# Late Yellow Rust
[*Pucciniastrum americanum* (Farl.) Arth.]
# of
# Red Raspberry (*Rubus idaeus* L.)

by
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**LATE YELLOW RUST OF RED RASPBERRY**

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# ABSTRACT

M.Sc. MARGIE LUFFMAN Plant Science

LATE YELLOW RUST [*Pucciniastrum americanum* (Farl.) Arth.]
OF RED RASPBERRY (*Rubus idaeus* L.)

Late yellow rust [*Pucciniastrum americanum* (Farl.) Arth.] has recently become a serious problem in *Rubus idaeus* L. (red raspberry) plantings of the Atlantic provinces of Canada. The alternate host is *Picea glauca* (Moench) Voss. (white spruce).

A program to determine the optimum schedule of fungicide applications for rust control based on the life cycle of the pathogen was established. Anilazine applications at the time of aeciospore release reduced leaf and fruit infections. Later applications at flowering time had a deleterious effect on fruit set and yield. Sulphur used late in the season did not improve disease control and had a negative effect on fruit yield. A program of three anilazine applications early in the season, terminating before flowering gave the best disease control.

Infection studies with different raspberry cultivars in the greenhouse resulted in foliar rust on all cultivars tested. There were considerable differences among the cultivars in the severity of rust that developed.

Field inoculations of fruit clusters of raspberry cultivars resulted in varying amounts of infection. Two cultivars did not become infected.

# RÉSUMÉ

M.Sc. MARGIE LUFFMAN Plant Science

## LA ROUILLE JAUNE TARDIVE [*Pucciniastrum americanum* (Farl.) Arth.] DE LA FRAMBOISE ROUGE (*Rubus idaeus* L.)

La rouille jaune tardive [*Pucciniastrum americanum* (Farl.) Arth.] est devenue, récemment, une maladie importante des plantations de *Rubus idaeus* L. (framboise rouge) dans les provinces maritimes du Canada. *Picea glauca* (Moench) Voss. (épinette blanche) est la plante-hôte intermediaire.

Des essais ont eté établi ayant pour but de determiner le meilleur plan pour l'application des fongicides pour le contrôle de cette rouille. Le plan d'application sera basé sur le cycle de vie de ce pathogene. Des applications d'anilazine, lors du relachement des éciospores ont réduit les infections foliaires et fruitières. Les applications tardives, c'est-à-dire lors de la fleuraison, ont eu un effet très délétère sur l'établissement et le rendement des fruits. Le soufre, appliqué tard durant la saison a eu aucun effet bénéfique pour le contrôle de ce pathogène et, de plus, a négativement affecté le rendement des fruits. Un programme de trois applications d'anilazine, débutant tôt la saison et terminant avant la fleuraison fut le meilleur pour le contrôle de cette maladie.

Plusieurs cultivars de framboises ont été étudie en serre pour leur susceptibilité à l'infection et ils furent tous atteints de rouille foliaire. Par contre, il y a eu des differences entre les cultivars dans la sévérité d'infection par la rouille.

Lors des inoculations au champs des fruits de plusieurs cultivars, il y a eu des effets variés dans l'intensité de l'infection. Deux de ces cultivars étudiés n'ont pas été atteint d'infection par cette rouille.

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## TABLE OF CONTENTS

## LIST OF TABLES

## LIST OF PLATES

# INTRODUCTION

Late yellow rust caused by *Pucciniastrum americanum* (Farl.) Arth. has occurred sporadically in red raspberry (*Rubus idaeus* L.) plantings in the Atlantic provinces of Canada for many years. In 1984 and 1985 it seriously affected a number of fields, rendering the fruit unmarketable and causing premature cane defoliation which can lead to a reduction in winter hardiness. This disease poses a threat to raspberry production in the Atlantic provinces of Canada.

Since its introduction from the Ottawa Research Station in 1972, the raspberry cultivar Festival has become one of the most widely planted cultivars in the region; it is highly susceptible to late yellow rust infection of both leaf and fruit. Other cultivars grown appear to have varying degrees of resistance to this rust.

The alternate host of *Pucciniastrum americanum* is white spruce [*Picea glauca* (Moench) Voss.]. The recent outbreak of late yellow rust resulted in severe cone infections and subsequent reductions in seed germinability. White spruce seed production is an important industry in the Maritimes.

Very little is known about the epidemiology of late yellow rust. In view of the variability of resistance exhibited by the different cultivars grown in the Atlantic provinces of Canada, a closer examination of this was warranted. Information is required to develop an optimum schedule of fungicide applications to control late yellow rust. This study was undertaken to elucidate the epidemiology of the disease and to develop an improved control program.

This thesis is submitted in the form of original papers suitable fcr journal publication. The thesis format has been approved by the Faculty of Graduate Studies and Research, McGill University, and follows the conditions outlined in the *Guidelines Concerning Thesis Preparation*, section 7 "Manuscripts and Authorship" which are as follows:

"The candidate has the option, subject to the approval of the Department, of including as part of the thesis the text of an original paper, or papers, suitable for submission to learned journals for publication. In this case the thesis must still conform to all other requirements explained in *Guidelines Concerning Thesis Preparation*. Additional material (experimental and design data as well as descriptions of equipment) must be provided in sufficient detail to allow a clear and precise judgement to be made of the importance and originality of the research reported. The abstract, full introduction and conclusion must be included, and where more than one manuscript appears, connecting texts and common abstracts, introduction and conclusions are required. A mere collection of manuscripts is not acceptable; nor can reprints of published papers be accepted.

While the inclusion of manuscripts co-authored by the candidate and others is not prohibited by McGill, the candidate is warned to make an explicit statement on who contributed to such work and to what extent, and Supervisors and others will have to bear witness to the accuracy of such claims before the Oral Committee. It should also be noted that the task of the External Examiner is made much more difficult in such cases, and it is in the Candidate's interest to make authorship responsibilities perfectly clear."

Although all the work reported here is the responsibility of the candidate, the project was supervised by Dr. D. Buszard, Department of Plant Science, Macdonald College of McGill University. The anilazine residue analyses were done by the Atlantic Provinces Pesticide Residue Lab, Kentville, Nova Scotia. For all four manuscripts, Dr. D. Buszard is the sole co-author. The first manuscript is *in press* in the Canadian Journal of Plant Science. The other manuscrips are being prepared for submission to the Canadian Journal of Plant Science.

# LITERATURE REVIEW

## 1.0 ASPECTS OF RASPBERRY PRODUCTION AND MORPHOLOGY

### 1.1 Production

#### 1.1.1 Canadian Production Areas

Raspberry production is a lucrative enterprise but the extreme perishability of the harvested fresh fruit makes it an increasingly scarce commercial commodity; 92% of the production is processed (Anonymous 1987). In British Columbia's Fraser Valley 75% of Canadian commercially grown raspberries are produced while 25% are grown in Ontario and Quebec (Shoemaker 1978).

#### 1.1.2 Maritime Raspberry Production Yields and Values

In the main production areas of the world raspberry yields of 10-14 tonnes per hectare are obtained (Shoemaker 1978). Maritime raspberry production is low and the farm size is small when compared to the rest of Canada. In 1983, 1984, and 1985 Canada produced 15387, 14999, and 14755 tonnes respectively. Their corresponding farm gate values were $19,377,000, $23,663,000, and $34,592,000 (Anonymous 1984, 1987).

#### 1.1.3 Types of Raspberries Grown and their Origin

All commercially grown raspberries in Canada are standard summer-bearing reds (Shoemaker 1978). According to Tozer (1985), the best modern red raspberry cultivars are derived from two *Rubus idaeus* subspecies, *R. idaeus* var. *vulgatus*, the European native and *R. idaeus* var. *strigosus*, the North American native. Although both are ecotypes of *Rubus idaeus* Jennings (1988) refers to the European raspberry as *R. idaeus* and the American as *R. strigosus*.

Even though this genetic base is relatively narrow, many highly productive cultivars have been developed.

## 1.2 Anatomy

### 1.2.1 Root and Cane Development

The red raspberry (*Rubus idaeus* L.) is an unusual horticultural species in that its aboveground part, the canes, are biennial while the root system is perennial. Bailey (1932) used the term primocane when referring to the first year mature vegetative shoot or cane and the term floricane in referring to the second year fruiting shoot or cane. Shoots which bear leaves can develop from axillary buds on the mother cane or from adventitious buds developing on the roots.

### 1.2.2 Fruit Classification

The fruit of red raspberry is of the aggregate type consisting of a collection of drupelets or fleshy sections containing a pyrene. Each pyrene has a hard endocarp enclosing one or two seeds (Reeve 1954). Each drupelet evolves from an individual ovary. Cultivars with a high degree of self-fertility show regularity of drupelet set while self-infertile types may result in berries which are one-sided or crumbly (Shoemaker 1978).

### 1.2.3 Flower Bud Formation

Waldo (1933) described the process of fruit bud formation on brambles. The differentiation process begins with the broadening of the growing point of a vegetative bud, followed by elongation of this growing point with axillary growing points appearing on each side. Following this, the terminal growing point becomes the terminal flower of the cluster while the illary points may become the terminal flowers of lateral flower clusters.

Generally, fruit bud differentiation proceeds from the cane terminals downward.

### 1.2.4 Fruit Growth Stages

Shoemaker (1978) described three definite fruit growth stages. The first is a period of rapid growth beginning at full bloom. This is followed by a period of slower growth during which the endocarp hardens. The third stage is one of rapid growth continuing to maturity. Carpellary tissue accounts for most of the growth of developing fruits.

