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Studies of environmental factors (temperature and relative humidity) effecting the development of pear rust (*Gymnosporangium fuscum*) and studies of control methods



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Examensarbete inom det dansk-svenska Hortonomprogrammet (300 hp, 300 ECTS) på Master-nivå avancerad E (30 hp, 30 ECTS)										
Titel:	Studies of environmental factors (temperature and relative humidity) effecting the development of pear rust (<i>Gymnosporangium fuscum</i>) and studies of control methods									
	Klimatstudier (temperatur och luftfuktighet) och hur dessa faktorer påverkar utvecklingen av päronrost (<i>Gymnosporangium fuscum</i>) samt studier av bekämpningsmetoder									
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PREFACE

This is the result of a master thesis performed as a Master of Science degree within the frame of the Danish-Swedish Horticulture Programme at the Swedish University of Agriculture, Alnarp. It is a continuation and an extension of my previous project, "The distribution of *Gymnosporangium fuscum* and its implication on pear cultivation in Sweden" (2008).

The purpose of writing this report was to study and evaluate how the local environment, temperature and humidity, influences the development of pear rust. The result would be a starting point for developing future control strategies and a tool in a potential forecasting system. The study was also supplemented with control procedures.

To my supervisor Guy Svedelius, thank you for all support, inspiriting discussions and cheerful calls in which we often ended up talking about something completely different. Special thanks to my examiner Professor Birgitta Rämert for your positive attitude and for inspire me to continue with the pear rust issue. I would also like to thank Partnerskap Alnarp for partly funding the trials and to FOR who initiated the pear rust project.

Tack min familj och mina vänner för all uppmuntran, support och roliga stunder, utan er hade jag nog inte blivit klar med denna uppsats. Tack snälla mamma för alla mysiga söndagsresor till Falköping vi gjorde under denna period.

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ABSTRACT

Pear rust is a serious pest of pears for the individual gardener and cause bright orange lesions on the leaves. Repeated infections cause poor vigour of the tree that with time cause reduced fruit set. The distribution of pear rust in Sweden has previously been limited to the southern parts of Sweden. In recent years the distribution of the pathogen have extended further north.

This study includes climate measurements of temperature and humidity conducted during the growing season of 2008, at three locations each representing a climate zone. Alnarp located in zone I, Lidköping in zone II and Skara in zone III. This combined with continuous observations of the juniper hosts to evaluate the release of spores. At the same time three different control methods of pear rust were evaluated as spraying of sulphur- and oil emulsion and fibre cloth covering of branches. During the previous season of 2007, pear rust infections were confirmed on the current tree and contaminated junipers were located in zone I. In zone II and III no infections of pear rust were supplied with infected branches brought from zone I.

During week 16 the first spore dispersing telial horns were developed in zone I. However no risk of infection of the pear leaves could be confirmed by the climate data. In week 18 next the occasion with developed telial horns occurred in zone I. Here could a high risk of infection be confirmed by the climate data. An infection was confirmed the following week, within reasonable time of incubation, as small yellow lesions on the pear leaves.

In week 22 another period with risk of infection was confirmed by the climate data. Here was no risk of reinfection of previous infected leaves due to age. But infections of newly emerged leaves could be confirmed later in the summer as the size of the lesions varied between the leaves.

No pear rust infection of pear leaves occurred in zone II, and only a few lesions occurred in zone III later in the season.

All control methods gave an affect on the infection rate of pear rust lesions in relation to untreated branches.

SAMMANFATTNING

Angrepp av päronrost förekommer främst i södra och västra Sverige och är ett problem för alla drabbade trädgårdsägare, nu ända upp i Mellansverige. Orsaken till spridningen antas vara tillgången av mottagliga enar i kombination med ett varmare och fuktigare klimat. Päronrosten orsakar rödorange fläckar på päronbladen och trädet försvagas successivt om smittan återkommer år efter år. Detta kan leda till att skörden minskar eller uteblir helt då karten kan falla i förtid.

Under säsongen 2008 gjordes klimatmätningar av temperatur och fuktighet samt försök på päronträd i tre olika växtzoner, Alnarp zon I, Lidköping zon II och Skara zon III. Försöken bestod av tre olika bekämpningsåtgärder på päronträden såsom besprutning av svavel-respektive oljelösning samt täckning av grenar med fiberduk. Under samma tid följdes även utvecklingen av gelérosten på enarna. Under säsongen 2007 bekräftades rikligt med päronrost på det aktuella päronträdet i zon I (Alnarp) och infekterade enar i omgivningen lokaliserades. Dock kunde ingen infektion konstateras på de aktuella päronträden i zon II och III. Här fanns heller inga infekterade enar i närområdet utan under försöksperioden applicerades här grenar från zon I med gelérost.

Redan under vecka 16 fanns det utvecklad och sporspridande gelérost på enarna i zon I men det fanns ingen infektionsrisk av päronbladen enligt klimatdatan. I vecka 18 var det åter utvecklad och sporspridande gelérost och vid detta tillfälle kunde klimatdatan bekräfta en stor infektionsrisk. Denna infektion av päronbladen kunde också bekräftas genom synliga bladfläckar veckan därpå vilket var inom rimlig inkubationstid.

I vecka 22 kunde ytterligare en period med infektionsrisk bli bekräftad. Här skedde dock ingen nyinfektion av de tidigare infekterade bladen utan det var de nyutvecklade bladen på skotten som löpte störst risk. Detta kunde också bekräftas senare på säsongen då bladfläckarna på dessa var mindre och senare i utvecklingen.

Ingen bladinfektion av päronrost kunde bekräftas i zon II. I zon III förekom endast ett fåtal fläckar under senare perioden av sommaren.

Alla bekämpningsförsök gav effekt på päronrosten i jämförelse med obehandlade grenar.

