PLANT RESEARCH INTERNATIONAL

Effects of heating and light exposure on the emergence, germination, viability and dormancy of weed seeds

Report of an inventory study concerning microwave energy, oven heating, hot water treatments and steaming

M.M. Riemens

Note 250

WAGENINGEN UR

PLANT RESEARCH INTERNATIONAL

Effects of heating and light exposure on the emergence, germination, viability and dormancy of weed seeds

Report of an inventory study concerning microwave energy, oven heating, hot water treatments and steaming

M.M. Riemens

Plant Research International B.V., Wageningen
July 2003

Note 250

WAGENINGEN UR

Plant Research International B.V.

Address : Droevendaalsesteeg 1, Wageningen, The Netherlands
: P.O. Box 16, 6700 AA Wageningen, The Netherlands
Tel. : +31 317 47 70 00
Fax : +31 317 41 80 94
E-mail : postkamer.pri@wur.nl
Internet : http://www.plant.wageningen-ur.nl

Table of contents

Summary

This note reports the results of an inventory study about different control methods and their influence on the viability, dormancy, germination and emergence of weed seeds. The inventory was made as a guideline for research aiming at the development of innovative weed control methods for organic intra-row weed control in Dutch agriculture. Using the available techniques there is still a notable amount of weed left in the row that has to be removed by hand. Bottlenecks concerning labour are mainly caused by the need to remove these weeds, especially in slowly growing crops. Focus in this study was on the weed seeds since the weed seed bank dynamics play a major role in regulating weed communities of agricultural land.

Heating in an oven, microwave energy, hot- water treatments, influencing the amount of light and steaming were the methods regarded. Furthermore, the effects of soil sterilisation on the microbial soil population were considered, since these organisms regulate biological and chemical properties of the soil. These properties can influence crop growth, both positively and negatively.

Temperatures needed for inhibition of germination and loss of viability mainly depend on the water content of the seeds and weed species. In general, temperatures ranging from 60-80 °C are necessary for a strong inhibition of seed germination and reduction of viability of imbibed seeds for most weed species. Dry seeds on the other hand need higher temperatures before germination is reduced and can even withstand heat exceeding 100 °C before their viability is reduced.

Reaching these temperatures is possible with microwave energy, although the effectiveness of this method is influenced by weed species, initial soil temperature, treatment duration, soil moisture, water content of the seeds, the power level and the microwave apparatus used.

Hot water treatments don't seem to be an option for the inhibition of weed emergence or germination, since these methods mostly enhance the germination, viability and emergence of the seeds, but may be used to enhance those factors when applying a false seedbed.

Although research is lacking on the effects of steaming on weed seeds, it appears to be a good method to reduce seedling emergence. Temperatures in the soil are thought to be high enough to reduce the viability, germination and emergence of seeds, but more information is needed on the effects on individual weed species, for instance *Chenopodium album* and *Stellaria media* before the method can be applied in practice. The susceptibility of micro-organisms to heating appears to be, in decreasing order, weed seeds, nematodes, fungi and bacteria. When applying one of the methods described in this paper, one should consider the possible effects on micro-organisms, since these can be important for crop growth or damage.

Most important mechanisms influencing the breaking or induction of dormancy of weed seeds conclude temperature, depth of burial and light. The flash of light during tillage is thought to be important for the breaking of dormancy of buried seeds, if temperatures are favourable for germination. However, a lot of weed species can also germinate without receiving light, although to a lower extent than in the light. Hence, methods to reduce or eliminate the amount of light received by the seeds during tillage may be a way to lower seedling emergence, but will not control weeds completely.

It can be stated that the methods described in this report have biological possibilities for organic intra-row weed control, alone or combined and can be taken into account during the development of innovative weed control methods in organic farming in the Netherlands.

1. Introduction and aim

Organic farming is regarded as a good answer to the shortcomings of the common agricultural practices. Shortcomings such as animal welfare, presence of pesticide-residues in food and environment, the loss of nature and landscape in agricultural areas and the mineral residues in air and water. A small, but growing amount of consumers has little faith in the way and rate the common agricultural sector will be able to tackle these shortcomings. Therefore, the Dutch government aspires a market share of 10% of the total agricultural sector in 2010 for organic farming. Achievement of these aspirations not only depends on the demand for organically grown products, but also on the physical and socio-economic infrastructure. Another important factor is the availability of required knowledge for manufacturers.

In the past, organically producing farmers were interviewed and asked to mention the biggest bottleneck for arable farming. Among others, they mentioned the weed control; although precautionary measures involving weed control are made, the amount of labour needed for intra-row weeding remains high, especially in crops such as carrot, onion and sugar beet.

The LNV-weed research programme 'Innovative weed control in favour of sustainable (organic) farming and public space' (397-V) aims at developing new insights to improve existing weed control systems and innovative methods in aid of weed prevention, non-chemical weed control and risk control fitting in the organic farming system.

As a part of this programme an analysis of the control of weed seeds was made and possibilities of innovative control methods with respect to the seed bank were regarded. Focus was on the weed seeds since the weed seed bank dynamics play a major role in regulating weed communities of agricultural land. Aim of the present study was to obtain new insights in weed control methods and their way of application in Dutch agriculture.

This report shows the results of a literature survey regarding different control methods and their influence on several properties of weed seeds. The effects of heating in an oven, microwave treatments, hot water treatments, steaming and light during tillage on the viability, germination, dormancy and seedling emergence of weed seeds in soil and possible side-effects on micro-organisms in soil are evaluated.

The overview of experiments described in literature in this report should be regarded as background information for the development of innovative weed control methods for intra-row weeding in favour of sustainable organic farming.

2. Effects of heating on weed seeds

2.1 Temperatures needed for loss of viability and inhibition of germination

Early work on effects of heating on survival of weed seeds representing seven species and five families, showed that seeds subjected to heat treatments for 15 min in sealed brass tubes immersed in an oil bath required temperatures varying from 85 to 100 °C for complete lethality (Hopkins, 1936). Since then several studies to determine the threshold temperature for germination (Egley, 1990; Horowitz & Taylorson, 1984; Thompson *et al.*, 1997) and viability (Egley, 1990; Horowitz & Taylorson, 1984; Moss, 1980) have been performed. The most important results will be mentioned in this chapter.

Regarding the seed survival and germination in both moist and dry heated soils, Egley (1990) exposed seeds of several weed species to temperatures of 40, 50, 60 and 70 °C in dry (2.1%) and moist soils (19.2%). Treatment durations were 0, 0.25, 0.5, 1, 2, 3, 5 and 7 days in an oven.

Exposure to continuous 40 or 50 °C for up to 7 days in dry soil did not reduce viable seeds of any species. Treatments at 60 °C for up to 7 days only slightly decreased seed survival of velvetleaf, spurred anoda and pitted morningglory. Seed survival of all species was progressively reduced during the 7-day period at 70 °C with the reduction varying with the species (Appendix I, Table 2).

Seed germination in response to 40, 50, 60 and 70 °C in dry soil varied with treatment time and species. Germination was progressively enhanced with time up to 70 °C for 6 days. This enhancement was attributed to the breaking of dormancy of hard seeds by heat because punctures through the coats of ungerminated seeds produced germination equal to the maximum viability of the seeds. Treatments at 70 °C for 6 days or longer decreased the germination because of the reduction of seed viability by heat. Soil temperatures of 40 °C for up to 7 days did not reduce the seed viability in moist soil. Responses to higher temperatures varied among species, but increasingly higher temperatures and longer treatment reduced the number of surviving seeds (Appendix I, Table 1).

Some relatively short heat treatments (<2 days) at 40 and 50 °C in moist soil increased seed germination, but longer treatments at higher temperatures decreased germination, apparently because of heat effects on seed viability. There was little or no germination after 2-5 days at 60 °C (depending on the species) or after any treatment at 70 °C. Most of the seeds in moist soil had imbibed water from the surrounding soil. Their moisture content was over 30 %, which made them probably more susceptible to high temperatures. Seeds in dry soil either had not imbibed or were only partially imbibed and were less susceptible to the higher temperatures.

Thompson *et al.* (1997) also conducted experiments with several weed species to determine the effect of heating in an oven on the germination of imbibed seeds. However, they discriminated between soil and oven temperatures and used shorter treatment durations than Egley (1990). The oven temperatures ranged from 102 to 262 °C and treatment durations from 0.5 to 10 min (Appendix I, Table 3). The average temperature of the soil, over the 10 min heating required to prevent over 90% germination, varied among species and ranged from 48 °C for *Avena fatua* to 65 °C for *Rumex obtusifolius* (oventemperatures ranged from 102 °C to 204 °C, respectively). The germination percentage of the weed species following treatments characterised by maximum and mean soil temperatures are shown in Appendix 1, Table 4.

Upon the experimental work that has been reported the threshold temperature required to prevent germination appears to be dependent on the weed species, moisture level of the seeds and the

treatment duration. Nevertheless, it can be stated that in general, temperatures of 60- 80 °C are necessary for a strong inhibition of seed germination and reduction of viability of imbibed seeds of most weed species. Dry seeds on the otherhand need higher temperatures before germination is reduced and can even withstand heat exceeding 100 °C before their viability is reduced.

