Selective Flame Weeding in Vegetable Crops

By

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Abstract

Flame weeding is a thermal weed control method that controls weeds through the application of extremely high temperatures. Field experiments were conducted from 2005 to 2007 to determine weed and crop tolerance to flame weeding and to investigate effects on plant development, crop yield, and crop quality. Dose-response curves were constructed for weeds common to horticultural fields in Québec. Flame weeding was more effective in controlling dicot weeds than monocot weeds. Flame doses that reduced common lambsquarters density by 95% (LD_{95}) ranged from 0.83 to 2.85 kg propane km^{-1} for plants at the cotyledon through the 6-leaf growth stage. LD_{95} values for redroot pigweed ranged from 1.19 to 2.72 kg propane km^{-1} for plants at the cotyledon through the 4-leaf growth stage. In shepherd's-purse, LD_{95} values for weeds at the cotyledon and the 2- to 5-leaf growth stage were 1.15 and 2.78 kg propane km^{-1}, respectively. Control of monocot weeds was poor, with survival greater than 50% for all flame doses evaluated. Onion and broccoli were tolerant of a single flame weeding treatment, with yield losses observed only when flamed within 20 days after transplantation (DAT). Among weed-free treatments, onion was able to withstand up to six flame treatments without any detectable loss in yield. However, flame treatments alone were not able to provide sufficient weed control to maintain yields. Flame weeding had minimal effects on time to reach maturity, leaf and bulb development, pungency or quercetin concentration in onion. Broccoli tolerated up to four flame treatments in weed-free plots without yield reductions. Flame-only treatments had lower yields than the flamed, weed-free treatments in one of two years. Flame treatments had limited effects on the number of days to maturity, leaf

development, and glucoraphanin concentration in broccoli. Yield losses in spinach and beets were observed when flamed at both the 4- and 6-leaf growth stages; however, no adverse yield effects were observed when spinach or beets were flamed preemergence. Selective flame weeding is a valuable weed control option and has the potential to reduce the amount of costly hand-weeding employed in organic production for many flame-tolerant crops.

Résumé

Le pyrodésherbage est une technique de contrôle des adventices qui utilise l'application de températures extrêmement élevées. Des expériences on été menées de 2005 à 2007 pour déterminer la tolérance au pyrodésherbage des adventices et de différentes cultures maraîchères et pour évaluer les effets sur le rendement, le développement et la qualité des cultures. Des courbes de réponses au pyrodésherbage ont été construites pour certaines adventices communes au Québec. La technique a contrôlé plus efficacement les dicotylédones que les monocotylédones. Les doses de pyrodésherbage qui ont réduit le chénopode blanc de 95% (DL_{95}) variaient de 0,83 à 2,85 kg propane km^{-1} pour les stades de croissance variant de cotylédons à 6 feuilles déployées. Les DL_{95} pour l'amarante à racine rouge se situaient entre 1,19 et 2,72 kg propane km^{-1} pour les stades de croissances variant de cotylédons à 4 feuilles déployées. Pour la bourse à pasteur, ces valeurs se situaient à 1,15 et 2,78 kg propane km^{-1} pour les stades de cotylédons et 2 à 5 feuilles déployées, respectivement. L'oignon et le brocoli ont toléré le pyrodésherbage; des pertes de rendements n'ont été observées que lorsque le traitement était effectué moins de 20 jours après transplantation. Dans les traitements sans adventices, l'oignon a été capable de résister jusqu'à six traitements de pyrodésherbage sans baisse observable de rendement. Toutefois, le pyrodésherbage seul a été insuffisant pour permettre un contrôle des adventices suffisant à maintenir le rendement. Le pyrodésherbage a eu un effet mineur sur la précocité, le développement des feuilles et du bulbe, l'âcreté ou la concentration de quercétine dans le bulbe. Le brocoli a été capable de résister jusqu'à 4 traitements de pyrodésherbage sans baisse observable de rendement dans les

parcelles sans adventices. Les traitements incluant seulement le pyrodésherbage ont eu un rendement inférieur aux traitements pyrodésherbés sans adventice et ce, dans une des deux saisons. Le pyrodésherbage a eu un effet limité sur la précocité, le développement foliaire et la concentration en glucoraphanine du brocoli. Dans l'épinard et la betterave, des pertes de rendements ont été observées lorsque pyrodésherbés aux stades 4 et 6 feuilles. Toutefois, aucun effet sur le rendement n'a été observé lorsque le traitement était effectué en pré-émergence. Le pyrodésherbage sélectif est un bon outil et offre la possibilité de réduire les coûts par rapport au désherbage manuel utilisé en production biologique dans plusieurs cultures tolérantes aux chaleurs extrêmes.

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Contributions of the authors

This document contains manuscripts which were co-authored by the candidate, Dr. Maryse Leblanc (Insitut de recherche et de développement en agroenvironnement, Saint-Hyacinthe, QC, Canada), Dr. Daniel Cloutier (Institut de malherbologie, Ste.-Anne-de-Bellevue, QC, Canada), Dr. Philippe Seguin (Department of Plant Science, Macdonald Campus, McGill University), and Dr. Katrine Stewart (Department of Plant Science, Macdonald Campus, McGill University). The candidate carried out the experiments and data analyses, planned the experiments along with the co-authors, and was the primary author of the manuscripts. Drs. Leblanc, Cloutier, Seguin, and Stewart provided funds for the research, planned the experiments along with the candidate, provided advisory guidance, and reviewed the manuscripts.

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1.0 General Introduction

Thermal weed control encompasses a variety of weed control methods that utilize very high or low temperatures to kill weeds. Thermal weed control methods may involve the use of steam, liquid nitrogen, carbon dioxide snow, infrared radiation, or flames as sources of temperature extremes (Fergedal, 1993; Rifai et al., 2003). In practice, however, thermal weed control usually involves the application of high temperatures; with flaming being the most commonly used method. In a comparison between infrared, flame, and steam systems, Rifai et al. (2003) found the open flame unit as possessing the best potential for thermal weed and pest management. As well, it has been reported that compared with flaming, freezing with liquid nitrogen or carbon dioxide snow used 3 and 6 times more energy, respectively (Fergedal, 1993). Flaming is typically fuelled by liquefied petroleum gas (LPG), normally propane or propane/butane mixtures. Prototype burners utilizing hydrogen as a fuel source have been developed as well (Andersen, 1997).

The idea of controlling weeds with extreme temperatures has been around since at least 1852, when the first implement for such use was patented. Flame cultivation was rarely used until the 1940's. However, interest in thermal weed control quickly diminished as effective and inexpensive chemical herbicides were discovered and became the dominant method of weed control (Vester, 1988). There has been a renewal of interest in flame weeding in recent years, due to situations where herbicides are not available or not desirable. Flaming has become popular in organic farming, in which the use of synthetic herbicides is prohibited and growers are interested in methods that can be used to replace or reduce laborious and costly hand-weeding. For example, in onion (*Allium cepa* L.) and other root crops grown under organic conditions, the labor requirement is often the largest production cost (Ascard, 1989; Mojžiš, 2002). The labor requirement for weeding the crop row by hand can take as much as 100-300 hours per hectare (Nemming, 1993).

Additionally, few new herbicides are being tested and approved for use on many vegetable crops. The cost of the testing required to register an herbicide for a crop is prohibitively expensive. Many agrochemical companies therefore see registration of herbicides for anything besides the major arable crops as uneconomical. Previously, producers relied on state-funded trials to test the safety and effectiveness of herbicides in vegetable crops. However, such trials are now receiving reduced funding (Brewster, 1994). Furthermore, effective herbicides may be removed from the market or discontinued. For example, the production of the herbicide Allidochlor (Randox) (N,N-diallyl-2-chloroacetamide) was discontinued, which was formerly used in onion production (Agriculture Canada, 1987). This could potentially leave producers of vegetable crops with limited weed control options.

Flaming leaves no chemical residues in the soil that could harm the environment or subsequent crops (Parish et al., 1995; Rifai et al., 2003). The soil is not disturbed, which avoids bringing new weed seeds to the soil surface and the resulting increased germination and emergence. Drawbacks of flame weeding include equipment purchase and fuel costs, as well as potentially high energy inputs (Hatcher and Melander, 2003) and carbon dioxide emissions, and insufficient information regarding use and recommendations.

Flame weeding is almost exclusively used to control weeds that occur within the crop row. Although the use of flaming to control weeds between crop rows has been explored in the past (Parker et al., 1965), more common today is to control inter-row weeds through conventional mechanical methods (Melander, 1998). Intra-row weeds can pose a problem as mechanical methods are ineffective in controlling weeds or cause too much damage to the crop plants, especially early in the growing season. Currently, flaming is used primarily preemergence in slow germinating row crops such as carrots (Ascard, 1995). When used in this manner, flaming is generally done with burners that are oriented parallel to and driven directly along the crop row. Such burners may or may not be covered with an insulated cover. Covers are used in an attempt to increase both flaming efficacy and fuel use efficiency. This type of flaming is referred to as non-selective, as it takes place prior to crop emergence so no distinction is made between targeted and non-targeted plants. In this method, the field is worked and the crop is direct seeded. The goal is to then wait as long as possible in order to allow the greatest number of weeds to emerge and flame just prior to crop emergence (Bond and Grundy, 2001). This method would kill the first group of emerging weeds, giving the crop a competitive advantage. Again, since the soil is not disturbed, preemergent flaming will not lead to increased weed emergence following treatment.

The alternative is selective flaming, where flame weeding occurs after transplantation or crop emergence. Flame weeding is selective if weeds can be controlled while damage to the crop is minimized. This can be accomplished by choosing flame doses that control weeds but do not harm the crop or by utilizing technological additions such as shields that protect the crop. Crop tolerance to flaming may be due to natural differences between the crop and the weed species, or by the crop being at a later stage of development such as in the case of transplants. In such a system, the burners are oriented perpendicular to the crop row, and directed at an angle towards the lower portion of the stem of the crop. Burners on either side of a row are staggered in order to keep flames from deflecting upwards and causing unnecessary damage to the crop (Huitink, 1972). The goal is then to determine flaming levels which are high enough to be able to effectively control weeds, but not higher than what the crop can withstand without undergoing unacceptable levels of damage. Although there have been some recent investigations into the use of selective flame weeding in some row crops (Ascard, 1989; Knezevic and Ulloa, 2007b, Knezevic and Ulloa, 2007c; Wszelaki et al., 2007), there is insufficient information available in the literature regarding the use and effects of selective flaming in many crop species.

Overall research hypothesis:

Flame weeding has the potential to be a valuable weed control tool for producers of horticultural crops. However, both crop and weed species can be expected to vary in tolerance to flame weeding treatments. Furthermore, flame weeding causes significant stress on plants, which may result in morphological, developmental, and physiological changes.

Sub-hypotheses:

1. Weed species differ in their susceptibility to flame weeding. Within species, response to flaming will be dependent upon flaming intensity and plant development. Dose response curves can be constructed to describe the response of plants to flame treatment.
2. Crop species will vary in their ability to tolerate flame weeding. Some species will be able to tolerate flame weeding in the intensity range required for weed control, while others receive unacceptable levels of damage.
3. Beyond yield considerations, flame damage is an extreme and acute stress and will cause other detectable developmental and physiological changes within crop plants.

The goals of this research project therefore are to:

1. Construct dose response curves for a number of weed species common to horticultural fields in Québec in order to determine flame doses which result in effective control.
2. Evaluate the feasibility for using selective flame weeding in four vegetable crops: onion, broccoli (*Brassica oleracea* L. var. *italica*), spinach (*Spinacia oleracea* L.), and common beet (*Beta vulgaris* L.) by testing a range of flaming intensities at different points during the season.
3. Further test promising crops identified in previous experiment with more intensive flaming regimens.
4. Evaluate crops for effects on developmental, physiological, and crop quality parameters due to flame weeding.

2.0 Literature Review

2.1 Weed control in minor crops

Many fruit, vegetable, and ornamental crops are considered minor crops (Agriculture and Agri-Food Canada, 2008), crops which are grown on much smaller areas than large acreage crops such as corn (*Zea mays* L.), wheat (*Triticum aestivum* L.), and soybean [*Glycine max* (L.) Merr.]. Producers of minor crops often have to depend upon a relatively small number of old herbicides, along with mechanical cultivation and hand-weeding for weed control. Because of this, managing weeds in many minor crops is more expensive than for large acreage crops (Fennimore and Doohan, 2008). Weed control in many specialty and high value crops require large labor inputs for hand-weeding. This poses a problem for regions with high labor costs such as Canada and the United States, as countries with lower labor costs have a decided advantage (Calvin et al., 2004; Oregon Department of Agriculture, 2007).

Many chemical companies are reluctant or unwilling to register new herbicides for use in minor crops, due to low sales capacity and the high value of many minor crops, which makes liability high (Baron et al., 2003; Bell et al., 2000; Bischoff, 1993). Nearly all efforts into the discovery of new herbicides is intended towards the use in major acreage crops due to the high costs associated with developing a new pesticide (Rotteveel and Powell, 2003). Chemical companies face increasingly high financial barriers in bringing a new herbicide active ingredient to market, with costs estimated at up to $184 million dollars (Gast, 2008). Combined with the fact that at least 20 herbicides (e.g., alachlor, asulam, cyanazine, EPTC, ioxynil, methyl bromide, monolinuron, propachlor, etc.) registered for use in minor crops in Canada and the United States have been removed from the market or had their use curtailed sharply, and it is apparent that weed control options are becoming more and more limited (Fennimore and Doohan, 2008; WSSA, 2002). In addition, the introduction of new herbicide activate ingredients has been decreasing in recent years (Gast, 2008).

