# HNS 152

# Rose and ornamental rosaceous trees: a short review of replant diseases and options for future research

July 2007

Project title:	Rose and ornamental rosaceous trees: a short review of replant diseases and options for future research
Project number:	HNS 152
Report:	July 2007
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Date project commenced	<b>1:</b> 1 January 2007
Date completion due:	31 July 2007
Key words:	Rose, rosaceous, tree, replant, disease

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# **GROWER SUMMARY**

#### Headline

Fungi and actinomycete bacteria appear to be the most likely causes of replant diseases in rose and rosaceous trees.

#### Background and expected deliverables

Replant diseases of rose and field-grown trees are long-known and complex problems. Previous research in the UK and overseas has investigated a wide range of possible causes and control strategies. However, there is no clear solution and they remain important problems both for rose growers and field tree producers. Currently in the UK there is interest in the potential of biofumigant crops, mycorrhizae, chloropicrin and other soil amendments as control treatments. Before undertaking any new field experiments, a review of the results of previous studies is required to aid development of a robust research strategy and to identify priority areas worthy of investigation.

This six-month project aims to review replant diseases affecting crop species in the Rosaceae family, with particular reference to rose and ornamental trees. It will draw information from scientific literature, trade press, growers, consultants and internet searches. Information will be interpreted to identify key findings and the relevance of these to the industry. We will make recommendations for future research and development, and summarise current control options.

#### Summary of the project and main conclusions

For the purposes of this review, replant diseases of the Rosaceae are defined as poor growth and performance of plants that have been planted in non-sterilised soil, usually where the same or related species has been grown previously, and where there is no clear cause. Attention is focussed on replant diseases that show at least some degree of species-specificity, as non-specific replant diseases usually have causes that are understood, and can be identified and/or controlled.

The species most commonly encountered during this literature review were almond, apple, asparagus, cherry, citrus species, grapevine, ornamental rose and pear. All of these are rosaceous species except asparagus, citrus and grapevine. Because of its

economic importance, apple (Malus  $\times$  domestica Borkh) is the species in which replant problems have been most studied. Apple replant disease is widespread and occurs in all the major production regions of the world.

#### Symptoms and diagnosis

Replant diseases exhibit no visible symptoms above ground other than a reduction in growth, stunting and a lack of vigour. The most severe growth retardation usually occurs in the first two years, after which there is often a gradual recovery, but affected plants do not catch up with unaffected plants. Yield of apple crops is reduced in the early years after planting. Below ground, roots are usually small, dark and compact and fine roots often show signs of decay.

An apple seedling bioassay has been developed to determine if a particular soil will lead to specific apple replant disease (SARD) in apple trees planted on the site. The test is based on a comparison of apple seedling growth in pots of soil that were either untreated or fumigated with chloropicrin. This is not currently available as a commercial test.

#### Causes

When a poor growth problem is identified, possible causes that are well known and can be easily investigated should first be excluded first – e.g. soil compaction, viruses and nematodes.

Causes of replant diseases in the Rosaceae are not fully understood. They could be simple (one dominating cause) or complex (interacting causes). The effective control of replant diseases by soil sterilisation is strong evidence that the principal causes are biotic. However, abiotic factors may interact with biotic factors, or be secondary or aggravating factors. No primary publications on the cause of replant disease in rose or ornamental rosaceous trees were found. Most research has concentrated on specific apple replant disease (SARD). This research is discussed below.

#### Fungi

It has been claimed that fungi have a causal role in replant diseases, but the evidence has either been circumstantial or related to a certain locality. In the UK, species of *Pythium*, notably *P. sylvaticum*, are reported to be one of the main causes of SARD in apple. It seems probable that in some situations at least, fungi are part of a complex group of causal agents.

#### Actinomycete and other bacteria

Actinomycete bacteria have been shown to be primary pathogens of apple seedling rootlets, and infection level was positively related to the degree of the specific apple replant disease symptoms. Some degree of species-specificity has been demonstrated. However, there is not clear evidence that actinomycetes are the predominant cause of replant diseases in most locations and for most rosaceous species. Actinomycetes cannot easily be cultured in isolation, making it difficult to prove cause and effect.

Other groups of soil bacteria can decrease plant growth, so have potential as causal organisms in replant diseases. However, there is not strong evidence that bacteria are the main cause in commercial situations.

#### Soil-borne viruses

Soil-borne viruses are not considered to be a cause of classic and well-described replant diseases of rosaceous species such as apple.

#### <u>Nematodes</u>

There is considerable evidence that nematodes are associated with replant disease in apples, peaches and cherries. Nematodes may predispose roots to attack by soil pathogens, but for roses and apples, only soil sterilisation prevents replant disease, and treatments that eliminate nematodes without soil sterilisation do not prevent replant diseases.

#### Nutrient deficiencies and pH

It is unlikely that a nutrient deficiency or imbalance is the primary cause of specific replant diseases, but it is possible that they could interact with other factors, and thus affect the severity of a replant disease. There have been beneficial effects of nutrients, especially mono-ammonium phosphate, but the mechanism of this is probably very complex, involving changes in soil micro-organism populations, effects of altered pH, effects on availability of other nutrients, and improved uptake of nutrients when mycorrhizal infections are poor.

Since there is strong evidence that replant diseases have a biotic cause, effects of soil pH are to be expected and do occur.

#### Soil structure

Soil structure may interact with other causes of replant diseases, such as effects of poor drainage on disease development in the presence of soil pathogens.

#### **Phytotoxins**

Phytotoxins released into soil from roots of a previous crop are an unlikely cause of replant disease. The presence of phytotoxic compounds, or the effects of such compounds on plant growth, is not sufficient evidence that these are the cause of replant diseases.

# **Reverse replant**

The term 'reverse replant' refers here to the reversal (or decreased severity) of replant disease symptoms when a plant with replant disease symptoms is replanted in soil in which the same species has not been previously grown. This phenomenon has been found in apple and cherry, but no reports have been found for other species.

# Physiological aspects

Plant diseases involve the action of causal factors and a response by the plant. The recognition of genetic resistance to replant diseases (eg in apple) suggests that there are physiological responses to causal factors, and that these vary between clones (e.g. different rootstocks). An important physiological aspect of replant diseases is the possible role of endogenous plant growth regulators (PGRs) in the infection of roots by pathogens.

# **Management and control**

Management options, for rose replant disease, include avoidance (plant roses somewhere else, or plant something else), soil sterilisation, soil replacement, use of cover crops and use of beneficial soil fungi.

# Soil sterilisation

Soil sterilisation, more correctly referred to as soil disinfestation, is falling out of favour with retailers and the most effective product available, methyl bromide, is no longer available for use in the UK for pre-plant use. This increases opportunities for new control methods.

Soil disinfestation either with a broad-spectrum biocide, or by steaming, has been the most popular and effective control strategy for replant diseases. This effective control measure is strong evidence that the principal causes are biotic.

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Chemicals used for disinfestation have included methyl bromide, formaldehyde (formalin), chloropicrin, and products that release methyl isothiocyanate (e.g. metam-sodium, dazomet). Current soil disinfestation options for rose and field-grown trees are listed in the literature review.

Soil disinfestation has dramatic effects on the ecology of soil microorganisms. Of particular interest is that mycorrhizal fungi are destroyed and this has led to studies of effects of soil sterilisation together with application of nutrients.

# <u>Fungicides</u>

Reported effects of fungicides on replant diseases of the Rosaceae are not consistent, reflecting the probable variation in causes between sites and species. However, some studies show benefits of fungicide use in soils affected by replant diseases.

# **Biological control**

There are three main types of biological control of replant diseases that have been investigated for rosaceous species:

- inoculation with mycorrhizae,
- inoculation with bacteria,
- soil amendment with organic materials, including incorporation of cover crops.

Within a complex soil ecosystem, it is not surprising that soil amendments or type of cover crop, including cultivar, can influence microflora composition, and that this can influence expression of soil-borne diseases. Modification of the soil microbial composition by biological means, is likely to be a more sustainable control method than chemical sterilisation.

# <u>Cultural control</u>

Cultural practices that can affect susceptibility to root pathogens and replant disease include fertility levels, cultivation practices (probably through degree of soil compaction), pH and soil moisture, but perhaps the most effective and control strategy of all is avoidance, through rotation or planting position.

# Research recommendations

This literature review has highlighted the need for further research into replant diseases of rose and ornamental rosaceous trees. The recommendations for further research work are summarised below:

#### <u>Cause</u>

- 1. Improve understanding of the causes of rose replant disease to enable the identification and development of reliable control methods other than soil disinfestation.
- 2. Explore the potential of modern molecular methods based on DNA extraction, analysis and quantification for investigating the cause of rose replant disease.
- 3. Determine the specificity of replant diseases in rose and ornamental rosaceous trees, including apple.

#### <u>Soil test</u>

4. Investigate the possibility and benefits of developing a rose seedling bioassay to determine before planting whether a particular soil would lead to rose replant disease, and the probable degree of symptoms.