2.2 Microwave energy

Irradiation with radiofrequency energy (RF), which includes microwaves, brings about dielectric heating of moist materials. Dielectric heating is the heating of electrically nonconducting materials, such as water and lipids, by conversion of electromagnetic energy into heat. Microwave frequencies are part of the RF portion, loosely consisting of frequencies above about 1 GHz.

Treatment of seeds of many plant species with RF (mostly at 39 MHz) increases permeability of seed coats without scarafication by abrasion, thereby increasing germination of seeds without diminishing their viability (Nelson *et al.*, 1984). It might be possible to use RF at 39 MHz to bring about the germination of the weed seeds prior to sowing or planting the crop. This germination prior to crop establishment would make mechanically weeding possible and thereby reduce the hours of hand weeding necessary to remove weeds in the row.

There are, however, certain species dependent threshold temperatures, above which germination declines, and temperatures above which thermal death occurs (Couture & Sutton, 1980). Therefore, weed control might be possible if soils containing weed seeds could be heated sufficiently to inhibit germination and/or emergence or cause the loss of viability of seeds. There are four frequencies in accordance with international agreements allowed, knowing 915; 2,450; 5,800 and 24,125 MHz. Although heating of the material is in proportion with the frequency, most of the papers that describe microwave experiments use a frequency of 2,450 MHz, since this is the highest frequency for which microwaves and other economically available equipment is made (Diprose *et al.*, 1984).

In a laboratory study, Barker & Craker (1991) evaluated the effectiveness of microwave heating of soils containing weed seeds on subsequent seedling emergence after conditions for germination had been met and maintained. Approximately 800 g of soil of variable wetness (10, 70, 150 and 280 g H_2O/kg) containing indigenous weed seeds was heated in a microwave (2,450 MHz) oven for 15 to 240s. The soil masses were 5 cm deep. The soil contained populations of galinsoga (*Galinsoga parviflora* Gav.), crabgrass (*Digitaria* sp L.), purslane (*Portulaca oleracea* L.), nutsedge (*Cyperus esculentus* L.), chickweed (*Stellaria media* [L.] Cyrillo), field pepperweed (*Lepidium campestre* [L.] R.Br.), Lambsquarters (*Chenopodium album* L.) and horseweed (*Conyza canadensis* [L] Cronq.). A temperature of 80 °C maintained for 30 sec. was needed to inhibit the emergence of seeds. It took about 120 s of heating to reach 80 °C for all soils. Heating was almost linear to 120 s, after which temperatures remained constant between 75 to 85 °C. Soils with 70 to 280 g H_2O/kg were not heated further by 240 s of irradiation, whereas air-dry soil (10 g H_2O/kg) was raised from 75 to 100 °C. They noted that shallowly planted seeds were not affected as much by microwaves as seeds placed at least 2 cm deep, data and experimental set up were unfortunately not shown. This could be due to cooling near the surface of the soil that prevented sufficient heating. Water content of the soil did not seem to effect the emergence of the weeds. Although more energy was probably absorbed in the wetter soils, the increased rate of heating of the soil with higher moisture contents was absent due to a larger amount of energy required to heat the larger masses of water (Barker & Craker, 1991).

However, when seeds have imbibed water from the soil surrounding them, their viability and thereby their germination percentage can be reduced after microwave treatment. Shafer and Smith (1974) for instance, treated dry and imbibed seeds of foxtail millet (*Setaria italica*) with 1 kW of 2.450 MHz radiation. Dry seeds had an LD_{50} of 30 minutes, and 4 and 16 hour imbibed seeds had LD_{50}'s of 35 and 25 seconds, respectively.

Experiments in the field, in which the energy levels needed to provide an efficient control of weed seeds were determined, gave different results.
Wayland *et al.* performed experiments in 1973 with seeds of wheat (*Triticum aestivum*) and radish (*Raphanus sativus*) and experiments in 1975 with wheat (*Triticum aestivum*), barley (*Hordeum vulgare* L.), oats (*Avena byzantina* K. Koch), mustard (*Brassica kirta* Moench), radish (*Raphanus sativus*) and turnip (*Brassica napus* L.). They determined the LD_{50} and germination percentage of the seeds after treatment in 1973 and the emergence of the weeds in 1975.

In 1973 seeds were buried after 10 hours of imbibition 2.5 cm deep in sandy loam soil of 6.8% moisture content. Microwaves were applied to the samples from a 1500 W (maximum) source at 2,450 MHz via a 10.2 cm^2, 5 cm deep, stainless steel radiator. The pre-treatment soil temperature was 25 °C. Wheat seeds were more susceptible (LD_{50} was reached at 100 J/cm^2) than radish seeds (LD_{50} at 180 J/cm^2). Germination was reduced to 15% for wheat and 40 % for radish after treatments of 210 J/cm^2 (Wayland *et al.*, 1973).

They continued their field experiments in 1975 with 2,450 MHz microwaves generated by a mobile microwave power unit consisting of four separate 1.5- kW magnetrons. A gasoline-engine-driven 60-Hz generator supplied the power. The microwave apparatus was drawn through the plots to give treatments of 35 to 325 J/cm^2. To provide 80 to 90% inhibition of emergence, energy- densities of 183 J/cm^2 were required at power levels of 4000 W (Wayland & Merkle, 1975). It was concluded that, for the power levels used, energy density and time of exposure were interchangeable, with respect to effectiveness, as long as total energy remained about the same.

Menges and Wayland (1974) determined the emergence of weeds after treatment with 2,450 MHz microwaves in the field. Seeds of several weed species were planted in the top 2 cm of irrigated and non-irrigated soils and treated with energy densities of 45 to730J/cm^2. In irrigated soils, all weed species except common purslane (*Portulaca oleracea* L.) had 96 to 100% control with 180 J/cm^2. Common purslane averaged 81 % at this energy level. Weeds were insufficiently controlled by 90 J/cm^2 and lower energy levels. Common purslane has small seeds that primarily germinate in the surface layers of the soil and may not have imbibed sufficient quantities of water and may have been resistant. The soil temperature was 80 °C 1 minute after irradiation with energy levels of 180 J/cm^2.

All weed species were controlled (100%) by 360 J/cm^2 on dry soil. Soil temperature after treatment increased with increasing energy levels and decreasing soil moisture and depth (Menges & Wayland, 1974).

According to these results an energy level of at least 180 J/cm^2 is required to provide an efficient inhibition of emergence of weed seeds of most species in moist soils. This level needs to be higher (up to 360 J/cm^2) on dry soils due to the enhanced resistance of non- imbibed seeds to microwave treatments. The germination inhibition in moist soils requires 210 J/cm^2 and viability can be reduced by 50 % at 100 J/cm^2 in these soils, both depending on the weed species.

Results from research performed in the field indicate that humidity of the soil and initial soil temperature are also critical for a low-cost use of microwaves for soil desinfestation. A soil with an initial temperature of 20 °C and a moisture content of 5.5 % requires 11 kWh/m^2 (= 3960J/cm^2) to increase the soil temperature to 61 °C, 54% less than a soil with the same initial temperature and a higher moisture content (15.5%). For soils with the same moisture contents (15% w.b.), but different initial temperatures (20 and 40 °C, respectively) colder soils require 14 kWh/m^2 (=5040J/cm^2) (42% less) (Mavrogianopoulos *et al.*, 2000).

The efficiency (from electricity to radiation) of a 2,450 MHz microwave varies from 30-60%, depending on its construction. The transmitted energy is converted into heat in the plant and soil. The efficiency of this process is about 40-60%. Both processes together (from electricity to radiation into

heat) have an efficiency of ± 20%. If microwave energy is to be used on the field, a mobile installation is needed. The electricity will than be supplied by for instance a diesel oil engine with a generator. The efficiency of the conversion of diesel oil into electricity is about 30%. The total efficiency from diesel oil to heat in plant and soil is than only 6%. The costs of a microwave generator are about Euro 1,150-1,800/kW (Vermeulen *et al.*, 2002).

So, regarding the results from these experiments described in literature, the practical use of irradiation to inhibit the weed seed viability, germination and emergence depends on a number of factors:

1. Weed species
2. Initial Soil temperature
3. Duration of heating
4. Soil- moisture
5. Moisture-content of seeds
6. Irradiation frequency
7. Power level
8. Microwave apparatus

2.3 Hot water treatments & steaming

Most studies on the effect of hot water treatments on weeds, such as steaming, focus on the control of aboveground plant parts, on both hard surface areas as in the field, or the germination improvement of seeds and not the inhibition of germination of seeds. Only few studies have been performed regarding the control of belowground plant parts.

To determine the effect of a hot-water treatment on germination-reduction of seeds, studies have been performed in the laboratory with velvetleaf (*Abutilon theophrasti*), *Commelina benghalensis* L and apple of Peru (*Nicandra physalodes* L).

Velvetleaf seeds were imbibed in a hot water bath of 40, 43, 49 and 52 °C up to 8h and 43, 45, 47 and 49 °C up to 5h. Significantly reduced germination was observed after 7h at 43 °C, 4h at 45 °C, 2h at 50 °C and 1h at 55 °C. Germination was completely inhibited after 6h at 55 °C, 8h at 50 °C and after 6h at 52 °C or 7 h at 49 °C (Horowitz & Taylorson, 1983).