2.2 Herbicide concerns and water contamination

Increasing public distrust over the safety of herbicides and their toxicity in the environment has contributed to the unease concerning their widespread use. Agricultural runoff, the transport of soil-adsorbed and dissolved herbicides in water from treated land areas, is the leading cause of contamination of surface waters by herbicides (Krutz et al., 2005; Leonard, 1990). High profile pesticide contamination, such as the finding of aldicarb in Long Island wells in the 1970's and DCBP (dibromochloropropane) in more than 2,000 wells in California, increased the public's attention on pesticide safety and pollution (Trautmann et al., 1990). Surveys have found pesticides in streams (Scribner et al. 2000; Senseman et al., 1997), rivers (Clark and Goolsby, 2000; Senseman et al., 1997; Thurman et al., 1996), lakes (Senseman et al., 1997; Thurman et al., 2000), and reservoirs (Thurman et al., 1996).

2.3 Vegetable crops in Canada

In 2002, the cultivated area of vegetables (excluding potatoes and greenhouse) in Canada was approximately 120,000 hectares. A bit over half of this land is planted with vegetables destined for processing. Roughly half of the cultivated vegetable area occurs in Ontario, with Québec having an additional 33%. There were 2,114 farms which produced vegetable crops in Québec in 2001, including 125 farms which reported producing organic vegetable, fruit, or greenhouse products (Agriculture and Agri-Food Canada, 2003). Overall in Canada, about 0.6% of land currently cultivated for commercial vegetable production is designated as organic (Agriculture and Agri-Food Canada, 2003). The number of farms in Canada reporting producing certified organic products increased from 2230 to 3555 from 2001 to 2006, an increase of 59% (Statistics Canada, 2008). In addition, the number of farms producing organic fruits, vegetables or greenhouse products increased from 614 to 916, a 49% increase.

2.4 Effects of flame damage in plants

It is not necessary to burn weeds to ashes during flame weeding. Rather, brief exposure to extreme temperature damages tissues beyond the point of repair. Research has shown that the primary cause of cell death due to supra-optimal temperatures in leaves is disintegration of the cellular membranes (Daniell et al., 1969). The effectiveness of flame weeding is due in part to the denaturisation of many proteins that occurs at temperatures of 50-70° C. Perhaps more important still is that the rapidly heated plant cells expand and damage the cellular membranes, leading to leakage of cellular contents (Vester, 1988). It has been determined that dehydration of tissue following cellular membrane damage was the primary cause of cellular death in flamed maize seedlings (Ellwanger et al., 1973a; Ellwanger et al., 1973b). Leaf temperatures required for this effect have been reported to be in the range of 55-100° C (Daniell et al., 1969; Hoffman, 1982 cited in Vester, 1988).

2.5 Variation in plant susceptibility to flame weeding

Plants vary in their susceptibility and tolerance to flame weeding. A number of morphological characteristics can affect a plant's ability to resist flame damage, including the thickness of the cuticle, level of pubescence, and extent of lignification. As the effect of flaming is in large part due to cellular expansion and dehydration, the water status of the plant would be expected to play a major role in a plant's ability to withstand flame damage. However, the implications of water status are unresolved. Most reports indicate that dry weather is more favorable for flaming than humid, moist weather (e.g.,, Vester, 1988). However, Ascard (1994) reported higher doses required in a drier year than in the two previous years with higher rainfall. On the other hand, Ascard (1990) noted that a drier year favoured flaming while a wetter year favoured chemical control, though this may have more to do with the need for some moisture with soil applied herbicides.

Plant size and development plays a significant role in a plant's susceptibility to flaming. Larger, mature weeds are more resistant to flaming and are more likely to survive flame damage than smaller individuals. More mature plants have a greater degree of pubescence and other morphological advantageous features than younger plants, have thicker, more robust leaves and stems, and an overall greater biomass which provides greater reserves in the event of partial flame damage. In a study utilizing a backwards facing, insulated burner, it was reported that doses above 40 kg propane ha^{-1} were needed to obtain 95% control of white mustard (*Sinapsis alba* L.) with 0 to 2 true leaves (Ascard, 1995). The dose required for the same level of control rose to 70 kg propane ha^{-1} when plants were at the 2 to 4 leaf stage.

Whether a plant ultimately survives, however, is largely a factor of the plant's faculty for regrowth following flame injury. The location of the growing point is very important in this regard (Ascard, 1995; Mayeux et al., 1968). Weeds with protected growing points are tolerant to flaming, whereas species with exposed growing points and sensitive leaves are susceptible (Ascard, 1998). This is illustrated in the relative susceptibilities of grasses and broadleaf weeds to flame weeding. In monocots, which tend to be harder to control with flaming than dicot species (Ascard, 1995; Knezevic and Ulloa, 2007a; Knezevic et al., 2008), the growing point is often at or just below the soil surface and protected by the surrounding leaves. Many broadleaf weed species, where the growing points are located at the tip of the stem and at leaf axils, tend to be more susceptible to flaming.

Ascard (1995) divided weeds into four groups based on tolerance towards flaming. The author reported that weed species with thin leaves and unprotected growing points such as common lambsquarters (*Chenopodium album* L.), common chickweed [*Stellaria media* (L). Vill.], and burning nettle (*Urtica urens* L.) were susceptible to flaming. Utilizing a covered, non-selective flamer oriented parallel to the row, plants of these species had a mortality rate of 95% when flamed with doses of 9-22 kg propane ha^{-1} at the one to four leaf stage. The second group consists of species moderately tolerant to flaming that could be completely killed at both early and later developmental stages, but which required higher rates than species in the first group. Placed in this group were species with a prostrate growth form and protected growing points or those with upright growth and possibly heat tolerant leaves, such as ladysthumb (*Polygonum persicaria* L.) and prostrate knotweed (*Polygonum aviculare* L.). The third group contained species where complete kill was only possible during early development stages, such as shepherd's-purse [*Capsella bursa-pastoris* (L.) Medik.]. Weeds in this category have a prostrate growth habit, especially early in the life cycle, and protected meristems. The last group is made up of species that, in that study, could not be killed by a single treatment no matter the flaming intensity or stage of development of the plant. Plants in this category may have a protected growing point or a creeping growth form. Annual bluegrass (*Poa annua* L.) was the only species in this category that appeared frequently in this study. Other grass species can be expected to belong in this latter category as well.

Domingues et al. (2008) modeled the response of barnyardgrass, green foxtail, velvetleaf, and morningglory to broadcast flaming. They found the monocot species to be more tolerant to flaming than the broadleaf species based on visual inspection. However, morningglory required the highest flaming dose to achieve 90% reduction in dry matter (41, 36, 20 and 24 kg propane ha^{-1} for morningglory, barnyardgrass, green foxtail, and velvetleaf, respectively). Cisneros and Zandstra (2008) evaluated the susceptibility of six weed species to flame weeding in a laboratory setting. They reported that weed susceptibility varied depending upon species and seedling size.

2.6 Flaming parameters

Flaming efficacy is influenced by a number of factors, including temperature, exposure time, and energy input (Ascard, 1997). Exposure time is primarily controlled by the driving speed of tractor mounted apparatuses, although fuel pressure and the width and overall size of the flame play a role as well. However, driving speed is a compromise between competing interests of reducing operating time as much as possible and exposing weeds to flames for an adequate length of time.

Minimizing fuel consumption is of great concern to producers. This is primarily to reduce operating costs, as one of the main advantages of flaming is the cost savings it can provide by reducing the amount of costly hand-weeding that is often required in organic production. High costs associated with flame weeding are largely a result of initial equipment purchases and the cost of fuel. As equipment purchase is unavoidable, fuel costs are therefore the area where the greatest cost reduction is possible, as well as in offering the most in terms of long term savings. Additionally, decreasing fuel input is of interest in order to reduce fossil fuel consumption and emissions associated with LPG combustion in regards to concerns about global warming. The amount of fuel consumed is dependent upon driving speed and fuel pressure. The goal is to minimize fuel usage while maintaining effective weed control; reducing fuel usage to the point where control is no longer achieved is counter productive and not a viable option.

There have been a number of studies which have demonstrated a correlation between increased fuel consumption and higher levels of weed control (Ascard, 1997; Lien and Robbins, 1967; Vester, 1988). In a study using covered, rear facing burners, Ascard (1995) reported that mortality of a number of weed species, including shepherd's-purse, burning nettle, and common chickweed, increased with increasing flaming dose, following a logistic pattern. Propane doses in the range of 10 to 100 kg propane ha^{-1} were used.

Burner angle can have an effect on the efficiency of a flaming apparatus. The angle and distance to the ground have a dramatic effect pattern of heat dispersal. In selective flaming, burners are normally angled and aimed at the base or lower part of the stem of the crop in order to reduce damage to the more sensitive leaf canopy. A burner angle between 30 and 45° is often reported and recommended (Ascard, 1989; Huitink, 1972; Mojžiš, 2002).

Holmøy and Storeheier (1993) conducted a set of laboratory experiments analyzing burner set-up. These experiments employed rear facing burners parallel to the direction of travel. Both shielded and unshielded designs were tested. It was reported that burners should be placed at an angle between 22.5 and 45° with respect to the horizontal, based on temperature, energy input, and exposure time (Holmøy and Storeheier, 1993)

Ascard (1998) evaluated five burner angles using a shielded parallel burner system in a non-selective weeding experiment: 45 and 67° directed both forwards and backwards with respect to the driving direction and 90° straight down (with respect to the ground). A backwards facing burner at an angle of 67° resulted in the most effective weed control, though differences between treatments were not significant. There was no correlation observed between temperature parameters measured in the laboratory (maximum temperature, exposure time, or temperature sum) for the different burner angles and weed control effects observed in the field. It was stressed, however, that this did not negate the value of temperature measurements; rather, that laboratory conditions need to better imitate conditions found in the field.

2.7 Selective flame weeding in crops

The predominant use of flame weeding has been non-selective, prior to crop emergence. However, there have been studies evaluating the use of selective flame weeding in a number of crops, where flame weeding treatments occur after crop plants are present. The petroleum industry sponsored a great deal of research into flame weeding in the USA during the 1960's, resulting in studies that examined selective flame weeding in a number of crops. Researchers conducted studies on the use of flame weeding in soybeans (Lalor and Buchele, 1966), grapes (Hansen et al., 1966), corn (Lalor and Buchele, 1966; Lien and Liljedahl, 1966; Reece et al., 1966), and cole crops (Wilson and Ilnicki, 1966), among others. As the use of herbicides became more and more widespread, funding for and interest in flame weeding waned. Interest in flame weeding was revived and research started again, predominantly in Europe. Vester (1988) selectively flamed onions, sweet corn, kale (*Brassica oleracea* var. *acephala*), common beet, and potatoes (*Solanum tuberosum* L.). Ascard (1989) compared flame weeding to chemical control and mechanical cultivation in onions. Holmøy and Netland (1994) experimented with selective flame weeding in cabbage (*Brassica oleracea* var. *capitata)*. More recently, Wszelaki et al. (2007) examined the use of selective flame weeding in tomato (*Solanum lycopersicum* L.) and cabbage. Knezevic and Ulloa (2007b) examined sensitivity of corn, soybean, sunflower

(*Helianthus annuus* L.), and sorghum [*Sorghum halepense* (L.) Pers.] to flame weeding. Heverton et al. (2008) evaluated the use of flame weeding in soybean and corn and concluded that soybean was more susceptible to flaming than corn. Ulloa et al. (2008) tested five flaming rates at three growth stages of winter wheat. Flaming in winter wheat was deemed unacceptable, as yield losses were 25% or greater at flaming rates that could be used to control most weed species. Knezevic and Ulloa (2007c) evaluated six crops for response to flame weeding: field corn, soybean, sorghum, sunflower, alfalfa (*Medicago sativa* L.), and red clover (*Trifolium pratense* L.). Field corn and sorghum were most tolerant to flame treatments, while red clover and alfalfa were the most susceptible.

Ascard (1989) conducted a trial to compare pre- and postemergence flame weeding to chemical control and hand-weeding treatments in both set and seeded onions. Seeded onions were selectively flamed once (onion height of 15 to 20 cm) while set onions were flamed twice (onion height of 20 to 25 and 40 to 50 cm). Flaming was reported to have a moderate effect on number of weeds, but a good effect on weed weight. This is consistent with observations in other studies that weed biomass is decreased much more easily than weed number with flaming. In set onions, the amount of hand-weeding required in the pre- and postemergent flaming treatments was 1/3 of that of chemically treated plots, and 1/4 of the mechanically controlled. The labor requirement in the chemically controlled plots was high due to only using preemergent herbicides. Yield was slightly higher in hand planted sets in the flamed treatments, but 20% lower in machine planted sets when compared to chemical control. In seeded onions, the labour requirement was lowest with the chemical control. Preemergent flaming halved the labour compared to the control, and postemergent flaming lowered that still. Yield in flamed treatments was not affected compared with the control in seeded onions.

2.8 Flaming effects on other naturally occurring biota

Given the observed effect of flame weeding on plants, many have wondered how flaming would affect other organisms. Soil microbes are essential for good soil health; therefore the effects of flaming on beneficial soil micro-organisms are an important consideration. However, it has been reported that only the top 5-10 mm of soil are heated up to any appreciable degree (Balsari et al., 1994; Rahkonen et al., 1999). When a relatively high flaming dose of 100 kg propane ha^{-1} was administered to bare soil, microbial biomass in the top 5 mm was reported to decline by 19% (Rahkonen et al., 1999). There was no significant effect of flaming on microbial biomass at a depth of 5-10 mm. Soil temperature was raised by 4.0° C at a depth of 5 mm and 1.2° C at 10 mm below the soil surface. In another study traveling at 0.42 m s^{-1} with a fuel pressure of 200 kPa (resulting in a comparable flaming dose of 116 kg propane ha^{-1}), temperatures reached 240° at the soil surface, but only 50° C at a depth of 5 mm (Balsari et al., 1994). Therefore, flaming can be expected to have a limited effect on microbial populations within the soil.