# <u>Control</u>

- 5. Determine the efficacy of biological methods, including mycorrhizae (eg Root-Grow), Agrobacterium radiobacter, cover crops, biological amendments and the use of specific pre-plant crops (eg wheat), in overcoming replant diseases in rose and ornamental rosaceous trees.
- 6. Determine the effect of mono-ammonium phosphate and some other nutrients (eg nitrogen fertilisers) on replant diseases in rose on non-sterilised soil.
- 7. Discuss with rose rootstock suppliers the potential for using rootstocks and ownroot roses with reduced susceptibility or tolerance to replant disease, if screening showed such to be available and transferable to commercial rootstocks.
- 8. Screen new apple rootstocks (eg CG.30 and CG.6210 from the USA), demonstrated to have increased resistance to replant disease overseas, for their resistance to replant disease in UK soils

- Determine which chemical fumigants and which soil steaming and heating methods provide effective control of replant diseases in rose and ornamental rosaceous trees.
- 10. Determine the soil heating (minimum temperature and duration) required to eliminate replant disease from soil.
- 11. Determine the effect of size of undisturbed root ball at planting on development of replant disease in rose.

It is recommended that priority in experimental work on disease control is given to options that are less expensive to conduct and implement.

# **Financial benefits**

Financial losses from replant problems extend across several horticulture sectors, including hardy nursery stock (especially roses), tree fruit (apples, pears, cherries, plums), soft fruit and the amenity sector (field-grown trees, especially *Prunus*). For example, field-grown *Prunus* trees on affected land may take 7 or 8 years, rather than 4, to reach a marketable size, and even then the value of individual trees may be reduced from £70 to £30 (H Ashardi, Hillier Nurseries, pers. comm.). Further losses result from the extra expense of planting more frequently than necessary, resulting in poor returns on high initial investment costs.

Following consultation, the HTA British Rose Group confirmed that 1% was a reasonable estimate of rose production losses to replant disease. With a 1% loss in marketable output, the direct financial loss is in excess of £240,000 per annum (based on 2004 farm-gate values of £23M for roses; Anon., 2006b). For field-grown ornamental trees, the extent of losses is not well known. Assuming the output is reduced by 1%, the annual financial loss here is £264,000 (based on 2004 farm-gate values of £25.2M for ornamental trees; Anon., 2006b). Industry consultation would be needed to determine a more reliable figure.

As well as unsatisfactory growth and quality losses suffered on the production site, replant disease can adversely affect the rose industry, in particular, through loss of repeat sales when purchasers experience poor growth in gardens and landscaped areas.

# Action points for growers

- Where there is stunted growth of rose and/or rosaceous trees, test for obvious known causes of stunting (e.g. nematodes, soil compaction) before concluding it is a replant problem.
- Rotate rose and rosaceous trees with non-rosaceous species to reduce the risk of replant disease developing in soil.
- For row crops where it is feasible (eg specimen tree rows), record the position of rows before you remove trees and plant the new crop midway between the old row positions.
- Where availability of land is restricted, consider soil disinfestation (eg chloropicrin, steam).
- There is evidence that incorporation of a mycorrhizal product after soil disinfestation will benefit growth of rose and some other trees and shrubs; consult an expert to ensure an appropriate mycorrhizal species is used.
- Use of mycorrhizae to overcome replant diseases has recently been widely promoted (e.g. Root Grow); anecdotal evidence suggests good efficacy.
- A range of measures to overcome replant diseases in home gardens or amenity situations have been recommended by the horticultural industry, including removal and replacement of soil and incorporation of peat, organic matter, green manures or mycorrhizae.
- Use of trickle irrigation has proved beneficial in some cases of apple replant disease.

# **SCIENCE SECTION**

#### Introduction

#### Background

Replant disease occurs when a susceptible crop is planted on non-sterilised land, usually on land that has previously grown that crop or a related species. Plants fail to grow satisfactorily even though recommended cultural practices are followed. The problem results in stunted growth, leading to inefficient use of land, labour and machinery. It is a particular problem in some species of the Rosaceae family.

Rose replant disease is well known to horticulturalists and there are many anecdotal reports about the nature of the disease and possible control measures. The problem affects both the rose producer and the retailer. Occurrence tends to be patchy and unpredictable. In home gardens, and especially for climbing roses where prime positions may be limited (e.g. adjacent to entrances), the only solution to overcome replant disease may be to replace the soil. However, the prospect of digging a large hole to replace the soil is not a practical option for many people and consequently a species other than rose may be planted. There are other sites (e.g. crematoria) where rose replant disease is a recurring problem and, for obvious reasons, the soil cannot be replaced. Both the above situations reduce retail demand for roses.

Discussion with growers on replant diseases in rose and trees triggered this proposal. Subsequent guidance from the HDC was that the scope should include the whole Rosaceae family, to allow information to be gathered from a wide range of plant species that are botanically related to rose and other ornamental species grown in the UK.

The Rosaceae is a very large family of plants, including over 100 genera and over 3000 species. Important genera include Cotoneaster, Crataegus, Fragaria, Malus, Potentilla, Prunus, Pyracantha, Rosa, Rubus, Sorbus and Spiraea. Species in several of these genera are known to be affected by replant disease. Most research attention has been given to orchard replant diseases, which have variable and complex causes, involving fungal pathogens, parasitic nematodes and possibly root exudates from the previous crop.

#### Scope and objectives

This project presents a summary science review of replant diseases of ornamental rosaceous crops (roses and rosaceous trees), drawing information from scientific literature, trade press, growers, consultants and internet searches. Information has been interpreted to identify key findings and the relevance of these to the industry. We have made recommendations for future research and development, and summarised current control options.

The specific objectives of the work presented here were:

- 1. deliver a science review of replant diseases/disorders affecting the Rosaceae;
- 2. provide interpretation to identify implications for UK ornamental species;
- make research recommendations with reference to rose and ornamental trees; and
- 4. provide a summary of current options for control of replant disease for growers of ornamental rosaceous species.

#### Methods and sources of information

Information was drawn from MAFF/Defra/HDC reports, scientific literature, trade press and internet searches and from discussion with growers and consultants. Information was interpreted to identify key findings and the relevance of these to the industry, using expertise from within the project group. We have made recommendations for future applied or strategic research and development.

Three types of approach were taken to deliver the science review.

1. <u>Literature search.</u> This covered the scientific and technical knowledge relating to replant diseases and disorders of the Rosaceae family.

The outcome of previous studies has been summarised, rather than a comprehensive description of the work undertaken.

- 2. <u>Analysis.</u> The information gathered was drawn together and expert analysis was undertaken by the project group to identify key findings and the relevance of these to the industry. Areas for future research and development are recommended based on current knowledge and a rational approach.
- 3. <u>Consultation with identified experts.</u> In the course of this work, the following experts have been approached for additional information and/or expert opinion.
  - John Adlam, Dove Associates
  - Peter Hingley, Certis
  - Andrew Tinsley, HDC
  - RHS Pathology Staff
  - Wadia Kandula (lincoln.ac.nz)
  - Royal National Rose Society
  - HTA British Rose Group (BRG), formerly the British Rose Growers Association (BRGA)
- 4. <u>Questionnaire</u>. A questionnaire (see Appendix 1) was sent to members of the British Rose Group through the Horticultural Trades Association (HTA), and to rose growers through the HDC.

# Literature review

#### Terminology and definition

Replant disease has been given many definitions, names and acronyms (Table 1), reflecting the complex and diverse nature of the problem.

Table 1.Some names and acronyms given to replant diseases or problems. There<br/>are many others that include a crop name other than apple: e.g. almond<br/>replant disease.

Name	Acronym	Example reference
Apple replant disease	ARD	Utkhede and Smith (1994)
Apple replant problem	ARP	Utkhede and Smith (1994)
Non-specific replant disease		Traquair (1984)
Orchard replant problem		Traquair (1984)
Poor growth disease	PGD	Anon. (1986)
Replant disease		Utkhede (2006)

Replant problem		Utkhede (2006)
Soil replant problem	SRP	Pitacco et al. (1994)
Soil sickness		Anon. (2006a)
Specific apple replant disease	SARD	Wittenmayer and Szabó (2000)
Specific replant disease	SRD	Savory (1966)

Replant diseases have been recognised for centuries (e.g. Worlidge, 1698, cited in Savory, 1966). Historically, replant diseases have been classified as either specific (i.e. species-specific) or non-specific.

- Specific replant diseases can be defined as being persistent, specific, not spreading in the soil, and not causing shoot symptoms other than a reduction in growth (Savory, 1966).
- Non-specific replant diseases are similar, but affect more than one species (Traquair, 1984).

However, more recently (especially during the last 30 years) the species-specificity of replant diseases classified as specific has been increasingly questioned. Sewell (1979) considered that the term specific replant disease was unjustified and a possible barrier to identification of the cause. There is conflicting evidence on this subject, but there is abundant evidence that some replant diseases are more specific than others.

In both apple and hardy ornamentals nursery stock, replant disease is not confined to replanting situations (Anon., 1986). The term poor growth disease (PGD) was suggested as a more appropriate name for this reason, but it is rarely used.

In this review we address replant diseases of the Rosaceae, defined as poor growth and performance of plants in non-sterilised soil, usually where the same or related species has been grown previously, and where there is no clear cause such as a recognised root disease, vascular wilt, nematode damage to roots or soil compaction.

# Species affected

Species most commonly encountered during literature searches for this review were almond, apple, asparagus, cherry, citrus species, grapevine, ornamental rose and pear. The Royal Horticultural Society (Anon., 2006a) list the following as frequently 12 of 47 © 2007 Horticultural Development Council affected, but state that only insufficient data has prevented the addition of many other species: apple, cherry (edible), cherry (flowering) on *Prunus avium* rootstock, citrus, peach, pear, plum, quince and rose on *Rosa canina* rootstock: all of these except citrus are members of the Rosaceae. *Sorbus* and *Malus* species are generally the most severely affected (N Dunn, pers. comm.).