In another study, *C. benghalensis* L. seeds were soaked in 70 °C hot water for 1 min. and their germination (germination percentage of 70%) was stimulated. This germination percentage was lowered to 10 % when treatment duration was elongated to 4 min. Soaking at this duration was apparently long enough to damage the imbibed seeds and subsequently reduce the germination (Kim *et al.*, 1990).

Soaking of *N. physalodes* L. seeds in preheated water at 50 °C for 10 minutes and incubation in the light at 25 °C did not affect the germination, but the same treatment followed by incubation in the dark instead of light significantly reduced the germination (Watanabe *et al.*, 2002).

The ability of hot water baths to inhibit seed germination appears to be depending on species, treatment duration and the availability and requirement of light after treatment. Besides, it has been reported that soaking seeds in water baths at temperatures ranging from 40 to 70 °C enhances germination of most plant species (Chacko & Chandrasekhara Pillai, 1997; Horowitz & Taylorson, 1984). Hence, the application of hot water it self seems not a good option for weed seed control.

A laboratory study on the effect of steaming on the emergence of weed seeds has been performed by Melander *et al.* (2002). They steamed soil in a 7x8 cm circular groove made in a wooden wheel with insulation in the bottom and at the sides. Soil was steamed by a timed flow of steam by four steam generators with a total effect of 8 kW. They measured the soil temperature while steaming and a short period after steaming. Different weed species were present in the soil samples; *Brassica napus*, *Lolium perenne*, *Capsella bursa-pastoris*, *Chenopodium album*, *Tripleurospermum inodorum*, *Polygonum* spp. and *Lolium perenne*. The maximum temperature at which no seedlings emerged any longer differed per species. In general at a maximum soil temperature of 80 °C, the amount of emerging seedlings was for all species tested nil, both after chilling and no chilling of the soil following treatment. It took approximately 100 sec. to reach a temperature of 80 °C and after steaming had been stopped the temperature dropped with 1 °C per 60 sec. This slow temperature decrease probably correlates with the good isolation of the soil and the temperature in the laboratory (Melander *et al.*, 2002; Melander *et al.*, march 2002).

In greenhouses steaming is used to control soil borne pathogens such as fungi and nematodes and a lot of research has been done to determine the effect of steaming on these organisms. The effects of steaming on weed seeds were never the first priority and data were not published in scientific journals. Nevertheless, some research groups have noted the effect of steaming in greenhouses on weed seeds and mentioned them in a report. In those experiments with several Dutch greenhouse soils, weeds were never observed after treatment at 70 °C or higher for 30 minutes. Specific attention was paid to *Senecio vulgaris, Poa annua* and *Stellaria media* and these weeds were found to need soil temperatures of 54, 58 and 51 °C, respectively during steaming for 30 minutes to prevent emergence (anonymous, 1992).

This temperature was also maintained for one hour in an experiment in 1988 in which weeds had to be removed from the field prior to a sugar beet experiment. However, the sugar beets were not able to grow in the field, probably due to negative effects on the soils chemical properties (Groeneveld, pers. comm.).

Few studies have been done on the effects of steam on weeds under field conditions. Field applications on aboveground plant parts have been studied in cropland weeds by Kolberg & Wiles (2002) and in forests by Norberg & Dolling (2003). Although weed species and energy doses (890 kJ/m^2; 3,200 kg H_2O/ha and 11,494 kJ/m^2; 12,300 kg H_2O/ha, respectively) differed in both studies and aboveground plant parts were removed, the vegetation always recovered after a certain amount of time. This recovery following steam treatment is probably linked to exposure time in that longer exposure times result in slower recovery (Kolberg & Wiles, 2002; Norberg & Dolling, 2003).

Soil temperatures in these studies during and after steaming are probably not high enough to affect the germination of the seeds in both studies. According to Kolberg steam at a temperature of 175 °C prior to application even stimulated the germination of other seeds than the ones they were focussing on (data not shown). Norberg & Dolling recorded the soil temperatures after treatment and found that a steam temperature of 100 °C, a treatment duration of 60 s and an initial soil temperature of 10 °C, can give soil temperatures of maximally 82 °C at a soil depth of 5 cm. This temperature is probably high enough to kill seeds of certain weed species, when it can be maintained long enough and moisture content of the seeds is high. However, soil temperatures rapidly decrease in the field after treatment and halvation of soil temperatures 5 minutes after treatment are not an exception (Norberg & Dolling, 2003). This is in contradiction with the result obtained by Melander *et al.* (2002) in the laboratory where soil temperature decreased very slowly.

Pinel *et al.* (2000) evaluated the effect of steaming on the emergence of native weed species, such as shepherds purse (*Capsella bursa-pastoris*), annual nettle (*Urtica urens*) and fat hen (*Chenopodium album*) in the field. They used a self-propelled steaming machine, developed by Regero (16, rue de Allemagne, BP 73427-44334 Nantes, Cedex 3, France) which raised temperatures to 100 °C in the top 10 cm of the soil. The temperature increase reduced with increasing depth. The steam was applied under pressure for 8 minutes beneath 3 metal pans, each 2.5 m x 1.5 m. The effect of field steaming on weed emergence was investigated in six trials at three sites. In each trial, a 30 m length bed was steamed and a similar length was left untreated for comparison. At site 1 and 2, steaming gave 95 % weed control on the surface of the bed. However, the weeds were not controlled on the shoulders of the bed and in the wheelings between beds outside the recording area. In the two trials at site 3, few weeds emerged on the untreated bed, but the weed control following steaming was still between 80 and 90 %. This is in accordance with the fact that, in general, the seeds from which weeds emerge are present in the top 5 cm of the soil (Grundy *et al.*, 1996). The only exception was clover (*Trifolium* spp.) with hard-coated seeds, which appeared to survive the treatments.

Furthermore, the steaming treatment killed most seeds in the surface soil but, not below the treated layer. Nevertheless, according to this study, field steaming to inhibit the emergence of weeds is a very effective control measure (Pinel *et al.*, 2000).

Although more information/research is needed regarding the soil temperatures during and after steaming and their effect on seed germination, viability and emergence it seems that steaming as a weed seed control method is possible. The biological effectiveness of steaming to control weeds and to obtain good crop growth depends on the temperature of the steam, weed species, treatment duration and initial soil temperature. For the practical use the amount of energy and water needed is of importance and should be taken into account.

3. Effects of heating on micro-organisms in soil

When using a control method in organic farming, it is important that it only affects target organisms and that there are no side effects causing further damage to living organisms either directly or indirectly. Furthermore, when sterilising soil possible effects on soil chemical and biological properties have to be regarded, since they influence the growth of the crop. These properties are for a large part regulated by soil micro-organisms and changes in the microbial population can change the soils biological and chemical characteristics. Besides that, soil sterilisation may have positive or negative effects on soil born plant pathogens. Therefore, an inventory was made to determine the effect of heating on micro-organisms in soils treated with sufficient heat to control weed populations.

3.1 Effects of microwave energy

Vela *et al.* (1976) concluded after conducting both laboratory and field studies that soil micro-organisms survived 2,450 MHz exposures at much higher dosage levels than those required for control of weed seeds in soil. They tested the effect of graded doses of microwave radiation on soil microflora and measured the effect by comparing the number of viable organisms in a given soil sample before and after irradiation. The following organisms were regarded: soil bacteria, bacterial spores, actinomycetes, fungi, nitrogen- fixing bacteria and nitrifying bacteria (Azobacter) and found to be resistant to 40,000 joules of microwave energy applied to each cm^2 of soil surface, in both wet and dry conditions, different soil types and localities. Fungi were more susceptible than bacteria (Vela *et al.*, 1976).

Wainwright *et al.* (1980) exposed small (20g) samples of various soils (an organic loam, Fitzwilliam and Chapeltown brown earths) to 1 kW of 2,450 MHz radiation in a cavity to measure the effect upon the micro-organisms. As the exposure times were increased from 10 to 30 seconds nitrification in organic loam soil was progressively reduced, with an accompanying rise in $NH4^+$-H. Respiration rates in the organic loam and Fitzwilliam brown earth were slightly depressed after a 20 second exposure but not by a statistically significant amount. Sulphur oxidation was stimulated in Chapeltown brown earth after a 20 second exposure to 2,450 MHz radiation and the number of S-ions in the organic loam increased two- and three-fold compared with the control 28 days after treatment for 20 seconds.

Plates prepared from the organic loam showed that fungi were reduced from 30,000 g^{-1} to zero after a 20-second exposure in the cavity while the numbers of heterotrophic bacteria remained unaffected. It was concluded that the microwaves had marked differential effects on the soil microorganisms, were unlikely to harm the soilstructure, and might stimulate seedling emergence and growth due to enhanced nutrient status of the soil (Wainwright *et al.*, 1980).

