0	0.0	22.5	0.8	2.4	0.8	5.1
7.0	0.2	8.3	0.0	0.0	0.0	0.2	0.0	2.1	0.9	0.0
3.4	0.0	10.0	0.0	0.4	0.0	1.4	1.9	0.0	0.0	3.3
13.7	0.6	19.8	0.0	3.6	0.0	2.0	0.8	0.0	0.0	0.0
Σ54.0	6.7	65.5	0.5	5.7	2.3	48.8	3.5	12.0	5.7	65.7
σ 4.2	1.7	5.6	.1	1.1	.7	6.8	.6	1.2	.8	5.1

C3 T9-6	C6 T9-9	C9 T9-12	C6 T12-6	C9 T12-9	C12 T12-12	C9 T3-6	C12 T3-9	C15 T3-12	C12 T6-6	C15 T6-9
86.0	40.9	12.2	31.2	1.8	8.9	30.8	0.5	91.2	0.0	9.5
80.0	7.0	31.8	83.6	16.6	8.1	32.2	0.0	20.0	0.8	23.1
1.4	12.7	50.0	78.2	4.0	0.0	0.8	0.0	26.8	0.0	18.9
82.0	88.7	4.6	47.8	4.2	2.2	10.1	1.6	36.8	0.0	7.2
0.6	9.7	21.0	16.5	9.1	1.5	11.4	0.9	2.4	4.4	3.1
72.0	7.8	0.0	18.6	6.9	0.0	10.0	5.4	29.8	6.2	20.1
26.1	30.0	15.8	8.2	3.9	9.0	35.9	4.8	6.4	1.1	19.8
81.3	13.8	8.7	15.9	0.0	0.0	32.4	2.0	17.9	0.0	3.5
83.5	27.4	79.8	10.4	82.3	0.0	85.8	0.8	5.2	0.0	8.4
56.9	25.3	83.2	7.2	5.5	1.1	62.4	0.0	7.3	0.0	15.6
Σ569.8	263.3	307.1	317.6	134.3	31.8	311.8	16.0	243.8	12.5	129.2
σ 34.5	24.6	30.4	28.6	24.6	4.1	26.2	2.0	26.2	2.2	7.4

C3	C6	C9	C12	C15
12.2	54.0	27.3	0.0	82.8
8.1	11.6	12.1	0.0	95.7
13.2	22.4	15.8	0.0	88.8
18.8	37.6	38.8	1.4	89.9
17.4	85.6	3.2	4.8	87.8
5.0	81.0	26.6	0.0	6.4
10.3	80.4	9.7	0.0	95.6
35.3	16.0	8.8	0.0	87.9
4.6	22.8	74.4	22.4	93.7
33.4	15.4	27.4	1.2	84.3
Σ158.3	426.8	244.1	39.8	813.9
σ 10.8	30.2	30.7	7.0	26.7

*Treatments codes explained in Table 3

Σ= Totals

σ = Standard deviation

Appendix Table 3. Explanation of treatment codes used in Appendix Tables 1 and 2 presenting stump treatment results.

Code	Explanation
C3, C6, C9,	Used by themselves to indicate the date of felling of control trees.
C12, C15	Used in conjunction with a chemical treatment code to indicate the felling of the treated trees. The numbers 3, 6, 9, 12, 15 represent the following months of felling: March 1968, June 1968, September 1968, December 1968 and March 1969
M9 and T9	M indicates MSMA and T Tordon 101. The numbers 9, 12, 3, 6,
M12 and T12	signify September 1967, December 1967, March 1968 and June 1968
M3 and T3	dates of chemical treatment
M6 and T6	
-6, -9-12	Dashed numbers following kind and date of treatment represent the time between chemical treatment and felling to the nearest month (i. e. six, nine, and 12 months)
Examples	
C3	Indicates the tree was poisoned with MSMA in September 1967 and
M9-6	felled six months later in March of 1968
C12	Indicates the tree was poisoned in December of 1967 with Tordon 101
T12-12	and felled 12 months later in December of 1968
C15	Indicates a control (living tree) was felled in March of 1969

Appendix Table 4. Fomes annosus infection of residual treated tree stumps expressed as a percent of the surface infected per treated stump over the mean percent area infected of control stumps felled on same date.

Stump number (1-10)	Month of treatment with herbicides: MSMA												Tordon 101											
	September			December			March			June			September			December			March			June		
	Months of incubation																							
	6	9	12	6	9	12	6	9	12	6	9	12	6	9	12	6	9	12	6	9	12	6	9	12
1	20	13	24	1	1	0	7	27	4	70	11	x	544	95	50	74	73	330	27	17	112	27	12	x
2	3	5	27	1	2	0	10	63	1	63	11		506	16	130	196	78	270	132	53	33	147	28	
3	9	1	16	0	4	0	14	27	1	26	9		9	28	205	183	16	73	3	30	45	207	23	
4	2	1	2	0	2	7	22	0	2	30	11		520	208	19	112	17	50	41	180	3	37	9	
5	41	1	32	0	15	0	42	0	3	0	21		4	23	86	37	28	300	47	160	37	0	4	
6	25	0	7	0	0	0	94	0	3	0	7		456	18	65	44	16	37	41	67	8	0	25	
7	69	0	5	0	0	0	1	0	0	0	6		165	70	36	19	337	0	147	26	22	0	24	
8	44	0	34	0	0	0	6	0	0	0	4		515	32	327	37	225	0	133	0	6	0	4	
9	22	0	41	0	0	0	9	0	0	0	0		529	65	341	24	0	0	352	0	9	0	101	
10	87	0	81	0	0	0	0	0	0	0	0		360	60	0	17	37	0	256	0	25	0	19	
Incubation total	322	21	269	2	24	7	205	117	14	189	80		3608	615	1259	743	817	1060	1179	533	300	418	249	
Month treatment totals		612			33			336			269			5482			2620			2012			677	
Mean		20.4			1.1			11.2			13.4			182.7			87.4			73.6			33.2	
Grand mean						11.5											94.2							