## 1.3 Seasonal Phases of Woody Shoot Growth

Hudson (1959) described the well-defined sequence of seasonal phases that each woody shoot or cane passes through during its life cycle. Shoots arise as root suckers from buds on roots, as stem suckers from axillary buds, or as lateral branches from aboveground axillary buds Shoots arising from root bud suckers pass through 9 distinct phases. In phase 1, root buds are initiated. As the root bud commences to grow, phase 2 occurs, involving the development of an underground sucker. Phase 3 occurs when this sucker emerges above ground. At this point, elongation ceases and leaf expansion begins, resulting in a tight rosette of leaves referred to as a primary rosette. Phase 4 is winter dormancy of the emerged shoots. Phase 5 begins after dormancy when environmental conditions (warmer temperatures and lengthening days) promote the rapid elongation of the shoot. Leaf initiation, internode elongation, and leaf expansion occur concurrently throughout phase 5. Phase 6 occurs at the end of the growing season when the shoot ceases to elongate and produces a secondary rosette of leaves at its tip. Flower primordia are initiated by most of the axillary meristems, but they are incapable of growing out at this time. Conversely, Williams (1959c, 1960) found that fall bearing raspberries initiate flower buds while shoots are still elongating. Fall bearing or primocane fruiting raspberries produce fruit at the tips of one-year old growth at the end of the first season. In the second season only the lower part of the cane produces fruit (Jennings 1988). At the end of phase 6 leaves senesce and fall. Phase 7 involves breaking dormancy of flower buds by meeting a cold requirement after which phase 8 begins with the promotion of flowering and then fruiting. Fruiting laterals developed from the fruit buds bear both leaves and flowers. Basal buds commence elongation to produce vegetative replacement shoots which will repeat the biennial cycle. Phase 9 involves senescence and death of the floricanes which die back to the position from which replacement shoots have grown.

## 1.4 Environmental Effects on Primocane Development

Williams (1959a) examined the relationship between environmental factors and the developmental habit of primocanes until the cessation of shoot elongation. He studied the behavior of autumn and spring emerging suckers. He found that during the autumn period of emergence, vegetative shoot elongation ceased; terminal rosettes of leaves formed and basal buds in the replacement zone enlarged. This period is characterized by shortening days and falling temperatures. Spring emergence of suckers occurred during lengthening days and rising temperatures. Newly emerged suckers elongated without rosetting and there was no clear differentiation of replacement zone basal buds. There was renewed activity in the apical meristem of previously emerged suckers, with new leaves opening and shoot elongation. It was also found that there was no new root sucker emergence during the period of longest days and highest temperature, but maximum shoot elongation occurred at this time.

## 1.5 Environmental Effects on Shoot Growth and Dormancy

Williams (1959b) and Jennings (1988) discussed the effects of daylength and temperature on the growth and dormancy of young shoots. The shorter days and lower temperatures of autumn induced dormancy and rapidly reduced the elongation rate in shoots. Long days and higher temperatures maintained shoots in a state of vegetative elongation. A period of low temperatures was found to be necessary to break dormancy so that the apical meristem could resume growth. Jennings et al. (1972) found that canes which were prematurely defoliated entered dormancy more slowly than canes allowed to naturally shed their leaves. It was felt that a dormancy inducing factor formed in the leaves.

### 1.6 Environmental Effects on Flower Initiation and Inflorescence Development

The effects of environment on flower initiation and inflorescence development was examined by Williams (1959c, 1960). He found that flower initiation in the axillary buds occurred in early autumn when days were shorter and temperatures were cooler. Flower initiation occurred later in axillary buds lower down the shoot. Basal buds remained vegetative and developed into vegetative shoots which replaced the fruiting shoot the following year. After flower initiation, low temperatures arrested development of the inflorescence. This would correspond to conditions encountered during the dormancy/overwintering stage. In the following year, warmer temperatures and longer days promoted inflorescence development. Since flowers of fall bearing raspberries are initiated in long days and high temperatures, Keep (1961) presumed that the variable character of fall fruiting resulted from the interaction of the developmental processes flower initiation in the terminal bud and cane elongation with each other and the environment.

## 2.0 RUST DISEASE PROCESS

### 2.1 Rust Taxonomy

Phylogenic classification of fungi ranks all plant rusts in the class Basidiomycetes and subclass Heterobasidiomycetidae. Rust fungi are in the order Uredinales which constitutes one of the largest groups in Basidiomycetes (Hiratsuka and Sato 1982). All rusts are obligate parasites in that they rely on host tissue for survival (Arthur 1929). Uredinales cover a wide range of obligate parasites which have been found to parasitize some ferns and virtually all groups of higher plants (McIntosh and Watson 1982).

## 2.2 Rust Life Cycles and Spore Forms

Rust fungi have three basic types of life cycles which are determined by the types of spores formed. Each species of rust consists of two stages of development which are usually designated by the gametophytic or haploid (N) state and the sporophytic or diploid (2N) state (Arthur 1934). There is variation in the complexity of rust life cycles with some rusts producing up to five different spore stages. When they occur, the typical progression of spore stages with accompanying nuclear status is basidiospore (N), pycniospore or spermatium (N), aeciospore (N+N), urediniospore (N+N), and finally teliospore (N+N-->2N-->N).

### 2.2.1 Rust Life Cycle Classification

Rusts which produce five spore stages are referred to as macrocyclic (Walker 1969). When the uredinial stage is absent, the rust is termed demicyclic; when both the uredinial and aecial stages are absent the rust is designated as microcyclic (Littlefield and Heath 1979). The urediniospores are functionally the conidial, repeating, or recycling spores. They have the ability to reproduce the uredial stage as long as suitable host material exists in appropriate environmental conditions. This stage is frequently responsible for the rapid spread of rusts. The teliospores are the ultimate sori of the sporophytic phase (Arthur 1929). Some species of macrocyclic and demicyclic rusts require two distinct alternating host plants for completion of their life cycle. Such rusts are termed heteroecious. Other species produce all spore forms on the same host and these nonhost alternating rusts are termed autoecious. Because of the abbreviated life cycle of microcyclic rusts, they are necessarily autoecious. Hiratsuka and Sato (1982) combined the three types of rust life cycle with their heteroecious or autoecious nature and proposed five basic variations as follows: microcyclic, heteromacrocyclic, automacrocyclic, heterodemicyclic, and autodemicyclic.

## 2.3 *Pucciniastrum americanum*

### 2.3.1 Taxonomy

Late yellow rust, *Pucciniastrum americanum* (Farl.) Arth. is classified in the family Melampsoraceae. Initially *P. americanum* was described under the name *Pucciniastrum arcticum* (Lag.) Tranz. var. *americanum* by Farlow (1908). However, in 1920 Arthur raised the fungus to specific rank due to differences in host range and morphology of the aecia and urediinia. Dodge (1923) listed *Rubus* species infected by *P. americanum.* These included *R. idaeus* var. *strigosus*, and *R. neglectus.* Because neither *P. americanum* nor *P arcticum* had been associated with their aecial forms, Darker (1927) conducted experiments to establish their connections with alternate hosts. He showed that *P. americanum* on *R.* idaeus var. *strigosus* has its aecial stage [*Peridermium ingenuum* (Arth.)] on white spruce, *Picea glauca* (Moench) Voss. Successful infections were obtained in both directions. No infection was obtained on *Rubus triflorus*. *P. arcticum* on *R. triflorus* also has its aecial stage (*Peridermium ingenuum*) on *Pices glauca.* Again, successful infections were obtained in both directions. No infection was obtained on *Rubus idaeus* var. *strigosus.* Therefore, while *Peridermium ingenuum* apparently designated the aecial stage of both *P. americanum* and *P. arcticum*, artificial infections established both as distinct species. Moss (1926) clearly delineated morphological differences in the characteristics of the uredinial peridium for both species. The peridium of *P. americanum* protruded beyond the epidermis and was found to be more sharply conical than that of *P. arcticum* which was low and broad.

### 2.3.2 Host Plants

Late yellow rust on red raspberry is also referred to as late leaf rust, autumn rust and late raspberry rust (Converse 1966). It is a heteroecious rust fungus which infects red raspberry (*Rubus idaeus* L.), purple raspberry (*R. neglectus* Peck) and wild red raspberry (*R. strigosus* Michx.). The alternate host is white spruce [*Picea glauca* (Moench) Voss ] (Arthur 1929).

### 2.3.3 Life Cycle and Spore Forms

*Pucciniastrum americanum* is a macrocyclic rust with five different spore forms. The gametophytic stage of the life cycle occurs on white spruce with the pycnia appearing in groups along the needles. Aecia usually develop on the needles of the current year and rarely on the cone scales (Ziller 1974). The sporophytic phase of the life cycle occurs on the *Rubus* host with uredinia and telia forming within rust pustules on the adaxial leaf surface, petioles, canes, and even fruit (Anderson 1956). Dodge (1923) and Darker (1929) described the life cycle of *P. americanum*. The subcuticular pycnia or spermagonia which are borne on the gametophytic mycelium on the spruce needles rupture for the escape of pycniospores. The gametophytic mycelium produce the aecia which enclose the masses of aeciospores. Aeciospores released by the spruce in late spring cause primary infections on red raspberry plants. Uredinial pustules which develop on the adaxial leaf surface contain urediniospores. These are the first spore form to arise from the sporophytic diploid mycelium. The urediniospores recycle on raspberry causing secondary infections on leaves and fruit as the growing season progresses. Later in the season teliospores develop within the pustules. The following spring the teliospore germinates and develops a basidium bearing basidiospores. Dodge (1923) surmised that teliospores overwinter on infected canes and leaf litter, giving rise to a source of inoculum for white spruce in early spring.

He also suggested that urediniospores may overwinter on raspberry in order that in the following growing season, renewal of infection occurs on raspberry, independent of white spruce.