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INTRODUCTION

Pears (*Pyrus cummunis*) are primarily cultivated in the southern parts of Sweden due the their limitations in hardiness (Fernqvist, 1993). There are a number of fungal diseases that may decrease the vitality of the tree, or destroy the harvest for the home gardener. Nectria canker, caused by *Neonectria galligena*, causes canker tissue which in time kills young trees or branches of older trees. Pathogens as pear scab, *Venturia pirina*, primary infects fruits and foliage, which become disfigured with black spots and lesions (Jones and Aldwinckle, 1990) and pear rust, *Gymnosporangium fuscum*, primary infects foliage and causes reduced fruit production.

To prevent infectious diseases the grower should avoid susceptible crops or varieties and make sure the growing conditions are good. If curative control is needed, the home gardener could use measures from the organic orchards. Soaps, oils, plant extracts and a few class III compounds are available (Pettersson and Åkesson, 1998). The most important factor affecting fungal distribution and infection is the environmental conditions. Mainly humidity and temperature on host surface, but also host susceptibility influence the infection rate. Plant susceptibility and pathogen infectivity remain essentially during a period of time, while environmental conditions change to various degrees and affect the development of the disease. These changes of the environment may favor either the host or the pathogen. Disease simulations models or forecasting systems, have been developed based on these environmental changes, Mills table and RIMpro for example (Agrios, 1997; Bio Fruit Advies, 2008).

The aim of this report was to study and evaluate how the local climate, influences the development of pear rust infections. This was conducted by making climate measurements of temperature and humidity during the growing season of 2008, at three locations each representing a climate zone. Alnarp located in zone I, Lidköping in zone II and Skara in zone III. This combined with continuous observations of the juniper hosts (*Juniperus chinensis, J sabinae*) to evaluate the release of spores. The study was also supplemented with control procedures suitable and possible for the use of home gardeners.

BACKGROUND

Casual organism

The European pear rust is caused by the fungi *Gymnosporangium fuscum* DC (syn. *Gymnosporangium sabinae* (Dicks) Wint.) and belongs to the Bacidiomycetes of the order Uredinales and family Pucciniaceae (Laundon, 1977a; Agrios, 1997). This is a genus primarily of northern temperate climate and there are about six species in northern Europe (Cummings and Hiratsuka, 2003). In a taxonomic account of the genus implemented by Kern (1973), 57 species of *Gymnosporangium* were recognized. Of species evaluated, 38 required *Juniperus* as host genera and 10 *Pyrus* (Kern, 1973). The genus of *Gymnosporangium* is unusual since its telial state occur on gymnosperms and the aecial state on dicotyledonous, predominantly on the Pomoideae of the Rosaceae. The gelatinizing pedicles of the teliospores characterize nearly all species (Cummings and Yasuyuki, 2003).

G. fuscum is an obligate parasite, attack only living tissue and alternates between species of *Pyrus* (the aecial host) and *Juniperus* (the telial host) to complete its lifecycle. The aecial hosts include *Pyrus communis*, *P. calleryana*, *P. elaeagrifolia*, *P. nivalis*, *P. salicifolia*, and *P. ussuriensis* (McCain and Rosenberg, 1961; Fitzner and Fischer, 2005). Whereas *P.* 8

korzhinskyi, *P. betulifolia*, *P. cordata* and the hybrid *P. salicifolia* 'Pendula' are the most resistant varieties according to Fitzner and Fischer (2005).

The telial hosts are predominantly cultivars within *Juniperus sabina*, *J. chinensis*, *J. media*, *J. scopulorum* and *J. virginiana* (Laundon, 1977a; Agrios, 1997). *Thuja* and *Chamaecyparis* are unaffected of pear rust as well is the common Swedish juniper, *Juniperus communis* (Laundon, 1977a).

Geographical distribution

G. fuscum is widely distributed throughout Europe with observations extending to Asia Minor (Lebanon, Syria and Turkey) and North Africa (Algeria and Morocco). The pathogen has also been introduced to North America (California, Washington, and British Columbia) probably through the importation of junipers from Europe (Laundon, 1977a; Hollebone, 2006).

The distribution of pear rust in Sweden has previously been limited to the southern parts, zone I-II, with expansions up on the western coast line (Pettersson and Åkesson, 1998; Svanfeldt, 2006). The majority of junipers cultivars within *J. sabina* are hardy up to zone V (Fernqvist, 1993). This access of host plants is a possible source for a further distribution of pear rust throughout the country.

In recent years has the distribution of the pathogen extended. According to an earlier study by the author, single observations have been made in Arvika and Gävle which represents zone III respectively IV of plant hardiness (Karlsson, 2008).

Symptoms and damage

Juniper host

Generally pear rust has insignificant affect on the juniper host. The infection is perennial and often it kills slender branches in three to four years (McCain and Rosenberg, 1961). The infections are inconspicuous except during moist conditions in spring. From mid April to mid May, telial horns swell up and become gelatinized, sizing about 10mm in width and 20mm in

high and spread basidiospores (Vukovits, 1980; Hilber and Siegfried, 1997). At dry conditions the swellings contract and become brown and hard. When shed, they leave small depressions in the distended stem tissue (Vu



Figure 1. Needles of juniper infected with pear rust.

leave small depressions in the distended stem tissue (Vukovits, 1980).

The pathogen infects young and succulent shoots and needles of nearby susceptible junipers by aeciospores instantly in the autumn. The first symptoms of infection occur as small telia of a few millimetres and can be discovered on juniper foliage as early as the following spring (Fig. 1) (Borno and van der Kamp, 1975; Ormrod et al., 1984).

Old perennial infections of the pathogen survive the winter as mycelium that breaks through the surface of the infected tissue in spring as small, firm and dark brown horns (Borno and van der Kamp, 1975).