Reduction of the microbial population was also observed in another study. In soil (moisture 5.2%) treated for 6 minutes in a microwave (650 Watt) no bacterial colony forming units were detected any more. After treatment for 5 minutes of a soil with a moisture content of 3.3% or 4 minutes for a soil of 5.2% no fungi were detected any longer (Chen *et al.*, 1995).

However, in a experiment in which 2.0 g of soil in sterile culture tubes, moistened with sterile water and pre- incubated for 48 h at 25 °C was treated for 10 minutes in a microwave oven at 2,450 MHz no significant reduction of the microbial populations in the soil was observed (Wolf *et al.*, 1989).

Since exposure to microwaves increases the temperature of moist materials (Barker & Craker, 1991; Wainwright *et al.*, 1980) and the effect of the radiation is thermal rather than direct radiation which kills

microorganisms (Vela & Wu, 1979), results in these studies probably differ due to different moister contents of the soils (Wolf *et al.*, 1989). Thus, the amount of water determines the temperature increase upon irradiation and thereby the occurrence of thermal death of the soil organisms.

Studies with other soil samples showed that exposures of small samples in a microwave oven had significant effect on micro-organisms, but that results depended on soil type, depth in soil, exposure time, amount of soil treated and soil water content (Ferris, 1984).

The susceptibility to control pests in soil by microwave heating appears to be, in decreasing order, insects, weed seeds, nematodes, fungi and bacteria. (Nelson, 1996).

3.2 Effects of steaming

The most important crops in which intra-row weeding is necessary and new methods are needed to reduce the amount of labour, are sugar beet (*Beta vulgaris*), carrot (*Daucus carota*) and onion (*Alium cepia*). In this part, attention will be focussed on the effects of steaming on the most important soil-borne pathogens of these crops, such as *Pythium* spp., *Fusarium* spp. and *Rhizoctonia* spp and their antagonists (Alabouvette, 1986; Davison & McKay, 1999; Wiseman *et al.*, 1996).

A disease-suppressive soil is one in which disease severity remains limited in spite of a high pathogen inoculum density. Since this disease suppressiveness of the soils is essentially microbiological in nature, i.e. it results from more or less complex microbial interactions, rather than from the direct effect of physiochemical factors on the pathogen (Alabouvette, 1986), the effects of steaming on the microbial community of the soil need to be regarded before steaming can be used to control weed seeds.

Wiseman *et al.* (1996) determined the suppressive characteristics of field suppressive soil to *Rhizoctonia solani*. Wheat plants were grown in soil suppressive or non-suppressive to *R. solani* steam pasteurised at 50, 60, 70 or 80 °C for 30 min or left untreated (control) and with or without *R. solani* inoculation. Plate counts of diluted steam pasteurised soil were used to measure the colony forming units (cfu) of total bacteria, fluorescent pseudomonads, actinomycetes, total fungi and *Trichoderma* spp. Results were calculated as cfu g^{-1} dry weight soil. Significantly lower disease occurred in untreated and 50 °C steam-pasteurised suppressive soil compared to non-suppressive for both the *Rhizoctonia* inoculated and non-inoculated treatments. Disease levels for inoculated pots were significantly higher following steam pasteurisation at 60, 70 or 80 °C compared to untreated or 50 °C. Counts for all five microbial groups were similar for both suppressive and non-suppressive soils following steam pasteurisation at 60,70 or 80 °C and significantly lower at these temperatures compared to untreated soil or steam pasteurisation at 50 °C (Table 1). The enhanced disease levels above 60 °C compared to untreated or 50 °C treatment is probably caused by differences in *Trichoderma* spp. This was the only group that was higher in both the untreated suppressive soil than the untreated non- suppressive soil as well as higher in the 50 °C pasteurised suppressive soil compared to the 50 °C pasteurised non-suppressive soil. Above 60 °C there was no difference between suppressive or non-suppressive soils (Wiseman *et al.*, 1996).

Table 1. *From Wiseman* et al. *(1996)*

Soil	Temp. (°C)	fungi	*Trichoderma* spp.	bacteria	Fluorescent Pseudomonads	actinomycetes
Suppr	Control	1.78×10^3 a	1.39×10^2a	1.29×10^6a	9.67×10^3a	1.03×10^5ad
	50	6.51×10^3b	71.1b	6.37×10^5b	3.06×10^4b	9.61×10^4a
	60	25.3c	3.62c	5.50×10^5c	0c	3.25×10^4c
	70	3.68c	0c	9.74×10^4c	0c	6.62×10^3c
	80	7.25c	0c	3.12×10^5c	0c	2.90×10^4c
Non-suppr	Control	3.53×10^3d	32.1d	1.81×10^6d	4.81×10^4d	1.07×10^5d
	50	3.62×10^3d	28.4d	1.10×10^6e	1.75×10^4e	1.49×10^5e
	60	14.4c	3.59c	3.36×10^5c	0c	1.65×10^4c
	70	7.13c	0c	2.14×10^4c	0c	7.14×10^3c
	80	10.8c	0c	8.67×10^4c	0c	1.81×10^4c
	LSD	1.26×10^3	20.1	3.00×10^5	1.01×10^4	3.29×10^4

Values are the means of 3 replica plates and those with the same letter are not significantly different (P=0.05). suppr=suppressive soil, non-suppr = non-suppressive soil, control is no soil treatment, 50, 60, 70 and 80 °C= steam pasteurisation temperature (30 min). Counts for fungi,* Trichoderma *spp., bacteria, fluorescent Pseudomonads and actinomycetes made on dichloran rose Bengal medium, a Trichoderma selective medium, tryptic soy agar, S1 medium and tryptic soy agar, respectively.*

Similarities also occur with *Fusarium*- suppressive soils (Alabouvette, 1986) and *Pythium*- suppressive soils (Knudsen *et al.*, 2002) in which the suppressive characteristics are removed by biocidal treatments such as steam.
The mode of suppression, however, can differ per pathogen and soil type, i.e. the suppression of plant pathogenic *Fusarium* spp. is most likely caused by intrageneric competition with non- pathogenic *Fusarium* spp. (Alabouvette, 1986), while the spread of *Rhizoctonia* spp. and *Pythium* spp. is probably inhibited by the interaction with *Trichoderma* spp. (Green & Jensen, 2000).

Pinel *et al.* (2000) used pure steam and obtained temperatures of 100 °C in the top layer of the soil. This temperature was found high enough to control most weed seeds and plant pathogens, but can produce undesirable soil biological and soil chemical side effects. Nitrification recovers slowly and, as a result, the ammonium and nitrate contents may rise to undesirable levels. At high temperatures and under anoxic conditions, manganese is mobilised rapidly; recovery of the Mn-oxidizing bacteria may take several months and in the mean time the Mn content of the crop may rise to an undesirable level (Raats, 1988; Runia, 2000).

Thus, when steam is applied as a control method for weed seeds, the possible effects on the microbial community in relation to soil- borne plant diseases and the soils chemical properties should be taken into account. Reestablishment of the microbial communities is possible by mixing the steamed soil with non- steamed soil. This is however not possible without introducing viable seeds from the non-steamed soil.

3.3 Effects of heating in an oven

It was found that oven drying (90 °C, 24h) of soil significantly reduced the microbial populations in soil compared to air-drying (Wolf *et al.*, 1989). Acea & Carballas (1999) determined the effect of the inoculation of heated (200 °C, 1h) soil with 0,5 % fresh soil in an oven. Inoculation increased the microbial counts 500- fold. The inoculated heated soil showed representatives of all the soil groups

studied, the relative proportions of each microbial group being similar to those found in this and other unheated soils. Only during the last week of incubation in the inoculated heated soil, the number of microbes, mainly bacteria, reached levels greater than that of the unheated soil. Ammonifiers and most of the other microbial groups showed a clear tendency to reach unheated soil values. An increase in microbial counts is common in soils whose microbiota has been totally or partially killed by heating. This is attributed to the improvement of the nutrient status, because newly available substrates are found in many heated soils (Diaz-Ravina *et al.*, 1993), their origin being partly the dead soil biota and partly the non-microbial soil organic matter (Acea & Carballas, 1999). The colonisation will in those cases be performed by the surviving organisms such as heat- resistant bacteria and some saprophytic fungi, together with micro-organisms with air-borne spores (Runia, 2000).

4. Influence of light and burial depth on weed seeds

Light is an important regulator of seed germination in many plants; it indicates that the environment is suitable for the early growth of the seedling. For some species, the light requirement for germination is fulfilled by very short light exposures; i.e. they have high light sensitivity (Andersson *et al.*, 1997). Light exposures of less than a minute and for some species less than a second of daylight is enough to induce germination in seeds of some species (Milberg *et al.*, 1996). It has been reported that a major source of light for buried weed seeds is the flash received during tillage (Buhler, 1997). In some agricultural weeds, sensitivity to short light exposures changes with the seed dormancy state during burial (Derkx & Karssen, 1993). Dormancy is weakened by the temperature, in many cases followed by the stimulation of a short light exposure during soil cultivation, thereby ensuring the germination of weed seeds at a time of year favourable for seedling growth (Andersson *et al.*, 1997).