2.9 Physiological effects of flame damage in plants

Flame damage is a significant acute stress that results in a number of physiological responses. After receiving flame damage from a butane lighter, tomato plants upregulated a number of genes involved in basic cell metabolism and defense response (Coker et al., 2005). Seven transcripts were systemically accumulated within one hour following flame damage. Levels of these transcripts subsequently returned to basal levels within 3 hours. Additionally, levels of proteinase inhibitor I, which is involved in plant defence, increased over 30-fold in the six hours following flame treatment. In another study, it was reported that a chloroplast mRNA-binding protein (CMBP) was systemically up-regulated after flame stimulus. Gene expression response involved a transient increase from 5-15 minutes after flame stimulus, followed by a decline to basal levels from 15-45 minutes, and finally another increase in expression levels after 60-90 minutes. This response is contrasted to that seen after mechanical damage, which produced the initial expression increase and decline, but not the second increase in gene expression (Vian et al., 1999).

In a study on the effects of flame exposure on maize, Ellwanger et al. (1973a) reported that transpirational water loss was 68% less in flamed seedlings than non-flamed seedlings when measured 16 hours after flame treatment. They also reported that carbon assimilation was much reduced in flamed seedlings compared to non-flamed seedlings, though it did not stop completely even in the most severely damaged leaves. These observations were attributed to stomatal closure due to dehydration and loss of cell turgor following flame damage.

Cellular damage in corn leaves exposed to flames was observed under a light microscope (Ellwanger et al., 1973b). Chloroplasts were distorted and exhibited ruptured envelopes. Tonoplasts were disrupted, allowing solute leakage between vacuole, plastids, and cytoplasm. With the tonoplast damaged, the vacuole could no longer help maintain cell turgor. Also, disruptions in the plasmalemma were visible, compromising cellular selective permeability and allowing solutes to leak between the apoplast and cell interior and ultimately leading to dehydration of the damaged tissue. Disruption to the cell membrane is what allows for the use of the “finger print test”, a quick method that can be employed in the field in order to determine if plant tissue has been effectively flamed. With this method, a flamed leaf is pressed between the thumb and finger. If a darkened fingerprint is visible on the leaf, cells have been ruptured and tissue dehydration and death can be expected.

2.10 Flavonoids in onion

Flavonoids are phenolic compounds distributed widely in the plant kingdom (Sellappan and Akoh, 2002). Flavonoids play a role in plant defense response to a number of stimuli, acting as a deterrent to herbivory, they play a role in the response to attack by pathogens (Stafford, 1997) and in response to physical damage caused by chemicals or UV light (Olsson et al., 1998). Flavonoids also have other important roles, being involved in the plant-microbe interactions associated with nitrogen fixation in legumes (Long, 1989; Olsson et al., 1998).

Onion has been reported to contain large quantities of flavonoids (Hertog et al., 1992a; Hirota et al., 1998; Patil et al., 1995). The major flavonoid in onion is quercetin, which is found within the plant predominately as glucosidic conjugates (Price and Rhodes, 1997; Sellappan and Akoh, 2002). The most prominent quercetin conjugates are 3,4’-O-diglucoside and 4’-O-monoglucoside, which together make up approximately 85% of the flavonoids found in onion (Price and Rhodes, 1997).

Flavonoid content in onion has been reported to vary with cultivar. In a comparison of 12 onion cultivars, total flavonoid concentration was found to vary from 486.9 to 979.1 mg kg^{-1} of fresh material (Marotti and Piccaglia, 2002). In another study evaluating flavonoid content in 4 onion cultivars, it was found that levels of 3,4'-O-diglucoside ranged from 50-1300 mg kg^{-1} and 4'-O-monoglucoside from 36-394 mg kg^{-1} of fresh onion tissue (Price and Rhodes, 1997). Environmental conditions have been observed to affect flavonoid concentrations as differences have been noted between locations (Patil et al., 1995).

Quercetin has garnered much interest due to its properties as an antioxidant and potential benefits for human health in regards to cardiovascular disease (Hertog et al., 1993; Hertog et al., 1995) and cancer (Hosokawa et al., 1990; Scambia et al., 1990). Quercetin levels in onion, therefore, may be important in relation to their value as a functional food. Alternatively, in light of the role of many flavonoids in defence response to many biotic and abiotic stresses, increased quercetin levels may impart a protective effect on the plant.

2.11 Onion flavour components

Despite possessing relatively low nutritional value, onions are highly valued in cuisines all around the world due to their distinctive flavour. Onions accumulate large quantities of sulphur compounds, particularly the gamma glutamyl peptides and S-alkenyl cysteine sulfoxides (ACSOs). In intact tissue, ACSOs are found in the cytoplasm while the enzyme alliinase is stored within the vacuole (Lancaster and Collin, 1981). When onions are eaten or chopped for cooking, the vacuole ruptures, releasing alliinase into the cytoplasm. ACSOs are then hydrolyzed by alliinase and form a number of volatile S-compounds responsible for the characteristic flavour and aroma associated with onions (Coolong and Randle, 2003).

Measurement of overall onion flavour and pungency is often carried out by measuring enzymatically produced pyruvic acid (EPY), a non-flavour product of the hydrolysis of ACSOs by alliinase (Kopsell et al., 1999). However, pyruvic acid is a common metabolic product found within plants. Therefore background pyruvic acid levels can upwardly bias measurements of onion pungency by overestimating pyruvic acid produced by the enzymatic hydrolysis of ACSOs by alliinase. For this reason, background levels of pyruvic acid are determined by thermal treatment of samples, resulting in enzymatic deactivation. Background pyruvic acid levels can then be subtracted from total pyruvic acid levels to yield EPY (Randle and Bussard, 1993). EPY has been shown to be highly correlated with overall taste perception (Wall and Corgan, 1992).

A number of factors are known to affect flavour intensity in onion, including temperature (Coolong and Randle, 2003) and sulphur fertilization levels (Randle et al., 2002) during plant growth. There is no information concerning the effects of flame weeding on flavour characteristics of onion.

2.12 Glucosinolates in broccoli

Glucosinolates are a class of compounds found in many plants, most prominently in the *Brassicaceae* (Fenwick and Heaney, 1983). Upon cell breakdown through maceration or mechanical means, glucosinolates are hydrolyzed by the enzyme mirosinase to yield a range of compounds including glucose, isothiocyanates, nitriles, and thiocyanates (Rosa, 1997). The breakdown products of the hydrolysis of glucosinolates are important components of the characteristic flavour and aroma of vegetables belonging to the *Brassicaceae* (MacLeod, 1976).

Glucosinolates may also play a role in plants' response to disease, as glucosinolate breakdown products have been shown to suppress a number of pathogens (Rosa et al., 1997). Bacterial soft rot is a serious disease of broccoli that can lead to total crop loss if severe and left uncontrolled. In a comparison of eight broccoli cultivars, it was determined that 48% of the differences between cultivars in suppression of *Pseudomonas marginalis*, one of the causal agents of bacterial soft rot, were due to differences in total glucosinolate content (Charron et al., 2002).

Glucosinolate levels in broccoli are affected by a number of factors, both genetic and environmental. Environment (year) had an impact on glucosinolate concentrations in nine genotypes tested in a three year trial (Farnham et al., 2004). Nearly 75% of the variation in the glucosinolate hydroxyglucobrassicin was due to environment. Significant environment × genotype interactions were observed as well. Effects due to genotype depended upon the individual glucosinolates. Shelp et al. (1993) reported that differences in glucosinolate concentrations in broccoli were greater between sites than between cultivars, indicating the degree to which glucosinolate content changes based growing conditions. Concentrations of individual glucosinolates varied widely between broccoli cultivars grown under identical conditions. Glucoraphanin concentrations were found to range from 0.8 $\mu mol\ g^{-1}$ DW in cv. EV6 to 21.7 $\mu mol\ g^{-1}$ DW in cv. Brigadier (Kushad, 1999).

Glucosinolates have garnered considerable interest in recent years due to their potential health benefits. Epidemiological data indicates that a diet rich in cruciferous vegetables may reduce the risk of certain cancers, especially those of the colon (Graham et al., 1978; Kohlmeier and Su, 1997) and prostate (Jain et al., 1999; Kolonel et al., 2000). Although their precise role is not yet fully understood, much of this effect is posited to be because their high glucosinolate content

2.13. Conclusion

Preemergence flame weeding has proven to be a valuable weed control tool for producers of many vegetable crops. Postemergence, selective flame weeding has the potential to be a valuable addition to the options available to vegetable producers. While studies have examined the response of many weed species to non-selective flaming methods, how this compares to selective methods is unknown. In addition, the tolerance of many crops to selective flame weeding has not been sufficiently determined. This study aims to advance the knowledge of the use of selective flame weeding in vegetable crops and contribute to providing a new weed control option for vegetable producers.

2.14 Connecting text

A slightly different version of the following chapter was published in Weed Technology (Weed Science Society of America, Allen Press Publishing) as "Weed Response to Flame Weeding at Different Developmental Stages" (Sivesind et al., 2009). The version included here has been modified to maintain consistent formatting and to reflect comments and suggestions of reviewers. The manuscript was co-authored by the candidate, Dr. Maryse Leblanc, Insitut de recherche et de développement en agroenvironnement, Dr. Daniel Cloutier, Institut de malherbologie, Dr. Philippe Seguin, Department of Plant Science, Macdonald Campus of McGill University, and Dr. Katrine Stewart, Department of Plant Science, Macdonald Campus of McGill University. The candidate carried out the experiments and data analyses and was the primary author of the manuscript. Drs. Leblanc and Cloutier provided funds, planned the experimental design, and reviewed the manuscript. Drs. Seguin and Stewart provided funds, supervisory guidance, and reviewed the manuscript.

3.0 Weed Response to Selective Flame Weeding at Different Developmental Stages

3.1 Abstract

Flame weeding is often used for weed control in organic production and other situations where use of herbicides is prohibited or undesirable. Response of several weeds to flame weeding was modeled using log-logistic equations. Dose-response curves were separately generated according to species and growth stage. Dicot species were more effectively controlled with flaming than monocot species. Common lambsquarters was susceptible to flaming, with doses required for 95% control (LD_{95}) ranging from 0.83 kg propane km^{-1} at the cotyledon stage to 2.85 kg propane km^{-1} for plants with 6 true leaves. LD_{95} values for redroot pigweed ranged from 1.19 to 2.72 kg propane km^{-1} for plants at the cotyledon and 4-leaf growth stage. Shepherd's-purse was 95% controlled with flame doses of 1.15 kg propane km^{-1} at the cotyledon stage and 2.78 kg propane km^{-1} at the 2-5 leaf stage. Monocot weeds were not able to be effectively controlled by any flame dose in this study, with survival greater than 50% at all growth stages evaluated for both barnyardgrass and yellow foxtail. Flame weeding can be an effective and labor-saving weed control method, the extent of which is partially dependent on the weed flora present. Knowledge of the local weed flora and their susceptibility to flame weeding is vital for the effective use of this method.

3.2 Introduction

Producers cite weed control as the most difficult problem they face when transitioning to organic production (Walz, 1999). Conventional agriculture makes widespread use of effective synthetic herbicides, which are prohibited under the rules of organic agriculture. Organic producers are forced to turn to other measures such as mechanical cultivation, which often are supplemented with laborious and costly hand-weeding. In less competitive crops such as onions, this added labor cost can be significant (Mojžiš, 2002). One way producers can attempt to reduce costs and labor requirements is through the use of flame weeding. Flame weeding is an allowed weed control option in organic production systems, often utilized prior to sowing as a stale seedbed technique or before crop emergence (Bond and Grundy, 2001). The latter method is often used with small seeded, slow-germinating crops such as onion and carrot (Ascard et al., 2007).

Directed flame weeding controls weeds in the crop row, as inter-row weeds can be effectively controlled through conventional mechanical methods (Melander, 1998). Intra-row weeds are more difficult to control as mechanical methods are ineffective or cause too much damage to the crop plants, especially early in the growing season. Many producers therefore are forced to rely on sometimes large amounts of hand-weeding. Hand-weeding can require a ready supply of field workers, and can be expensive for large areas or for less competitive crops that require multiple hand-weedings. The labor requirement for weeding the crop row by hand is considerable and can take as many as 200-300 hours per hectare in seeded onions (Ascard and Fogelberg, 2008). Flame weeding provides organic producers effective weed control in the crop row where cultivation is difficult and reduces the amount of costly hand-weeding.

Ascard (1994, 1995, 1997, 1998) conducted a comprehensive series of trials on the effectiveness of flame weeding. These studies evaluated the role of different biological factors on weed flora susceptibility, as well as technical aspects of the burner apparatus that had an effect on flame weeding efficacy. These studies utilized the type of system used for preemergent flaming; namely covered burners oriented parallel to the crop row. Cisneros and Zandstra (2008) evaluated the response of six weed species (three monocots and three dicots) to a covered flamer in a laboratory setting. Ascard (1994) constructed dose-response curves of various weed species according to plant size and density which demonstrated differences in susceptibility between species tested. Knezevic et al. have conducted field studies on the susceptibility of a number of dicot and monocot weed species to broadcast flame weeding using a flaming apparatus mounted on an ATV (Knezevic et al., 2008; Knezevic and Ulloa, 2007a; Knezevic and Ulloa, 2007b). These previous studies found that weed susceptibility to flaming varied among species and seedling size. In general, dicot species are reported to be controlled more effectively with flaming than monocot species.

Flaming can alternatively be used after crop emergence or planting in tolerant species. Flaming with crop plants present requires a different system, where uncovered, angled burners are staggered and set perpendicular to the crop row. Many of the recent studies on flame weeding have utilized a covered, parallel burner system as is used in preemergent flaming. It is unknown how well the results of those studies translate to the uncovered, cross-flaming burners required for application with crop plants present.

Experiments were conducted to evaluate the effect of flame weeding on a variety of weed species common to horticultural fields in south-western Québec under field conditions. A broad range of flaming intensities were tested on dicot and monocot weeds of differing maturity stages. A cross-flaming system as is used in selective postemergence flaming was used for flame treatments. Dose-response curves were then constructed in order to determine the correct dose to apply based on the weed flora present. Dose-response curves for weeds are important so that the lowest effective dose can be applied, which saves energy and results in lower production costs for the producer.