Apple (*Malus* × *domestica* Borkh), because of its economic importance, is the species in which replant problems have been most studied. Apple replant disease is widespread and occurs in all the major production regions of the world (Traquair, 1984). Most experimental investigations have used plant species producing edible crops, but replant problems have been studied in a wide range of plant species. Indeed, most cultivated plants perform less well when replanted compared with the preceding crop, and the benefits of a crop rotation are adequate evidence for this.

#### Symptoms and diagnosis

There are no visible symptoms above ground other than a reduction in growth (Savory, 1966), stunting and a lack of vigour (Anon., 2006a). These are common features of replant diseases in different hosts. The most severe growth retardation usually occurs in the first two years, after which there is often a gradual recovery, but affected plants do not catch up with unaffected plants (Savory, 1966). The yield of top fruit crops is reduced, particularly during the early years after planting (Berrie, 1987). Rowan (Sorbus aucuparia) is reported to show particularly severe growth retardation, but even here replant disease per se does not cause plant death (N Dunn, pers. comm.).

Below ground, roots are usually small, dark and compact (Anon., 2006a) and fine roots often show signs of decay (Savory, 1966). Roots of apple trees affected by replant disease had extensive sloughing away of epidermal and cortical layers (Caruso *et al.*, 1989).

For apple, a soil test to identify the likelihood of a SARD problem was previously offered by ADAS. The growth of apple seedlings over a three month period in pots of untreated and sterilised orchard soil was compared. The disease was considered to be present when there was a 50% or more increase in growth of seedlings in the sterilised soil compared with the unsterilised (Anon., 1983; Sewell, 1992). We found no other reports of soil tests to identify those with a replant problem.

# Causes

#### **Introduction**

Taking an overview of diseases of crop plant species causing poor growth, often the cause of the problem is known: an example is club root (*Plasmodiophora brassicae*) of brassica species (Savory, 1966). These growth problems that have a known cause are not usually referred to as replant diseases. However, there are many replant diseases for which the cause is less clear and these include the replant diseases or problems that affect members of the Rosaceae, which are the subjects of this review.

We emphasise here that the causes of replant diseases in the Rosaceae are not fully understood (Anon., 2006a; Spethmann and Otto, 2003). Causes could be simple (one dominating cause) or complex (interacting causes). Utkhede (2006) concluded that "soil sickness is a very complex phenomenon and research is needed to sort out various factors responsible before lasting methods of control can be developed". Because of the complex and varied nature of replant diseases, studies at different sites do not necessarily investigate precisely the same disease, leading to variation in reported causes.

The effective control of replant diseases by soil sterilisation (see section below on management and control) is strong evidence that the principal causes are biotic. However, abiotic factors have also been studied and may interact with biotic factors, or be secondary or aggravating factors. The main reported factors that are possible causes are considered under separate sub-headings below.

#### <u>Fungi</u>

There are many reports of fungi being the main cause of replant diseases in rosaceous species. In a recent review paper, Utkhede (2006) provides a concise summary of the main fungi reported as causal agents. Causal fungi identified were from three taxonomic groups: *Oomycetes*, *Hyphomycetes* and *Basidiomycetes*. Fungal species or groups reported to cause replant diseases are many, and important ones are listed in Table 2 with example references. We found no primary publications of work to determine the cause(s) of replant disease in rose.

**Table 2.**Fungi reported to cause replant disease in rosaceous species.

Fungal species or genus	Host plant(s)	Example reference	

Armillaria mellea	Apple	Sutton et al. (1981)
Cylindrocarpon spp.	Apple	Braun (1995)
Fusarium spp.	Peach	Utkhede (2006)
Mortierella spp.	Apple	Utkhede et al. (1992)
Peniophora sacrata	Apple	Taylor and Wallace (1970)
Penicillium claviforme	Apple	Čatská et al. (1988)
Penicillium janthinellum	Apple	Utkhede and Li (1988)
Phytophthora cactorum	Apple	Mazzola (1998)
Pythium sylvaticum	Apple	Anon. (1983)
Pythium spp.	Apple	Mazzola (1998)
Rhizoctonia solani	Apple	Mazzola (1997)
Torulomyces lagena	Apple	Utkhede et al. (1992)
Trichoderma hamatum	Apple	Utkhede et al. (1992)

Sewell (1981) argued that *Pythium* spp. are a possible cause of apple replant disease. However, the evidence presented is mainly circumstantial. For example, negative growth responses of seedlings grown in soil inoculated with *Pythium* spp. were of a similar magnitude to growth increases obtained by soil sterilisation in replanted orchards. Several other authors have also demonstrated that *Pythium* spp. are soil pathogens of apple (Mazzola, 2002).

Mazzola (1998) claimed to clearly demonstrate that fungi are the main cause of apple replant disease in Washington State. The studies were systematic and used selective biocides, soil pasteurisation, isolation of microorganisms from roots, and pathogenicity tests (Table 3). It was concluded that a fungal complex caused disease development, and that the main fungi were Cylindrocarpon destructans, *Phytophthora cactorum*, *Pythium* spp. and *Rhizoctonia* spp.

In the UK, results with the soil fumigant dazomet (Basamid) suggest that soil microorganisms other than *Pythium* species contribute to poor growth of apple. Dazomet, which has little effect on actinomycetes and other bacteria, was less effective than heat in controlling the disease in pot tests using a mixture of soils severely affected by SARD (Anon., 1986).

Table 3.	Biomass of 'Gala apple seedlings after 4 weeks of growth in soil amended
	with fungal isolates (from Mazzola, 1998).

Fungal isolates(s)	Plant weight (mg)
Control	678
Cylindrocarpon destructans 8-13-11	441
Fusarium solani D44-1	577
Pythium ultimum A30-2	238
Pythium ultimum A30-2 + F. solani D44-1	320
Pythium ultimum A30-2 + C. destructans 8-13-11	171
Rhizoctonia solani AG 5 5-103	320
Rhizoctonia solani AG 5 5-103 + F. solani D44-1	367
Rhizoctonia solani AG 5 5-103 + C. destructans 8-13-11	268
LSD (P=0.05)	126

Many other studies have demonstrated that soil fungi can cause growth retardation. For example, Braun (1995) showed that *Cylindrocarpon lucidum* and *Pythium irregulare* caused symptoms like replant disease on apple and pear, but not on plum or peach. The fungal species or genera involved vary between studies, host species, geographic regions, and individual orchards.

It should be recognised that the ability of a fungal pathogen to cause growth retardation does not provide proof that the same pathogen is the cause of replant disease, even when the pathogen is isolated from a problem soil.

#### <u>Actinomycetes</u>

Actinomycetes (also known as Actinobacteria) are a group of Gram-positive bacteria that are common in soil and play an important role in decomposition of organic matter. They produce hyphae with a diameter lower than 1  $\mu$ m, are aerobic, and prefer neutral pH (Spethmann and Otto, 2003). Actinomycetes are also sometime known as micromycetes (e.g. Čatská, 1994).

Otto and Winkler (1998) state that "actinomycetes are considered to be the cause of specific replant disease in some species of Rosaceae as they invade the cortex of rootlets by penetrating the epidermal cells". They showed damage to root hairs and cortical cells of rootlets, leading to rootlet death. Szabó et al. (1998) presented evidence (using electron microscopy) for the pathogenicity of actinomycetes in rootlets of apple seedlings, and concluded that these microorganisms may be the pathogens responsible for specific apple replant disease. In support of this hypothesis they showed that many apple root pieces were exclusively infected by actinomycetes, which would not be the case if infection with actinomycetes were secondary. Furthermore, the proportion of rootlets infected with actinomycetes was positively related to the degree of the specific apple replant disease symptoms (i.e. the extent of growth retardation).

Further evidence for actinomycetes as the main causal pathogen for apple replant disease was gained from soil heating experiments in which the minimum temperature needed to remove replant disease from problem soils was compared with the temperature tolerance of groups of microorganisms (Spethmann and Otto, 2003). The temperature tolerance of some strains of actinomycetes isolated from problem soils was very close to the minimum temperature needed to remove the replant disease from the soil.

Otto *et al.* (1994a) demonstrated some degree of specificity in the infectivity of actinomycetes from a soil with a specific apple replant disease problem. Actinomycete infection was found in the rootlets of apple, pear and rowan (*Sorbus aucuparia*), but not in those of cherry, plum and rose. Interestingly, cherry and plum are less severely affected by replant disease. They concluded that the host range of actinomycete apple pathogens is not restricted to *Malus*, but does not include all genera of the Rosaceae.

#### Bacteria (other than actinomycetes)

There have not been many studies to test the possible roles of other bacteria as causes of replant diseases. Just over 40 years ago, in a long and detailed review, Savory (1966) pointed out that there had been little or no attention to bacteria as possible causes, but that bacteria could fulfill all the known requirements of a pathogen causing replant disease. By 1968 Hoestra (1968, cited in Utkhede, 2006) had shown the failure of nematicides and fungicides to control apple replant disease, suggesting a possible role for bacteria.

Two strains of Bacillus subtilis have been shown to decrease growth of apple trees (Utkhede and Smith, 1994), and changes in the composition of other bacteria, the

fluorescent pseudomonad community, have been shown in response to treatments that improved growth of apple in a problem soil (Gu and Mazzola, 2003).