The growth of the pathogen is generally restricted to the cortex and the hyphae are intercellular (Schmid, 1954). The mycelium stimulates increased cell formation of the cambium and cause enlargement of the branch tissue. From these stem swellings, bright tongue-shaped telial horns appear as columns (Vukovits, 1980).

Pear host

Infection of pear leaves occur at the time of basidiospores release on junipers, usually from the mid of April to end of May. Young and succulent pear leaves are most susceptible to infection. According to Jones and Aldwinckle (1990) are the apple leaves most susceptible to infection of *G. juniperi-virginianae* (apple cedar rust) when they are 4-8 years old (Jones and Aldwinckle, 1990). No similar facts could be found according to pear leaves and infection of *G. fuscum*.

The first symptoms appear as yellow spots on the upper side of young leaves, generally seven days after the infection (Fig. 4) (Dong et al., 2006). Gradually, these circular shaped spots become thickened bright orange sizing up to two centimetres in diameter. These lesions are very conspicuous and are not to be confused with other pathogens (Ormrod et al., 1984). One individual leaf may have several lesions depending on the infection pressure, leaf age and the susceptibility of the variety (Juhásova and Praslièka, 2002).

In the centre of the lesions, black dots of fruiting bodies, spermagonia, are formed. These appear within 13-17 days after infection (Vulkovits, 1980). Opposite the spermagonia, aecia are formed in groups of 4-16 aggregated cluster cups onto small swollen areas of tissue on the underside of the leaf. These are pale coloured, cylindrical structures 2-5mm high, 1-3mm wide with longitudinal splits that remain closed at apex (Heinze, 1978). The aecia require four month for development and appear from the end August, depending on when the infection occurred. However, other factors that affect the time of aecia development are weather influences, soil conditions and internal conditions of the tree (Bernaux, 1947). Aeciospores are released from late August until November or until all infected pear leaves are shed in fall (Ormrod et al., 1984).

Heavy infected leaves may curl and drop prematurely (Hilber and Siegfried, 1997; Naqvi, 2004). If infection pressure is high over many years the pear tree may lose its vitality and predispose it to attacks by secondary pathogens. The fertility rate of the tree could also be affected, resulting in poor fruit set or premature fruit drop (Gram and Weber, 1944; Phillips and Burdekin, 1992; Hilber and Siegfried, 1997).

November - March



The infection of the juniper host is perennial and appear as swellings of the stems with scars from previous telial horns.

April



The pathogen survive the winter as mycelium which breaks through the surface of the infected tissue in spring as small, firm and dark brown horns

September - October



Aecia are formed opposite the spermagonia on the underside of the leaf. These produce and disperse aeciospores that infect tissue of juniper.

August



Characteristic symptoms of pear rust.



June - July



Spermagonia are formed in the middle of the lesions that later become visible as small black dots.

April - May



In moist conditions the telial horns swell up and disperse basidiospores that infect susceptible pear tissues.

May - June



When the telial horns are shed, they leave small depressions in the distended stem tissue.

June



The first symptom of pear rust infection on pear leaf occurs as small yellow dots one week after infection.

Life-cycle

G. fuscum survives the winter as perennial dikaryotic mycelium on stems of its juniper host and produce telia annually in spring. When the conditions are moist and temperature favourable, the telial horns swell and form the characteristic yellow-brown, tongue-shaped structure, formed by columns of teliospores and pectin (Phillips and Burdekin, 1992; Butin, 1995; Agrios, 1997). The teliospores are thick-walled and two-celled by transverse septum sizing 42-56 x 22-32µm. The shape is ellipsoid with a yellowish colour. They are borne singly on long pedicles, which absorb water and cause the gelatinized swellings (Laundon, 1977a; Cummings and Yasuyuki, 2003).

The teliospores germinate and produce a four-celled and club-shaped basidia from which four basidiospores are released (Phillips and Burdekin, 1992). Basidiospores are dispersed by air and infect succulent pear leaves primarily at night when humidity is high. They require free water on plant foliage to germinate. If deposited on dry foliage their vitality remains for approximately a day if humidity is high (Hilber et al., 1990; Wayne and Howard, 2005).

The distance for how long the basidiospores may go by wind could be rather long, 300-500 meters, but then with small infections as a result. Within a distance of 50 meter severe infections are to be expected. Other factors that influence the distribution and infections rate of basidiospores are wind direction, topography and the severity of the infection on the juniper host (Hilber and Siegfried, 1997; Siegfried and Viret, 2004).

When the basidiospores infect the young pear leaf, haploid mycelium is produced that forms spermagonia on the upper side of the leaf. The spermagonia are immersed in host tissue and apparent as small black sticky dots in the centre of the lesion (Phillips and Burdekin, 1992; Agrios, 1997). These contain haploid spermatia and receptive hyphae. Insects are involved in the distribution of these haploid spermatia and they are attracted to the lesions by the nectar with sticky content excreting from the spermagonia (Kotte, 1958; Heinze, 1978).

The spermatia fertilize the receptive hyphae, which result in the production of dikaryotic mycelium and dikaryotic spores (Agrios, 1997). The dikaryotic mycelium forms aecia on the underside of the leaves. The aecia produce aeciospores, which are one-celled sizing 23-37 μ m diameter and are broadly ellipsoid (Laundon, 1977a; Sinclair and Lyon, 2005).

The aeciospores are released from the pear leaves at the time of maturation of aecium, normally from September, until all infected leaves are shed. Air currents to junipers carry the aeciospores. After the aeciospores have been released, the mycelium in the infected pear tissue normally dies out. Occasionally it infects and survives the winter at the base of the pear buds (McCain and Rosenberg, 1961; Hunt and O'Reilly, 1978; Vulkovits, 1980; Butin, 1995).