3.3 Materials and methods

3.3.1 Field management

Field experiments were conducted at the Institut de recherche et de développement en agroenvironnement (IRDA) in Saint-Hyacinthe, QC, Canada (45° 38' North, 72° 57' West) in 2005 and 2006. Four experiments were conducted over two years, each containing one of the following crops: onion (*Allium cepa* L. 'Vaquero'), broccoli (*Brassica oleracea* L. var. *Italica* 'Everest'), beet (*Beta vulgaris* L. 'Rosette'), and spinach (*Spinacia oleracea* L. 'Unipack 151'). For broccoli and onion experiments, the soil type was a Duravin loam; for spinach and beet the soil type was a St-Damase sandy loam.

Fields were fertilized as indicated by soil tests in accordance with local recommendations (Centre de Référence en Agriculture et Agroalimentaire du Québec, 2003). In 2005, onion received a broadcast application of 383 kg ha^{-1} of 14-20-24 NPK on May 17 and a banded application of 100 kg ha^{-1} 27-0-0 NPK on July 7. In 2006, onions received a broadcast application of 420 kg ha^{-1} of 13-11-22 NPK on May 28 and a banded application of 80 kg ha^{-1} of 27-0-0 NPK on July 6. Sixty day old onion transplants were planted May 18, 2005 and May 28, 2006 with 15 cm spacing. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

For the broccoli experiment in 2005, 573 kg ha^{-1} of 14-21-21 NPK and 1.5 kg ha^{-1} of boron was applied May 24, 2005, and banded with 100 kg ha^{-1} of 27-0-0 NPK on July 8. In 2006, broccoli received a broadcast of 685 kg ha^{-1} of 14-21-21 NPK and 1.5 kg ha^{-1} boron on May 29, and 80 kg ha^{-1} of 27-0-0 NPK was banded on July 6. Sixty day old broccoli transplants were planted May 25, 2005 and May 30, 2006 with 30 cm spacing between plants. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

Spinach was broadcasted with 346 kg ha^{-1} 23-8-23 NPK on May 31, 2005 and banded with 100 kg ha^{-1} 27-0-0 NPK on July 8, 2005. In 2006, spinach received a broadcast application of 465 kg ha^{-1} of 17-6-17 NPK on May 3, and a banded application of 80 kg ha^{-1} of 27-0-0 NPK on June 15. Spinach was direct seeded at a rate of 2.5 kg ha^{-1} with 4.5 cm spacing on June 1, 2005 and May 5, 2006. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

Beets received 608 kg ha^{-1} of 18-8-28 NPK and 1.5 kg ha^{-1} of boron broadcast at seeding in 2005 and 696 kg ha^{-1} of 15-7-18 NPK with 1.5 kg ha^{-1} boron in 2006. Beet was direct seeded at 5 kg ha^{-1} with 2.5 cm spacing on June 1, 2005 and May 8, 2006. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

3.3.2 Experimental design

In order to evaluate weed response at a range of plant developmental stages, flame treatments were applied at numerous points over the course of the growing season. Main plots were factorial combinations of flame dose and time point of flame treatment. There were twelve flame doses: 0.54, 0.68, 0.86, 0.90, 1.08, 1.18, 1.35, 1.43, 1.48, 1.97, 2.15, and 2.95 kg propane km^{-1}. Onion was flamed at five time points: 15, 21, 33, 40, or 49 days after transplantation (DAT) in 2005 and 9, 20, 34, 51, or 60 DAT in 2006. Broccoli received flame treatments at five time points: 14, 26, 33, 41, or 49 DAT in 2005 and 10, 20, 30, 41, or 50 DAT in 2006. Spinach and beet were each flamed at three growth stages: one preemergence and two postemergence at the 4- and 6-leaf stages. The onion and broccoli experiments each contained 244 plots (4 repetitions of 12 flaming doses × 5 time points + 1 control). The spinach and beet experiments each contained 148 plots (4 repetitions of 12 flaming doses × 3 stages + 1 control). Experimental plots received only a single flame treatment at the designated maturity stage.

Quadrats (20 × 50 cm) were placed along the center of each plot before flame treatments and weeds were recorded according to species and maturity stage.

Between one and three days following flame treatment, quadrats were reassessed for weed survival. To eliminate difficulties in assessment and to control for contamination of the data by post-treatment weed emergence, in 2006 quadrats were replaced with tagging of individual weeds. Weeds along the crop row were marked by placing a metal marker around, but at a distance from the base of the plant so as not to affect the response. Surrounding weed flora was then removed. In 2006 it was decided to limit evaluation of weed response to four weed species in order to ensure sufficient weed numbers for accurate data. Subsequently, each of the four blocks was seeded with one of four weed species: redroot pigweed (*Amaranthus retroflexus* L.), common lambsquarters (*Chenopodium album* L.), barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.), or yellow foxtail (*Setaria pumila* (Poir.) Roemer & J.A. Schultes). Weed response data was not collected from the beet experiment in 2006.

3.3.3 Flaming specifications

Flame treatments were performed using a tractor mounted Red Dragon flaming system utilizing two unshielded model LT 1 ½ × 6 Liquid Torch liquid phase burners (Flame Engineering, Inc., LaCrosse, KS) directed perpendicularly to the crop row (Figure 3.1). Burners were staggered to avoid flames intersecting and deflecting upwards. Burners were set at an angle of 30° with respect to the horizontal 18 cm from the row measured along the angle. Flaming dose treatments were controlled by fuel pressure and tractor driving speed. Three pressures (138, 241, and 345 kPa in 2005; 117, 214, and 310 kPa in 2006) and four driving speeds (2, 3, 4, and 5 km h^{-1}) were combined to yield the 12 flame doses. The amount of propane burned per hour was recorded for each fuel pressure and used to convert treatments to a linear scale of kilograms of propane consumed per kilometer of treated row. Fuel pressures were different in 2006 than in 2005 because a fuel regulator had to be replaced between the two years. In order to account for differences between regulators, fuel pressures were modified to maintain propane consumption rates equal year to year, thus ensuring consistent flaming doses.

3.3.4 Flaming dose calculation

Flame weeding doses are often given in terms of mass of fuel used per area of coverage (e.g. kg propane ha^{-1}). In situations utilizing non-selective systems using covered burners oriented parallel to crop row, the width of coverage is assumed to be the width of the burner cover. The flaming dose per area can then either be represented as coverage of the entire field area, or else as the actual fuel usage per hectare flamed. Presenting doses on a broadcast basis is difficult when using non-covered cross-flamers, as the width of coverage would be somewhat arbitrarily decided as coverage would decrease gradually with distance from the crop row. Care must be taken with the latter approach as well, as any dosage given is dependent upon row spacing and must be converted if row spacing is not consistent. For this reason, we have decided to present the flaming doses used in this study as propane burned per unit row length (i.e. kg propane km^{-1}). We feel this is prudent as it accurately represents the fuel used, and would be simple to accurately compare dosages used in separate studies by authors using different equipment. Also, this avoids the problem of determining exact width of coverage for uncovered cross-flamers. In order to determine the actual amount of fuel required for a given field, all that is required is to multiply the linear rate by 10 and divided by the row width in meters. For example, in this study we used a row spacing of 0.90 meters, so a dose of 0.9 kg propane km^{-1} would be equal to 10 kg propane ha^{-1}. For a row spacing of 0.60 meters, this same example would be equal to 15 kg propane ha^{-1}. This approach simplifies the comparison of rates used by different parties, and makes it easy to calculate the actual amount of fuel that is required for any given field.

3.3.5 Statistical analyses

Regression analyses were performed using GraphPad Prism v. 5.02 (GraphPad Software, Inc., La Jolla, CA). Percent weed survival was initially regressed over flame dose using a four-parameter log-logistic equation (Seefeldt et al., 1995) as shown in equation 1:

$$y = (D - C)/(1 + \exp[b*(\log(x) - \log(LD_{50}))]) + C \qquad [1]$$

where y is the percent survival, x is the flame dose, C is the lower asymptote, D is the upper asymptote, LD_{50} is the effective dose that reduced response 50%, and b is the slope of the curve at the LD_{50}. As the dependent variable in this study is on a percentage scale, the upper asymptote can reasonably be set at 100, resulting in equation 2.

$$y = (100 - C)/(1 + \exp[b*(\log(x) - \log(LD_{50}))]) + C \qquad [2]$$

If the lower asymptote C is equal to zero, then the four-parameter model is reduced to a three-parameter model as shown in equation 3.

$$y = 100/(1 + \exp[b*(\log(x) - \log(ED_{50}))]) \qquad [3]$$

Furthermore, equation 2 can be re-parameterized to incorporate values other than the LD_{50} (Schabenberger et al., 1999):

$$y = (100 - C)/(1 + w*\exp[b*(\log(x) - \log(ED_k))]) + C \qquad [4]$$

$$w = k/(100 - k)$$

where k is the response of interest. For example, k = 95 if the dose that results in 95% response is of interest. Note that if k = 50, the added term w = 1 and equation 4 is reduced to equation 2. Equation 3 can be similarly re-parameterized, resulting in equation 5.

$$y = (100)/(1 + w*\exp[b*(\log(x) - \log(ED_k))]) \qquad [5]$$

$$w = k/(100 - k)$$

Dose response curves were generated separately for each weed species and growth stage. Growth stages among weed species tended to be highly clustered within flaming time points, as earlier time points contained weeds at earlier growth stages, while later time points contained more advanced weeds (e.g., data on the response of cotyledon weeds was collected at the first time point, whereas the response of 6-leaf weeds occurred at a later time point). Therefore, within each experiment weed data was combined across time points within repetitions.

Dose response curves were independently generated using replicate data for each experiment (crop). Extra-sum-of-squares *F*-tests at the 5% significance level were used to test whether equation 4 or equation 5 best described the data. Extra-sum-of-squares *F*-tests were then used to test whether a single dose response curve could be made to describe the data across all experiments or if response curves differed among experiments (Seefeldt et al., 1995). If *F*-tests indicated a single response curve could be made across all experiments, data from each experiment was pooled and a single dose-response curve was generated for the entire data set. Lack-of-fit *F*-tests were used to check the fit of the model to the data. LD_{50} and LD_{95} values of different growth stages were compared within a weed species using 95% confidence intervals.

3.4 Results and discussion

Weed response to flaming varied and was dependent upon species and maturity stage. Extra-sum-of-squares *F*-tests indicated that dose response curves did not differ between experiments for any weed species; therefore a single dose response curve was generated for each weed species and growth stage across experiments. The flaming doses used in this study were in the appropriate range with weed survival, depending on species and growth stage, varying from 100% to complete kill.

Equation 5 was preferred over equation 4 for all growth stages of dicot weeds according to extra-sum-of-squares *F*-tests. Lack-of-fit *F*-tests were not significant for any of the curves tested at the 95% confidence level, indicating that the model was appropriate for the data.

Common lambsquarters response to flame weeding was successfully modeled by equation 5 for the cotyledon through the greater-than-6-leaf stage. As common lambsquarters development progressed, increased flaming doses were required for comparable levels of control (Figure 3.1). LD_{50} values for common lambsquarters increased from 0.37 kg propane km^{-1} at the cotyledon stage to 1.05 kg propane km^{-1} at the greater-than-6-leaf stage, though LD_{50} values for the 4- and 6-leaf growth stages did not differ based on 95% confidence intervals (Table 3.1). LD_{95} values increased from 0.83 kg propane km^{-1} at the cotyledon stage to 2.85 kg propane km^{-1} for 6-leaf lambsquarters.

Overall, the response of redroot pigweed was similar to that of common lambsquarters. Equation 5 accurately described the response up to the six-leaf stage (Figure 3.2). Response of redroot pigweed to flame weeding in general shifted to the right as developmental stage increased, although some overlap was observed. Weeds at the cotyledon stage were controlled with moderate doses (LD_{50} = 0.32 kg propane km^{-1}), while successively higher doses were required for similar levels of control in more advanced stages (Table 3.1). LD_{50} values increased from 0.32 kg propane km^{-1} at the cotyledon stage to 0.97 kg propane km^{-1} for 6-leaf redroot pigweed. Based on 95% confidence intervals, LD_{50} values for adjacent growth stages were often similar (Table 3.1), but differences were observed when comparing growth stages of greater disparity (e.g., 1-leaf and 3-leaf redroot pigweed). LD_{95} values for redroot pigweed at the cotyledon, 3-leaf, and 6-leaf growth stages were 1.19, 2.36, and 2.41 kg propane km^{-1}. Few differences between LD_{95} values for the different growth stages were observed in redroot pigweed based on 95% confidence intervals (Table 3.1).

Response of shepherd's-purse decreased as flame dose increased (Figure 3.3). Shepherd's-purse had a higher LD_{50} value at the cotyledon stage (0.58 kg propane km^{-1}) than common lambsquarters (0.37 kg propane km^{-1}) and redroot pigweed (0.32 kg propane km^{-1}) (Table 3.1). This is likely due at least in part to shepherd's-purse being in rosette form at this point in its growth and close to the ground, as opposed to the upright growth habit of common lambsquarters and redroot pigweed. Evaluation of other species with similar growth forms is necessary to examine this further. At the 2-5 leaf stage, a flaming dose of 0.85 kg propane km^{-1} was required to reduce plant density by 50% (Table 3.1). Flaming doses required for control of shepherd's-purse were in the same range of those required for similar levels of control of common lambsquarters and redroot pigweed. This is despite the fact that shepherd's purse is generally smaller at these growth stages than common lambsquarters and redroot pigweed plants. The small size and rosette growth form of shepherd's-purse may help some individuals avoid flame damage, thus increasing overall survival.