Results of a complex study of effects of nematodes, fungi and bacteria on young apple trees growing in apple replant disease soil (Utkhede, Vrain and Yorston, 1992), showed effects of bacteria alone (strains of *Bacillus subtilis*), or in combination with other factors.

These publications show that soil bacteria can decrease growth of apple, and have potential as causal organisms in replant diseases. However, they do not show that bacteria are the main cause in commercial situations.

#### <u>Viruses</u>

It has frequently been observed that plants affected by replant disease recover when transferred to fresh soil (see Reverse replant section below). This would seem to exclude soil-borne viruses as a cause of the classic and well-described replant diseases of rosaceous species such as apple. Further evidence that viruses are not the cause has come from grafting experiments and the gradual improvement in growth of affected plants over a long period of time (Savory, 1966).

# <u>Nematodes</u>

In the case of roses, as with apples, only soil sterilisation prevents replant disease, and treatments that eliminate nematodes without soil sterilisation do not prevent replant disease (Spethmann and Otto, 2003). This is supported by other work with apple, showing that elimination of nematodes with a nematicide did not improve growth of apple in soils with a specific apple replant disease (Hoestra, 1994). This provides evidence to exclude nematodes as a causal factor in apple.

Despite this, Utkhede (2006) states that there is considerable evidence that nematodes are associated with replant disease in apples, peaches and cherries. Genera and species of nematode mentioned in that review include: *Pratylenchus penetrans*, *Pratylenchus vulnus*, *Xiphinema* spp., *Cricoemella* spp., and *Meloidogyne* spp.

Nematodes would be expected to retard growth of rosaceous species, as is the case with many crop plant species. However, this is a problem that is relatively well understood (compared with replant disease), and is not considered here in detail.

#### Nutritional problems

Reports of effects of nutrients on replant diseases are many, and often conflicting. Many reports have shown that added major and minor nutrients do not improve growth of plants affected by replant disease (see reviews by Savory, 1966; Utkhede and Smith, 1994; Utkhede, 2006). Savory (1996) also reports that a few authors have reported responses of replanted crops to added nutrients, particularly under acid conditions. Clearly, as with soil pH, it is very difficult to interpret results of field or pot experiments with nutrients because of the many interacting affects of the treatments, the difficulty of good experimental control, and the differing conditions between experiments. To repeat individual experiments under the same conditions is usually impossible. Savory (1966) concluded that it would seem most unlikely that a nutrient deficiency or imbalance can be the primary cause of specific replant diseases.

Any nutritional problems are likely to be non-specific, but it is possible that they could interact with other factors, and thus affect the severity of a replant disease. Utkhede and Smith (1991) presented results showing that application of nitrogen to problem soils, with or without phosphate, suppressed the growth of fungi that can cause apple growth retardation (*Penicillium janthinellum* and *Constantinella terrestris*) and promoted the growth of bacteria (*Bacillus subtilis*) antagonistic to those fungi. It has also been shown with a *Prunus* species that repeated applications of nitrogen fertiliser prevented replant disease in a problem soil (Anon., 2006a). In contrast, Sadowski *et al.* (1988) attributed failure of replanted apple orchards to excessive nitrogen fertilisation.

Effects of phosphate, particularly mono-ammonium phosphate, have been more consistent. Mono-ammonium phosphate has been shown to be beneficial to apple plantings in sterilised soil. This is because in soil that has been sterilised, vesicular-arbuscular mycorrhizal infections are eliminated, decreasing phosphate uptake (Gur, Luzzati and Katan, 1998). However, reports by Utkhede and Smith (1993), Gur, Luzzati and Katan (1998), Wilson, Andrews and Nair (2004), and Wojcik and Klamkowski (2005) all showed improved growth of apple in a non-sterilised problem soil, in response to mono-ammonium phosphate fertilisation. Wilson, Andrews and Nair (2004) did leaf analysis for nutrients and concluded that growth responses were not associated with any nutritional effect.

Interestingly, Wojcik and Klamkowski (2005) pointed out that mono-ammonium phosphate can decrease soil pH, and their results showed this effect in the second year of study.

Clearly there have been beneficial effects of nutrients, especially mono-ammonium phosphate. However, the mechanism of this effect is probably very complex, involving changes in soil micro-organism populations, effects of altered pH, effects on availability of other nutrients, and improved uptake of nutrients when mycorrhizal infections are poor.

In summary, it seems likely that responses to nutrients do not indicate a cause of replant disease, but rather are interacting with other factors in a complex soil environment.

#### <u>Soil pH</u>

Effects of soil pH on the occurrence and severity of replant diseases are complicated by the effects of soil pH on the growth of the plant species in question and on nutrient availability. Reports on this subject are generally about apple, which prefers a soil pH of between 5.5 and 6.5 (Jonkers and Hoestra, 1978).

It has been reported that growth responses of apple to soil fumigation increased with increasing soil pH (Savory, 1967; Hoestra, 1968; Sewell, Roberts and Elsey, 1992), but a report by Sewell, Preece and Elsey (1988) did not support this. Pot experiments have shown that apple replant disease was less severe when the soil pH was lowered (Hoestra, 1994), but the pH values are not given in this paper.

In ADAS pre-planting pot tests for SARD, equally good growth of apple seedlings in both sterilised and unsterilsed soil tended to occur where the pH was<6.0, although not all acidic soils gave a low growth response (Anon., 1986).

Jonkers and Hoestra (1978) briefly reviewed effects of pH on specific apple replant disease and concluded that a low pH of the soil (values of 4.0–4.5) prevents specific apple replant disease. However, some reports quoted in this review suggested that the acidity required to prevent specific apple replant disease would be too acidic for favourable growth.

Results of studies such as these are difficult to interpret, because of the multiple effects of changing soil pH (e.g. effects on root physiology, nutrient availability, plant

growth and the microorganism community). However, the general weight of evidence suggests soil pH influences replant disease severity in at least some cases. It is known that soil pH influences soil microorganism ecology: for example, actinomycetes prefer a neutral pH (Spethmann and Otto, 2003). Since there is strong evidence that replant diseases have a biotic cause, effects of soil pH are to be expected.

#### Soil structure

Soil compaction can be defined as a reduction in soil pore volume and structure, and an increase in soil bulk density. At a small scale this can be seen as a reduction in the size and number of macropores and a general change in the shape and continuity of pores (Soane *et al.*, 1981). Compaction leads to reductions in hydraulic conductivity, permeability and diffusivity of water and air through the pore system: excess water cannot drain away. Root growth is restricted, decreasing uptake of nutrients and water, and thus canopy growth and yield are impaired. Poor soil aeration can lead to de-nitrification (loss of nitrogen to the atmosphere).

Soils are more prone to compaction if soil structural stability is poor, or if they are excessively wet (beyond the plastic limit). Generally the worst compaction occurs on sands and silts because of their poor structural stability. Heavier soils are more stable when dry but become less stable when wet.

Traffic of machinery through orchards may cause compaction between the rows, and further compaction could be caused during grubbing, but this would usually be removed during preparation for replanting. Compaction could also occur during any period of arable cropping before replanting.

Savory (1966) considered that compaction of orchard soils was not a serious problem, is easily diagnosed, and is quite non-specific in action. However, Traquair (1984) states that adverse effects of poor soil structure can be a factor in the development of non-specific replant problems. These authors agree that any replant problem caused by poor soil structure would not be expected to show any species-specificity, and the cause of the problem can be easily investigated.

The most important aspect of soil structure as a factor influencing replant diseases is the possible interaction with other causes, such as effects of poor drainage on disease development in the presence of soil pathogens.

#### **Phytotoxins**

Many studies have investigated possible toxic substances that could be produced during decomposition of previous crop or plant residues, especially roots. Utkhede and Smith (1994) briefly reviewed abiotic causes of replant problems and stated that phytotoxins have been shown to be the primary cause of peach replant problem. The toxins, produced during decomposition of peach roots were hydrocyanic acid, benzaldehyde, condensed tannins, prunasin, and other compounds. However, Savory (1966), in a more detailed (but earlier) review argued against a causal role of phytotoxins in replant diseases: the toxin would need to be highly species-specific in action, be resistant to microbial decomposition for several years, retard growth in the presence of root residues. It was concluded that phytotoxins are an unlikely cause of replant disease.

Many reports are cited by Savory (1966), Traquair (1984) and Utkhede and Smith (1994), showing either production of phytotoxins in orchard soils, and/or effects of phytotoxins that can be produced in orchard soils. However, the presence of phytotoxic compounds, or effects of such compounds on plant growth are not sufficient evidence to conclude that these are the cause of replant diseases. For example, Adamska and Politycka (2001) demonstrated that toxic phenolic compounds are released in soil when apple roots are decomposing. However, the phenolic compounds decreased in the soil over a two month period, indicating that they would be unlikely to persist for between 5 and 20 years, the range of persistence reported for replant diseases (Savory, 1966).

#### **Reverse replant**

The term 'reverse replant' is used here to mean the reversal (or decreased severity) of replant disease symptoms when a plant with replant disease symptoms is replanted in soil in which the same species has not previously been grown.

This phenomenon has been shown for apple and cherry (Savory, 1966), but we have not found reports for other species.

Savory (1966) points out some important implications of the reversibility of replant diseases, including the following:

• the causal factor must be soil based;

 either the causal factor is not transferred to the new site with the roots, or it fails to establish in the new environment, or it must be present in the soil before planting to cause replant disease.