Environmental requirements of pear rust infection

Teliospores of *G. fuscum* in mature telial horns germinate when free water from rain is available and produce basidiospores, which infect succulent pear leaves. And the infection of pear leaves by basidiospores is dependent on the duration of moist periods which and the temperature during such. The cedar apple rust (*G. juniperi-virginianae* Schwein) is an important rust of apples in eastern North America. During rain the teliospores germinate to produce basidiospores within 4 hour at 11-25°C and within 5-7 hour at 7-11°C but not at 26-30°C. Basidiospores of *G. juniperi-virginianae* only infect the host if there is a film of water present for a sufficient time (Jones and Aldwinckle, 1990).

Effect of temperature and Relative humidity

A Swiss study by Hilber et al. (1990) was conducted to evaluate the influence of temperature on telio- and basidiospore germination of *G. fuscum* in vitro. The study also evaluated the effects of temperature, inoculums concentration and leaf wetness periods (LWP) on potted seedlings of pear, grown in plastic pots in the greenhouse (Hilber et al., 1990).

The germination rate of teliospores of *G. fuscum* was evaluated in vitro by incubate inoculated Petri dishes at 100 % RH at 0°C, 5°C, 10°C, 15°C, 20°C, 25°C and 30°C. The most favourable temperature for teliospores to germinate occurred between 10-25°C with an optimum at 15-20°C. No germination occurred at 0°C, 5°C and 30°C. This shows that the temperature influence the development of the pathogen on the junipers in spring. After 12 hour the germination rate at 10°C was 71% and 85% at 20°C (Hilber et al., 1990).

Germination rates of basidiospores was examined in the same way as for the teliospores and occurred at 5-25 °C, with an optimum at 20 °C. No germination was observed at 0 °C and 30 °C (Hilber et al., 1990).

The study indicates 15° C as the optimum temperature for infection of pear leaves. However, the most important factor affecting infection rate is the LWP (leaf wetness period). At the optimum temperature $(15^{\circ}$ C) a LWP of 3 hour was enough to have 3 to 10 lesions per leaf. Infections could be considered severe even at temperatures as low as 0-4°C, but it required an increasingly longer LWP. At 4°C a LWP of 7 hour gave 1 to 2 lesions per leaf. The infection rate of the pear seedlings increased with increasing LWP and increasing inoculums concentration (Hilber et al., 1990).

In 2006 another study by Dong et al. was conducted to evaluate the effect of environmental conditions (temperature, RH and duration of free water) on germination and survival of telioand basidiospores of the Japanese pear rust, *Gymnosporangium asiaticum* Miyabe ex G. Yamada. *G. asiaticum* is a genus native to Asia and alternates between junipers (*Juniperus chinensis* and *J. procumbens*) and pear species (among others both *Pyrus communis* and *P. sinensis*). The Japanese pear rust is distributed through China, Japan and perceived in the USA (Laundon, 1977b).

Teliospores germinated within the temperature range 5-28°C, with an optimum between 16°C and 20°C. At these temperatures the minimum time for basidiospores production was 3 hour. According to the study the telial horns needed to be soaked in water for initiating germination. For as little as 30 seconds initiated production of basidiospores. After this primary soaking, RH had little effect except at extreme temperatures (Dong et al., 2006).

Basidiospores germinated at 5-30°C with an optimum at 15°C and required free water or saturated moisture to germinate. In free water from rain the germination was eight times greater than at RH 100%. According to the study the basidiospores appeared to be tolerant of dry periods. They survived for at least six days with RH as low as 45% (Dong et al., 2006).

Control measures

Cultural control

Elimination of either host will control pear rust. Removal of the juniper host is preferably done within a distance of at least 500m from the pear tree to accomplish result. Infection severity will decrease with increased distance between the hosts. To achieve this, a voluntary cooperative effort between neighbours must be made to prohibit further development and spread of the disease. If susceptible cultivars were removed and replaced with resistant ones, the disease cycle of the fungi would be broken and the pathogen thereby controllable. It is not

possible to cure a diseased juniper. But, removal of infected branches could give result if made before the time of spore release or before gelatinisation of the telial horns (Hilber and Siegfried, 1997).

No long-term effect is achieved by removing the infected leaves of the pear tree since the infection is annual, i.e. the tree will be reinfected next spring if diseased junipers still occur within the vicinity. Since the shed leaves are no source of infection, they could be left on the ground without further processing (Hunt and O'Reilly, 1978; Hilber and Siegfried, 1997).

Fungal control

No curative chemical control measures are available for the individual gardener against pear rust. Within the commercial and IP fruit production chemical control is an important management tool of fungal attack. The fungicides approved and registered in Sweden has shown to have an affect on *G. fuscum* as well, especially those for control of scab, *Venturia sp* (Juhlin, 2006b).

Sulphur is commonly used and probably the oldest fungicide known (Agrios, 1997). Today it is an important tool against scab, *Venturia sp.* in other countries. In Sweden is the sulphur-granulate Kumulus DF registered and allowed in organic production for control of powdery mildew, *Podosphaera leucotricha* (Ell. & Ev.) E. S. Salomon. It has also shown side effects on *Venturia*, (Juhlin, 2006a).

Kumulus DF is a class III fungicide and approved for public use. Thereby could it be an alternative for the individual gardener. Preventative applications of Kumulus in spring during time with risk of infection ought to decrease infections of pear rust. However, repeated use is necessary since sulphur evaporates and easily washed off by rains. Kumulus should not be applied in bloom since it may intimidate pollinators (Sandskär et al., 2005; Juhlin, 2006a).

Vegetable oil emulsions have shown to have an effect on *Podosphaera*. Primary it operates by contact with physical impact on the pathogen (Sandskär et al., 2005). Oil emulsions also seem to reduce infections by changing the characteristics of the leaf surface. This would eventually prohibit spore penetration and prevent infection (Agrios, 1997).