The light requirement for germination has attracted attention during the last decade as a way of controlling agricultural weeds. In field experiments, dark harrowing has been tested as a method of reducing the amount of weed seedlings emerging after cultivation (Jensen, 1995). Although the results suggest that the method has promise, variation in emergence creates great uncertainty as to the effect of such a treatment. Part of the variation is due to environmental factors, such as temperature, soil moisture and nutrient contents, that affect the germination of seeds (Andersson *et al.*, 1997). Besides, the effect of dark harrowing will depend strongly on the weed flora composition since there are great differences between species regarding light requirements (Milberg *et al.*, 1996) and seasonal dormancy cycles (Baskin & Baskin, 1988).

Andersson *et al.* (1997) tested germination of 42 weed species in light of long duration, in darkness and in darkness after a short light exposure (SDLE). They used a photon flux density at seed level of 230 μmol $m^{-2}s^{-1}$(red light = 19 μmol $m^{-2}s^{-1}$ and far-red light =23 μmol $m^{-2}s^{-1}$). From the 42 species tested, seeds of 26 germinated to significantly higher percentages ($p<0.05$) after a 5-s light exposure than that kept in constant darkness. They used an exposure time of 5 s, which was estimated to correspond too less than 1 s of full daylight (full sun at noon). Of the summer annuals, 76% and of both the winter annuals and unclassified annuals, 65% germinated to higher percentages after the short-light treatment than those kept in the darkness. (See appendix II for Table 1; an overview of germination percentages of seeds after treatment). They defined the level of light requirement as the proportion of seeds requiring light to germinate, i.e. showing a difference in germination between darkness and short light exposure, or between darkness and full light. In 17 species tested, germination in full light was less than, or not significantly different from, the short light exposure. Thus, in many cases the short light exposure seemed to fulfil the light requirement for germination. In no case did germination in darkness exceed germination in the short- light treatment.

Milberg *et al.* (1996) performed a similar experiment with 70 weed species, of which 44 gave comparable results. They used a photon flux density at seed level of 210 μmol $m^{-2}s^{-1}$ (red light = 18 μmol $m^{-2}s^{-1}$ and far- red light = 23 μmol $m^{-2}s^{-1}$). From the 44 species germination was stimulated by SDLE in 24, while the other 20 showed no or inconsistent responses. Eight of 11 plant families contained species that responded to SLDE. Light sensitivity was shown equally by summer annual, winter annual and perennial species (See appendix II for Table 1; an overview of germination percentages of seeds after treatment).

In both studies it is concluded that although the light responses of seeds change continuously with the annual dormancy cycle and changes in temperature requirements, the light requirement shown by a

large portion of the species tested implies a great potential for integrating photocontrol of seed germination into weed management programmes (Andersson *et al.*, 1997; Milberg *et al.*, 1996).

Since these results suggest a wide-spread extreme light-sensitivity which is not restricted to species adapted to regularly disturbed land, short exposures of light are not likely to have been the only or even the main factor causing the evolution of the response to short light exposures. According to Milberg *et al.* (1996) there has not been selection for sensitivity to short periods of high light intensity, but selection has favoured the ability to detect very low photon fluence rates which, for instance occur during daytime at a few millimetres depth in soil.

The amount of light penetrating the toplayer of the soil is very low. A number of factors affect the transmittance of light through soil; particle size, moisture content, particle colour and presence of organic matter (Tester & Morris, 1987).

Baumgartner (1953) found that when the diameter of particles of quartz sand are 0.2-0.5 mm, a depth of 1-2 mm was needed to reduce the radiation by 95 %, but for very large particles, of 4-6 mm in diameter, 10 mm of sand were needed for the same reduction in radiation. Other studies also note a decrease in soil transmittance with decreasing particle size (Bliss & Smith, 1985). However, their have been no detailed measurements of the penetration of light through soil mixtures of widely different particle sizes, as would often been found in field situations.

Depending on the soil type, moisture content either increases or decreases the light transmittance of the soil. When sand is saturated the transmittance will increase, whereas saturation of clay and loam decreases the transmittance of light. This difference is probably attributed to a reduction in the reflection of light from the soil particles. When the particles are translucent, as in sand, transmission can increase through the particles; but in dark soil, reduced reflection only leads to increased absorption by the particles (Bliss & Smith, 1985). The darker particles are thought to adsorb the light. Another explanation is the increased reflection of light between particles of the lighter coloured soils, whereas the reflection in dark soils is lower (Tester & Morris, 1987). The role of organic matter has not been investigated yet, but was mentioned by Tester *et al.* (1987) as a factor possibly influencing light transmission.

Based on different studies regarding the penetration of light through soil, it can be concluded that physiologically and significantly amounts of light rarely penetrate more than 4-5 mm through soil, and that only 0.01% light is transmitted through to a depth of 3 mm. Hence, the sensitivity of the seeds after burial needs to be enhanced to achieve high germination levels (Tester & Morris, 1987). In studies on seed germination periodicity, it has been demonstrated that large changes in light sensitivity can be induced when seeds are buried in a soil seed bank (Baskin & Baskin, 1985) which may be linked with seasonal patterns such as temperature changes (Derkx & Karssen, 1993; Pons, 1991).

This enhanced sensitivity can cause the breaking of dormancy and thereby germination when soil is disturbed and seeds receive a flash of light, for instance during tillage. It may be an option to cover the machine while disturbing the soil to prevent breaking of dormancy and subsequent germination. The effect of this method, however, will not only depend on weed species and climate circumstances, but also will also depend on the particle size, moisture content and colour of the soil.

5. Conclusions & Recommendations

It became clear from the inventory study that the temperature threshold above which emergence, viability and germination of seeds was inhibited depends on weed species, treatment duration and moisture level of the seeds. Nevertheless, for most species the germination and viability of imbibed seeds can be reduced by temperatures ranging from 60- 80 °C. Temperatures to affect the germination of dry seeds are significantly higher (>80 °C) and to cause a viability reduction of dry seeds temperatures exceeding 100 °C are necessary.

High temperatures can be obtained by heating in an oven, with the use of microwave energy, steaming and a hot water bath.

The treatment duration needed to inhibit the viability and germination of the seeds by heating them in the oven is very long and varies between experiments. Egley (1990) found a viability reduction of moist seeds after 1 day in the oven at 60°C and no effect after heating for 7 days in dry soil at this temperature. According to Thompson *et al.* (1997) 10 minutes is enough to reduce the germination of the seeds at very high oven temperatures (≥ 155 °C). Although these results show the ability to inhibit the establishment of weeds in soil, the energy requirements to take up a layer of soil and heat it in an oven will probably be too high; treatment durations will be too long. Therefore, application of this method in the field with known equipment appears to be impractical.

Treatment durations needed to inhibit the germination, viability and subsequent emergence are shorter with the use of microwave energy. Good results were obtained in laboratory experiments with 2,450 MHz microwaves. Two minutes of heating were necessary to reach temperatures of 80 °C and cause the inhibition of seedling emergence (Barker & Craker, 1991). However, the effect of moisture content of the seeds on the method's effectiveness is high. The treatment duration to reduce the viability of seeds by 50 % for dry seeds can be 60 times longer (30 minutes) than for 4 to 16 hours imbibed seeds (30 seconds) (Shafer & Smith, 1974).

The importance of moisture level of seeds on the effectiveness was also noticed in field experiments. Germination of moist seeds was reduced at energy levels of 210 J/cm^2 (Wayland *et al.*, 1973), viability was reduced by 50 % at 100-180 J/cm^2, depending on the weed species and emergence by 80-90 % at energy densities of 180 J/cm^2. Levels to reduce seedling emergence from dry seeds needed to be higher (360 J/cm^2) (Menges & Wayland, 1974; Wayland & Merkle, 1975; Wayland *et al.*, 1973).

The amount of energy needed to produce these levels varies with the soil moisture content and initial temperature of the soil. Differences in energy requirement of 40 % were recorded (Mavrogianopoulos *et al.*, 2000). According to Vermeulen *et al.* (2002), the efficiency of microwave generator in the field will be 6%. Although this makes the method at this moment very expensive and impractical, an option to reduce the loss of energy would be to pick up the top soil layer, containing most weed seeds, in a microwave, heat it and place it back. It may be useful to cover the machine during application to prevent the possible germination of weed seeds remaining in the soil. Future research has to be focussed on the reduction of energy loss and thereby the costs, treatment of wet soil or reduction of energy requirements for treatment of dry seeds.

The best option of the methods regarded appears to be steaming. Temperatures and moisture contents that can be reached with this method are high enough to inhibit the germination and emergence of weeds and the affected soil layer can be deep enough (>5 cm) (Grundy *et al.*, 1996). Additionally, according to laboratory studies, treatment time is relatively short (100 seconds are needed to reach a temperature of 100 °C) (Melander *et al.*, 2002).

Experimental results from the field are also promising (90 % weed control after 8 minutes steaming with steam of 100 °C) (Pinel *et al.*, 2000), thereby giving this method good prospects for future use. Steaming one small strip in which the crop is sown to reduce weed seedling emergence in the row seems possible. Temperatures should be high enough to inhibit the emergence of weeds (in general, 80 °C) and low enough to prevent undesirable soil chemical side effects due to killing certain micro-organisms (≥100 °C).