The benefits of replanting apple trees affected by a replant disease in soil sterilised by fumigation with chloropicrin, were studied by Ross *et al.* (1984). They concluded that the benefit of improved growth after replanting into sterilised soil (compared with growth after replanting into non-fumigated soil) was offset by the shock to the tree of lifting and replanting. After 4 years, trees replanted into fumigated soil had not grown faster than undisturbed trees. This work was done in Nova Scotia, in a soil of pH 5.8, which is more acidic than the pH preferred by actinomycetes. No indication was given of the likely cause of the replant disease on the experiment site.

Spethmann and Otto (2003) state that for replant disease in roses "there is no evidence that transplanting roses from a 'sick' soil to a virgin soil will improve the symptoms". However, The Royal Horticultural Society (Anon., 2006a) referring generally to replant diseases, advise that it is a characteristic of replant diseases that vigour can be restored by transferring an affected plant to fresh soil.

#### Physiological aspects

Plant diseases involve the action of causal factors and a response by the plant. It is not surprising that there has been little reporting of physiological responses of plants to factors causing replant diseases, because neither the causal factors, nor replant diseases, are well defined. In many cases, replant diseases may have only moderate symptoms and be difficult to identify in the absence of unaffected plants. However, the recognition of genetic resistance to replant diseases (see section above on genetic resistance) suggests that there are physiological responses to causal factors, and that these vary between clones (e.g. different rootstocks).

An important physiological aspect of replant diseases is the possible role of endogenous plant growth regulators (PGRs) in the infection of roots by pathogens. The long soil-persistence of apple replant disease led to the hypotheses that propagules can be induced to germinate by particular plant species, and that compounds exuded from rootlets are involved in this effect (Otto *et al.*, 1994b). It was shown that infection by actinomycetes could be increased by seedling decapitation, and by application to the shoot of a synthetic auxin, a synthetic cytokinin or gibberellins (Otto *et al.*, 1994b). However, the same authors point out

that other woody plants also exude PGRs through their roots, but do not suffer from the same replant disease. Wittenmayer and Szabó (2000) used radioactively labeled PGRs to quantify roots exudation of these, and found no difference between susceptible and non-susceptible plants, and detected no substance that occurred exclusively in seedlings susceptible to replant disease.

There is a relationship between infection by actinomycetes and both metabolic activity of the host plant and application of some PGRs to the shoot (Spethmann and Otto, 2003), but the signaling process has not been identified.

#### Management and control

#### Introduction

Management options, for rose replant disease, include avoidance (plant roses somewhere else, or plant something else), soil sterilisation, soil replacement, use of cover crops, and use of beneficial soil fungi.

Soil sterilisation, more correctly referred to as soil disinfestation, is falling out of favour with retailers and the most effective product available, methyl bromide, is no longer available for use in the UK for pre-plant use. This increases opportunities for new control methods.

The main established and novel methods for management and control of replant diseases are considered under separate sub-headings below.

#### Soil disinfestation

Soil disinfestation either with a broad-spectrum biocide, or by steaming, has been the most popular and effective control strategy for replant disease (Traquair, 1984; Spethmann and Otto, 2003; Anon., 2006a; Utkhede, 2006). This effective control by soil disinfestation is strong evidence that the principal causes are biotic.

Chemicals used for replant disease control have included methyl bromide, formaldehyde (formalin), chloropicrin, and products that release methyl isothiocyanate (e.g. metam-sodium, dazomet) (Table 4 and see reviews by: Savory, 1966; Traquair, 1984; Utkhede, 2006). Some of the chemicals given in Table 4 have also been used successfully in combinations: e.g. Browne *et al* (2006) used combinations of 1,3-dichloropropene and chloropicrin, and iodomethane and chloropicrin.

Chemicals	Plant species in which replant diseases were controlled	References
Calcium cyanamide	Apple (Malus x domestica)	Wilson, 2005
Chloropicrin	Apple (Malus × domestica) Almond (Prunus dulcis)	Oehl, 1979 Browne et al., 2006
Dazomet	Apple (Malus × domestica) Apple (Malus × domestica)	Neilsen and Yorston, 1991 Otto and Winkler, 1993
1,3-dichloropropene	Almond (Prunus dulcis)	Browne et al., 2006
Formaldehyde (formalin)	Apple (Malus x domestica)	Neilsen and Yorston, 1991
	Peach (Prunus persica)	Bingye and Shengrui, 1998
lodomethane	Almond (Prunus dulcis)	Browne et al., 2006
Metam-sodium	Apple (Malus × domestica) Apple (Malus × domestica)	Otto and Winkler, 1993 Smith, 1994
Methyl bromide	Apple (Malus x domestica)	Smith, 1994
	Almond (Prunus dulcis)	Browne et al., 2006

Table 4.Chemicals used effectively as broad-spectrum biocides for control of<br/>replant diseases, in example studies reported in research papers.

Current soil disinfestations options for rose and field-grown trees are shown in Table 5 and Table 6. Key characteristics of chemicals available in the UK for soil disinfestation are given in Appendix 2.

Active ingredient	Product(s)	
Chloropicrin	Chloropicrin Fumigant; K & S Chlorofume	
Dazomet	Basamid	
Formaldehyde	Formalin	
Metam-sodium	Discovery 510, Fumethan, Metam 510, Metham Sodium 400, Sistan, Sistan 38, Vapam	
1,3-Dichloropropene	Telone II	

Methyl bromide, is no longer available for use in the UK for pre-plant use.

Formalin is a chemical treatment that has been widely investigated for control of replant disease in top fruit and nursery stock, but was never widely adopted as a treatment in the UK. However, it now appears likely that formalin may become unavailable in the EU in the near future due to failure of suppliers to support reregistration of this active ingredient under Regulation 91/414 as either a Biocide or a Pesticide (V Powell, HDC; pers. comm.).

Chloropicrin is reported to be the most consistent treatment for control of SARD in the UK (Berrie, 1987). Treatment can only be applied by a certificated contractor and it is obligatory to cover the soil with sheets of low gas permeability for at least 4 days to retain the vapour. Currently it is not widely used on apple replanting sites; treatments other than soil disinfestation, such as placing peat in the planting hole and trickle irrigation, have proved beneficial in some situations.

There are many reports of studies that have shown benefits of soil disinfestation methods. For example, Smith (1994) showed that both methyl bromide (applied at 454 g per tree site by probe, or across the site at 450 to 675 kg/ha) and metamsodium (applied at 1120 L/ha) greatly improved long-term performance of apple, cherry and pear trees; dazomet improved long-term performance of apple and pear trees in France (P Hingley, pers. comm.); chloropicrin improved performance of ornamental rosaceous trees (N Dunn, pers. comm.).

Savory (1966) reports variability in the efficacy of sterilants (e.g. methyl bromide), and indicates that different application rates of chloropicrin are needed on different soil types. It is to be expected that benefits of soil disinfestation will vary between sites because replant diseases are very variable in severity, and soil conditions and application methods would be expected to affect efficacy.

Soil disinfestation has dramatic effects on the ecology of soil microorganisms (Savory, 1966; Traquair, 1984). Of particular interest is that mycorrhizal fungi are destroyed (Traquair, 1984) and this has led to studies of effects of soil sterilisation together with application of nutrients (e.g. Neilsen and Yorston, 1991; Sewell *et al.*, 1992). In an examination of the results of 506 soil bioassays using pot-grown apple seedlings, Sewell *et al.* (1992) reported that addition of phosphorus (to compensate for eradication of mycorrhizae) increased economically significant growth responses to chloropicrin fumigation from 39% to 68%.

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Although soil disinfestation kills mycorrhizae, the treatment is only partial, even if very thorough, because it is effective to a limited depth. Thus, some microorganisms are left to recolonise the soil and roots, and mycorrhizae are usually re-introduced on the planting material. Fumigation of soil has been shown to increase root colonisation with mycorrhizae in some cases (D.R.W. Kandula, pers. comm.), but not others (e.g. Kandula *et al.*, 2006).

In summary, it is well established that soil disinfestation is a good control method for replant diseases of rosaceous plants.

# Steaming and other forms of soil heating

Steaming has not been used widely as a means of controlling replant diseases, most probably due to the high cost of applying steam on a field scale. Moyls and Hocking (1994) showed 120% growth improvement in apple after 2 minutes steaming. With the loss of methyl bromide however, an increasing range of soil steaming and heating methods are becoming available (Table 6). It would be useful to know the minimum temperature and duration required to eliminate a replant disease from soil. Reduced-cost treatment methods warrant investigation for their effect on replant diseases. Table 6.Soil steaming and other forms of soil heat treatment available in the UK or<br/>under development.

Method	Comment
Sheet steaming	Widely used in UK glasshouse cut flower production
Steam plough	Occasionally used in UK glasshouse crop production
Vacuum steaming (negative pressure steaming)	Occasionally used in UK glasshouse crop production
Self-propelled plate steamer	Available in the UK; tested for weed control in field crop production (leafy salads). Shallow treatment only.
Short-duration, low temperature steaming	A prototype machine has been developed (van Loenen et al., 2003)
Direct soil heating	Soil removed, heated in a rotating drum, and beds re-laid (UK Sterilizers Ltd)
Crop debris burners	Used in UK glasshouse crop production. Disinfests the soil surface only.
Radio frequency and microwave heating	A prototype machine (`Agritron`) has been developed in the Netherlands for glasshouse crop production
Hot air	A hot-air steriliser (800°C) that is used with a soil spading machine; for field or protected crops. Reported to be used in the Netherlands (`Cultivit`) and Israel.