The response of the two monocot species examined in this study to flame weeding was quite different from what was observed in the dicot species. Flame dose was not a good predictor of weed survival for either barnyardgrass or yellow foxtail and response could not be accurately described by the log-logistic equation. Neither barnyardgrass nor yellow foxtail was able to be effectively controlled at any flaming rate tested. For barnyardgrass, mean survival at all stages was greater than 75% for all flaming doses tested (Figure 3.4). Yellow foxtail was controlled somewhat more effectively, but control was still poor as mean survival exceeded 50% for all growth stages and flaming doses (Figure 3.5). The low levels of control observed in the two monocot species studied were not due to tolerance to the flame treatment. Rather, the high survival rate was due to these species' much greater ability to recover following flaming. In earlier maturity stages (e.g., 1-2 leaves) higher flaming rates killed nearly all above ground tissue. However, after 2-3 days, visible regrowth would occur. This was due to the meristem in monocots being located near or below ground level protecting it from flame damage. Additionally, the growing point is surrounded by a protective sheath of leaves, further protecting it from damage. These phenomena can result in an increased percentage of weed flora being monocot species in the weeks following flame treatment, as dicots are largely killed and monocots survive.

The results of this study largely agree with data previously reported in the literature. Wszelaki et al. (2007) reported that monocots and weeds with fleshy leaves were more difficult to control with flaming than most dicot species. Ascard (1995) divided weed species into four groups based on susceptibility to flame weeding. The most susceptible species were those with unprotected meristems and thin leaves, such as common lambsquarters and common chickweed [*Stellaria media* (L.) Vill.]. Redroot pigweed and common lambsquarters would be placed in this category, and in this study were able to be effectively controlled through the 4- and 6-leaf stages, respectively. The final group contained the least susceptible species, which were not able to be controlled with a single flame treatment. The only species in Ascard's study that was included in this final group was annual bluegrass (*Poa annua* L.), though the author notes that other monocot species may be expected to belong to this group as well. The results presented here suggest that the monocot species tested, barnyardgrass and yellow foxtail, do in fact belong in this grouping. Other studies, however, have reported greater control of grass species than we were able to obtain in our study. Knezevic and Ulloa (2007b) reported up to 80% control of barnyard grass and green foxtail from flaming, though flame doses required were double that required to obtain equivalent control in dicot species. One possible explanation for the discrepancy between our study and the results reported by Knezevic and Ulloa is that in their study response is based upon visual injury ratings, while our study measured weed survival. The capacity of grasses for regrowth has been well documented (Ascard, 1994). Biomass of grass weeds may be dramatically diminished in the days following flame treatment without resulting in extensive weed mortality.

Dose-response curves of weeds to flame weeding had previously been explored in a series of experiments carried out by Ascard (1994, 1995). This study was designed in part to confirm and expand on the results of those studies. Weeds were flamed at a greater number of specific growth stages in order to achieve a more exact picture of the dose-response relationship. In this study, rates required to achieve 95% control were found to be 1.25 kg propane km^{-1} for common lambsquarters at the 2-leaf stage, and 2.78 kg propane km^{-1} for shepherd's-purse in the 2-5 leaf stage. Ascard (1995) was able to achieve 95% control of common lambsquarters and shepherd's-purse at rates of 0.4 and 0.7 kg propane km^{-1}, respectively, for common lambsquarters and shepherd's-purse at the 2-leaf growth stage (using a 20 cm width of coverage for both studies). The considerably lower flaming rates required for equivalent control in the latter study are likely due to the use if an insulated cover, which retained heat and improved efficiency. Although the rates can't be directly compared due to differences in methodological and technical aspects of these experiments (e.g., uncovered cross-flaming system in current study compared to a parallel, covered apparatus used by Ascard), the trends present between species in both studies are similar. Monocot species were not effectively controlled in either study. Ascard (1995) noted that 100% control of annual bluegrass was not achieved at either the 1-2 leaf stage or the greater than 6-leaf stage. The lower limit of survival of annual bluegrass was found to be 31%. In addition, increased emergence of annual bluegrass was observed after treatment with higher flaming doses. In the current study, barnyardgrass mean survival was greater than 75% for all flame doses and growth stages, and mean survival of yellow foxtail was never less than 50%. Any changes in emergence patterns were not recorded, as we evaluated only weeds that were present prior to flame treatments.

Cisneros and Zandstra (2008) evaluated the effectiveness of flame weeding on several weed species in a laboratory setting using a parallel mounted, covered flaming system. They reported greater variability in response between monocot species than was found in our study. The authors reported little control in numbers of barnyardgrass at either the 0-2 leaf or the 2-4 leaf stage (even an increase in numbers reported in all treatments), regardless of flaming intensity. However, a substantial reduction in plant biomass 14 days after treatment was observed in all treatments.

Although control was somewhat better in our study, these results are largely in agreement with the results of our study, where satisfactory control of barnyardgrass was never attained. Substantially better control was observed with green foxtail [*Setaria viridis* (L.) Beauv.] where stand reductions of 70-99% were observed at the 0-2 leaf stage depending upon flaming dose. Though no differences were seen between treatments, all were significantly lower than the control. At the 2-4 leaf stage, reductions of 14-77% as compared to the non-flamed control were observed depending upon flaming dose. A significant reduction in plant numbers of large crabgrass [*Digitaria sanguinalis* (L.) Scop.] at both the 0-2 and 2-4 leaf stage was only achieved with the highest dose tested (49 and 32% reductions, respectively). All treatments resulted in reduced fresh weights 14 days after treatment in the 0-2 leaf stage. At the 2-4 leaf stage, no significant reductions in fresh weight were observed in any treatment. Much greater differences were seen in responses to flaming between monocot species in their study than we found in ours. The reason for this discrepancy is unclear. Cisneros and Zandstra also examined three dicot weeds: redroot pigweed, common ragweed (*Ambrosia artemisiifolia* L.), and common lambsquarters. At the 0-2 leaf stage, the authors reported no differences between treatments in reductions of plant stands, but all were significant from the untreated control (92, 82, and 93%, respectively, averaged over treatments). At the 2-4 leaf stage, reductions of 95, 93, and 99% were observed, respectively, when averaged over treatments. Of note is the fact that for common lambsquarters and to a greater extent common ragweed, flaming was more effective on plants with 2-4 leaves than those with 0-2 leaves, whereas in redroot pigweed, flaming was approximately of equal effectiveness with either stage in three of the four treatment levels tested. The authors speculated that the reason for this could be a larger surface area of the more mature seedlings in which the flame to contact. This observation contradicts the results of this study, which found, with few exceptions, a steady decrease in the effectiveness level of a particular flaming dose on dicot weeds of increasing maturity. It should be noted, however, that Cisneros and Zandstra found this phenomenon to be most pronounced in common ragweed, which was not included in our study.

3.5 Summary and conclusions

This study was designed to construct dose-response curves for a number of weed species common to horticultural fields in a field environment, utilizing a cross-flaming system appropriate for use when crop species are present. The range of flaming doses used in this study were appropriate for the construction of response curves for the dicot species, with weed survival ranging from 0 to 100% depending upon species and plant development. The similarity in response of common lambsquarters and redroot pigweed to flame weeding suggests that weeds of similar morphology and growth form should be able to be controlled at comparable flame doses. Previous reports support this hypothesis (Ascard, 1994, Cisneros and Zandstra, 2008; Knezevic et al., 2008). The monocot species evaluated were not able to be effectively controlled with any flaming dose tested, as mean control of yellow foxtail and barnyardgrass never exceeded 50 and 30%, respectively. The data generated overall agrees with the information available in the literature, with upright, dicot species more easily controlled than monocot species. The information provided in this study should further the understanding of weed response to flame weeding and help producers to more effectively utilize this weed control tool.

3.6 Acknowledgements

The authors would like to thank Germain Moreau, field technicians, and summer students for assistance with field operations and data collection. This research was supported by a grant from the Conseil des Recherches en Pêche et Agroalimentaire du Québec (CORPAQ).

3.7 Tables and figures

Table 3.1. Predicted values and standard errors of regression parameters for models describing the response of selected weed species at different growth stages to flame weeding treatments. Data is from four experiments in 2005 (beet, broccoli, onion, and spinach) and three experiments in 2006 (broccoli, onion, and spinach). Data for the response of shepherd's-purse to flame weeding collected in 2005 only. Regression parameters were estimated using equation 5.

		Regression parameters (± SE)				
Weed species	Growth stage	b [a]	LD_{50}		LD_{95}	
Common lambsquarters	cotyledon	3.56 ± 0.43	0.37 ± 0.02	a[b]	0.83 ± 0.04	a
	2 leaves	3.35 ± 0.45	0.52 ± 0.03	b	1.25 ± 0.11	b
	4 leaves	3.66 ± 0.42	0.69 ± 0.02	c	1.55 ± 0.13	bc
	6 leaves	2.25 ± 0.41	0.77 ± 0.06	c	2.85 ± 0.59	c
	> 6 leaves	2.16 ± 0.46	1.05 ± 0.08	d	4.10[c] ± 1.16	c
Redroot pigweed	cotyledon	2.23 ± 0.56	0.32 ± 0.07	a	1.19 ± 0.25	a
	1 leaf	3.13 ± 0.68	0.56 ± 0.05	b	1.43 ± 0.23	ab
	2 leaves	1.97 ± 0.40	0.61 ± 0.07	bc	2.71 ± 0.64	ab
	3 leaves	2.55 ± 0.40	0.74 ± 0.05	cd	2.36 ± 0.37	ab
	4 leaves	2.68 ± 0.39	0.91 ± 0.05	de	2.72 ± 0.41	b
	6 leaves	3.25 ± 0.66	0.97 ± 0.06	e	2.41 ± 0.43	ab
Shepherd's-purse	cotyledon	4.34 ± 0.92	0.58 ± 0.04	a	1.15 ± 0.14	a
	2-5 leaves	2.48 ± 0.49	0.85 ± 0.06	b	2.78 ± 0.59	b

[a] b, the slope of the curve at the LD_{50}; LD_{50}, the dose giving a 50% response; LD_{95}, the dose giving a

[b] LD_{50} and LD_{95} values followed by the same letter within a weed species do not differ based on 95% confidence intervals.

[c] Uncertain estimate; out of range of observed values.

Figure 3.1. Response of common lambsquarters seedlings at the cotyledon, 2-leaf, 4-leaf, 6-leaf, and greater-than-6-leaf growth stages to a single flame weeding treatment. Flame doses ranged from 0.54 to 2.95 kg propane km^{-1}. Weed response measured as percent of plants that survive flame weeding, as determined one to three days following flame treatment. Regressions constructed using equation 5. Parameter estimates are found in Table 3.1. Points represent mean values of data from four experiments in 2005 and three experiments in 2006. Error bars represent one standard error of the mean (SE).

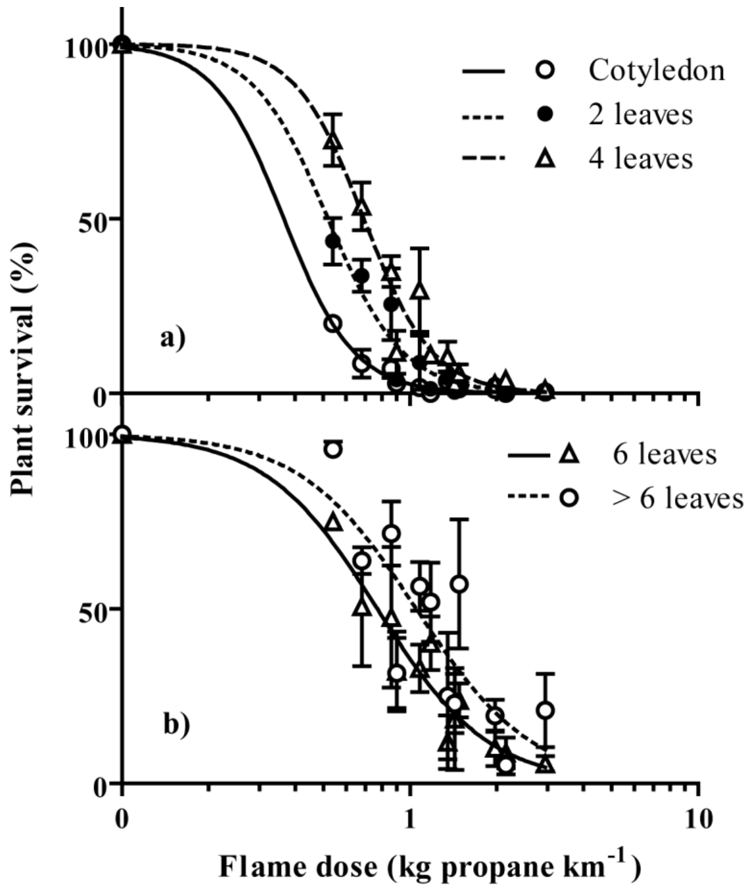

Plant survival (%)
100
50
0
Cotyledon
2 leaves
4 leaves
a)
6 leaves
> 6 leaves
b)
0
1
10
Flame dose (kg propane km-1)

Figure 3.2. Response of redroot pigweed seedlings at the cotyledon, 1-leaf, and 2-leaf growth stages (a) and at the 3-leaf, 4-leaf, and 6-leaf growth stages (b) to a single flame weeding treatment. Flame doses ranged from 0.54 to 2.95 kg propane km^{-1}. Weed response measured as percent of plants that survive flame weeding, as determined one to three days following flame treatment. Regressions constructed using equation 5. Parameter estimates are found in Table 3.1. Points represent mean values of data from four experiments in 2005 and three experiments in 2006. Error bars represent one standard error of the mean (SE).