### 2.3.4 Symptoms on Spruce

Visual symptoms of *P. americanum* on white spruce are much less pronounced than those on raspberry. It is normally a needle rust (Plate 1) which occasionally occurs on cone scales. Infected needles have an orange rust coloured appearance and eventually drop (Arthur 1920). Severe cone infection is evidenced by orange-brown discolourations on the cone scales (Plate 2) and subsequent reduction in seed germinability (Smith et al. 1986). In the past, the occurrence of *P. americanum* as a cone scale rust on white spruce was more of a curiousity than a problem. However, extremely high levels of cone infection in 1984 could reflect a change in its status from an unimportant disease (T. Renault, personal communication).

### 2.3.5 Symptoms on Raspberry

Symptoms of *P. americanum* on raspberry can occur on the leaves, petioles, shoots, fruit calyces (Plate 3), and even the fruit (Anderson 1956). Uredinial pustules on the adaxial leaf surface appear as powdery yellow-orange spore masses in midsummer (Plate 4). Curling of the leaflet (Plate 5) and eventual defoliation occurs. These spore masses may appear on other infected parts. Fruit can be infected at all stages of development (Arthur 1929). Individual infected drupelets are converted into yellow-orange uredinial pustules (Plates 6 and 7), rendering the fruit unsaleable (Ellis and Ellett 1981).

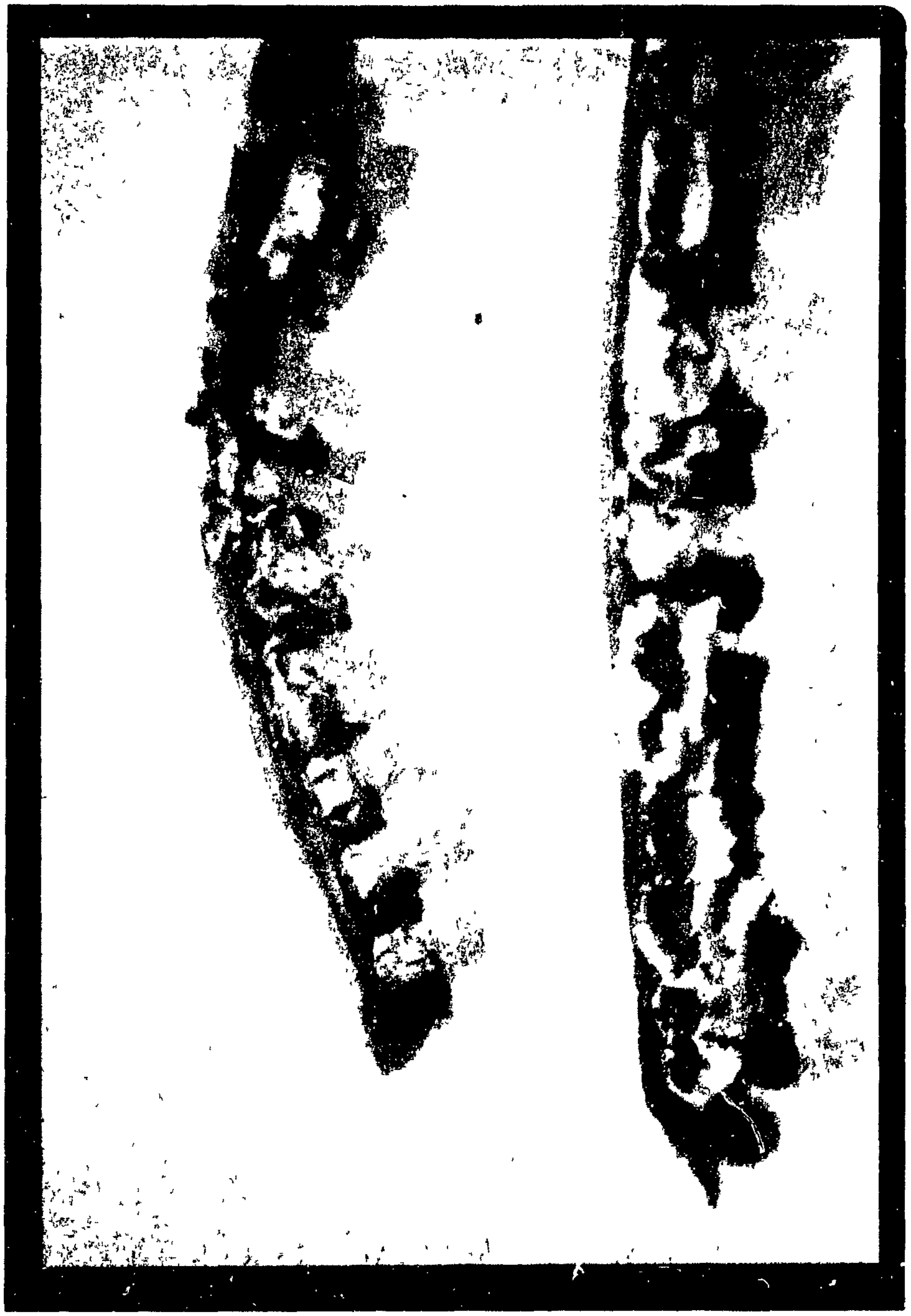

**Plate 1** White spruce [*Picea glauca* (Moench) Voss.] needles infected with *Pucciniastrum americanum* (Farl.) Arth. 40X Mag.

**Plate 2** **White spruce [*Picea glauca* (Moench) Voss.] cones infected with *Pucciniastrum americanum* (Farl.) Arth. Note orange-brown discolourations on cone scales.**

Plate 3 Fruit calyx of red raspberry (*Rubus idaeus* L.) cv. Festival infected with *Pucciniastrum americanum* (Farl.) Arth. 80X Mag.

**Plate 4** Adaxial leaf surface of red raspberry (*Rubus idaeus* L.) cv. Festival infected with *Pucciniastrum americanum* (Farl.) Arth. Uredinial pustules are filled with yellow-orange urediniospores.

Plate 5 Abaxial leaf surface of red raspberry (*Rubus idaeus* L.) cv. Festival infected with *Pucciniastrum americanum* (Farl.) Arth. Note curling of the leaf, general chlorosis and necrotic flecks.

Plate 6 Partially ripe red raspberry (*Rubus idaeus* L.) cv. Festival infected with *Pucciniastrum americanum* (Farl.) Arth. Note individual infected drupelets are converted into uredinial pustules. 40X Mag.

**Plate 7** **Fully ripe red raspberry (*Rubus idaeus* L.) fruit cv. Festival infected with *Pucciniastrum americanum* (Farl.) Arth. Note individual drupelets are converted into uredinial pustules. 80X Mag.**

# 3.0 LATE YELLOW RUST ON RED RASPBERRY

## 3.1 Disease Incidence

### 3.1.1 Current Prevalence in the Atlantic Provinces of Canada

Late yellow rust has occurred sporadically in red raspberry plantings in the Atlantic provinces of Canada for many years. However, in 1984, 1985, and 1986, it seriously affected a number of fields, rendering the fruit unmarketable.

### 3.1.2 History of Occurrence

Very little is known about the epidemiology of late yellow rust presumably because of its former rarity. Anderson (1956) felt that late yellow rust was so insignificant that no special control measures were warranted. Connors (1967) discussed dynamic changes in predominate pathogens caused by fluctuations in temperature and rainfall as well as shifts in cultivars and changes in agricultural practice. He cited the introduction of the raspberry cultivar Viking into commercial production and the subsequent economic importance of late yellow rust in raspberry plantings, particularly in the Maritime provinces. The alternate host, white spruce, was used extensively in windbreaks there. Not only did Viking become severely rusted, but other cultivars became moderately rusted. The Canadian federal government began publishing annual reports of the Canadian Plant Disease Survey in 1920. The Forest Insect and Disease Survey was first published in 1951. Late yellow rust on raspberry was reported in the following years: 1930-1933, 1936, 1938, 1941-1943, and 1958. On spruce it was reported in 1941, 1942, 1951, 1952, 1954, 1960, 1962, 1965-1969, and 1973 (Connors 1976). With the exception of 1941, these reports of late yellow rust incidence were of minor consequence. In 1941, aecia were seen on spruce in New Brunswick on 04 June and nearby raspberry plantings were heavily rusted by late July.

In Prince Edward Island, one large planting of Viking was destroyed while others were seriously affected.

### 3.2 Cultivar Susceptibility and Environmental Influence

The red raspberry cultivar Festival was introduced in 1972 from the Ottawa Research Station. Its productivity and hardiness made it an excellent choice for Maritime growers. Since its introduction, it has become one of the most widely planted cultivars in the Atlantic provinces of Canada. Festival appears to be quite susceptible to late yellow rust in this region (Nickerson and Mahar 1987). The infection process of an epidemic begins when the dispersal unit (spore) comes into contact with a suitable host plant under suitable environmental conditions (Zadoks and Schein 1979). Possibly the wet humid weather from April to June in 1984, combined with the abundance of a susceptible host (Festival) led to a serious outbreak of late yellow rust.

### 3.3 Control Measures

Berger (1971) discussed three epidemiological strategies to minimize losses due to disease. These included: eliminating or reducing the initial inoculum or delaying its appearance, shortening the exposure time of the crop to the pathogen, and slowing the rate of disease increase. A disease control program for late yellow rust would be based on the latter technique. This would include chemical control and the use of resistant varieties. In 1986 the Atlantic Provinces Raspberry Production Guide (Anonymous 1986) recommended six different fungicides for control of various raspberry diseases. These sprays were to be applied from the green tip stage through until harvest; they were, however, ineffective in controlling the rust. The cultivar Nova, which was introduced in 1981 by Kentville, N.S. has shown a high degree of resistance to late yellow rust. This indicates that prospects are good for long term rust control through the use of resistant cultivars.