Another way of preventing pear rust infection is to stop the spores from reaching the leaves by hooding the tree with fibre cloth. No previous studies are to be found evaluating its effect on pear rust. However, fibre cloth covering are used in organic growing systems to protect the crop from pathogens (Grundberg, 2003).

Forecasting systems

There are a number of different simulation models used for recreation of disease outbreaks building on the correlation between local climate data and pathogen. Apple scab, *Venturia inaequalis* (Cooke) Wint., is an ever-present fungal disease in orchards and different warning systems have been developed over the years to forecast its density of spores. Mills table present the correlation of duration of rain required at each temperature for infection of apple scab to take place. It also suggests the approximate days of incubation. By these means the grower decides control measures (Agrios, 1997; Jones and Aldwinckle, 1990).

Today is computer simulation models, building on local climate data used in orchards as forecasting systems. Such a system is RIMpro, a warning system, containing models for apple scab, codling moth and sooty blotch. RIMpro is used in integrated and organic orchards throughout Europe. In practice, growers use these systems to modify the timing of applications of fungicides (Bio Fruit Advies, 2008).

FIELD EXPERIMENT

Climate and control measurements

The climate was studied during the growing season of 2008 by position climate loggers in pear trees with the aim to estimate the correlation between climate and pear rust infection. Pear trees at three different locations were studied, each representing a climate zone. Alnarp located in zone I, Lidköping in zone II and Skara in zone III. This combined with continuous observations of the juniper hosts to evaluate the release of spores. At the same time three different control methods of pear rust were evaluated within the current trees. Fibre cloth covering was chosen as method of encapsulating and thereby protecting the leaves from infection. Oil emulsion was used as a control method since it has contact and physical impact on the fungi. Kumulus was used as a preventative control method.

Material and method

Observation of telial horns on juniper host

In the location of Alnarp five junipers were selected within the adjacent range, in this case of 75-250 meters from the studied pear tree (Fig 4). The junipers chosen were confirmed with telial horns. Of these were two represented by cultivars within *Juniperus sabina*, two *J. chinensis* and one *J. media*. Observations were made on weekly basis estimating the release of basidiospores. The observations started the 7th of April, just before pear bloom and ended when the occurrence of telial horns was over the 29th of June.





Figure 3. Telial horns on juniper branch applied artificially in Skara and Lidköping.

Figure 4. Map over the location of Alnarp campus and the distribution of basidiospores from the junipers to the pear trees.

S1: Juniperus sabina (100 meter from pear tree)
S2: Juniperus sabina (250 meter from pear tree)
C1: Juniperus chinensis (220 meter from pear tree)
C2: Juniperus media (220 meter from pear tree)

C3: Juniperus chinensis (220 meter from pear tree)

In the locations of Lidköping and Skara the pathogen was artificially introduced due to lack of juniper hosts in the neighbouring area. Only few lesions of pear rust have previously been observed on the trees. Therefore juniper branches with telial horns were brought from Alnarp and Lidköping and mounted 25 meters from the pear tree (Fig. 3). These branches were subsequently replaced to ensure good condition of the telial horns and production of basidiospores. Swelling of telial horns confirmed production of basidiospores.

Monitoring climate loggers

The pear trees were selected regardless of variety. At each location one climate logger was positioned close to the middle of the tree at about 150 centimetres above ground. A paper cap was enfolded around the loggers as protection against sun radiation (Fig. 5).

The climate loggers registered temperature (°C) and humidity (RH %) every fifth minute. The data was transferred by the Diligence of windows software and used to calculate dew point and risk of leaf wetness used in this investigation. The climate registration with logger in Alnarp started at the 16th of April, just before bloom and in connection with the appearance of telial horns on the juniper host. Climate registration with logger in Lidköping and Skara started the first week in



Figure 5. Climate logger positioned in Alnarp seen from below and enfolded by paper cap as protection.

May. During the season observations were made on regular basis to follow the development and growth of the spore stages.

Trials of control procedures

Three different control procedures were used during the study to evaluate its effect on pear rust infection. Branches chosen were equally distributed over the tree and treated with either a sulphur emulsion of Kumulus DF (0.30% = 3 g/l = 5 kg/ha), oil emulsion (2%) and fibre cloth covering. Kumulus DF (BASF) was applied with a hand sprayer. Rape oil (cold-form) was used in the oil emulsion and applied by hand sprayer. The fibre cloth (13 g/m^2) was wrapped around the entire branch and fixed with stapler (Fig. 6).



Figure 6. Applications of fibre cloth covering as a control procedure in Alnarp.

The procedures were compared with untreated branches. The first treatment was conducted at the

29th of April in Alnarp, just after bloom in week 18. The treatments were repeated at the 5th of May, 12th of May and the 24th of May, totally four occasions. In Lidköping and Skara the first treatment was conducted at the 18th of May, just before bloom. The treatments were repeated 18th of May and 25th of May with a break during bloom.

Each treatment was repeated five times on each tree. Ten leaves from each replicate were randomly graded, infected (1) and uninfected (0). Totally 200 leaves of each tree were evaluated.

Calculations

Calculations of climate data

To estimate the accumulated time of potential leaf wetness and condensation the dew point temperature was calculated using the data of humidity and air temperature received from the loggers. The following formulas were used calculating saturation vapour pressure (Es) and actual vapour pressure (E) to obtain the dew point temperature (Palmer, 2000).

First saturation vapour pressure Es was calculated: Es = $6.11 * 10.0 \land (7.5 * T / (237.7 + T))$ Where T is the temperature (°C) at a given time.

The actual vapour pressure E: E = (RH * Es) / 100Where RH is the relative humidity in percent at a given time.