Before it can be applied, more information is necessary regarding the soil temperatures during and after steaming and their effects on seed germination, viability and emergence of weed species that cause the most problems in the Netherlands. Since we know that certain organisms beneficial to crop growth such as antagonists of pathogens, can also be killed by steaming, the risk of soil borne pathogen establishment should be determined. These pathogenes such as *Pythium*, *Fusarium* and *Rhizoctonia* may reestablish in the steamed strip prior to their the antagonists and thereby affect crop growth.

One should also consider the combined use of the methods. It may for instance be possible to create a false seedbed and at the same time apply radiofrequency energy at 39 MHz or steam at low temperatures (40 °C) to bring about the enhanced germination and emergence of weeds. These weeds can then be mechanically removed prior to planting or sowing the crop. In such and other systems, information on the light requirement of seeds to break dormancy can be of great importance.

Another option that is not mentioned in this report that may provide solutions for the intra-row weed control in organic agriculture is electroporation. A method using electroporation to control weed seeds was investigated at the Swedish University of Agricultural Sciences. Electroporation is a well-known technique to incorporate specific genes into cells. Exposing the cell to high-intensity electric field pulses temporarily destabilize the cell membrane making it highly permeable to exogenous molecules. An increase in the electric field strength will however result in permanent pores in the cell membrane which are lethal to the cell. This effect may then be used as a weed control method. Fogelberg (2000) exposed weed and crop seeds to electric fields of 3-5 kV cm^{-1}. The electric field was obtained with electric high-voltage pulses with a duration of 1 ms. The seeds were either treated in peat soil, natural soil or in cuvettes filled with tap-water. Survey experiments with weed seeds treated in cuvettes showed control effects of 80 -100 %.

Experiments in natural soil obtained from vegetable fields showed a less pronounced weed control effect compared to the laboratory trials. About 40 % weed reduction was achieved in soils with a flora of *Urtica urens*, *Capsella bursa-pastoris* and *Chenopodium album*. In these trials two series of each 50 pulses were given using 3kV cm^{-1} and 4 kV cm^{-1}. Crop seeds were also treated with high-voltage pulses. The results showed that the sensitivity to electroporation varies with time. Some common crops such as peas, are controlled to about 100 % while other are less sensitive.
The results of the treatments in natural soil show a potential to use the method for weed control in e.g. vegetable crops. However, additional work must be carried out in order to evaluate the effects of electroporation in field experiments and possible negative impact on soil microflora (Fogelberg, 2000).

Literature

Acea, M.J., & T. Carballas, 1999.
Microbial fluctuations after soil heating and organic amendment. *Bioresource Technology* 67: 65-71.

Alabouvette, C., 1986.
Fusarium-wilt suppressive soil from the Chateaurenard region: review of a 10-year study. *Agronomie* 6: 273-284.

Andersson, L., P. Milberg & A. Noronha, 1997.
Germination response of weed seeds to light, light of short duration and darkness after stratification in soil. *Swedish Journal of Agriculture* 27: 113-120.

Anonymous, 1992.
Stomen. Technische handleiding bij het stomen van grond en substraat., pp. 46-49, Informatie en kennis centrum akker- en tuinbouw. Afdeling glasgroente en bestuiving, Afdeling bloemisterij.

Barker, A.V. & L.E. Craker, 1991.
Inhibition of weed seed germination by microwaves. *Agronomy Journal* 83: 302-305.

Baskin, C.C. & J.M. Baskin, 1988.
Germination ecophysiology of herbaceaous plant species in a temperate region. *American Journal of Botany* 75: 286-305.

Baskin, J.M. & C.C. Baskin, 1985.
The annual dormancy cycle in buried weed seeds: a continuum. *BioScience* 35 No.8: 492-498.

Baumgartner, A., 1953.
Das Eindringen des Lichtes in den Boden. Forstwisschenschaftlisches Zentrallblatt 72:172 In Tester *et al. (*1987).

Bliss, D. & H. Smith, 1985
Penetration of light into soil and its role in the control of seed germination. *Plant, Cell and Environment* 8: 475-483.

Buhler, D.D., 1997.
Effects of tillage and light environment on emergence of 13 annual weeds. *Weed Technology* 11: 496-501.

Chacko, K.C. & P.K. Chandrasekhara Pillai, 1997.
Storage and hot-water treatments enhance germination of guanacaste (*Enterolobium cyclocarpum*) seeds. *International Tree Crops Journal* 9: 103-107.

Chen, S.Y., D.W. Dickson & D.J. Mitchell, 1995.
Effects of soil treatments on the survival of soil microorganisms. *Journal of Nematology* 27: 661-663.

Couture, L. & J.C. Sutton, 1980.
Effect of dry heat treatments on survival of seed borne *Bipolaris sorokiniana* and germination of barley seeds. *Canadian Plant Disease Survey* 60: 59-61.

Davison, E.M. & A.G. McKay, 1999.
Reduced persistance of metalaxyl in soil associated with its failure to control cavity spot of carrots. *Plant Pathology* 48: 830-835.

Derkx, M.P.M. & C.M. Karssen, 1993.
Changing sensitivity to light and nitrate but not to gibberellins regulates seasonal dormancy patterns in *Sisymbrium officinale* seeds. *Plant, Cell and Environment* 16: 469-479.

Diaz-Ravina, M., M.J. Acea & T. Carballas, 1993.
Microbial biomass and its contribution to nutrient concentrations in forest soils. *Soil Biology & Biochemistry* 25: 25-31.

Diprose, M.F., F.A. Benson & A.J. Willis, 1984.
The effect of externally applied electrostatic fields, microwave radiation and electric currents on plants and other organisms, with special reference to weed control. *The Botanical Review* 50: 171-223.

Egley, G.H., 1990.
High-temperature effects on germination and survival of weed seeds in soil. *Weed Science* 38: 429-435.
Ferris, R.S., 1984.
Effects of microwave oven treatment on microorganisms in soil. *Phytopathology* 74: 121-126.
Fogelberg, F., 2000.
Electroporation- can we control weed seeds by the use of electric pulses applied in soil? *In* '4th EWRS workshop on physical weed control', Elspeet, The Netherlands.
Green, H. & D.F. Jensen, 2000.
Disease progression by active mycelial growth and biocontrol of *Pythium ultimum* var. ultimum studied using a rhizobox system. *Phytopathology* 90: 1049-1055.
Hopkins, C.Y., 1936.
Thermal death point of certain weed seeds. Canadian Journal of Research 14 (C): 178-183. In Tester, *et al.*, 1987.
Horowitz, M. & R.B. Taylorson, 1983.
Effect of high temperatures on imbibition, germination, and thermal death of velvetleaf (*Abutilon theophrasti*) seeds. *Canadian Journal of Botany* 61: 2269-2276.
Horowitz, M. & R.B. Taylorson, 1984.
Hardseededness and germinability of velvetleaf (*Abutilon theophrasti*) as affected by temperature and moisture. *Weed Science* 32: 111-115.
Jensen, P.K., 1995.
Effect of light environment during soil disturbance on germination and emergence pattern of weeds. *Annals of Applied Biology* 127: 561-571.
Kim, S.Y., S.K. De Datta & B.L. Mercado, 1990.
The effect of chemical and heat treatments on germination of *Commelina benghalensis* L. areal seeds. *Weed Research* 30: 109-116.
Knudsen, I.M.B., K.M. Larsen, D.F. Jensen & J. Hockenhull, 2002.
Potential suppressiveness of different field soils to *Pythium* damping-off of sugar beet. *Applied Soil Ecology* 21: 119-129.
Kolberg, R.L. & L.J. Wiles, 2002.
Effect of steam application on cropland weeds. *Weed Technology* 16: 43-49.
Mavrogianopoulos, G.N., A. Frangoudakis & J. Pandelakis, 2000.
Energy Efficient Soil Disinfestation by Microwaves. *Journal of Agricultural Engineering Research* 75: 149-153.
Melander, B., T. Heisel & M.H. Jorgensen, 2002.
Aspects of steaming the soil to reduce weed seedling emergence. *In* 'European Weed Research Society' (L. B. Bastiaans, D.T.; Christensen, S.; Hatcher, P.E.; Kudsk, P.; Grundy, A.C.; Marshall, J.C.; Streibig, J.C.; Tei, F., Ed.), pp. 236-237, Grafisch Service Centrum Van Gils BV, Wageningen.
Melander, B., T. Heisel & M.H. Jorgensen, March 2002.
Band-steaming for intra-row weed control. *In* '5th EWRS workshop on Physical Weed Control', Pisa, Italy.
Menges, R.M. & J.R. Wayland, 1974.
UHF electromagnetic energy for weed control in vegetables. *Weed Science* 22: 584-590.
Milberg, P., L. Andersson & A. Noronha, 1996.
Seed germination after short-duration light exposure: implications for the photo-control of weeds. *Journal of Applied Ecology* 33: 1469-1478.
Nelson, S.O., 1996.
A review and assessment of microwave energy for soil treatment to control pests. *Transactions of the American Society of Agricultural Engineers* 39: 281-289.
Nelson, S.O., L.E. Stetson & W.W. Wolf, 1984.
Long-term effects of RF Dielectric heating on germination of Alfalfa seed. *Transactions of the American Society of Agricultural Engineers* 27: 255-258.