## 4.0 OUTLINE OF PROPOSED RESEARCH

### 4.1 Introduction

This project on control of late yellow rust was undertaken because the disease poses a serious threat to both the raspberry industry and the white spruce industry in the Maritimes. The objectives of the research were to study variations in susceptibility to rust infection of raspberry cultivars and to develop a chemical control program based on an understanding of the life cycle of the pathogen in order to achieve an improved disease management system.

### 4.2 Importance of Late Yellow Rust on Raspberry

#### 4.2.1 Disease Incidence

From 1984-1986 incidence of the disease ranged from a trace to 100% of the fruit infected in fields examined in Nova Scotia (Nickerson and Mahar 1987). Rusted fruit is unsaleable and represents a substantial loss to the growers. In 1984 and 1985 the farm gate value of raspberries was $174,000 and $172,000 respectively for New Brunswick and Nova Scotia combined (Anonymous 1987).

#### 4.2.2 Effects of Defoliation

Doughty et al. (1972) have shown that premature cane defoliation can increase the susceptibility of raspberries to winter injury. Their work showed that freeze injury to raspberry buds was significantly greater after two-thirds of the leaves were removed and after complete defoliation when compared to the control. In severely rusted plantings defoliation of the canes occurs (Dodge 1923). Reduction of winter hardiness due to defoliation could impose severe limitations on raspberry growing in cold regions.

### 4.2.3 Cultivar Susceptibility & Life Expectancy of Plantings

In the Atlantic provinces of Canada, winter hardiness is of prime concern in raspberry culture. Cultivar testing at various locations has led to the recommendation of Festival as a sufficiently hardy and productive cultivar for the region. Therefore, it has been widely planted over a period of years. Raspberry establishment requires a substantial investment and growers are faced with three successive years of establishment costs prior to receiving any revenue; these are the fallow, planting, and the training years. Grant (1982) estimated the total expense for these years along with their respective interest charges accumulated to the fourth year (the first crop year) to be $13,650.13. This total compounded amount represents the cost of establishment per hectare which must be repaid over the harvesting years. Properly maintained blocks can be expected to produce well for several years. It is quite unrealistic to consider abandoning previously established plantings even though they consist mainly of Festival which has been shown to be highly susceptible to late yellow rust.

## 4.3 Importance of Late Yellow Rust to the Forestry Industry

### 4.3.1 Impact of Cone Rust and Levels of Infection

Cone rusts have a direct impact on all short and long term aspects of forestry because of their potential to interfere with seed production. In 1984 levels of infection were so high that tree tops viewed from a distance actually appeared yellow because of excessive spore production (Magasi 1985).

### 4.3.2 Effects of Cone Rust

Severe infections of late yellow rust on white spruce caused infected cones to produce only 35% of their complement of sound seed while up to 80% of the seed that appeared to be sound failed to germinate (Smith et al. 1986).

Lack of control in a given year has serious consequences in terms of losses in seed production. The extremely high value of Maritime seed orchards warrants special control measures for any factors which directly or indirectly affect seed production.

## 4.4 Control Measure Proposals

### 4.4.1 Source of Inoculum

It is virtually impossible to eliminate the alternate host from the standpoint of both the raspberry industry and the forestry industry. It is also impossible to eliminate the wild *Rubus* host growing abundantly throughout the Atlantic provinces of Canada. Therefore, a continuous source of inoculum is ensured for the recycling of the disease. Since host eradication is out of the question, other control measures must be considered.

### 4.4.2 Fungicide Applications

Fungicide evaluation tests carried out in Bouctouche, New Brunswick in 1985 by Luffman (1986) indicated that anilazine (4,6-dichloro-N-(1-chlorophenyl)-1,3,5-triazin-2-amine) was promising for the control of late yellow rust. Sulfur was also effective but appeared to have a phytotoxic effect. Further experimentation was required to determine the optimum schedule of spray applications using these two fungicides and to determine whether early season sprays at the time of aeciospore release from the spruce would be effective.

### 4.4.3 Anilazine and Sulfur

Anilazine is a foliar fungicide developed by the Ethyl Corporation and the Pittsburgh Coke and Chemical Company. It is manufactured under license by the Chemagro Corporation. MacDougall et al. (1964) described its biological and physical properties. The fungicidal effectiveness of anilazine is well established for several plant diseases. It is used as a protective spray and applied before disease occurs. Pure anilazine is a white crystalline solid and it is insoluble in water, but soluble in toluene at 30°C (5% w/v) and in acetone at 30°C (10% w/v). It is available as a 50% wettable powder and a 5% dust. The chemical structure is presented in Appendix 5.

Tweedy (1969) stated that elemental sulfur is the oldest of the pesticides and is still widely used for its fungicidal properties. Its use as a pesticide was documented as early as 1000 B.C. (Mellor 1930). Forsyth (1802, cited by Tweedy 1969) first suggested that it be used for disease control. McCallan (1967) referred to the period ending in 1882 as the sulfur era in his discussion of the history of fungicides.

Sulfur is a yellow solid which is insoluble in water and slightly soluble in ethanol and diethyl ether. It is a broad spectrum fungicide which controls several diseases. The reasons for its toxicity to fungal organisms are not understood. It is a stable chemical compatible with other pesticides (Tweedy 1969, Raw 1970 and Martin 1972). Sulfur phytotoxicity is temperature-dependent and has occurred on several plants. It is most prevalent during warm and humid periods Typical symptoms include scorching, burning, dwarfing of the foliage, yield reduction and sometimes premature defoliation (Tweedy 1969, Buchel 1983). Physical properties are presented in Appendix 5.

Sulfur may be formulated as a dust, wettable powder, or a vapour (Tweedy 1969).

### 4.4.4 Cultivar Differences

In view of the high degree of variability in susceptibility to late yellow rust exhibited by different raspberry cultivars, closer examination of this was warranted. Field screening of selected cultivars was done at the green fruit and ripe fruit stage to assess susceptibility to infection. An evaluation of cultivar susceptibility and the infection process was made in the greenhouse. A study of these aspects forms the basis of an improved disease management system.

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## CONTROL OF LATE YELLOW RUST
## [*Pucciniastrum americanum* (Farl.) Arth.]
## OF RED RASPBERRY

### INTRODUCTION

Late yellow rust caused by *Pucciniastrum americanum* (Farl.) Arth. has occurred sporadically in red raspberry (*Rubus idaeus* L.) plantings in the Atlantic provinces of Canada for many years. In 1984 and 1985 it seriously affected a number of fields, rendering the fruit unmarketable.

Symptoms of *P. americanum* can occur on the leaves, petioles, shoots, fruit calyces, and the fruit (Anderson 1956). Uredinial pustules on the adaxial leaf surface appear as powdery yellow-orange spore masses in midsummer. Curling of the leaflet and eventual defoliation occur (Arthur 1929). Individual infected drupelets are converted into yellow-orange uredinial pustules, rendering the fruit unsaleable (Ellis and Ellett 1981).

Doughty et al. (1972) showed that premature cane defoliation increases the susceptibility of raspberries to winter injury. This reduction of winter hardiness due to premature defoliation imposes severe limitations on raspberry growing, particularly in the Atlantic provinces of Canada where winter hardiness is a prime concern in raspberry culture. The red raspberry cultivar Festival has been recommended as a sufficiently hardy and productive cultivar for this region. Since its introduction from the Ottawa Research Station in 1972, it has become one of the region's most widely planted cultivars. It is also highly susceptible to late yellow rust infection of both leaf and fruit (Nickerson and Mahar 1987).

The alternate host of this rust is white spruce [*Picea glauca* (Moench) Voss.] (Arthur 1929). The aecia usually develop on the current year's needles and rarely on the cone scales (Ziller 1974). Dodge (1923) and Darker (1929) described the life cycle of *P. americanum*. Aeciospores released by the spruce in late spring cause primary infections on red raspberry plants. Uredinial pustules which develop on the adaxial leaf surface contain urediniospores which recycle on raspberry causing secondary infections on leaves and fruit as the growing season progresses.

Fungicide evaluation tests carried out by Luffman (1986) in Bouctouche, N.B. in 1985 indicated that anilazine (4,6-dichloro-N-(1-chlorophenyl)-1,3,5-triazin-2-amine) was most effective in controlling late yellow rust. Anilazine is a wettable powder fungicide which is used as a protective spray and applied before disease appears. It is registered for use on raspberries in Canada and has a minimum 14 days to harvest interval spray restriction. This paper discusses the results of 2 years (1986 and 1987) of field work at the Senator Hervé J. Michaud Experimental Farm at Bouctouche, New Brunswick using anilazine to determine the optimum schedule of spray applications. This forms the basis of an improved disease management system.

## MATERIALS AND METHODS

The trial was carried out in a block of red raspberries originally established in 1981 for cultivar evaluation. Only the cultivar Festival was used for the experiment. The design was completely randomized. Treatments were applied to a 3 metre length of row and for all treatments the rate of anilazine applied was 3.5 kg $ha^{-1}$. All treatments were applied with a single nozzle handgun sprayer. As per the manufacturer's recommendation a 7 day interval between fungicide applications was observed. The period of aeciospore release was determined by inspecting daily all white spruce trees in close proximity to the raspberry plots from the first week of June until the third week of July.

In 1986 four treatments were applied, replicated four times. These were:

(A) a control with no chemical treatment;

(B) four weekly sprays of anilazine from June 26 - July 17 ;

(C) eight weekly sprays of anilazine from June 26 - August 14 ;

(D) four weekly sprays of anilazine from June 26 - July 17, followed by four weekly sprays of sulphur at 4.0 kg $ha^{-1}$ from July 24 - August 14 .

The start of the fungicide applications (June 26) corresponded to aeciospore release.