The dew point temperature (Tdc) was obtained: Tdc = $(-430.22 + 237.7 * \ln (E)) / (-\ln (E) + 19.08)$

The accumulated time (h) during measured period with high risk for condensation and leaf wetness was calculated by reducing the temperature at a given time, with the dew point temperature at a given time. A temperature difference within the range of $0.3-1.0^{\circ}$ C from dew point temperature was calculated. The deviation of 0.8 degrees from dew point is used as an estimating point in figures below (see table 1).

Statistical analyses

The results from the control procedures were analysed using Mann-Whitney U test and Kruskal-Wallis test to distinguish if the different methods had given statistical differences. The Kruskal-Wallis test is a ranking test with the data replaced by their ranks. Both tests are non-parametric tests and are a measure of equality of two population distributions when there is no assumption of normal distribution (Dytham, 1999). In this case, one nominal variable and one measurement variable is used and the measurement variable does not meet the normality curve. The statistical analyses were conducted in Minitab.

RESULTS

Observations of telial horns

The first symptoms of emerging telial horns were observed in the beginning of April (week 14) and present on all juniper varieties in Alnarp. In Lidköping telial horns were observed the same period of time, during week 14, on a *J. chinensis* variety.

The first symptom of emerging telial horns appeared as dark brown eyes in the previous season scars. In the time just before pear bloom (week 16), the telial horns started to maturate and teliospores attain ability to germinate. After successive rainfall these telial horns fell off and left scars within a few days. No additional telial horns emerged from the current infection. Instead new telial horns emerged constantly during ten weeks in sequence (until week 24). Except on *J. sabina* on which the development of telial horns was completed in the middle of

May (week 20).

A visible difference was experienced in the general behaviour of the pathogen between the juniper varieties. On the *J. sabina* varieties the duration of telial horns appeared to be shorter and the infection rate was less compared to *J. chinensis* and *J. media*. The *J. chinensis* and *J. media* varieties appeared to have the most established infections and the largest amounts of infected branches. The period of mature and spore dispersing telial horns was also more constant on these varieties.

Climate

The climate in Alnarp was studied from the 16th of April until the 30th of June. Climate registration in Lidköping and Skara started the first week in May. Climate data and results from the different climate zones I-III are presented and analysed below.

Condensation and leaf wetness

During the first two weeks of measurements, week 16 and 17, only data of week 16 showed a short period with risk of infection of the newly emerged pear leaves in Alnarp. In week 16 the air temperature deviation 0.8° C from dew point was 0.6 hours (Fig. 7). The accumulated time of RH at 90% was 12 hours and normally appeared in the sunrise (Fig. 8).

The first occasion with a high risk of spore dispersal and infection occurred in week 18 where the accumulated time with risk of condensation and leaf wetness was 23 hour (Fig. 7). In this week was also the gathered time of RH at 90% as much as 44.5 hours and 20 hours at 95% RH (Fig. 8).

The next occasion with risk of leaf wetness and spore infection of pear leaves occurred in week 22. The accumulated time with risk of condensation was 12 hours (Fig. 7), 22 hours with RH at 90% and 11 hours with RH at 95% (Fig. 8).



Accumulated time with risk of condensation 0.8 degree C from dew point per week in Alnarp

Figure 7. Accumulated time with risk of condensation (air temperature deviation 0.8° C from dew point) per week in Alnarp.



Accumulated time with RH at 90% and 95% in Alnarp

Figure 8. Accumulated time with RH at 90% and 95% per week in Alnarp.

Table 1. The table below shows the accumulated time (in hours) with risk of condensation and leaf wetness per week in Alnarp. The accumulated time with air temperature deviation (temperature (T) - dew point temperature (Tdc)) are presented within the range of $0.3-1.0^{\circ}$ C from dew point.

Accumulated time (h) per week with calculated risk of condensation in Alnarp													
Temperature deviation													
(T-Tdc ⁰C)	16	17	18	19	20	21	22	23	24	25			
0.3	0	0	5.4	0	0	0	3.7	0	0	0			
0.4	0	0	5.8	0	0	0	3.8	0	0	0			
0.5	0	0	12.6	0	0	0	5.2	0	0	0			
0.6	0	0	16.8	0	0	0	9.3	0	0	0			
0.7	0	0	19.9	0	0	0	10.7	0	0	0			
0.8	0.6	0	23.0	0	0	0	12.1	0	0	0			
0.9	0.7	0	25.7	0	0	0	13.7	0	0	0			
1	1.4	0	26.9	0	0	0.08	16.4	0	0	1.5			

In Lidköping and Skara the measurements started in week 19. During the fist three weeks the measurements showed periods with risk of infection of the emerging pear leaves. In week 19 the accumulated time of risk of condensation was 14 respectively 11 hours (Fig. 9). The accumulated time of RH at 90% was 23 hours in Lidköping and 24 in Skara (Fig. 10 and 11).

In week 20 there was a higher risk of spore dispersal and infection in Skara compared to Lidköping. The accumulated time with risk of condensation and leaf wetness was 36 hours and RH at 90% 54 hours (Fig. 9).

During week 21 the risk of leaf wetness and infection of pear leaf continued with the accumulated time of 18 respectively 17.5 hours of leaf wetness (Fig. 9).

The following weeks, 22 and 23 the accumulated time with risk of condensation was reduced but to increase again in week 24 and 25 (Fig. 9).



Figure 9. The accumulated time 0.8° C from dew point presented per week in Lidköping and Skara.



Accumulated time with RH at 90% and 95% in Lidköping

Figure 10. Accumulated time with RH at 90% and 95% per week in Lidköping.



Accumulated time at RH 90% and 95% in Skara

Figure 11. Accumulated time with RH at 90% and 95% per week in Skara.

Temperature and relative humidity

The following figures 12, 13 and 14 present the temperature and RH data from the location of Alnarp as mean day and night for each week in a time line. The pictures correspond to the observations made the current week. Table 2,3 and 4 present the corresponding climate data for Lidköping and Skara week19 to 25.