Norberg, G. & A. Dolling, 2003.
Steam treatment as a vegetation management method on a grass-dominated clearcut. *Forest Ecology and Management* 174: 213-219.
Pinel, M.P.C., W. Bond & J.G. White, 2000.
Control of soil- borne pathogens and weeds in leaf salad monoculture by use of a self-propelled soil- steaming machine. *In* 'Proceedings of the fifth international symposium om chemical and non-chemical soil and substrate desinfestation' (M. L. Gullino, and J. Katan, Eds.), pp. 125-130, Acta Horticulturae, Torino, Italy.
Pons, T.L., 1991.
Induction of dark dormancy in seeds: its importance for the seed bank in the soil. *Functional Ecology* 5: 669-675.
Raats, P.A.C., 1988.
Disinfection of soils with steam. *In* 'Fertilization of vegetables', pp. 117-119, Acta Horticulturae, Wageningen, Netherlands.
Runia, W.T., 2000.
Steaming methods for soils and substrates. *In* 'Proceedings of the fifth international symposium om chemical and non-chemical soil and substrate desinfestation' (M. L. Gullino, and J. Katan, Eds.), pp. 115-123, Acta Horticulturae, Torino, Italy.
Shafer, F. & D. Smith, 1974.
Influence of internal and external moisture levels on toxicity of microwaves to seedlings. In Abstract no. 224, pp 97 in Proceedings of 1974 Meeting Weeds Science Society of America. North Texas State University In Diprose *et al.* (1984).
Tester, M. & C. Morris, 1987.
The penetration of light through soil. *Plant, Cell and Environment* 10: 281-286.
Thompson, A.J., N.E. Jones & A.M. Blair, 1997.
The effect of temperature on viability of imbibed weed seeds. *Annals of Applied Biology* 130: 123-134.
Vela, G.R. & J.F. Wu, 1979.
Mechanism of lethal action of 2,450- MHz radiation on microorganisms. *Applied and Environmental Microbiology* 37: 550-553.
Vela, G.R., J.F. Wu & D. Smith, 1976.
Effect of 2,450 MHz microwave radiation on some soil microorganisms in situ. *Soil Science* 121: 44-51.
Vermeulen, G.D., R.P. Zuydam & D.A.G. Kurstjens, 2002.
Toepassingsmogelijkheden van niet-chemische technieken voor onkruidbestrijding op verhardingen., pp. 51-52, IMAG, Wageningen.
Wainwright, M., K. Killham & M.F. Diprose, 1980.
Effects of 2450 MHz microwave radiation on nitrification, respiration and S- oxidation in soil. *Soil Biology & Biochemistry* 12: 489-493.
Watanabe, H., Y. Kusagaya & M. Saigusa, 2002.
Environmental factors affecting germination of apple of Peru. *Weed Science* 50: 152-156.
Wayland, J. & M. Merkle, 1975.
Control of weeds with UHF electromagnetic fields. *Weed Research* 15: 1-5.
Wayland, J.R., F.S. Davis & M.G. Merkle, 1973.
Toxicity of an UHF device to plant seeds in soil. *Weed Science* 21: 161-162.
Wiseman, B.M., S.M. Neate, K.O. Keller & S.E. Smith, 1996.
Suppression of *Rhizoctonia solani* anastomosis group 8 in australia and its biological nature. *Soil Biology & Biochemistry* 28: 727-732.
Wolf, D.C., T.H. Dao, H.D. Scott & T.L. Lavy, 1989.
Influence of Sterilization methods on selected soil microbiological, physical and chemical properties. *Journal of Environmental Quality* 18: 39-44.

Appendix I.
Temperatures needed for seed destruction

Table 1. *Percentage viable seeds remaining after the seeds were heated at 40, 50, 60 or 70 °C for 0 to 7 days in moist soil (Egley, 1990).*

		Viable seeds as affected by days in soil (%)							
Species	Soil temperature (°C)	0	0.25	0.5	1	2	3	5	7
S.s.	40	100	100	100	100	100	100	100	100
	50	100	100	100	45	30	22	14	35
	60	100	7	1	0	0	0	0	0
	70	100	0	0	0	0	0	0	0
A.t.	40	98	95	98	100	99	99	100	100
	50	100	98	64	47	48	52	44	38
	60	100	35	23	15	10	14	9	6
	70	100	2	1	0	0	0	0	0
A.c.	40	100	100	100	100	99	99	100	98
	50	100	99	100	99	93	82	59	49
	60	100	98	91	53	28	26	18	11
	70	99	26	14	13	2	1	0	0
I.l.	40	99	97	92	95	95	92	99	96
	50	97	97	85	38	39	38	36	49
	60	100	35	39	38	32	33	29	22
	70	97	21	26	15	8	4	0	0
X.s.	40	100	100	100	96	100	92	97	97
	50	100	100	100	96	1	0	0	0
	60	100	0	0	0	0	0	0	0
	70	97	0	0	0	0	0	0	0
P.o.	40	98	98	99	100	98	98	98	100
	50	94	93	95	95	93	89	81	82
	60	96	74	78	27	38	39	26	30
	70	93	21	8	9	7	5	4	0
A.r.	40	100	98	98	99	100	100	100	100
	50	99	99	99	99	99	96	46	44
	60	100	23	16	12	16	12	5	4
	70	99	17	16	22	8	12	7	5
S.h.	40	93	92	96	93	96	96	96	98
	50	90	89	85	84	54	20	8	17
	60	97	92	35	11	11	10	7	4
	70	98	48	20	21	0	1	0	1

S.s= Sida spinosa, A.t.= Abutilon theophrasti, A.c.= Anoda cristata, I.l.= Ipomoea lacunosa, X.s.= Xanthium strumarium, P.o.= Portulaca oleracea, A.r.= Amaranthus retroflexus, S.h.= Sorghum halepense.

Table 2. Percentage viable seeds remaining after the seeds were heated at 40, 50, 60 or 70 °C for 0 to 7 days in dry soil (Egley, 1990).

		Viable seeds as affected by days in soil (%)							
Species	Soil temperature (°C)	0	0.25	0.5	1	2	3	5	7
S.s.	60	100	100	100	100	100	100	100	99
	70	100	100	100	98	68	54	52	8
A.t.	60	98	99	97	98	93	98	100	92
	70	95	97	88	94	66	27	24	4
A.c.	60	100	100	89	99	98	94	98	80
	70	98	100	94	85	59	29	23	0
I.l.	60	100	95	90	100	87	80	96	87
	70	99	97	55	84	85	54	29	26
X.s.	60	100	100	100	100	100	100	100	100
	70	100	100	100	100	100	90	86	8
P.o.	60	99	100	99	100	99	100	99	100
	70	99	99	99	99	100	100	99	99
A.r.	60	100	100	100	100	100	100	99	99
	70	100	100	100	100	100	98	100	100
S.h.	60	98	99	95	97	96	96	98	100
	70	100	100	99	99	100	90	96	87

S.s= Sida spinosa, A.t.= Abutilon theophrasti, A.c.= Anoda cristata, I.l.= Ipomoea lacunosa, X.s.= Xanthium strumarium, P.o.= Portulaca oleracea, A.r.= Amaranthus retroflexus, S.h.= Sorghum halepense.

Table 3. Maximum and mean temperatures of the soil (Thompson et al., 1997).

Oven temperature (°C)	Heating time (min)	Maximum temperature (°C)	Mean temperature over treatment (°C)
102	0.5	28	26
	1	30	28
	2	35	30
	5	49	41
	7.5	56	45
	10	59	48
155	0.5	28	26
	1	33	29
	2	42	33
	5	61	49
	7.5	68	55
	10	70	58
204	0.5	28	27
	1	36	30
	2	50	37
	5	74	58
	7.5	81	65
	10	83	69
262	0.5	29	27
	1	39	31
	2	60	40
	5	94	71
	7.5	98	71
	10	99	84

Table 4. Germination percentage of the weed species following treatments characterised by maximum and mean soil temperatures (Thompson et al.*, 1997).*

Temp. (°C)		% germination (100%= 50 seeds)									
Max.	Mean	A.f.	A.m.	C.alb.	C.arv.	G.a.	P.p.	R.o.	S.a.	S.v.	V.p.
28	26	20	11	14	39	25	0	77	26	32	25
28	26	10	12	40	57	27	0	80	40	51	26
28	27	16	14	20	44	20	10	79	35	39	24
29	27	10	7	21	46	16	0	79	37	42	26
30	28	15	5	22	40	18	0	80	31	31	23
33	29	10	11	26	49	26	0	93	35	38	28
35	30	22	10	25	51	31	0	76	46	37	25
36	30	12	11	24	43	28	8	68	36	34	30
39	31	16	11	24	46	22	0	74	16	28	15
42	33	13	8	28	50	29	0	87	35	38	25
49	41	20	14	28	43	18	0	74	30	36	32
50	37	16	13	26	34	21	10	84	39	32	18
56	45	32	16	29	49	28	0	74	19	21	17
59	48	3	10	28	18	12	0	86	16	9	9
60	48	2	7	11	13	5	0	67	7	8	11
61	49	6	4	22	11	17	0	73	7	5	17
68	55	2	3	12	0	3	0	60	0	0	0
70	58	1	1	4	0	0	0	61	0	0	0
74	58	0	0	20	0	3	1	58	0	0	6
81	65	0	0	0	0	0	0	9	0	0	0
>83	>69	0	0	0	0	0	0	0	0	0	0

A.f.= Avena fatua, A.m.= Alopecurus myosuroides, C.alb.= Chenopodium album, C.arv.= Cirsium arvense, G.a.= Galium aparine, P.p.= Polygonum perisicaria, R.o.= Rumex obtusifolius, S.a.= Sonchus asper, S.v.= Senecio vulgaris, V.p.= Veronica persica.