In 1987 five treatments replicated three times were applied. These were:

(E) a control with no chemical treatment ;

(F) one application of anilazine at the beginning of the aeciospore release period on June 19;

(G) one application of anilazine on July 7, 14 days prior to the commencement of harvest;

(H) two applications of anilazine, the first on June 19, and the second on July 7 ;

(I) three applications of anilazine on June 19, June 29, and July 7.

Originally treatment I was to be weekly sprays from the beginning of aeciospore release until 14 days before harvest, but an unusually early harvest season and inclement weather prevented this.

The data recorded in both trials for each harvest included the weights of 25 berries chosen at random, the weights of rusted fruit, and a total fruit weight which comprised all fruits harvested. The data were analyzed by analysis of variance (Appendix 1) and Duncan's multiple range test was used to separate the means. A count of the rusted leaves and an assessment of the severity of foliar rust on primocanes after the removal of the fruiting canes was also made. Fifty leaves per replicate for each treatment were sampled at random from the mid portion of the canes, approximately 0.75-1.5 m above ground. This method avoided selecting very young developing leaves at the tips of canes and older leaves on the lower portion of the canes which would be more likely to be infected, since rust first appears on lower older leaves. The number of leaves infected were counted for each treatment and these data were transformed to square roots prior to analysis of variance. Disease symptoms were scored on each leaf using a 0-3 severity scale. A value of 0 was assigned when there was no rust present, 1 for rust present on lower epidermis, 2 for rust abundant on lower surface with scattered chlorotic flecks on upper epidermis, and 3 for rust very abundant on lower surface with chlorosis general on upper epidermis plus necrotic flecks. The number of leaves in each category was multiplied by the assigned rank and added. This total was divided by the number of leaves infected to obtain a severity index. Within each year the individual observations were ranked and the means of the ranks were determined for four treatments (1986) and five treatments (1987). For this variable with its ordinal scale of measurement a nonparametric method of statistical analysis (Daniel 1978) was used because of its general applicability. The Kruskal-Wallis one-way analysis of variance (Appendix 2) followed by a multiple comparison test was used to determine the equality of the treatments. The method described by Dunn (1964) for calculating the critical values to compare rank means of the treatments was employed and an experimentwise error rate of 15% was used.

## RESULTS

In 1986 aeciospore release from white spruce trees in the vicinity of the trial occurred from June 23 - July 10. Rust first appeared on the calyces and the lower leaves of fruiting laterals of raspberries on July 20. The harvest period was from August 4 - 21. Among the treatments there were no significant differences ($p < 0.05$)

for the variable total fruit harvested (Table 1). The mean weight of 25 berries was significantly ($p < 0.05$) lower for treatments C and D when compared to the control (Table 1). The control (treatment A) produced significantly more ($p < 0.05$) rusted fruit than the other treatments and rusted fruits were harvested from August 7 until the end of harvest. No rusted fruits were harvested from either treatment C or D (Table 1) and rusted fruit were harvested on only one date (August 18) in treatment B. In assessing foliar rust each treatment was significantly ($p < 0.05$) different for the variable "Number of infected leaves". Treatment A (control) had the most infected leaves followed by treatments D, B, and C respectively (Table 2). In the severity index the individual observations were ranked and the means of the ranks of the four treatments are given in Table 2. The rank means of the control and B were significantly (15% experimentwise error rate) higher than C and D.

In 1987 aeciospore release from nearby white spruce occurred from June 18 - July 6. Rust first appeared on the calyces and lower leaves of the fruiting laterals on July 18. Rusted fruit was harvested after August 7 in treatments E and G and after August 11 in treatments F and H. The harvest period was from July 21 to August 20. Treatments did not affect either the weight of 25 berries or total yield (Table 1). All fungicide treatments controlled rust on the fruit. Treatment I was the best for rust control but H was not significantly distinguishable from it in terms of rusted fruit. Rusted fruit was harvested from treatment F on August 11 - 17, from G on August 7 - 14, and on August 11 - 14 for treatment H. In treatment E (control) rusted fruit was harvested from August 7 until the end of harvest. In assessing foliar rust treatment I had significantly ($p < 0.05$) fewer total leaves infected than all other treatments (Table 2). In the severity index the individual observations were ranked and the means of the ranks are given in Table 2. The rank means of treatments E and G were significantly (15% experimentwise error rate) higher than treatment I.

Table 1. Effect of fungicide applications on yield, rusted fruit and berry weight

| Treat-ment | Dates of fungicide applications* | Yield per plot (g) | Rusted fruit (g) | Wt. of 25 berries (g) |
|---|---|---|---|---|
| 1986 | | | | |
| A | Control | 1206.40ns | 37.04a | 93.17a |
| B | June 26 July 3, 10, 17 | 1121.40ns | 4.88b | 88.59ab |
| C | June 26 July 3, 10, 17 July 24, 31 Aug. 7, 14 | 849.30ns | 0.00b | 85.67b |
| D | June 26 July 3, 10, 17 July 24, 31 Aug. 7, 17 | 1076.50ns | 0.00b | 87.67b |
| | SEM | 104.56 | 2.42 | 1.52 |
| | Significance | n.s. | 0.05 | 0.05 |
| 1987 | | | | |
| E | Control | 624.60ns | 43.10a | 66.78ns |
| F | June 19 | 564.10ns | 2.60b | 65.33ns |
| G | July 7 | 750.80ns | 3.07b | 70.67ns |
| H | June 19, July 7 | 525.47ns | 1.90bc | 67.00ns |
| I | June 19, 29, July 7 | 531.60ns | 0.00c | 63.11ns |
| | SEM | 111.07 | 1.27 | 3.31 |
| | Significance | n.s. | 0.05 | n.s. |

* All fungicide treatments were anilazine at 3.5 kg $ha^{-1}$ except for D in which the last four sprays were sulphur at 4.0 kg $ha^{-1}$.

a-c Within each column means followed by the same letter were not significantly different at 5% according to Duncan's multiple range test.

Table 2. **Effect of fungicide applications on foliar rust**

| Treatment | Number of infected leaves | Rank mean of severity index | Treatment | Number of infected leaves | Rank mean of severity index |
|---|---|---|---|---|---|
| 1986 | | | 1987 | | |
| A | 6.69a | 14.50a | E | 7.03a | 13.00a |
| D | 3.85b | 6.75b | G | 6.99a | 12.00a |
| B | 2.33c | 10.25a | F | 6.86a | 5.67ab |
| C | 0.71d | 2.50b | H | 6.61a | 7.33ab |
| | | | I | 3.27b | 2.00b |

Means subjected to square root transformation; transformed means presented.

Within each column means followed by the same letter were not significantly different at 5% according to Duncan's multiple range test.

Within each column means followed by the same letter were not significantly different at the 15% experimentwise error rate.

Critical values for comparing rank means among treatments for 1986 and 1987 were 7.53 and 8.87 respectively.

## DISCUSSION

In 1986 the three fungicide regimes used all effectively controlled fruit and foliar rust. All treatments included applications during the initial infection period (June 23 - July 10). Treatments C and D were continued for 5 weeks beyond the initial infection period, suppressing secondary infection inoculum so that no fruit became infected. The negative effect of later fungicide applications on berry weight is not surprising since fungicides may be phytotoxic and have been shown to reduce pollen viability and thereby fruit size and yield of other crops (Bristow and Shawa 1981, Eaton and Chen 1969a, Eaton and Chen 1969b, Khanizadeh and Buszard 1987). Increasing the anilazine applications from four to eight reduced the number and severity of leaves infected whilst adding sulphur to the program appeared to increase the number of leaves infected compared with anilazine alone.

In 1987, again all fungicide treatments controlled fruit rust but only treatment I reduced the number of leaves infected and the severity of infection. A single application of anilazine after the initial infection period (treatment G) was the least effective fungicide treatment in controlling foliar rust. The 1987 growing season was unusually dry (Anonymous 1987) and it is likely that natural rust infection was low due to the lack of humidity during the spore release period; in a more typical year, the incidence of rust would probably be higher. In 1987 when all of the treatments ended at least 14 days prior to harvest there was no effect on berry weight. This emphasizes the importance of avoiding fungicide applications during the main flowering period. Thus early season applications appear to be important to control primary infection. Anilazine, which is already registered for use on raspberries could be included in an early season spray program to control late yellow rust.

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## CONNECTING TEXT

A disease control program for late yellow rust is based on chemical control and the use of resistant cultivars. The preliminary research dealt first with chemical control based on an understanding of the life cycle of the pathogen. The next step was to study the resistance of cultivars grown in the Atlantic provinces of Canada.

Considerable differences among raspberry cultivars in foliar resistance to late yellow rust have been observed in the field (Nickerson and Mahar 1987). Festival, one of the most widely grown cultivars, appeared to be very susceptible while Nova, a cultivar which is becoming very popular is apparently more resistant. A series of inoculation experiments were undertaken *in vitro* to study the infection process of foliar rust on a range of cultivars.

## REFERENCE

Nickerson, N.L. and Mahar, J. 1987. Late yellow rust of raspberries in Nova Scotia. Kentville Research Station Annual Report 1986. 17.

## SUSCEPTIBILITY OF PRIMOCANES OF SIX RED RASPBERRY CULTIVARS TO LATE YELLOW RUST
*Pucciniastrum americanum* (Farl.) Arth.

## INTRODUCTION

Late yellow rust [*Pucciniastrum americanum* (Farl.) Arth.] on red raspberry (*Rubus idaeus* L.) is a sporadic problem in the Atlantic provinces of Canada (Luffman and Buszard 1988). In Nova Scotia, Nickerson and Mahar (1987) noted that the cultivars Carnival and Festival appeared susceptible to the rust while Nova and Boyne showed some resistance; similar observations were made in 1984 and 1985 in New Brunswick (Luffman, unpublished data). The mechanics of this resistance have not been previously studied. This paper describes the response of primocanes of six raspberry cultivars to infection by *Pucciniastrum americanum* in controlled inoculation experiments in a greenhouse.