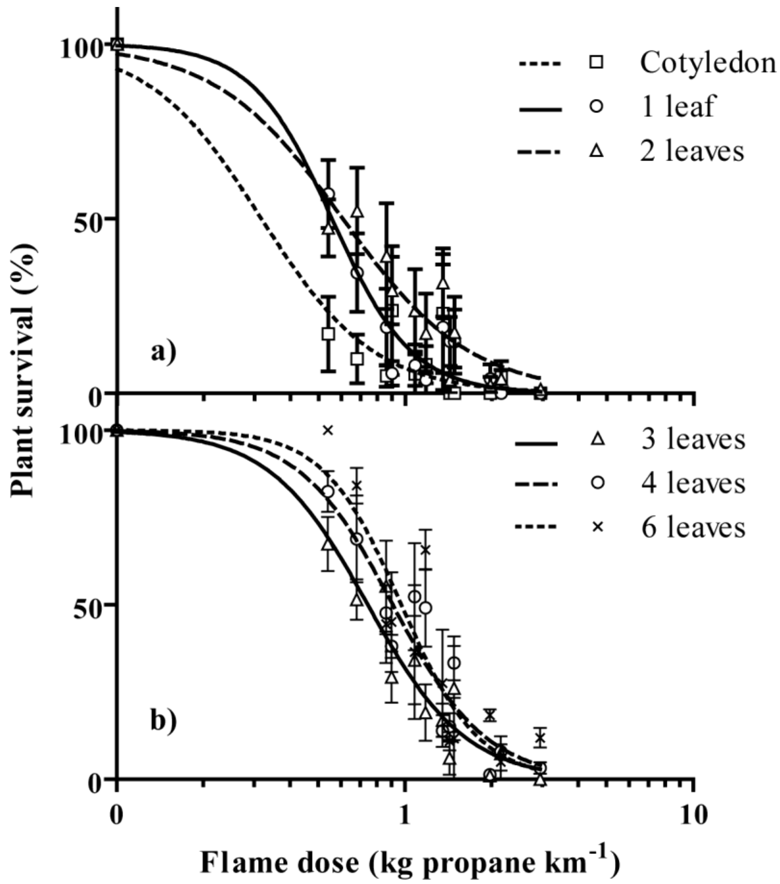
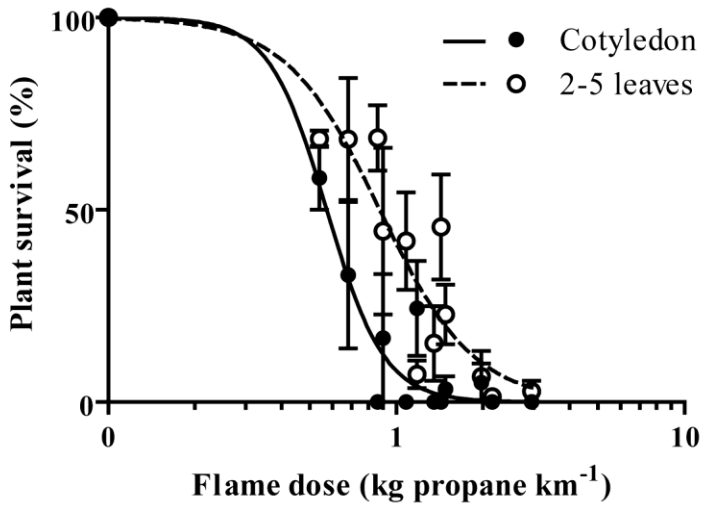
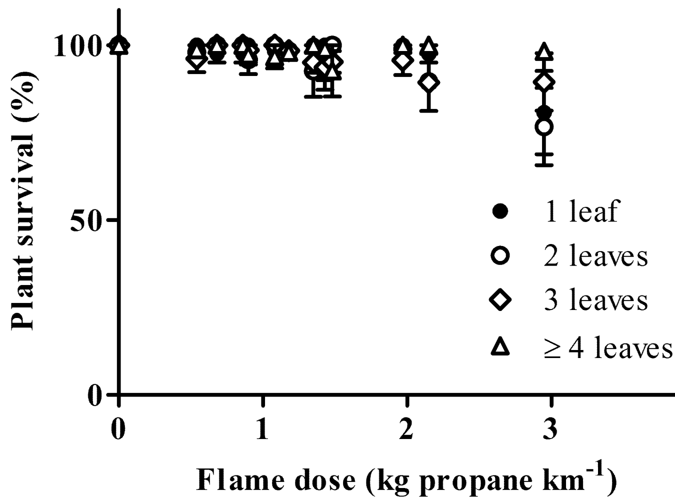

Plant survival (%)
100
50
0
Cotyledon
1 leaf
2 leaves
a)
3 leaves
4 leaves
6 leaves
b)
0
1
10
Flame dose (kg propane km-1)

Figure 3.3. Response of shepherd's-purse seedlings at the cotyledon and 2-5 leaf growth stages to a single flame weeding treatment. Flame doses ranged from 0.54 to 2.95 kg propane km^{-1}. Weed response measured as percent of plants that survive flame weeding, as determined one to three days following flame treatment. Regressions constructed using equation 5. Parameter estimates are found in Table 3.1. Points represent mean values of data from four experiments in 2005. Error bars represent one standard error of the mean (SE).



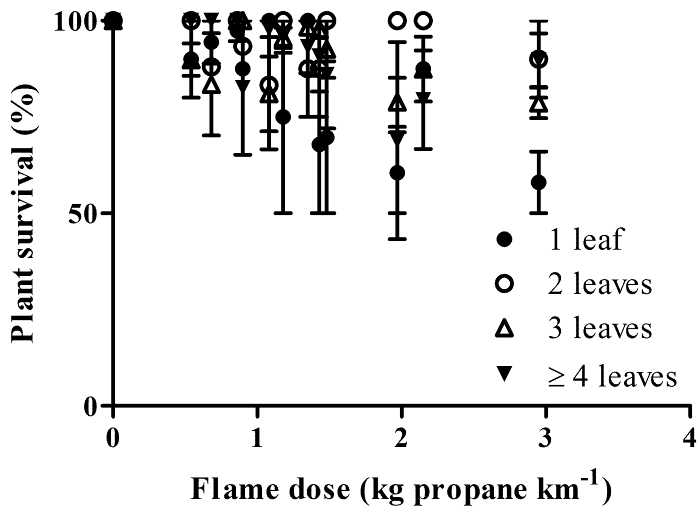
100
50
0
Plant survival (%)
0
1
10
Flame dose (kg propane km-1)
Cotyledon
2-5 leaves

Figure 3.4. Response of barnyardgrass seedlings with one leaf to greater-than-four leaves to a single flame weeding treatment. Flame doses ranged from 0.54 to 2.95 kg propane km^{-1}. Weed response measured as percent of plants that survive flame weeding, as determined one to three days following flame treatment. Points represent mean values of data from four experiments in 2005 and three experiments in 2006. Error bars represent one standard error of the mean (SE).



100
50
0
Plant survival (%)
0
1
2
3
4
Flame dose (kg propane km-1)
1 leaf
2 leaves
3 leaves
≥ 4 leaves

Figure 3.5. Response of yellow foxtail seedlings with one leaf to greater-than-four leaves to a single flame weeding treatment. Flame doses ranged from 0.54 to 2.95 kg propane km^{-1}. Weed response measured as percent of plants that survive flame weeding, as determined one to three days following flame treatment. Points represent mean values of data from four experiments in 2005 and three experiments in 2006. Error bars represent one standard error of the mean (SE).


Plant survival (%)
100
50
0
0
1
2
3
4
Flame dose (kg propane km-1)
1 leaf
2 leaves
3 leaves
≥ 4 leaves

3.8 Connecting text

In the previous study, response to flame weeding was modeled for a number of weeds common to horticultural fields in Québec. Results suggest that some weeds can be effectively controlled with flame weeding, with response dependent upon flame dose, weed species, and morphological development. However, the tolerance of many crop species to flame weeding has not been well established. The following study was conducted in order to evaluate four vegetable crops as possible candidates for use in flame weeding programs, and to compare the flaming rates these crops could withstand with those required to effectively control weeds.

The manuscript was co-authored by the candidate, Dr. Maryse Leblanc, Insitut de recherche et de développement en agroenvironnement, Dr. Daniel Cloutier, Institut de malherbologie, Dr. Philippe Seguin, Department of Plant Science, Macdonald Campus of McGill University, and Dr. Katrine Stewart, Department of Plant Science, Macdonald Campus of McGill University. The candidate conducted the experiments and data analyses and was the primary author of the manuscript. Drs. Leblanc and Cloutier provided funds, planned the experimental design, and reviewed the manuscript. Drs. Seguin and Stewart provided funds, supervisory guidance, and reviewed the manuscript.

4.0 Response of Onion, Broccoli, Spinach, and Beet to Selective Flame Weeding

4.1 Abstract

Field experiments were conducted to evaluate the use of pre- and postemergence flame weeding in onion, broccoli, spinach, and beet. Treatments consisted of a single flame weeding treatment at one of 12 flaming doses ranging from 0.54 to 2.95 kg propane km^{-1}. Flame weeding treatments were applied at one of five times throughout the season for onion and broccoli and at one of three growth stages for spinach and beet. Yield parameters were recorded and regressed against flame dose using log-logistic equations. Onion and broccoli yields were only affected when flamed less than 15 days after transplantation (DAT), with 10% yield loss occurring in onion at a dose of 1.78 kg propane km^{-1} and in broccoli at a dose of 1.19 kg propane km^{-1}. Preemergence flaming did not affect yields in either spinach or beet. However, yields in both spinach and beet were reduced by flame weeding at either the 4- or 6-leaf growth stages. A 10% loss in total spinach yields occurred from flame doses of 0.75 and 0.68 kg propane km^{-1} applied at the 4- and 6-leaf growth stages, respectively. Total beet yields were reduced 10% at the 4-leaf by flame doses of 0.98 and 0.56 kg propane km^{-1} in 2005 and 2006, respectively. Total beet yields were reduced by 10% at the 6-leaf stage by flame doses of 0.83 and 1.25 kg propane km^{-1} in 2005 and 2006. Onion and broccoli were tolerant of flame weeding at most time points evaluated. Based on the results of this study, only preemergence flame weeding can be recommended for spinach and beet.

4.2 Introduction

Weed control options available to producers under the rules of organic agriculture are limited. To control weeds, producers may utilize crop rotation, mulches, and a relatively small number of allowable contact natural herbicides. Once weeds have emerged, a large percentage of weed control is achieved by mechanical cultivation and through hand-weeding. One alternative to mechanical cultivation and hand-weeding for inter-row weeds is flaming weeding.

Flame weeding is commonly used in organic production for non-selective, preemergence control of weeds in row crops such as carrots and onions (Ascard et al., 2007). There are many advantages of flame weeding. Unlike mechanical cultivation, flaming does not stir up the soil, which can lead to increased germination and weed emergence. Again unlike repeated mechanical cultivations, flaming does not have detrimental effects on soil texture and organic matter. For organic producers, one of the most attractive aspects of flame weeding is the possibility to eliminate or reduce the amount of costly hand-weeding that they must employ. The amount of hand-weeding required is dependent upon the level of weed pressure and the competitive ability of the crop, but can increase production costs dramatically (Mojžiš, 2002). The most common use of flame weeding in row crops has been in non-selective situations, either before planting or preemergence of the crop. Accordingly, most flame weeding research has focused on non-selective flaming and aspects related to weed susceptibility. Studies have been undertaken that examine the effect of plant density (Ascard, 1994), and fuel pressure and burner setup (Ascard, 1997) on weed susceptibility. Investigations of flaming doses on different species and maturity stages have been conducted in a laboratory setting (Cisneros and Zandstra, 2008) and produced dose-response curves by testing in the field (Ascard, 1995; Knezevic and Ulloa, 2007b; Sivesind et al., 2009).

Alternatively, flame weeding can be used postemergence, in a selective manner using cross flamers. This is a possibility if crop plants are sufficiently tolerant of flame applications. Selective flaming could be very beneficial to producers, as mechanical cultivation of the crop row can be difficult and can cause damage to plants and their roots if cultivation is too deep. There is relatively little concrete information available in the literature regarding the use of selective flaming for many crop species. In the 1960's the petroleum industry sponsored research on a wide range of potential uses for flame weeding. Investigations into the use of flaming on cole crops (Wilson and Ilnicki, 1966), grapes (Hansen et al., 1966), corn (Lalor and Buchele, 1966; Lien and Liljedahl, 1966; Reece et al., 1966), sorghum (Reece et al., 1966) and soybeans (Lalor and Buchele, 1966) were undertaken. After this initial flurry of activity, flaming research slowed down for a number of years. After little activity in flaming research in the 1970's, flaming research resumed in Europe in the 1980's. Vester (1988) selectively flamed onions, sweet corn, kale, beet, and potatoes. Ascard (1989) compared pre- and postemergence flaming to mechanical and chemical control in set and seeded onions. Field trials comparing the use of flame weeding to band spraying in cabbage were conducted (Holmøy and Netland, 1994; Netland et al., 1994). Wszelaki et al. (2007) investigated the effects of selective flaming on transplanted cabbage (*Brassica oleracea* var. *capitata*) and tomato (*Solanum lycopersicum* L.). Recently, research into selective flaming in winter wheat (Ulloa et al., 2008), soybean (Knezevic and Ulloa, 2007b; Knezevic and Ulloa, 2007c; Heverton et al., 2008), corn (Knezevic and Ulloa, 2007b; Knezevic and Ulloa, 2007c; Heverton et al., 2008), sorghum, sunflower, alfalfa, and red clover (Knezevic and Ulloa, 2007c) have been conducted.

The following study was conducted to evaluate the potential for the use of flame weeding in four vegetable crops: onion (*Allium cepa* L.), broccoli (*Brassica oleracea* L. var. *italica*), spinach (*Spinacia oleracea* L.), and beet (*Beta vulgaris* L.). A range of flaming doses were applied at different points during the growing season. Single flame treatments were applied in order to determine how crop tolerance to flame weeding changed as plants developed over the course of the growing season.

4.3 Materials and methods

4.3.1 Field management

Field experiments were conducted at the Institut de recherche et de développement en agroenvironnement (IRDA) in Saint-Hyacinthe, QC, Canada (45° 38' North, 72° 57' West) in 2005 and 2006. Four experiments were conducted over two years, each containing one of the following crops: onion, broccoli, beet, and spinach. For broccoli and onion experiments, the soil type was a Duravin loam; for spinach and beet the soil type was a St-Damase sandy loam.