16/4 Start climate measure

29/4 Start of treatments

			Ocular risk o	f infection	Low risk of	infection	High risk of infection		
	W.	15	w.	16	W.	17	w. 18		
	Temp °C	RH %	Temp °C	RH %	Temp °C	RH %	Temp °C	RH %	
mean day	-	-	10.9	60.0	12,1	47,2	16	60	
mean night	-	-	3,2	82,7	5,3	73,7	10,2	73,3	
temp diff.			7,7		6,8		5,8		

Figure 12. A time line presenting temperature and RH data as mean of day and night for week 16-18 in Alnarp. The pictures correspond to the observations made the current week.



	9/5 first sympto	m of pear rust	Distinct sympton	ms	leave	s			
	Low risk of i	nfection	Low risk of in	fection	Low risk of in	fection	High risk of infection		
	w.	19	w. 2	20	w. 2	1	w. 22		
	Temp °C	RH %	Temp °C	RH %	Temp °C	RH %	Temp °C	RH %	
mean day	20,4 51,6		17,3 47,3		16,3 48,7		17,8	53,4	
mean night	11,2 80,8		8,7	68,7	8,5	74,3	10	77,7	
temp diff.	9,2		8,6		7,8		7,8		

Figure 13. A time line presenting temperature and RH data as mean of day and night for week 19-22 in Alnarp. The pictures correspond to the observations made the current week.



					Only scars left	of telial horns	spermagonia		
	Low risk of i	nfection	Low risk of in	nfection	Low risk of in	fection	Low risk of infection		
	w. 23		w. 2	24	w. 2	25	w. 26		
	Temp °C	RH %	Temp °C	RH %	Temp °C	RH %	Temp °C	RH %	
mean day	12.1	47.2	15.2	57.6	17.3*	61.4*	15.6	61.2	
mean night	5.3	73.7	11.2	79.1	13.6*	78.0*	12.3	68.5	
temp diff.	6.8		4		3.7*		3.3		

Figure 14. A time line presenting temperature and RH data as mean of day and night for week 23-26 in Alnarp. The pictures correspond to the observations made the current week.

* Data presented in week 25 are received from Trädgårdslaboratoriet at Alnarp.

Table 2: A time line presenting temperature and RH data as mean of day and night for each week 19-25, in Skara.

	w. 19		w.	20	w.	21	w.	22	w.	23	w.	24	w.	25
	Temp	RH	Temp	RH	Temp	RH	Temp	RH	Temp	RH	Temp	RH	Temp	RH
	C°	%	C°	%	C°	%	C°	%	C°	%	C°	%	C°	%
mean day	18.8	41.5	11.1	64.4	13.6	47.9	20.5	33.1	16.4	65.1	19.1	45.9	15.0	54.3
mean night	7.8	83.0	6.5	86.2	6.0	81.3	10.4	70.2	9.3	85.4	10.1	71.9	8.0	76.6
temp diff.	11.0		4.6		7.6		10.1		7.1		9.0		7.0	

Table 3: A time line presenting temperature and RH data as mean of day and night for each week 19-25, in Lidköping.

	w. 19		w.	20	w.	21	w.	22	w.	23	w.	24	w. 1	25
	Temp	RH	Temp	RH	Temp	RH	Temp	RH	Temp	RH	Temp	RH	Temp	RH
	C°	%	C°	%	C°	%	C°	%	C°	%	C°	%	C°	%
mean day	18.6	44.8	12.6	64.9	14.2	49.4	20.2	37.0	23.3	37.8	15.8	10.5	14.5	71.0
mean night	8.8	76.4	5.7	86.2	6.8	78.1	10.5	70.9	13.6	72.8	10.5	83.3	9.3	92.9
temp diff.	9.8		6.9		7.4		9.7		9.7		5.3		5.2	

Control procedures

The interference between the control procedures was analysed using Mann-Whitney U test. The test results indicate that there was a significant difference between treated and untreated branches independent of procedure. This shows that all control methods gave an affect on the infection rate. But there were no significant variation between the different procedures according to the Mann-Whitney U test.

The variation between the procedures was also analysed using Kruskal-Wallis test. The result ranked fibre cloth first, then Kumulus, oil and untreated last. However, the Kruskal-Wallis test shows how the procedures relate and are ranked to each other. Not their effect on pear rust.

In the procedures of oil emulsion and fibre cloth covering, burn damages occurred on the leaves.

DISCUSSION

Variation in occurrence of telial horns between the varieties

The duration of telial horns appeared to be shorter and the infection rate was lower on the *J*. *sabina* varieties compared to *J*. *chinensis* and *J*. *media*. This could be due to differences in infection rate, growing pattern or morphological differences. The period of symptoms and developing telial horns lasted from week 14 until week 24, which is 10 weeks in a sequence. However, there was a variation in occurrence of the telial horns between the varieties. On the varieties of *J*. *sabina* the development of telial horns was completed in the middle of May. This was almost a month earlier than for the other varieties.

The measurement of the climate started in the middle of week 16, before bud break of pear and in correlation with the appearance of telial horns on the juniper host. Before this week (week 15, see figure 12) no gelatinised telial horns occurred and no leaves were fully developed on the pear host. Thereby no infection could occur. In the end of week 15 there was a short rainfall generating the telial horns to swell. Unfortunately there was no climate data available. This short period of rain triggered the telial horns to be gelatinised during the following period (week 16) until they fell off. During week 16 the accumulated time of RH over 90% was only 12 hours. According to the study by Dong et al. (2006) the telial horns that were soaked in water for as little as 30 seconds, initiated production of basidiospores. After this primary soaking, RH has little effect except at extreme temperatures (Dong et al., 2006). This explains why there could be gelatinised telial horns during periods of less favourable environment as during week 16.