## MATERIALS AND METHODS

The source of *P. americanum* used was infected leaves of the cultivar Festival collected from the field in Bouctouche, New Brunswick. Urediniospores were brushed from the adaxial leaf surface of the infected leaves onto the adaxial surface of leaves of three-month old virus-free greenhouse grown plants of the same cultivar. The plants were then placed in a darkened mist chamber at 20°C for 48 hours. When these leaves became infected (7 days), the resulting urediniospores were used to infect more leaves in order to build up the source of inoculum. This step was repeated to maintain a supply of inoculum.

Urediniospores were vacuum harvested into a vial; distilled water was added to make up a spore suspension. The concentration of the suspension was determined using a haemocytometer (Tuite 1969) and distilled water was added to produce a suspension of 30,000 spores $ml^{-1}$. Previous research showed this concentration resulted in successful infection of susceptible cultivars (Nickerson, personal communication).

Urediniospore viability *in vitro* was determined using a modified version of the method used by Anthony et al. (1985). Spore suspension was sprayed on four petri dishes for each test. The dishes contained 1.5% distilled water agar (DWA) and were placed in the dark at 20°C. After eight hours percent germination was determined by counting the germ tubes. When the length of the germ tube equalled the diameter of the spore, the spores were considered to have germinated (Zadoks and Schein 1979). Nickerson (personal communication) observed spore germination in as little as 6 hours. On each plate four fields were chosen at random. The total number of spores in each field were counted and the percent germination was then calculated for each field The average of the four fields on the four petri plates determined the overall germination

The urediniospore suspension was used in experiments to determine the relative susceptibility of the cultivars Festival, Carnival, Boyne, Nova, Heritage, and Royalty by spraying onto the adaxial leaf surface of the two youngest fully expanded leaves of four-month old virus-free greenhouse grown plants. Each trifoliolate leaf received 0.5 ml of spore suspension. The plants were then placed in a darkened mist chamber at 20°C for 48 hours. Following this they were kept in a greenhouse at 20°C under a 16-hour photoperiod with supplemental lighting (0.8-1.0 klx, high pressure sodium lamps). For each test there were four replicate plants of each cultivar, and the test was repeated four times.

Leaf tissue changes such as chlorosis and necrosis resulting from infection were recorded 28 days after inoculation. Development of the uredinia was also measured; this included the latent period (the time from inoculation to the beginning of sporulation), and the leaf area covered by lesions both at the beginning of the infectious period (at the first appearance of spores) and 28 days after inoculation. The actual leaf area affected, ie. covered by uredinia, was drawn on a transparent acetate sheet and the area was determined using a planimeter. After 28 days the leaves were detached and photocopied. The planimeter was used to determine the total leaf area. Percent leaf area affected by the rust was calculated when symptoms first appeared (ie. at the end of the latent period, the onset of sporulation) and after 28 days. A test of homogeneity of variance showed no differences among the four tests so the results were pooled before analysis of variance (Appendix 3) was performed on the data and Duncan's multiple range test was used to separate the means.

## RESULTS

In the *in vitro* spore viability tests the germination of the urediniospores was in the range of 51-60% in all four tests. The average germination was 57, 56, 55, and 57% for tests 1, 2, 3, and 4 respectively. The germ tubes tended to grow straight up from the DWA plate, so it was sometimes difficult to see them

All six cultivars produced sporulating uredinia as a result of inoculation with the spore suspension. Nova had a significantly longer latent period than all other cultivars; Boyne had the second longest latent period and was followed by Royalty. Festival, Carnival, and Heritage had the shortest latent periods (Table 1).

On the cultivar Nova there were significantly fewer and smaller uredinia produced at the onset of sporulation; only a few of these uredinia sporulated; most simply produced a hypersensitive reaction, an area of dead tissue which spread no further. Boyne developed more and larger uredinia than Nova, but significantly less than the other four cultivars. Carnival had the greatest area covered by uredinia at the onset of sporulation. Festival followed Carnival with significantly more and larger uredinia developed at this stage compared with the other cultivars.

At the end of the 28-day observation period Nova and Boyne still had the smallest and fewest uredinia. Since the onset of sporulation the area affected on Nova had increased from 0.12% to 0 20%. Boyne also showed a small increase (from 0.41% to 1.08%) in diseased area. Carnival had the greatest area (71.96%) affected while Festival was second with 60.53% of the total leaf area covered by sporulating uredinia. These uredinia were filled with masses of yellow-orange urediniospores. The hypersensitive reaction on Nova persisted throughout the experiment and was not

Table 1

**Response of six red raspberry cultivars to inoculation with urediniospores of *Pucciniastrum americanum* (Farl.) Arth. under controlled environment conditions**

| Cultivar | Latent period (days) | Percentage of leaf area diseased at onset of sporulation* | Percentage of leaf area diseased 28 days after inoculation* |
|---|---|---|---|
| Nova | 15.50a | 0.12a | 0.20a |
| Boyne | 12.19b | 0.41b | 1.08a |
| Royalty | 8.81c | 0.90c | 41.51b |
| Heritage | 7.63d | 0.99d | 49.59c |
| Festival | 7.19d | 1.36e | 60.53d |
| Carnival | 7.19d | 1.39f | 71.96e |

* Arcsin transformation used for analysis; untransformed means in table.

a-f Within each column means followed by the same letter are not significantly different at the 5% level according to Duncan's multiple range test.

observed on any other cultivar.

No symptoms were present on the abaxial leaf surface of any of the six cultivars at the onset of sporulation. By the end of the 28-day period considerable changes had occurred on all cultivars except Nova and Boyne. These two had some very small chlorotic lesions and there was some necrotic flecking on Boyne. Almost all of the abaxial leaf surfaces of both Carnival and Festival were generally chlorotic and there were spreading necrotic lesions. Curling of the leaves was also evident. Heritage and Royalty were similarly affected, but to a lesser extent.

## DISCUSSION

The observed differences among the six cultivars in response to artificial inoculation with urediniospores of *Pucciniastrum americanum* clearly established a ranking order from the least to most resistant. In this collection Carnival was least resistant, followed by Festival, Heritage, Royalty, Boyne, and Nova respectively. The ranking of Carnival and Festival as least resistant and Boyne and Nova as most resistant confirms observations of field infection previously mentioned.

Nelson (1978) classified disease resistance into two major types. In the first, the host resists the establishment of a successful parasitic relationship by restricting the infection site and the infection process. This resistance to infection is often referred to by the term hypersensitivity. In the second type, following successful infection, the host resists subsequent colonization and reproduction of the parasite. This is characterized by the terms partial resistance and slow rusting.

By applying the principles of Nelson's definitions, it is possible to assess the resistance shown by the six cultivars. Nova was the only cultivar used which showed a hypersensitivity reaction, a type of resistance. The range of latent periods exhibited by Carnival, Festival, Heritage, and Royalty is similar to that found by Darker (1929) in his inoculation work with *Pucciniastrum americanum* on red raspberry. Darker used aeciospores from the alternate host, *Picea glauca* (Moench) Voss, and the latent periods ranged from 7-11 days. The longer latent period and fewer and smaller uredinia developed on Nova and Boyne indicate that both are highly resistant to this rust. Following successful infection, Boyne and Nova appeared to have resisted subsequent development of the pathogen thereby slowing the rate of disease increase. The longer latent period and fewer and smaller uredinia are consistent with the type of resistance shown by slow rusting cereals (Wilcoxson 1981). Since these tests were done by mass inoculation using field inoculum, information was not obtained on different races of *P. americanum* and their interaction with the cultivars.

Parlevliet (1979) assumed that disease symptoms usually quantitatively reflected growth of the pathogen in the host. Therefore disease severity (the area of plant tissue affected by disease expressed as a percentage of the total area assessed) can be used to assess disease resistance. Disease severity is usually the cumulative result of the components infection frequency, latent period, spore production, and infectious period. Royalty and Heritage appear to have partial resistance to this rust, with Royalty having more than Heritage. Development of the pathogen was quantitatively hindered on these resistant cultivars. Both spore production and the subsequent percentage of leaf area affected both at the onset of sporulation and 28 days after inoculation were reduced.

These results give some insight into cultivar interactions of red raspberries with *Pucciniastrum americanum*. However, more information is needed on different races of the pathogen in order to study the differential interactions between host and pathogen genotypes and to establish the types of resistance exhibited by the raspberry cultivars.

Based on the results of this study the use of cultivars with some resistance to late yellow rust is recommended in areas where this disease is a problem.

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## CONNECTING TEXT

After studying foliar primocane infections, the next step was to examine the infection process of fruit rust on cultivars grown in the Atlantic provinces of Canada. Field observations indicated that there were considerable differences in fruit rust among cultivars (Nickerson and Mahar 1987). Since it was not feasible to study fruit infections on floricanes in a controlled environment, artificial inoculations were done *in vivo*. Information was thus obtained on resistance of fruit to infection by late yellow rust by a range of cultivars.

## REFERENCE

Nickerson, N.L. and Mahar, J. 1987. Late yellow rust of raspberries in Nova Scotia. Kentville Research Station Annual Report 1986. 17.

## A NOTE ON: THE SUSCEPTIBILITY OF SIX RED RASPBERRY CULTIVARS AND TAYBERRY TO FRUIT INFECTION BY LATE YELLOW RUST [*Pucciniastrum americanum* (Farl.) Arth.]

### INTRODUCTION

The occurrence of late yellow rust [*Pucciniastrum americanum* (Farl.) Arth.] on red raspberries (*Rubus idaeus* L.) in the Atlantic provinces of Canada has been sporadic for many years. In 1984 and 1985 a serious outbreak occurred (Luffman and Buszard 1988). Nickerson and Mahar (1987) in Nova Scotia observed that the cultivar Carnival had less rust on the fruit than Festival, but the foliage was equally or more rusted. Similar observations were made in New Brunswick (Luffman, unpublished data).