Experiments were fertilized according to local recommendations and based on results of soil tests (Centre de Référence en Agriculture et Agroalimentaire du Québec, 2003). The onion experiment was broadcasted with 383 kg ha^{-1} 14-20-24 NPK on May 17 and banded with 100 kg ha^{-1} 27-0-0 NPK on July 7. In 2006 onions were broadcasted with 420 kg ha^{-1} of 13-11-22 NPK fertilizer on May 28 and banded with 80 kg ha^{-1} of 27-0-0 NPK on July 6. Sixty day old onion transplants (cv. Vaquero) were planted May 18, 2005 and May 28, 2006 with 15 cm spacing. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

The broccoli experiment was broadcasted with 573 kg ha^{-1} of 14-21-21 NPK and 1.5 kg ha^{-1} of boron on May 24, 2005, and banded with 100 kg ha^{-1} of 27-0-0 NPK on July 8, 2005. In 2006, broccoli received a broadcast of 685 kg ha^{-1} of 14-21-21 NPK and 1.5 kg ha^{-1} boron on May 29, and 80 kg ha^{-1} of 27-0-0 NPK was banded on July 6. Sixty day old broccoli transplants (cv. Everest) were planted May 25, 2005 and May 30, 2006 with 30 cm spacing between plants. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

Spinach was broadcasted with 346 kg ha^{-1} 23-8-23 NPK on May 31, 2005 and banded with 100 kg ha^{-1} 27-0-0 NPK on July 8, 2005. In 2006, spinach received a broadcast application of 465 kg ha^{-1} of 17-6-17 NPK on May 3, and a banded application of 80 kg ha^{-1} of 27-0-0 NPK on June 15. Spinach (cv. Unipack 151) was direct seeded at a rate of 2.5 kg ha^{-1} with 4.5 cm spacing on June 1, 2005 and May 5, 2006. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

The beet field in 2005 received 608 kg ha^{-1} of 18-8-28 NPK and 1.5 kg ha^{-1} of boron broadcast at seeding and in 2006 was broadcast 696 kg ha^{-1} of 15-7-18 NPK with 1.5 kg ha^{-1} boron on May 8. Beet (cv. Rosette) was direct seeded at 5 kg ha^{-1} with 2.5 cm spacing on June 1, 2005 and May 8, 2006. Plots consisted of a single row 2.5 m in length with 0.90 m between adjacent rows.

4.3.2 Experimental design

All experiments were set up in randomized complete block design (RCBD) with four replications. Main plots were factorial combinations of flame dose and time point of flame treatment. There were twelve flame doses: 0.54, 0.68, 0.86, 0.90, 1.08, 1.18, 1.35, 1.43, 1.48, 1.97, 2.15, and 2.95 kg propane km^{-1}. Onion was flamed at five time points: 15, 21, 33, 40, or 49 days after transplantation (DAT) in 2005 and 9, 20, 34, 51, or 60 DAT in 2006. These time points corresponded to BBCH stages of 12 (2 leaves), 13-14 (3 to 4 leaves), 15-16 (5 to 6 leaves), 17 (7 leaves), and 19 (9-10 leaves), respectively, in 2005 (Feller et al., 1995). The number of leaves at flaming was not recorded for 2006. Broccoli received flame treatments at five time points: 14, 26, 33, 41, or 49 DAT in 2005 and 10, 20, 30, 41, or 50 DAT in 2006. These time points corresponded to BBCH stages of 15-16 (5 to 6 leaves), 19 (9 or more leaves; 9 leaves), 19 (9 or more leaves; 10 leaves), 19 (9 or more leaves; 11 leaves), and 19 (9 or more leaves; 12 leaves), respectively, in 2005. The number of leaves at flaming was not recorded for 2006. Spinach and beet were each flamed at three growth stages: one preemergence and two postemergence at the 4- and 6-leaf stages. The 4- and 6-leaf stages correspond to 14 and 16 on the BBCH-scales for spinach and beet. For all experiments, one control plot was included that received no flame treatment. The onion and broccoli experiments each contained 244 plots (4 repetitions of 12 flaming doses × 5 time points + 1 control). The spinach and beet experiments each contained 148 plots (4 repetitions of 12 flaming doses × 3 stages + 1 control). Experimental plots received only a single flame treatment at the designated maturity stage. All plots were maintained weed-free throughout the season with periodic hand-weeding. Inter-rows were weeded as necessary through mechanical cultivation.

4.3.3 Flaming specifications

Flame treatments were performed using a propane fuelled, tractor mounted unshielded Red Dragon system equipped with two liquid phase Model LT 1 ½ × 6 Liquid Torches (Flame Engineering Inc., LaCrosse, KS). Burners were set up in a cross flaming manner perpendicularly facing and staggered along the crop row. Each burner was set at a 30° angle with respect to the ground, 18 cm from the center of the row measured along the angle. Flaming dose treatments were controlled by fuel pressure and tractor driving speed. Three pressures (138, 241, and 345 kPa in 2005; 117, 214, and 310 kPa in 2006) and four driving speeds (2, 3, 4, and 5 km h^{-1}) were combined to yield the 12 flame doses. The amount of propane burned per hour was recorded for each fuel pressure and used to convert treatments to a linear scale of kilograms of propane consumed per kilometer of treated row. Fuel pressures were different in 2006 than in 2005 because a fuel regulator had to be replaced between the two years. In order to account for differences between regulators, fuel pressures were modified to maintain propane consumption rates equal year to year, thus ensuring consistent flaming doses.

4.3.4 Assessment of crop dose response

Onions were harvested at crop maturity, when a majority (at least 75%) of tops had fallen. A 1.5 m section was harvested in the center of each plot, which corresponded to 10 plants per plot. Onions were pulled and cleaned of excess dirt, placed in mesh onion bags, and left in the field to dry in the sun. Bags were removed from the field later the same day and brought indoors to cure at ambient temperature under circulating air. The curing process was deemed complete when the skins were dry and necks were closed and dry, which took 2-3 weeks. Cured onions had tops and roots removed, graded according to established guidelines, and weighed.

Broccoli plants were monitored and plots harvested when mature, when beads were still tight before flowering. In 2005, all plots were harvested on July 21. In 2006, because of greater variability in maturity between plants, two harvests were taken, on July 21 and July 24. The terminal head from five plants in the center of each plot were cut to a length of 20 cm, placed in a plastic bag, and removed from the field and placed at a 4 °C. Broccoli heads were graded, weighed, and measured for head diameter using a digital calliper. All grading and measurements were completed within two days of harvest.

Spinach plants were harvested on July 12, 2005 and July 3, 2006. Plants from a 1.5 m section in the center of each plot were pulled by hand, placed in polyethylene bags, and stored at 4 °C. Spinach plants were counted and leaves graded according to established guidelines and weighed.

Beet was harvested on August 11 and 12, 2005 and August 1, 2006. Beets from a 1.5 m section in the center of each plot were pulled by hand, placed in polyethylene bags, and stored at 4 °C. Beet tops were removed, and roots graded according to established guidelines and weighed.

4.3.5 Flaming dose calculation

Flame weeding doses are often expressed in terms of fuel used per area of coverage (e.g. kg propane ha^{-1}). We have presented flaming doses used in this study as propane burned per unit row length (kg propane km^{-1}). We used this method as it describes the flaming dose effectively and would be simple to accurately compare dosages used in separate studies by authors using different equipment and field design. To convert the flame doses used in this study to kg propane ha^{-1}, all that is required is to multiply by 10 and divide by the row width in meters. For example, in this study we used a row spacing of 0.90 meters, so a dose of 0.54 kg propane km^{-1} would be equal to 6 kg propane ha^{-1}. For a row spacing of 0.60 meters, this same example would be equal to 9 kg propane ha^{-1}. This approach simplifies the comparison of doses used by different parties, and makes it easy to calculate the actual amount of fuel that is required for any given field.

4.3.6 Statistical analyses

Data collected were subjected to analyses of variance (ANOVA) using the MIXED procedure of the Statistical Analysis System to identify significant treatment effects and interactions (SAS, 2003). Homogeneity of variance was tested using the Brown-Forsythe test; data transformations were not necessary and analyses were conducted on original data. Flame doses, flaming time points, and years were considered fixed effects. Replication and interactions with replication were considered random. Repetitions were nested within years. The control was not included in the initial

ANOVA in order to allow for analysis of the factorial structure of the experiment. Significance was declared at $P \leq 0.05$.

When ANOVA indicated significant flame dose effects, regression analyses were performed using GraphPad Prism v. 5.02 (GraphPad Software, Inc., La Jolla, CA). Yield parameters were initially regressed over flame dose using a four-parameter log-logistic equation (Seefeldt et al., 1995) as shown in equation 1:

$$y = (D - C)/(1 + \exp[b^*(\log(x) - \log(ED_{50}))]) + C \qquad [1]$$

where y is the response variable, x is the flame dose, C is the lower asymptote, D is the upper asymptote, ED_{50} is the effective dose that reduced response 50%, and b is the slope of the curve at the ED_{50}. If the lower asymptote C is equal to zero, then the four-parameter model is reduced to a three-parameter model as shown in equation 2.

$$y = D/(1 + \exp[b^*(\log(x) - \log(ED_{50}))]) \qquad [2]$$

In situations where the lower asymptote is not well-defined, the three-parameter equation can often improve model fit and parameter estimation (Knezevic et al., 2007). Data sets that lacked data points defining the lower asymptote were often better fit with the three-parameter model than the four-parameter model that estimated the lower asymptote.

The log-logistic equation is often very successful at describing dose-response relationships. However, it does not allow for comparisons between separate regressions at any value except the dose causing 50% response. Furthermore, the dose that causes 50% response is not of particular interest in studies examining the effect on crop species, since yield reductions or injury significantly less than 50% are already intolerable. In such studies, doses that cause 5 or 10% response are of greater interest. To solve this problem, equation 1 can be re-parameterized as follows in order to incorporate values other than the ED_{50} (Schabenberger et al., 1999):

$$y = (D - C)/(1 + w*\exp[b*(\log(x) - \log(ED_k))]) + C \qquad [3]$$

$$w = k/(100 - k)$$

where k is the response of interest. For example, $k = 95$ if the dose that results in 95% response is of interest. Note that if $k = 50$, the added term $w = 1$ and equation 3 is reduced to equation 1. The three-parameter log-logistic equation (equation 2) can be re-parameterized in a similar fashion resulting in the following equation.

$$y = D/(1 + w*\exp[b*(\log(x) - \log(ED_k))]) \qquad [4]$$

$$w = k/(100 - k)$$

A stimulatory effect has been observed in plants treated with very low doses of some herbicides (Miller et al., 1962; Freney, 1965; Wiedman and Appleby, 1972). This phenomenon, dubbed hormesis, can interfere with use of the log-logistic equation where the maximum response occurs when $x = 0$. When yield means were plotted against flame dose, the possibility of a hormetic effect became apparent in some instances. Brain and Cousens (1989) suggested modifying equation 1 as follows in order to model a dose-response relationship when a hormetic effect is present:

$$y = (D - C + gx)/(1 + \exp[b*(\log(x) - \log(ED_{50}))]) + C \qquad [5]$$

where g indicates the rate of increase in response present at low doses. Equation 5 can be further re-parameterized to incorporate any ED_k value (Schabenberger et al., 1999).

$$y = (D - C + gx)/(1 + w*\exp[b*(\log(x) - \log(ED_k))]) + C \qquad [6]$$

$$w = k/(100 - k) + (100/(100 - k)*[(g*ED_k)/(D - C)$$

The estimate of g can be used to test for the significance of a hormetic response. If the 95% confidence interval for g does not surround 0, then the hormetic response is significant and a model incorporating a term for the hormetic response should be used (Schabenberger, et al., 1999).

Lack-of-fit F-tests were used to check the fit of the model to the data. Extra sum-of-squares F-tests at the 5% significance level were used to determine the model that best fit the data (Motulsky and Christopoulos, 2004). If none of the log-logistic equations were able to successfully model the data, a linear regression was fit using the GLM procedure of SAS (SAS, 2003) testing linear and quadratic parameters.

4.4 Results and discussion

4.4.1 Onion

No interactions between dose and year were observed for either marketable or total onion yield; data was therefore pooled over years (Table 4.1). A dose × time point interaction was present for both marketable and total onion yields, as a significant effect due to flame dose was present only at the first time point for both marketable and total onion yields. Marketable and total onion yields declined as flaming dose increased at the first time point (Figure 4.1). Yield losses of 5% were observed at a flame dose of 1.27 kg propane km^{-1}, while 10% loss in yield occurred at a flame dose of approximately 1.78 kg propane km^{-1} (Table 4.2). At the highest flame dose tested, yields were reduced approximately 25% for marketable and total yields when flamed at the first time point. Onion yields were not affected by flame dose at the second through fifth time points.

There was no singular cause for yield losses observed in the field. Diseases observed either in the field or in harvested bulbs include slippery skin, sour skin, soft rot, and white rot. Slippery skin, sour, skin, and soft rot are bacterial diseases caused by *Pseudomonas gladioli* pv. *allicola*, *Pseudomonas cepacia*, and *Erwinia carotovora* subsp. *carotovora*, respectively. These bacteria are often secondary invaders, taking advantage of injury or primary infection to gain entry into the host plant (McDonald, 1994). This would likely be the primary method by which flaming would contribute to increased disease incidence in cultivated crops. The danger for this occurrence would be present for the entire growing season, as disease infection may occur anytime including during mechanical topping at harvest. This fact could be a danger for flame treatments occurring late in the growing season. However, we do not have any evidence to suggest that flame treatments increased disease prevalence in onion.