Appendix Table 5. Rates of vertical colonization of the stumps of the various treatments by fungal competitors of Fomes annosus.

Treatments (MSMA)		Number of disks per infection class*																																	
		MSMA																																			
		Treated			Sept. 67						Dec. 67									March 68									June 68								
		Felled			March 68			June 68			Sept. 68			June 68			Sept. 68			Dec. 68			Sept. 68			Dec. 68			March 69			Dec. 68			March 69		
Disk levels depths 2", 4", 6"			2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6
Disk percent*	1.	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0
Colonization	2.	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0
Class	3.	9-	8-	8	0-	0-	0	0-	0-	0	0-	1-	2	0-	0-	0	0-	0-	0	0-	1-	3	0-	0-	1	0-	0-	0	0-	1-	2	0-	0-	0	0-	0-	0
	4.	10-	2-	2	3-	3-	3	3-	3-	3	3-	2-	1	3-	3-	3	3-	3-	3	3-	2-	0	3-	3-	2	3-	3-	3	3-	2-	1	3-	3-	3	3-	3-	3

Treatments (Tordon)		Tordon																																	
		Treated			Sept. 67						Dec. 67									March 68									June 68								
		Felled			March 68			June 68			Sept. 68			June 68			Sept. 68			Dec. 68			Sept. 68			Dec. 68			March 69			Dec. 68			March 69		
Disk levels depths 2", 4", 6"			2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6
Disk percent*	1.	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	3-	3	0-	0-	1	0-	0-	3	0-	0-	0	3-	3-	3	0-	0-	0			
Colonization	2.	0-	5-	6	0-	0-	0	0-	0-	0	0-	0-	0	0-	2-	3	3-	0-	0	0-	3-	2	3-	0-	0	0-	0-	0	0-	0-	0	0-	0-	1			
Class	3.	0-	4-	4	0-	0-	2	0-	2-	2	0-	0-	2	0-	1-	0	0-	0-	0	0-	0-	0	0-	0-	0	0-	1-	2	0-	0-	0	0-	1-	2			
	4.	10-	1-	0	3-	3-	1	3-	1-	-	3-	3-	1	3-	0-	0	0-	0-	0	3-	0-	0	0-	0-	0	3-	2-	1	0-	0-	0	3-	2-	0			

Treatment (No chemical)																	
	Felled	March 68			June 68			Sept. 68			Dec. 68			March 69			
Disk levels depths 2", 4", 6"		2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	2-	4-	6	
Disk percent*	1.	0-	0-	4	0-	0-	0	0-	2-	3	3-	3-	3	0-	0-	3	
Colonization	2.	0-	7-	6	0-	0-	3	0-	1-	0	0-	0-	0	0-	3-	0	
Class	3.	0-	3-	0	0-	1-	0	0-	0-	0	0-	0-	0	0-	0-	0	
	4.	10-	0-	0	3-	2-	0	3-	0-	0	0-	0-	0	3-	0-	0	

*Percent area of disk surface colonized, 1. 0-25%, 2. 25-50%, 3. 50-75%, and 4. 75-100%. March 68 felling has ten replicates, the rest three. Length of stump incubation prior to sampling by felling date; March 68-16 weeks, June 68-16 weeks, Sept. 68-25 weeks, Dec. 68-22 weeks, March 69-16 weeks.

EFFECT OF MSMA IN MALT AGAR ON GROWTH OF *Fomes annosus*

Methods

MSMA was added to autoclaved 2.5% malt agar to give media containing 500, 5,000 and 50,000 p.p.m. of active ingredients of MSMA. The media was poured into petri plates and after cooling was inoculated with agar disks containing *Fomes annosus* cut from seven day old cultures growing in petri dishes on malt agar. Twenty replicates of each concentration were made.

Results

In no case did *Fomes annosus* succeed in growing on the MSMA agar. Many of the plates were contaminated with molds. It is believed the mold spores were in the herbicide solution since some of the growth initiated below the surface of the agar. At any rate at concentrations 500 ppm the molds attained luxuriant growth and even obtained some growth at 50,000 ppm.

Discussion

These results are consistent with observations of heavy growth of molds on MSMA treated axe cuts and reports in literature of molds being the only fungi capable of assimilating organic arsenical compounds (Alexander, 1966).