Influence of temperature and relative humidity

Alnarp

Already in week 16 there was an ocular risk of spore dispersal and infection, because there were gelatinised telial horns spreading spores. However, this risk of leaf infection could not be confirmed by the climate data within the pear tree canopy. There were no risk of leaf wetness by condensation and the relative humidity did not reach above 95%. The accumulated time with RH at 90% was 12 hours, however, this is not considered as a risk of infection.

The next time with ocular risk of infection occurred in week 18 with gelatinised spore spreading telial horns. Here could risk of infection be confirmed by the climate measurements with an accumulated risk of a leaf wetness period of 23 hours. From the middle of the week the accumulated time of RH at 95% was 20 hours. In contrast to the previous week the mean day and night temperatures was higher, 16° respectively 10.2° C. All factors indicating that leaf infection most certainly occurred. According to Hilber et al. (1990) 15° C is the optimum temperature for infection of pear leaves. However, the most important factor affecting infection rate is the leaf wetness period, as the basidiospores require free water on plant foliage to germinate. At 15°C a period of 3 hours would be enough to have several lesions per leaf. Week 18 supplied several hours of leaf wetness risk making it possible to cause infection at lower temperatures.

The climate data achieved from week 18 indicated a high risk of leaf infections, which was confirmed the following week. The first symptoms of pear rust occurred the 9th of May in the end of week 19. Generally the first symptoms appear seven days after infection (Dong et al., 2006). Here the first symptom could be confirmed nine days after the initiated moist period. This time of incubation is most likely affected by temperature and relative humidity but also by the age of the pear leaves (Vulkovits, 1980).

In week 22 the climate data confirmed an additional risk of pear rust infection. The accumulated time with risk of leaf wetness was 12 hours and the RH at 95% was 10.8 hours. Still telial horns developed at the junipers distributing basidiospores. At this time the previous infections had developed apparent symptoms as small yellow dots. These leaves should not risk an additional infection due to age. However, still new leaves are developed from shoots risking infection. Infections of newly emerging leaves could be confirmed later in the summer as the size of the lesions varied between the leaves of the shoot. The leaves at the base had more developed lesions then those at the top. One explanation could be that they were infected in different periods.

The most important factor affecting pear rust infection is the environmental conditions, mainly humidity and temperature on host surface. Plant susceptibility and pear rust infectivity remain essentially during a period of time in spring (Agrios, 1997). In this study, during ten weeks with developing telial horns while the climate conditions alter. Forecasting systems have been developed based on these environmental changes. A resembling system would therefore be possible as a measuring tool for pear rust. However, developing these models demands research and assessment for years (Agrios, 1997; Bio Fruit Advies, 2008).

Lidköping and Skara

The climate data achieved from Lidköping and Skara during the weeks 19, 20 and 21, indicated several occasions with risk of spore infection. During week 20 the risk of pear rust infection could be considered very high in Skara. This week the accumulated time with risk of leaf wetness was 36 hours and the RH at 95% was 30.2 hours. In the same week the mean day and night temperatures was 11° respectively 6.5° C, which would be considered low. However, the long period of moisture during these weeks is the most affecting factor on leaf infection (Hilber et al., 1990).

Unfortunately no infections of pear rust were developed in Lidköping. In Skara were the leaf infections limited and symptoms were spotted in week 24. These lesions were at that time about five millimetres in diameter that would be a sign of that the infections were about four

weeks old and that an infection occurred during week 20 or 21.

The climate data indicated several occasions with risk of infection, which defaulted. No or very limited infections were spotted. One possibility is that the limited amount of telial horns (only one branch per location was applied) restricted the spore dispersal. The severity of the infection on the juniper host affects the distribution and infection rate according to Hilber and Siegfried (1997). Siegfried and Viret, 2004.

Control procedures

Fibre cloth covering

There was a noticeable difference between the treated and untreated branches independent of procedure. The fibre cloth cover gave an unexpected significant reducing effect of infection according to the Mann-Whitney U test. It was also positioned first by the Kruskal-Wallis test, which ranked the procedures in relation to each other. The effect of the fibre cloth was probably due to divergent environment inside. This affected leaf wetness and made the conditions unfavourable for the pathogen.

Unfortunately the covering cloth was removed to late in the season. It should have been removed when the risk of infection ended. The result of the prolonged covering was burn damage of the leaves and malformed shoot growth. These injuries would have been limited if the cloth was removed earlier.

The fibre cloth covering can only be possible as a control method of pear rust on young and small trees. In this study only individual branches was covered which gave an effect on the micro climate inside.

Kumulus DF

The Kumulus control significantly reduced the infection rate and was ranked second according to the Kruskal-Wallis test. It is most effective used preventively (Sandskär et al., 2005; Juhlin, 2006a). In this study it was applied at least once a week from the time after bloom until the risk of infection was over. It was applied repeatedly since it easily is washed off by rains and in order to give new emerging leaves protection.

Applications of Kumulus DF can be an alternative for the individual gardener as a control method of pear rust. However, the applications must be preventative and no curative effect is to be expected. Still, Kumulus should be used with care since it may affect the pollinators. It could also, in high concentration, cause burn damages on leaves and flowers. Another issue for the gardener is the application technique. As it works by contact it requires good coverage of the leaves, which would be complicated in large trees.

Oil emulsion

Also the oil emulsion gave a significant effect of the pear rust infection rate but was ranked according to the Kruskal-Wallis test as the least good of the procedures. Though, this method gave distinct burn damages on the leaves, which affected the ornamental appearance of the tree. This would not be an option as control method. However, it was applied as frequent as the Kumulus procedure. This seems to be unnecessary and maybe one application would be enough as protection. But new emerging leaves must be preventatively protected.

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