#### <u>Fungicides</u>

Reported effects of fungicides on replant diseases of the Rosaceae are not consistent, reflecting the probable variation in causes between sites and species. Some reviews have not included consideration of fungicides as an effective control option. For example, Spethmann and Otto (2003) state that effective control is only possible with soil fumigation, and Utkhede (2006) has not included fungicides in a section on chemical control.

However, some studies show benefits of fungicide use in soils affected by replant diseases. Mazzola (1998) used problem soils collected from orchards in Washington State, USA, and reported that difenconazole or metalaxyl enhanced growth of apple seedlings in all five soils tested, and fludioxinil enhanced growth in the two soils tested. Meanwhile, in the same study, benomyl improved apple growth only in soil from one orchard, in two out of three experiments. Szczygiel and Zepp (1998) also tested growth of apple in problem soils in pot experiments, and found that the effectiveness of two fungicides (Aliette and Captan, full product details not given) varied according to the experiment, and were less effective than additions of biohumus, mono-ammonium phosphate, peat or decomposed bark. Both fungicides were effective in one out of three experiments.

These variable results are not surprising since fungicides usually have a narrow spectrum of activity, and there is no clear evidence that any one fungus is the cause of a replant disease. For example, Aliette and metalaxyl are known to provide some control of *Phytophthora* and *Pythium* spp., two of the fungal groups reported as possible causes of apple replant diseases. Mazzola *et al.* (2002) showed that metalaxyl sensitivity varied among species of *Pythium*.

#### Genetic resistance

Clear evidence has been presented by Leinfelder and Marwin (2006), of genetic variability in susceptibility of apple to a replant disease. In an experiment on an old apple orchard site in New York State, USA, rootstocks were a more important factor affecting expression of replant disease than fumigation with Telone C-17 (1,3-dichloropropene, 78% v/v, and chloropicrin, 17% v/v) or planting position. Rootstocks CG.6210 and CG.30 were more resistant than G.16, M.26 and M.7. This study supported earlier work by Isutsa and Merwin (2000), which showed differences in tolerance of rootstocks to apple replant disease. This work also used soil from New York orchards. They concluded that use of resistant materials in rootstock breeding programmes could provide new management options for replant diseases of apple.

In the UK, apple cultivars Cox's Orange Pippin and Golden Delicious are reported to be much affected by the disease, while Bramley's Seedling is less affected and pears are not appreciably affected. The rootstocks M9 and MM106 are particularly susceptible, while less dwarfing rootstocks such as MM11 have the vigour to overcome low levels of SARD but may be affected by high levels (Anon., 1983). M26 also has partial resistance to replant disease (N. Dunn, pers comm.).

Browne et al. (2006) also found variation in susceptibility to a replant disease between rootstocks of almond (*Prunus dulcis*).

Given that a number of different commercial rootstocks are available for grafting roses, and that other roses are grown on their own roots, the variation in susceptibility 29 of 47 © 2007 Horticultural Development Council among roses warrants investigation. The predominant rootstock currently used in UK rose production is *Rosa dumetorum* 'Laxa'. However small quantities of *R. canina* selections such as 'Inermis' are sometimes used. HDC trials (HNS 6a, 1990–1993; Burgess, 1995) showed that *R. canina* 'Inermis' and 'Uniform' were more vigorous and could give a better grade-out than *Rosa* 'Laxa', and were also much less susceptible to rust (*Phragmidium mucronatum*). However, they produced more suckers than 'Laxa' which was considered a distinct disadvantage both for growers and gardeners, and this is why 'Laxa' remains the most popular rootstock. Some *R. multiflora* rootstock selections are also used for budding some rose types where incompatibility problems arise on 'Laxa' stocks.

#### **Biological control**

There are three main types of biological control of replant diseases that have been investigated for rosaceous species:

- 1. inoculation with mycorrhizae,
- 2. inoculation with bacteria,
- 3. soil amendment with organic materials, including incorporation of cover crops.

Some examples of mycorrhizal fungi and bacteria that have been studied as biological control agents of apple replant diseases are given in Table 7.

Vesicular-arbuscular mycorrhizal fungi form a symbiotic association with roots of most plants. The plants benefit from improved uptake of nutrients, especially those with low soil mobility such as phosphorus. Inoculation of sterilised soil with mycorrhizae has already been discussed (see "Soil sterilisation (disinfestation)" above). Mycorrhizae have also been studied as possible biological control agents for replant diseases in unsterilised soil. Apple grown in a replant disease soil has a lower level of colonisation with vesicular-arbuscular mycorrhizae than in soil not affected by replant disease (Caruso et al., 1989; Čatská and Taube-Baab, 1994; D.R.W. Kandula, pers. comm.).

In a study with pot-grown apple seedlings, inoculation of a problem soil with the mycorrhizal fungus *Glomus mosseae* significantly increased growth (Utkhede *et al.,* 1992), but the mechanism of this response was unclear. Other studies on mycorrhizae (outside the scope of this review) have shown that mycorrhizal associations can protect against fungal root diseases by improving phosphorus uptake (discussed in Utkhede *et al.,* 1992).

Taube-Baab and Baltruschat (1993) worked with five *Glomus* isolates, including *G. mosseae*, and found improved growth of young apple trees after inoculation with *G. intraradices* and *G. spec.* D13, but not after inoculation with *G. mosseae*. Total shoot length was greater than that for plants grown in sterilized soil (by steaming), indicating that the mycorrhizal fungi provided a benefit that more than out-weighed the negative effect of the replant disease. Čatská (1994) also showed that apple replant disease was suppressed by inoculation of apple-tree seedlings with *G. fasciculatum* and *G. macrocarpum*.

Table 7.	Examples of fungi and bacteria that have been shown to have activity as	
	biological control agents for apple replant diseases.	

Fungi or bacteria	References
Mycorrhizal fungi	
Glomus mosseae	Utkhede et al., 1992
Glomus intraradices	Taube-Baab and Baltruschat (1993); Utkhede and Smith, 2000
Glomus spec. D13	Taube-Baab and Baltruschat (1993)
Glomus fasciculatum	Čatská (1994)
Glomus macrocarpum	Čatská (1994)
<u>Bacteria</u>	
Agrobacterium radiobacter	Čatská and Hudská, 1993; Čatská and Taube-Baab, 1994
Fluorescent-putidia type Pseudomonas strains	Biró et al., 1998
Bacillus subtilis	Utkhede, 1999; Utkhede and Smith, 1994; Utkhede and Smith, 2000
Enterobacter agglomerans	Utkhede and Smith, 2000

Clearly, inoculation with vesicular-arbuscular mycorrhizal fungi can improve growth of apple in problem soils, although the effects of individual fungal species are variable, probably because of differences between sites/soils in the precise and complex cause of the replant problems.

A range of mycorrhizal products are available and some are being used on rose (e.g. Root Grow) with the aim of overcoming replant diseases (Chambers, 2005).

Inoculation with certain bacteria has also been shown to improve growth in plants affected by replant diseases (Table 7). Čatská and Hudská (1993) suggested that inoculation with Agrobacterium radiobacter may affect growth of apple in a problem soil by changing the composition of the rhizosphere microflora, and decreasing the population of phytotoxic fungi (e.g. *Penicillium claviforme*), for which *in vitro* inhibition was demonstrated. Apple seedling shoot dry weight was increased by 33% compared with uninoculated, but there was no comparison with seedlings not affected by the replant disease.

Biró *et al.* (1998) presented evidence that fluorescent-putidia type *Pseudomonas* strains can "remove" apple replant disease, but concluded that the effect was dependent on soil and environmental conditions, as effectiveness varied between trials.

Bacillus subtilis can promote growth of apple trees in a problem soil, when applied as a root-dip treatment at the time of planting (Utkhede, 1999), or as a soil drench (Utkhede and Smith, 1994). As with studies of other biological treatments, it was not clear whether this effect was through growth promotion or control of pathogens.

Biological control of replant diseases has been demonstrated by soil amendment with organic materials and by growing cover crops before replanting.

Gu and Mazzola (2003) grew apple seedlings in apple orchard soils in pots, after three previous 28-day growth cycles with ryegrass or one of 11 wheat cultivars. At the end of each cycle the shoots were cut and discarded and the soil in each pot was mixed to prepare a seedbed. Some wheat cultivar treatments, but not others or ryegrass, enhanced subsequent growth of the apple seedlings. Five cultivars consistently improved growth compared with growth in untreated soils, and two cultivars did not improve growth. It was shown that wheat cultivars which enhanced seedling growth modified the genetic and species composition of the fluorescent pseudomonad populations in the soils. These populations (from soils cropped with wheat cultivars that enhanced apple seedling growth) inhibited *in vitro* growth of a fungal complex that had previously been shown to cause disease development in Washington State (Mazzola, 1998) more than populations from soils cropped with other wheat cultivars. The main fungi were *Cylindrocarpon destructans*, *Phytophthora cactorum*, *Pythium* spp. and *Rhizoctonia* spp. (see earlier section on fungi). Application of wheat root exudates also modified the composition of fluorescent pseudomonad populations in a wheat-cultivar-specific manner. This work has shown that plant cultivar, as well as species, is important when evaluating benefits of cover crops for control of soil-borne fungal diseases.