In order to examine more closely any differences between these two cultivars, grown extensively in the Maritime provinces, and others, fruit infections were studied *in vivo*.

### MATERIALS AND METHODS

Fruit clusters of six field-grown red raspberry (*Rubus idaeus* L.) cultivars and Tayberry (a blackberry-raspberry hybrid) were artificially inoculated in the field with *Pucciniastrum americanum* (Farl.) Arth. The raspberry cultivars were Carnival, Festival, Fall Red, Royalty, Boyne, and Nova. Four fruit clusters of each cultivar were chosen at random for each inoculation from a plot originally established for cultivar evaluation. Two inoculations were made, the first with aeciospores from infected needles of the alternate host, white spruce [*Picea glauca* (Moench) Voss.], and the second using urediniospores from infected red raspberry leaves.

White spruce needles infected with *P. americanum* were collected on June 30, 1987. The yellow-orange aeciospores were harvested onto a glass slide and then brushed onto the fruit clusters of the six red raspberry cultivars and Tayberry. These fruits were harvested on July 24, 1987.

Red raspberry leaves of the cultivar Festival infected with *P. americanum* were collected on July 21, 1987. Urediniospores were brushed first from the adaxial leaf surface onto a glass slide and then onto the fruit clusters. These fruits were harvested on August 4, 1987.

An overhead irrigation system was used to wet the fruit thoroughly for 1 hour before inoculation. After inoculation the fruit clusters were sprayed with a fine mist of distilled water and covered with a polyethylene bag to maintain high humidity. Inoculations were made at dusk so that the fruit would not overheat in the bags, which were removed the following morning.

Data recorded included the total number of fruits per cluster, the number of fruits which were infected at harvest (expressed as a percentage) and the latent period (the time from inoculation until the appearance of sporulating uredinia). The statistical design was a randomized complete block. The variable "percentage of fruit infected" was transformed to the arcsin prior to analysis. Each of the four fruit clusters inoculated was treated as a replicate and analysis of variance (Appendix 4) was performed on all data. Duncan's multiple range test was used to separate the means. Cultivars which did not become infected were not included in the analyses.

## RESULTS AND DISCUSSION

Two raspberry cultivars, Nova and Royalty, and Tayberry were apparently resistant to fruit infection by this rust. The other cultivars exhibited different susceptibilities to the rust (Table 1).

The range of latent periods exhibited by the fruit of the infected cultivars is quite similar to that found in previous research on foliar rusting (Darker 1929, Luffman and Buszard, unpublished data). Boyne, which had the longest latent period, also had the lowest percentage of fruit infections in both the aeciospore and urediniospore inoculations (Table 1).

**Table 1** **Response of fruit clusters of red raspberry cultivars to inoculation with aeciospores and urediniospores of *Puccinastrum americanum* (Farl.) Arth. in the field**

| Cultivar | Aeciospore inoculation | | Urediniospore inoculation | |
|---|---|---|---|---|
| | Latent period (days) | Percent fruit infected | Latent period (days) | Percent fruit infected |
| Boyne | 11.25a | 18.50a | 11.50a | 15.38a |
| Fall Red | 7.75b | 27.98b | 7.50b | 25.89b |
| Carnival | 7.25b | 29.79b | 7.25b | 30.83b |
| Festival | 7.25b | 40.63c | 7.50b | 40.21c |
| Nova | --- | --- | --- | --- |
| Royalty | --- | --- | --- | --- |
| Tayberry | --- | --- | --- | --- |

* Arcsin transformation used for analysis; untransformed means presented in table.

a-c Within columns means followed by the same letter are not significantly different at the 5% level according to Duncan's multiple range test.

Examination of individual infected fruits showed no apparent differences among cultivars; if fruits became infected they appeared to be equally rusted. Susceptible cultivars comprised three distinct groups in terms of rusting. Festival had the highest percentage; Carnival and Fall Red ranked second; Boyne had the least amount of rusted fruit. This ranking and the percentages of rusted fruit were remarkably similar for both inoculations. Losses of up to 30% of the fruit of the cultivar Heritage have been reported (Ellis and Ellett 1981). In this study the cultivars examined ranged from completely resistant to highly susceptible with losses of up to 40% of the fruit. In areas where late yellow rust is a problem the use of resistant cultivars of summer and fall-fruiting raspberries is recommended.

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Nickerson, N.L. and Mahar, J. 1987. Late yellow rust of raspberries in Nova Scotia. Kentville Research Station Annual Report 1986. 17.

## CONNECTING TEXT

Research (Luffman 1986, Luffman and Buszard 1988) indicated that anilazine applied during the period of aeciospore release from the alternate host, white spruce [*Picea glauca* (Moench) Voss.], effectively controlled late yellow rust of red raspberry. To check the safety of anilazine applications on raspberry up to 14 days before harvest as recommended by the manufacturer, residue analyses were done.

Pesticides are known to persist on plant surfaces. Evans (1968) noted that deposit building implies that greater quantities of active ingredient are retained on the plant surface as a result of consecutive applications of the same concentration of fungicides. Luffman and Buszard (1988) found that three applications of anilazine were effective in controlling late yellow rust. Fungicides are subject to the weathering effects of wind, rain, and temperature variations (Hartley and Graham-Bryce 1980).

Anilazine applications were made *in vivo* such that residues reflected the normal practice of a raspberry grower.

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## EFFECT OF PREHARVEST INTERVAL AND RATE OF ANILAZINE APPLICATIONS ON RESIDUES FOUND ON RED RASPBERRY (*Rubus idaeus* L.) FRUITS

### INTRODUCTION

Anilazine [4,6-dichloro-N-(1-chlorophenyl)-1,3,5-triazin-2-amine] is a wettable powder fungicide which is used as a protective spray and applied before disease occurs (MacDougall et al. 1964). It is registered for use on raspberries in Canada and has a minimum 14 days to harvest interval spray restriction. The maximum residue limit permitted is 5 ppm in Canada and 10 ppm in the United States. This chemical has become of interest recently because of an outbreak of late yellow rust [*Pucciniastrum americanum* (Farl.) Arth.] on red raspberries (*Rubus idaeus* L.) in the Atlantic provinces of Canada. Previously the occurrence of this disease was sporadic and there was no need for special control measures (Luffman and Buszard 1988). However, the lack of control during the recent outbreak of late yellow rust led to research (Luffman 1986, Luffman and Buszard 1988) which has shown anilazine to be effective in controlling this disease.

### MATERIALS AND METHODS

The experiment was carried out in 1986 and 1987, in a raspberry field of the cultivar Festival. The guidelines established by the Food and Agriculture Organization of the United Nations for pesticide residue trials were followed (Anonymous 1981). The statistical design was a randomized complete block and plots were 3 m of linear row. There were three replications of each treatment and an unsprayed control in each year.

Fruit samples of at least 1 kg were weighed, bagged, labelled, and frozen immediately after harvest. They remained frozen until the residue analyses were performed using the methodology described by Zweig (1972) at the Atlantic Provinces Pesticide Residue Laboratory, Kentville, Nova Scotia.

In 1986 there were three weekly anilazine applications with four preharvest intervals: 1, 3, 7, and 14 days. The rate of application was 3.5 kg $ha^{-1}$ (1.75 kg $ha^{-1}$ a.i.). Fruit samples were harvested on August 21, 1986.

In 1987 there were four weekly anilazine applications with only two preharvest intervals: 7 and 14 days. Two rates of anilazine were applied: 3.5 kg $ha^{-1}$ (1.75 kg $ha^{-1}$ a.i.) and 2.5 kg $ha^{-1}$ (1.25 kg $ha^{-1}$ a.i.). These two rates reflect the upper and lower limits recommended by the manufacturer. Fruit samples were harvested on August 12, 1987.

The anilazine residue data were subjected to analysis of variance and Duncan's multiple range test was used to separate the means.

## RESULTS

In 1986 and 1987 all samples of fruits from the plots which received anilazine were found to contain unacceptably high levels of pesticide residue (Table 1). In 1986 residues were higher after the 7 day than the 3 day interval. Very low levels of residue were detected in the fruit from the unsprayed control treatments in 1986, probably as a result of spray drift. In 1987 use of the lower rate of anilazine resulted in significantly lower residues on fruit harvested after 7 or 14 days, but residues were always greater than the permissable limits.

Table 1 Levels of anilazine residues on red raspberry (*Rubus idaeus* L.) fruits sprayed at different preharvest intervals

| Preharvest interval (days) | No. of appli-cations | Date of application | Application rate (kg $ha^{-1}$ a.i.) | Anilazine residues (ppm)* |
|---|---|---|---|---|
| 1986 | | | | |
| 1 | 3 | Aug. 7,14,20 | 1.75 | 86.80a |
| 3 | 3 | Aug. 5,11,18 | 1.75 | 27.67c |
| 7 | 3 | July 31, Aug. 7,14 | 1.75 | 36.43b |
| 14 | 3 | July 24,31, Aug. 7 | 1.75 | 9.00d |
| Control | - | ---- | - | 0.30e |
| 1987 | | | | |
| 7 | 4 | July 15,22,29 Aug.5 | 1.75 | 30.00a |
| 7 | 4 | July 15,22,29 Aug.5 | 1.25 | 20.63b |
| 14 | 4 | July 8,15,22,29 | 1.75 | 17.03c |
| 14 | 4 | July 8,15,22,29 | 1.25 | 13.53d |
| Control | - | ---- | - | n.d. |

n.d. = not detected

* Analyses for 1986 and 1987 were done separately. Within each column means followed by the same letter are not significantly different at the 5% level according to Duncan's multiple range test.