Appendix II.
Seed germination affected by light

Table 1. *Germination percentage of seeds tested in full light, darkness and in darkness after a short duration light exposure (SDLE). Differences in germination between light-SDLE and SDLE- darkness are given by T1 and T2, respectively. Differences between populations (P) and population-treatment (PxT) interactions are presented for the SDLE-darkness comparison only (Andersson* et al.*, 1997),(Milberg* et al.*, 1996). A stands for Andersson result, M for Milberg result.*

	Germination[a]			Difference[b]			
Species	Light	SDLE	Darkness	T_1	T_2	P	PxT_2
Summer annuals							
Urticura urens (A)	86.3	90.1	23.5	ns	***	***	ns
Urticura urens (M)	87.0	53.4	7.5	***	***	***	ns
Bilderdykia convolvulus (A)	64.1	58.0	48.6	ns	ns	***	ns
Bilderdykia convolvulus (M)	94.4	62.3	61.9	ns	ns	***	ns
Polygonum aviculare (A)	60.5	52.1	32.8	ns	***	***	ns
Polygonum aviculare (M)	69.2	27.8	10.1	***	***	***	ns
Polygonum lapatifolium (A)	76.1	42.8	23.4	***	***	*	ns
Polygonum lapatifolium (M)	74.1	12.3	4.8	**	**	*	**
Chamomilla recutita (M)	49.8	0.8	0	x	x	x	x
Chamomilla suaveolens (M)	75.0	4.0	2.1	x	x	x	x
Chenopodium album (A)	99.5	92.3	46.7	**	***	ns	ns
Chenopodium album (M)	75.1	52.4	2.9	***	***	ns	**
Chenopodium suecium (A)	97.9	76.7	18.9	*	***	ns	**
Chenopodium suecium (M)	51.9	17.7	1.7	***	***	x	x
Chenopodium polyspermum (M)	75.4	31.4	13.8	***	***	x	x
Chaenorrhinum minus (M)	52.3	56.4	24.6	***	***	***	**
*Flaginella uliginosa (*M)	97.8	4.3	0.8	***	***	x	***
Spergula arvensis (A)	33.6	35.2	1.8	ns	***	*	ns
Fumaria officinalis (A)	14.2	11.6	12.0	ns	ns	*	ns
Descurainia sophia (A)	6.8	1.4	4.4	d	d	d	d
Sinapsis arvensis (A)	67.1	75.9	57.6	ns	***	***	*
Sinapsis arvensis (M)	28.4	32.8	27.8	**	**	***	***
Euphorbia helioscopia (A)	16.5	29.0	30.7	ns	ns	*	ns
Anchus arvensis (A)	58.1	53.7	22.3	***	***	***	*
Galeopsis bifida (M)	45.3	56.0	20.1	***	***	x	**
Galeopsis speciosa (A)	37.5	68.6	26.9	***	***	**	*
Galeopsis speciosa (M)	29.7	46.5	33.8	***	***	***	***
Galeopsis tetrahit (A)	82.7	77.5	70.9	ns	***	***	*
Galeopsis tetrahit (M)	19.1	17.5	2.2	***	***	x	***
Matricaria matricarioides (A)	99.7	87.3	11.2	***	***	***	**
Galinsoga ciliata (A)	74.9	7.8	0.9	***	***	ns	ns
Galinsoga ciliata (M)	99.5	1.5	1.9	ns	ns	ns	ns
Sonchus asper (A)	88.7	84.5	79.8	ns	ns	***	ns
Sonchus asper (M)	99.3	25.3	18.3	*	*	***	ns
Sonchus oleraceus (A)	100.0	99.4	74.6	ns	***	ns	ns

Species	Germination[a]			Difference[b]			
	Light	SDLE	Darkness	T_1	T_2	P	PxT_2
Sonchus oleraceus (M)	100.0	50.6	21.2	***	***	***	ns
Avena fatua (A)	3.5	1.3	2.6	d	d	d	d
Winter annuals							
Papaver argemone (A)	0.7	2.2	0.2	d	d	d	d
Papaver dubium (A)	0.9	2.3	0.5	d	d	d	d
Papaver rhoeas (A)	10.0	11.4	6.2	d	d	d	d
Erodium cicutarium (A)	37.1	7.4	9.7	***	ns	ns	ns
Erodium cicutarium (M)	15.7	13.1	20.3	ns	ns	***	**
Myosotis arvensis (A)	36.6	7.9	1.5	***	ns	x	ns
Myosotis arvensis (M)	20.3	3.9	5.6	ns	x	x	x
Buglossoides arvensis (A)	78.9	68.4	63.1	***	ns	***	*
Buglossoides arvensis (M)	87.8	80.4	61.6	***	***	***	*
Lapsana communis (A)	91.0	53.8	1.2	***	***	ns	ns
Lapsana communis (M)	96.7	7.1	5.3	x	x	x	x
Silene noctiflora (A)	99.8	99.5	85.7	ns	ns	***	**
Silene noctiflora (M)	100	71.3	18.9	***	***	***	***
Stellaria media (A)	89.3	96.5	33.6	***	***	ns	ns
Stellaria media (M)	45.9	52.4	17.5	***	***	***	**
Capsella bursa-pastoris (A)	34.1	20.9	9.0	***	*	***	ns
Thlaspi arvense (A)	41.2	60.8	0.2	***	***	ns	ns
Viola arvensis (A)	41.2	71.9	34.1	***	***	***	ns
Galium aparine (A)	47.2	32.7	28.9	***	ns	***	ns
Lamium amplexicaule (A)	99.1	94.3	3.2	***	***	**	*
Lamium purpureum (M)	38.3	16.5	17.8	ns	ns	***	ns
Veronica agrestis (A)	88.5	59.8	6.7	***	***	***	ns
Veronica agrestis (M)	16.3	11.9	9.0	ns	ns	***	ns
Veronica arvensis (A)	75.5	9.9	0	***	**	ns	ns
Veronica arvensis (M)	49.5	0	0	x	x	x	x
Centaurea cyanus (A)	80.9	48.9	43.7	***	ns	***	ns
Matricaria chamomilla (A)	98.7	32.3	2.8	***	***	*	ns
Matricaria perforata (A)	99.6	79.9	4.2	***	***	***	ns
Matricaria perforata (M)	99.0	4.4	3.1	***	***	x	***
Senecio vulgaris (A)	98.6	93.2	64.7	**	***	***	ns
Senecio vulgaris (M)	92.4	9.5	4.1	ns	ns	**	ns
Unclassified annuals							
Apera spica-venti (M)	40.9	17.8	9.9	**	**	***	***
Bromus tectorum (M)	84.3	99.0	100.0	x	x	x	x
Berteroa incana (M)	23.3	14.6	14.3	ns	x	x	x
Conyza canadensis (M)	18.6	7.8	0.8	***	***	ns	ns
Galium spurium (A)	8.7	47.0	25.7	***	***	***	*
Poa annua (M)	90.0	46.7	45.3	ns	ns	***	ns
Lactua seriola (M)	988.7	84.2	54.2	***	***	x	***
Lamium hybridum (A)	83.3	57.4	14.0	***	***	***	**
Lamium hybridum (M)	87.2	75.6	65.2	***	***	***	***

Species	Germination[a]			Difference[b]			
	Light	SDLE	Darkness	T_1	T_2	P	PxT_2
Perennial							
Bunias orientalis (M)	18.1	17.4	14.0	ns	ns	***	**
Cerastium fontanum (M)	88.0	59.2	16.7	***	***	x	x
Melilotus alba (M)	32.7	24.1	23.6	ns	ns	***	ns
Taraxacum officinale group (A)	98.1	97.8	95.3	ns	ns	ns	ns
Taraxacum officinale group (M)	100.0	100.0	65.2	x	x	x	x
Verbascum thapsus (M)	49.9	32.3	0	x	x	x	x
Rumex crispus (M)	46.5	0.5	0	x	x	x	x
Rumex longifolius (M)	98.0	21.5	0.2	***	***	***	x
Rumex obtusifolius (M)	94.5	26.0	0.2	***	***	x	***

[a] *Values are the means of three populations with two replicates.*

[b] **, **, ***= significant differences, or significant interactions ($p<0.05$, $p<0.01$ and $p<0.001$, respectively). ns= not significant, d= strong dormancy, x= analysis not possible due to missing value.*