Marigold (Tagetes patula cv. Harmony) has been shown to decrease Pythium spp. populations on sites in the Winfield-Oyama area of Canada, where apple or pear trees had been recently removed (Edwards *et al.*, 1994). Mazzola and Mullinix (2005), working in Washington State, USA, showed that benefits of wheat cover crops or *Brassica napus* green manure crops were inferior to soil sterilization with methyl bromide. Soil amendment with *B. napus* seed meal had variable results and results were influenced by nematode (*Pratylenchus* spp.) infestation.

Gur *et al.* (1998) suggested that the effectiveness of soil additives, including activated charcoal and compost, may have been caused by the absorption of ethylene and other harmful compounds excreted by microorganisms causing the replant disease. These treatments did decrease ethylene concentration in soil and roots, but there was no clear link between ethylene and disease expression.

Within a complex soil ecosystem, it is not surprising that soil amendments or type of cover crop can influence microflora composition, and that this can influence expression of soil-borne diseases. Modification of the soil microbial composition by biological means, is likely to be a more sustainable control method than chemical sterilisation.

## Other cultural practices – nutrients, irrigation, rotation

Cultural practices that can affect susceptibility to root pathogens and replant disease include fertility levels, cultivation practices (probably through degree of soil compaction), pH and soil moisture (Traquair, 1984), but perhaps the most effective and control strategy of all is avoidance, through rotation or planting position.

In an experiment on an old orchard site in New York State, USA, Leinfelder and Marwin (2006) found that trees planted in the old grass lanes performed better than those planted in the old tree rows. There are also many anecdotal reports of spatial patterns in the performance of trees in replanted orchards, where the pattern relates to planting positions of the old trees (John Adlam, pers. comm.). Such reports support the widely-accepted view that the causal factors are not very mobile in the soil (Savory, 1966).

There are many reports that mono-ammonium phosphate (MAP) application (either alone or in combination with another treatment such as a biological control agent) improves growth of apple in a problem soil. Phosphorus is not very mobile in the soil, so plants are susceptible to sub-optimal phosphorus nutrition when root growth is impaired.

Effects of MAP application have already been considered in the earlier section of this report on causes, and in the subsection on nutritional problems. Responses to nutrients may help as a practical treatment for plants affected by a replant disease, but do not indicate a cause of replant disease.

Examples of reports that show positive effects of MAP application include Gur *et al.* (1998), Szczygiel and Zepp (1998), Utkhede and Smith (1993), Utkhede and Smith (2000), Wilson (2005); Wilson *et al.* (2004) and Wojcik and Klamkowski (2005). However, Utkhede (1998) found that MAP reduced growth but not yield, and attributed the discrepancy with other results to soil infestation with *Phytophthora cactorum*.

Wilson *et al.* (2004), found that MAP can be toxic because of high concentration of salt (measured by electrical conductivity) above 2 g of MAP per litre of soil. Wojcik and Klamkowski (2005) supported this, and concluded that apples given MAP applications must be watered to avoid osmotic stress.

It has also been shown with a *Prunus* species that repeated applications of nitrogen fertiliser prevented replant disease in a problem soil (Anon., 2006a). However, Szczygiel and Zepp (1998) applied "NPK" fertiliser (15 g ammonium nitrate, 0.45 g triple superphosphate and 0.22 g potassium sulphate per litre of soil) to pot-grown apple seedlings in soil from orchards, and found that this treatment was ineffective. In the same study, magnesium limestone application (2 g or 4 g per litre of soil) was also ineffective.

Utkhede (1998) found no benefit of nitrogen application to apple trees in soil infested with *Phytophthora cactorum*. This study also showed that irrigation practice affected tree performance, with a benefit of a soil-drying period between irrigation events.

Although it has been shown that a low pH of the soil prevents specific apple replant disease the pH required would be too low for favourable growth (Jonkers and

Hoestra, 1978). Thus, there is no scope to use soil pH modification as a replant disease control method. To control common scab of potatoes (caused by an actinomycete), some growers use sulphur to modify the soil pH in tuber zone, which is localised compared with the extent of the root system. To attempt control of a replant disease by this method would be impractical because pH modification would be required throughout the root zone.

Trickle irrigation was reported to be beneficial in promoting good growth and cropping of apple on SARD-affected soil (Anon. 1883).

## Implications for roses and ornamental rosaceous trees

This review is based predominantly on studies of apple replant disease. No primary publications that investigated replant diseases of rose or ornamental rosaceous trees were found. Hence it is necessary to draw conclusions for rose and ornamental rosaceous trees based on other species. The following implications for rose and ornamental rosaceous trees are proposed:

- 1. Known causes of stunted growth (e.g. soil compaction, nematodes) should be excluded before poor growth in a crop is ascribed to a replant disease.
- 2. Nematodes can be a problem in rose (Spethmann and Otto, 2003), but they are not the cause of rose replant disease under the definition of the term used in this review. If roses or ornamental rosaceous trees show stunted growth, the roots should be checked for nematode infestation, and species involved identified to determine their role in the problem.
- 3. Observations suggest that the symptoms and characteristics of replant disease in rose are similar to those for apple. However, it is not clear if reverse replant occurs with rose (i.e. if affected plants grow normally when transferred to fresh soil).
- 4. There is some evidence that soils that result in replant disease in apple also result in replant disease in rose (Spethamann and Otto, 2003).
- 5. There is good evidence of variation in susceptibility to specific replant disease among different apple rootstocks. Given that a number of different commercial rootstocks are available for grafting roses, and that other roses are grown on their own roots, the variation in susceptibility among roses warrants investigation. The predominant rootstock currently used in UK rose production is *Rosa dumetorum* 35 of 47

'Laxa'. However small quantities of *R. canina* selections such as 'Inermis' are sometimes used.

- 6. It will be essential to improve understanding of the causes of rose replant disease to enable the identification and development of reliable control methods other than soil disinfestation.
- 7. Best options for future control methods that are more sustainable than soil disinfestation are biological control and breeding to utilise genetic resistance.

# Current industry view on replant disease in rose

A total of 23 completed questionnaires were received from rose growers (see Appendix 1). The main conclusions were:

- 1. Most growers think rose replant disease is a significant problem either in production or as a knock-on effect from reduced sales.
- Crop rotation is a very important strategy for most growers, and generally sufficient new land is available, although some rose growers are now having difficulty finding fresh land. (This will decrease and is already a problem for some ornamental tree growers; moving staff long distances is costly and impacts on the environment).
- 3. Soil disinfestation, mycorrhizae or green manures are rarely used in field scale production. (Some ornamental tree growers already use soil disinfestation and green manures each year on a field scale).
- 4. Most respondents reported that replant disease is a very significant problem for customers and gardeners, with a negative effect on potential sales. Customers frequently ask for advice on how to manage replant disease.
- 5. Soil replacement is very widely recommended for amateurs, as is using wellrotted compost or manure. Until recent label changes that restrict its field of use, soil disinfestation with Armillatox was also recommended.
- 6. Recently the use of mycorrhizae (mainly RootGrow) has been very widely recommended and promoted, and anecdotal evidence supports good efficacy.

7. Any future research work should include testing RootGrow and other mycorrhizal products more thoroughly, and the active organisms be identified.

# Current options for rose and field-grown trees

- 1. Use clean land (rotate crops).
- 2. Disinfest the soil with a chemical treatment (Table 5).
- 3. In the longer term biological options may have the greatest applicability. These include:
  - mycorrhizae (eg Root-Grow)
  - growing and incorporating specific cover crops,
  - biological amendments (e.g. manures, micro-organisms such as Agrobacterium radiobacter), and
  - devising rotations that use specific varieties of crops shown to reduce replant diseases (e.g. wheat for SARD in Washington, USA).

# **Research recommendations**

## <u>Cause</u>

- Improve understanding of the causes of rose replant disease to enable the identification and development of reliable control methods other than soil disinfestation. The relative importance of fungi and actinomycetes as causes of replant diseases in rose and ornamental rosaceous trees should be determined following the methods used in apple, including fungicides with specific activity (see Spethmann and Otto, 2003; Mazzola, 1998).
- 2. The potential of modern molecular methods based on DNA extraction, analysis and quantification for investigating the cause of rose replant disease should be explored. For example, the use of real-time PCR (polymerase chain reaction) to quantify specific organisms (Suarez *et al.*, 2005), or T-RFLP (terminal restriction fragment length polymorphism) to quantify the main fungal and bacterial groups associated with roots of affected and unaffected plants. The latter technique has recently been used to analyse Actinomycete populations on roots (Conn

and Franco, 2004) and to assess fungal diversity of microbial communities (Lord et al., 2002).

3. Determine the specificity of replant disease in rose and ornamental rosaceous trees.

## <u>Soil test</u>

4. Investigate the possibility and benefits of developing a rose seedling bioassay (either with susceptible rootstock seedlings or ex-micropropagated roses on their own roots), similar to the SARD apple seedling bioassay, to determine preplanting if a particular soil would lead to rose replant disease, and the probable degree of symptoms.