## DISCUSSION

In New Brunswick the present 14 day preharvest interval required between the application of the chemical and harvest is too short.

Burchfield (1967) stated that the persistance of a pesticide is affected by chemical and photochemical deterioration and erosion by wind and rainfall which cause deposits to disappear. The tenacity of a fungicide is reduced by wind and rain since it can be mechanically dislodged from plant surfaces. Hartley and Graham-Bryce (1980) stated that evaporation and the action of rain can cause the active ingredient to be lost from chemical deposits. Weather conditions in 1987 were unusually dry (Anonymous 1987). Rain fell on only three occasions during the course of the experiment. This may account for the high residue level on the fruits In 1986 the lower residue levels on the 3 day preharvest interval compared to the 7 day treatment may be explained by a heavy rainfall immediately after the 3 day treatment It is likely that some of the chemical washed off since it hadn't had a chance to penetrate to the fruit.

While Luffman and Buszard (1988) have shown that a three anilazine application treatment program applied during the initial infection period of late yellow rust, the time of aeciospore release from the alternate host, white spruce, [*Picea glauca* (Moench) Voss.] can control late yellow rust, the current interval between the last spray application and harvest is unacceptable. A safe interval needs to be established.

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# CONCLUSION

At the outset of this project it seemed apparent that improved disease control of late yellow rust would be achieved through the use of appropriate chemicals based on an understanding of the life cycle of the pathogen and the use of resistant raspberry cultivars. The research has confirmed this.

It has been determined that spray applications of anilazine timed to coincide with aeciospore release from the alternate host, white spruce [*Picea glauca* (Moench) Voss.], effectively reduce both foliar and fruit rust.

Epidemiological studies of foliar and fruit infections of different cultivars indicated that prospects are good for longterm rust control through the use of resistant varieties. In a raspberry breeding program the *in vitro* screening technique would be useful.

Residue analysis tests indicated that in New Brunswick the present 14 day interval required between the application of anilazine and harvest is too short. Further work is required to establish a safe interval.

## SUGGESTIONS FOR FUTURE RESEARCH

Very little is known about the epidemiology of late yellow rust, presumably because of its former rarity. A research program of temperature/wetness studies to determine the optimum conditions for the development of late yellow rust would be useful. In addition to this, infection studies with different races of *Pucciniastrum americanum* would provide information on differential interactions between host and pathogen genotypes.

Infection studies have thus far confirmed the susceptibility of *Rubus idaeus* and *Rubus neglectus* to *P. americanum.* Testing of different *Rubus* species would identify others which are susceptible and those which are immune.

It has become apparent that the levels of anilazine residues on the fruits are above the maximum residue limit allowed at the currently recommended 14 day preharvest interval Further testing should be done at preharvest intervals greater than 14 days to recommend a safe interval. Testing should also be done to see if lower rates of anilazine effectively control late yellow rust.

APPENDIX 1

ANALYSIS OF VARIANCE FOR NO. OF LEAVES INFECTED (TRANSFORMED DATA), RUSTED FRUIT, TOTAL FRUIT, AND BERRY WEIGHT IN FUNGICIDE TRIAL: MEAN SQUARES AND SIGNIFICANCE

FUNCICIDE TRIAL 1986

| SOURCE | DF | NO. OF LEAVES INFECTED MS | RUSTED FRUIT MS | TOTAL FRUIT MS | WT. OF 25 BERRIES MS |
|---|---|---|---|---|---|
| Treatment | 3 | 25.8879*** | 1275.4653*** | 93094.2425ns | 40.2847* |
| Error | 12 | 0.0841 | 23.4618 | 43731.3501 | 9 2569 |

FUNGICIDE TRIAL 1987

| SOURCE | DF | NO. OF LEAVES INFECTED MS | RUSTED FRUIT MS | TOTAL FRUIT MS | WT. OF 25 BERRIES MS |
|---|---|---|---|---|---|
| Treatment | 4 | 7.9026*** | 1022.9717*** | 26149.5227ns | 22.8778ns |
| Error | 10 | 0.1606 | 4.8387 | 37012.0367 | 32.9481 |

*, ***, ns - significant at 5% and 0.1% and not significant respectively

APPENDIX 2

## KRUSKAL-WALLIS ONE-WAY ANALYSIS OF VARIANCE FOR SEVERITY INDEX RANKING CLASSIFIED BY THE VARIABLE TREATMENT

WILCOXON SCORES (RANK SUMS)

| LEVEL | N | SUM OF SCORES | EXPECTED UNDER HO | STD DEV UNDER HO | MEAN SCORE |
|---|---|---|---|---|---|
| 1986 | | | | | |
| A | 4 | 58.00 | 34.00 | 8.19 | 14.50 |
| B | 4 | 41.00 | 34.00 | 8.19 | 10.25 |
| C | 4 | 10.00 | 34.00 | 8.19 | 2.50 |
| D | 4 | 27.00 | 34.00 | 8.19 | 6.75 |

KRUSKAL-WALLIS TEST (CHI-SQUARE APPROXIMATION)
CHISQ= 13.99 DF= 3 PROB>CHISQ= 0.0029

WILCOXON SCORES (RANK SUMS)

| LEVEL | N | SUM OF SCORES | EXPECTED UNDER HO | STD DEV UNDER HO | MEAN SCORE |
|---|---|---|---|---|---|
| 1987 | | | | | |
| E | 3 | 39.00 | 24.00 | 6.93 | 13.00 |
| F | 3 | 17.00 | 24.00 | 6.93 | 5.67 |
| G | 3 | 36.00 | 24.00 | 6.93 | 12.00 |
| H | 3 | 22.00 | 24.00 | 6.93 | 7.33 |
| I | 3 | 6.00 | 24.00 | 6.93 | 2.00 |

KRUSKAL-WALLIS TEST (CHI-SQUARE APPROXIMATION)
CHISQ= 12.43 DF= 4 PROB>CHISQ= 0.0144

APPENDIX 3

## ANALYSIS OF VARIANCE FOR PERCENT AREA DISEASED AT ONSET OF SPORULATION AND 28 DAYS AFTER INOCULATION (TRANSFORMED DATA) AND LATENT PERIOD IN FOLIAR INOCULATIONS:

MEANS SQUARES AND SIGNIFICANCE

| SOURCE | DF | LATENT PERIOD MS | PERCENT AREA DISEASED AT ONSET OF SPORULATION MS | PERCENT AREA DISEASED 28 DAYS AFTER INOCULATION MS |
|---|---|---|---|---|
| TIME | 3 | 0.0000001ns | 0.55555ns | 0 000581ns |
| CV | 5 | 0.0008332*** | 368.20000*** | 3 524900*** |
| TIME*CV | 15 | 0.0000001ns | 0.48888ns | 0.000905ns |
| ERROR | 96 | 0.0000003 | 0.00000 | 0.000344 |

***, ns - significant at 0.01% and not significant respectively

APPENDIX 4

ANALYSIS OF VARIANCE FOR PERCENT FRUIT INFECTED(TRANSFORMED DATA), AND LATENT PERIOD IN FRUIT CLUSTER INOCULATIONS:

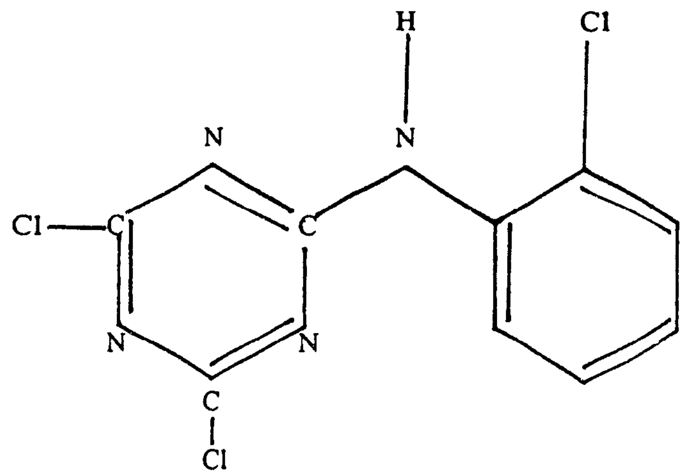
MEANS SQUARES AND SIGNIFICANCE

| | | AECIOSPORE INOCULATION | | UREDINIOSPORE INOCULATION | |
|---|---|---|---|---|---|
| SOURCE | DF | PERCENT FRUIT INFECTED MS | LATENT PERIOD MS | PERCENT FRUIT INFECTED MS | LATENT PERIOD MS |
| BLK | 3 | 0.0016ns | 0.0833ns | 0.0066ns | 0.2292ns |
| CV | 3 | 0.0365** | 14.9167*** | 0.0469*** | 16.7292*** |
| ERROR | 9 | 0.0031 | 0.3056 | 0.0017 | 0.3403 |

**, ***, ns - significant at 1%, 0.1% and not significant respectively

APPENDIX 5

## ANILAZINE - CHEMICAL STRUCTURE
## BIOLOGICAL AND PHYSICAL PROPERTIES




Anilazine is synthesized by the reaction of cyanuric chloride with o-chloroaniline. It is rapidly hydrolyzed under reflux with 1N sodium hydroxide.

| | |
|---|---|
| TOXICITY: | Acute oral $LD_{50}$ for female rats is 2710 mg $kg^{-1}$. Dermal toxicity is low, although precautions against excessive dermal contact should be made. |
| MELTING POINT: | 159-160°C |

## ELEMENTAL SULFUR: PHYSICAL PROPERTIES

| | |
|---|---|
| TOXICITY: | Non-toxic to man and warm-blooded animals. |
| MELTING POINT: | 115°C (becomes a yellow mobile liquid) |