## <u>Control</u>

- 5. Determine the efficacy of biological methods (see Current Options, above) in overcoming replant diseases in rose and ornamental rosaceous trees.
- 6. Determine the effect of mono-ammonium phosphate and some other fertilisers (eg nitrogen) on replant disease in rose grown on unsterilised land.
- 7. Discuss with rose rootstock suppliers the potential for using rootstocks and ownroot roses with reduced susceptibility or tolerance to replant disease, if screening showed such to be available and transferable to commercial rootstocks.
- 8. Screen new apple rootstocks (eg CG.30 and CG.6210 from the USA), demonstrated to have increased resistance to replant disease overseas, for their resistance to replant disease in UK soils
- Determine which chemical fumigants and which soil steaming and heating methods provide effective control of replant diseases in rose and ornamental rosaceous trees.
- 10. Determine the soil heating (minimum temperature and duration) required to eliminate replant disease from soil.
- 11. Determine the effect of size of undisturbed root ball at planting (into a soil affected by replant disease) on development of replant disease in rose (eg bare root plants compared with container-grown plants with different sized root balls).

12. Write a Factsheet on replant diseases of rose and ornamental rosaceous trees suitable for both growers and the home-garden and amenity sectors.

# Acknowledgements

We are grateful to all who completed the questionnaire on rose replant disease.

# Technology transfer

- Project final report (the review)
- Article in HDC news, summarising key findings with implications for the UK industry (in press)
- Presentation at HTA British Rose Group Seminar, Cambridge, 29 November 2007.

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# Appendix 1. Rose replant disease – grower survey summary results

## Response

A total of 13 replies received from the version distributed by HTA.

A total of 12 replies received from the version distributed by HDC.

## 1. How important is rose replant disease to you as a grower?

General response was: very important, but two said 'little impact' or 'learned to live with it'. A British Association of Rose Breeders (BARB) officer mentioned seeing lots of poor roses attributed to replanting on same land, however rotation appears to be used by most growers. One grower buys in bare root roses for potting, so ownproduction section not relevant.

## 2. Have you experienced poor vigour / establishment attributable to replant?

Yes 6 No 13 Again, some commented problem not seen because they rotate crops.

## 3. Do you need to rent land?

Most people rent land, though 3 said they did not.

## 4. Is availability of fresh land a problem?

Yes 4 No 15. Clearly most producers currently have access to fresh land.

## 5. Do you use soil sterilisation or other measures?

Yes 0 No 17. Obviously soil sterilisation etc. rarely practiced for production – again probably because most rotate. One reply mentioned they steamed 20 yrs ago under glass.

## 6. Rotations used?

All those that answered this question said Yes 14

## 7. Interval before re-cropping with roses?

This varied (no. of responses in brackets if more than 1)

3 yrs	4 yrs	4 – 5 yrs	4 – 7 yrs	5 yrs (5)	5 - 8 yrs

6 – 8 yrs 7 – 8 yrs 10 yrs >10 yrs (2) 12 yrs

or a mixture (1 yr break with barley, then 5 yr break with grass).

#### 8. What crops used for rotation (only asked on HDC form)?

Following mentioned: sheep grazing; pigs; grass; veg / arable; cereals; arable crops; wheat/grass; mainly sugarbeet; considering mustard.

#### 9. Is there a 'knock-on' effect on sales because of perceived replant problem?

Yes 16 No 3 One grower warns customers of potential problem first.

## 10. If Yes, Score significance 1 – 5 (5 high) (only asked on HDC form)

5 responses score 5; 2 response score 3; 1 response score 1.

#### 11. Do Customers ask advice about managing replant?

Yes 17 No 1

#### 12. What control measures recommended?

Number of respondents mentioning:

Soil replacement 15				
Armillatox	10			
Rootgrow	8			
Mycorrhizae (not specific product)		6		
Addition of well rotted compost / manure				
Jeyes Fluid	2			
Break crop / rotation		3		
Plant container roses (good root system) 1				
Plant in cardboard box with new soil				

#### 13. Some specific comments on recommendations to customers:

- Leave 1 yr, double dig & add rotted manure.
- Sterilise with Jeyes Fluid, or dig out old soil to 18", spray Jeyes Fluid, put new soil on top.
- Armillatox at 2%.
- Withdrawal of Armillatox means replacing soil necessary.
- Used to recommend Armillatox but had to stop due to EU regulations. Now recommend Rootgrow or soil change.
- Leave bed for 1 or 2 years; add lots of organic matter, replace soil 2 ft x 2 ft.
- Clear out old roses; give good dose lime, use break crop (e.g. dahlias).
- Lots specifying soil changing, Rootgrow, Armillatox.

## Other general comments or information:

- Industry needs a solution.
- Not one customer who used Rootgrow has returned and said it doesn't work.
- No one has come back to say Rootgrow doesn't work.
- Rootgrow successfully trialled at Whittington on old beds.
- Local nursery dipped ornamentals in Rootgrow before potting for container sales and achieved twice the growth!
- Rootgrow is very effective.
- One garden reckoned his roses had become 'over-vigorous' since using Rootgrow!

- Has recommended Armillatox for at least 5 years, but in last 2 yrs recommends
  Rootgrow no negative feedback so far.
- Important to use Rootgrow in combination with well rotted manure / compost.
- Awaiting HDC recommendation before using Rootgrow.
- Hampton Court Palace and Kew Gardens likely to be using Rootgrow.
- Mycorrhizae probably need addition of OM to be effective, but only a tentative observation.
- I've yet to find a decent chemical that works.
- Reckon it's important to clear whole bed of old roses replanting odd plants less effective due to disease pressure from nearby old roses.
- Has used MeBr effectively once a 100 yr old rose garden. Replanted 400 roses and only 2 suffered any problems.
- Considering soil sterilisation as availability of new land a problem for me.
- Occasionally suggested customers try Tagetes to cleanse soil as used by Narcissus trade on Scilly Isles.
- Nursery recommendation sheet soil replacement if roses been there for > 7 years, or alternatively use Armillatox (dil 1:40) recommending 35–45 cm deep holes at 15-20 cm intervals to aid penetration. 1 litre diluted Armillatox for 3 m<sup>2</sup>. Leave 2 3 weeks before planting the bed.

## Overall conclusions from Grower Questionnaire:

- 1 Most growers think rose replant disease is a significant problem either in production or knock-on effect for customers / sales.
- 2 Crop rotation a very important strategy for most growers. Generally sufficient new land is available.

- 3 Soil sterilisation or other chemical treatment, mycorrhizae or green manures rarely used in field-scale production.
- 4 Most think replant disease is a very significant problem for customers / gardeners with a negative effect on potential sales. Customers frequently ask for advice on how to manage replant disease.
- 5 Soil replacement is very widely recommended for amateurs, also using well rotted compost / manure. Also soil sterilisation using mainly Armillatox.
- 6 Recently use of mycorrhizae (mainly Rootgrow product) has been very widely recommended and promoted, and anecdotal evidence supports good efficacy.
- 7 Any future research work should certainly include trialling Rootgrow &/or other mycorrhizae product more thoroughly (with identification of active organisms involved).
- 8 Armillatox is now marketed as a Soap Based Outdoor Cleaner and does not have pesticide registration status. It has no label recommendation for use as a disinfectant for rose replant or other diseases, and so cannot now be legally recommended by nurseries or garden centres for this purpose.

# Appendix 2. Key characteristics of chemicals available for soil disinfestation

	Chloropicrin	Dazomet	Metam-sodium	1-3-dichloropropene	Formaldehyde
Approval status	LTAEU* from full approval (Outdoor use only)	Full approval (Outdoor and protected use)	Full approval (Outdoor and protected use)	LTAEU from full-label approval (Outdoor use only)	Commodity substance approval (Outdoor and protected use) Due to be revoked.
Rate of use	150-400 L/ha	220-570 kg/ha (outdoor) 220-760 kg/ha (protected)	400-1000 L/ha	225 L/ha	0.5 L/m <sup>2</sup> diluted with 2.5 L water
Physical form	Liquid	Granules	Liquid	Liquid	Liquid
Preferred temperature for effective treatment	Above 10°C	Above 7°C	Above 10°C	Above 5°C	Effective above 0°C
Application method	Injected into soil applied in water or through drip tape irrigation. Can only be applied by contractor	Best applied using specialist applicator	Injected into soil using specialist applicator	Injected into soil. Can only be applied by contractor	Soil drench. Terragator for field scale use
Requirement for polythene cover after treatment	Must be sealed with sheets of low gas permeability for at least 4 days.	Polythene cover preferred, but surface can be sealed by smearing or frequent watering	Polythene cover preferred, but surface can be sealed by smearing or frequent watering	Polythene cover preferred, but surface can be sealed by smearing	No cover required
Preferred Interval required prior to planting	At least 14-20 days (depending on product), but a cress test is recommended	Usually 6 weeks, but a cress test is recommended	Usually 7 weeks, but a cress test is recommended	Usually 6 weeks, but a cress test is recommended	At least 4 weeks
Spectrum of activity	Good fungicide. High doses needed for good weed and nematode control	Good fungicide. Controls many soil pests, nematodes and weeds	Good control of nematodes, weeds and fungal diseases at higher doses	Mainly nematicidal but some evidence that it controls verticillium wilt	Good fungicide and general biocide. Limited effect against weeds and nematodes
Human toxicity	Highly toxic. A chemical subject to	Harmful in contact with skin and if swallowed. Irritating to eyes skin and	Irritating to eyes, skin and respiratory system	Toxic if swallowed. Harmful in contact with skin and by inhalation. Irritating to eyes,	Toxic if swallowed. Harmful in contact with skin or by

			the Poisons Law	respiratory system		skin and respiratory system	inhalation
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\*LTAEU Long Term Arrangements for Extension of Use