Nemming (1993) conducted an experiment evaluating the use of pre- and postemergence flaming in onion. Onions were flamed 20 days after sowing for the preemergence treatment and 27 days after sowing for an early postemergence flame treatment; no significant yield reductions were reported for onions flamed pre- or postemergence. Ascard (1989) conducted a study comparing flaming with mechanical and chemical methods in sets and direct seeded onions. In set onions, no yield reductions were observed when flamed three times (at crop heights of 5, 20 and 40 cm) postemergence in hand-planted sets, but reductions were seen when onions were planted more erratically due to mechanical planting. The difference in effect was attributed to the hand-planted onions being more uniformly planted than those planted mechanically. In direct seeded onions flaming at preemergence and then once at a height of 15 cm resulted in no yield reduction. In these trials, the selective flaming dose used was in the range of 1.15-1.53 kg propane km^{-1}, which falls in the middle of the range of doses used in the current study. Because of differences in experimental design, it is difficult to accurately compare the results of these two sets of studies. In the present study, yields were reduced 5% at 1.27 kg propane km^{-1}, and 10% at 1.78 kg propane km^{-1}, but only at the first time point during the season (15 and 9 DAT in 2005 and 2006). In Ascard's study, yield reductions were observed in mechanically planted set onions from doses in this range. The main difference, besides the number of flame applications, is that the first selective flaming Ascard applied was when onions were only 5 cm tall, which would be earlier than was ever applied in the present study. These two studies support the idea that onions are able to withstand selective flame weeding without adverse yield effects, but this is dependent upon dose, timing, and other agronomic factors.

4.4.2 Broccoli

Effects on yield parameters in broccoli were limited to treatments that were flamed at the first time point. A three-way interaction between flame dose, time point, and year was observed for broccoli head diameter (Table 4.1). In 2005, the log-logistic equation was not able to successfully describe the response of broccoli head diameter to flame dose at the first flaming stage. Polynomial regression was therefore used to describe the response, with only a linear term significant (Figure 4.2). In 2005, broccoli head diameter declined from 144 mm in the non-flamed control to 109 mm when flamed with a dose of 2.95 kg propane km^{-1} at the first flaming time point (14 DAT), a reduction of 24%. In 2006, the log-logistic equation was best able to describe the response of broccoli head diameter to flame doses when flamed at the first flaming time point (Figure 4.2). Head diameters were reduced 5% at a flame dose of 1.40 kg propane km^{-1}, 25% at 1.81 kg propane km^{-1}, and 50% at 2.11 kg propane km^{-1}. A flame dose × time point interaction was present for marketable and total broccoli yields, as flaming only affected yields when applied at the first flaming time point in 2005 and 2006 (Table 4.1). When flamed at the first time point, marketable and total broccoli yields declined as the flame dose increased (Figure 4.2). ED_{50} values for marketable and total broccoli yields flamed at the first maturity stage were 2.56 and 2.65 kg propane km^{-1}, respectively, averaged over years (Table 4.2).

Marketable yield in broccoli flamed at the first time point was reduced by 5, 10, and 25% by flame doses of 0.75, 1.02, and 1.62 kg propane km^{-1}, averaged over years. Total broccoli yield was reduced 5, 10, and 25% by flame doses of 0.90, 1.19, and 1.77 kg propane km^{-1} at the first time point. No negative effects on broccoli yield parameters were observed when broccoli was flamed at the second through fifth time points.

There have been few studies evaluating the use of selective flame weeding in broccoli. Wilson and Ilnicki (1966) conducted experiments on flame weeding in broccoli and other cole crops. They reported no yield loss in broccoli due to one or two flame weeding treatments. Although not many studies have been conducted into the use of flame weeding in broccoli, there have been experiments conducted on the use of flame weeding in other cole crops. Netland et al. (1994) reported no injury to cabbage due to two selective flame weeding treatments. Wselaki et al. (2007) reported that flame weeding delayed crop harvest in cabbage by two weeks and reduced yields compared to a hand-weeded control. Damage to 5-10 cm kale plants was dependent upon flame dose, with more damage apparent from a higher dose (Vester, 1988).

Overall, broccoli was quite tolerant of post-emergent flame weeding in our study. Negative effects on yield parameters were restricted to the first time point; no negative effects were observed when broccoli was flamed at the second through fifth time points with flame doses ranging from 0.54 to 2.95 kg propane km^{-1}. When flamed at the first time point, total yield in broccoli was reduced by 5 and 10% at flame doses of 0.90 and 1.19 kg propane km^{-1}, respectively, doses that would be within the range necessary to control annual weeds depending upon the species makeup and development of the weed population (Sivesind et al., 2009). It would therefore be of interest to delay the first flame weeding as long as possible to avoid yield losses.

4.4.3 Spinach

A flame dose × growth stage × year interaction was present for plant number in spinach. Therefore, dose response regressions were performed in growth stages separately in each year. No regressions of plant number over flame dose were significant for spinach flamed at preemergence in either year. However, plant number was affected by flame dose at both the 4- and 6-leaf growth stages in both 2005 and 2006 (Figure 4.3). Plant number was reduced markedly by moderate flame doses at the 4- and 6-leaf growth stages. When flamed at the 4-leaf growth stage, the number of spinach plants was reduced by 50% by flame doses of 1.66 and 1.32 kg propane km^{-1} in 2005 and 2006, respectively (Table 4.2). Further, plant number was reduced 5% at flame doses of 0.66 kg propane km^{-1} and 10% at flame doses of 0.81 kg propane km^{-1} when flamed at the 4-leaf growth stage. When flamed at the 6-leaf growth stage, plant number was reduced 5% at flame doses of 0.65 and 1.02 kg propane km^{-1} in 2005 and 2006, respectively (Table 4.2). Plant number was reduced 10% at flame doses of 0.80 and 1.52 kg propane km^{-1}, and 25% at flame doses of 1.08 and 2.73 kg propane km^{-1}. Plant numbers were reduced by the same amount by similar flame doses in the 4- and 6-leaf growth stages (Table 4.2). At both growth stages, the number of spinach plants was noticeably reduced by flame doses at the lower end of the range tested.

A dose × growth stage interaction was present for marketable and total spinach yields (Table 4.1). There was no effect on marketable or total spinach yields when flamed preemergence, but yields were affected by flaming at the 4- and 6- leaf growth stages (Figure 4.4). Marketable and total spinach yields were reduced by half when flamed with moderate doses of 1.28 and 1.34 kg propane km^{-1} at the 4-leaf growth stage (Table 4.2). Yields were negatively affected by even lower doses, with yields reduced by 5, 10, and 25% by flame doses of 0.63, 0.75, and 0.99 kg propane km^{-1} at the 4-leaf growth stage. Similar to what was observed when spinach was flamed at the 4-leaf growth stage, marketable and total spinach yields were affected by flame dose at the 6-leaf growth stage (Figure 4.4). Marketable and total spinach yields were reduced by 50% at flame doses of 1.34 and 1.43 kg propane km^{-1} (Table 4.2). Negative effects on yields were present at the lowest flaming doses tested, with 5, 10, and 25% yield losses occurring when spinach was flamed with doses of 0.52, 0.66, and 0.96 kg propane km^{-1} at the 6-leaf growth stage.

Flame weeding spinach preemergence did not negatively affect yields in this study. Flaming preemergence has been used successfully in other crops, such as onion, carrot, and beets (Ascard, 1989; Nemming, 1993; Rasmussen, 2003). Bàrberi et al. (2008) reported positive results when preemergence flaming was combined with other physical weed control methods in spinach. Preemergence flaming in conjunction with a precision hoe resulted in increased yield over a standard regimen utilizing biodegradable mulch, despite increased weed pressure. We are not aware of any studies that have evaluated the use of postemergence flame weeding in spinach. When spinach was flamed postemergence, however, yield loss occurred at relatively moderate flame doses. Yield reductions of 10% occurred at less than 1.0 kg propane km^{-1} at both the 4- and 6-leaf growth stage. Depending upon the size and composition of the weed flora, flame doses needed to control weeds would be high enough to cause a 10% yield loss (Sivesind et al., 2009). Yield reductions of this magnitude would be unacceptable to most producers. Postemergence flame weeding in spinach therefore does not appear to be a viable weed control option. However, preemergence flaming can be recommended as a method for giving spinach a competitive early season advantage against emerging weeds.

4.4.4 Beets

As a dose × growth stage × year interaction was present for both marketable and total yields in beet, regressions were performed separately for each growth stage and year (Table 4.1). Preemergence flaming had no effect on beet yields in either year of this study. Marketable beet yields were reduced by flaming at the 4-leaf growth stage in 2005 and 2006 (Figure 4.5). Marketable beet yields were reduced by 50% at flame doses of 1.37 and 1.31 kg propane km^{-1} in 2005 and 2006, respectively (Table 4.2). Doses that resulted in lesser degrees of yield loss were lower in 2006 than in 2005. Flame doses that resulted in 5, 10, and 25% marketable yield loss were 0.61, 0.74, and 1.01 kg propane km^{-1} in 2005, compared to 0.17, 0.29, and 0.61 kg propane km^{-1} in 2006. Total beet yields were reduced by flame weeding at the 4-leaf growth stage in 2005 and 2006 as well (Figure 4.5). ED_{50} values for total beet yield were 1.63 kg propane km^{-1} in 2005 and 2.04 kg propane km^{-1} in 2006. Still, yield losses of 10% or greater still occurred at flame doses less than 1.0 kg propane km^{-1}. Yields of marketable beets were reduced by flaming at the 6-leaf growth stage (Figure 4.5). Five percent yield loss occurred at a flame dose of 0.77 kg propane km^{-1} in 2005 and 0.26 kg propane km^{-1} in 2006 , while yield losses of 25% were present when beets were flamed with doses of 1.55 and 0.92 in 2005 and 2006, respectively (Table 4.2). As was observed for marketable beet yields, total beet yields were affected by flame weeding at the 6-leaf growth stage in both 2005 and 2006 (Figure 4.5). Five percent total yield loss was observed when beets at the 6-leaf growth stage were flamed with doses of 0.54 and 0.91 kg propane km^{-1} in 2005 and 2006, respectively (Table 4.2). Flame doses that resulted in reduction of total beet yields of 10 and 25% were 0.83 and 1.58 kg propane km^{-1} in 2005, and 1.25 and 1.99 kg propane km^{-1} in 2006.

Vester (1988) flamed beets at the 4- and 6-leaf stages and reported withering of leaves and other visually determined injury. However, the author noted that regrowth occurred, compensating for the damage. Nemming (1993) found that flaming beets preemergence reduced weeds by 50% and reduced the time required for subsequent hand-weeding. Effects on yield were not determined and no postemergence flaming was attempted. Rasmussen (2003) evaluated preemergence flame weeding in beets, and reported a 50% reduction in weed densities compared to non-flamed treatments. The author urged caution when considering crop yield effects in that study due to weed interference and low beet densities.

No effects on marketable or total beet yields were observed when beets were flamed preemergence. Total beet yield losses of 10% were observed when beets were flamed at the 4-leaf growth stage with doses of 0.98 kg propane km^{-1} in 2005 and 0.56 kg propane km^{-1} in 2006. Ten percent loss in total beet yields occurred at similar doses at the 6-leaf growth stage, from doses of 0.83 kg propane km^{-1} in 2005 and 1.25 kg propane km^{-1} in 2006. Flaming doses in the range that caused 10% loss in yield in beets would be similar to that required for effective weed control (Sivesind et al., 2009). Flame weeding in beets at the 4- or 6-leaf growth stage could not be recommended based on the results of this study. However, pre-emergence flaming in beets could still provide a valuable early season competitive advantage for the crop.

4.5 Summary and conclusions

Flame weeding affected the four crops evaluated in this study to differing degrees. Onion and broccoli were negatively affected by flame weeding only at the first time point in each species, and were unaffected at subsequent time points. Yield losses of 10% in onion occurred at a flaming dose of 1.78 kg propane km^{-1} at the first time point. Broccoli was less tolerant of flame weeding at the first time point, as 10% yield loss occurred when flamed with a dose of 1.19 kg propane km^{-1}. Neither spinach nor beet was negatively affected by preemergence flame weeding. However, yield loss was observed for both spinach and beet when flamed at the 4- and 6-leaf growth stages. Total spinach yields were reduced 10% by similar flame doses at the 4- and 6-leaf growth stages; 0.75 kg propane km^{-1} at the 4-leaf growth stage and 0.68 kg propane km^{-1} at the 6-leaf growth stage. Beet response to flame weeding was more variable year to year. Total beet yields were reduced 10% by doses ranging from 0.56 to 0.98 kg propane km^{-1} at the 4-leaf growth stage, depending on the year, and 0.83 to 1.25 kg propane km^{-1} at the 6-leaf growth stage.

The results of this study suggest that onion is highly tolerant of flame weeding and would likely tolerate flame weeding treatments at most time points during the growing season. Broccoli was highly tolerant of flame weeding at all flame doses tested beginning 20 DAT; flame weeding treatments prior to that point would be possible if flame doses were kept below approximately 1.0 kg propane km^{-1}. Postemergence flaming in spinach caused too great of yield loss at both the 4- and 6-leaf growth stages to be able to recommend its use. Though somewhat more tolerant to flaming than spinach, postemergence flaming in beet caused too much damage at both the 4- and 6-leaf growth stages to be a viable weed control option. As a single flame weeding treatment is not sufficient to impart season long weed control, further studies are necessary to evaluate multiple flame weeding treatments in onion and broccoli.

4.6 Acknowledgements

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4.7 Tables and figures

Table 4.1. Significant main effects and interactions of year, flame dose, and time point (onion and broccoli) or growth stage (spinach and beets) on yield parameters in onion, broccoli, spinach, and beets in 2005-06.

	Onion		Broccoli			Spinach			Beets	
Effect	Marketable yield	Total yield	Diameter	Marketable yield	Total yield	Plant number	Marketable yield	Total yield	Marketable yield	Total yield
Year (Y)	**[a]	**	**	**	**	**	NS	NS	*	*
Flame Dose (D)	*	*	***	**	***	***	***	***	***	***
Y×D	NS	NS	NS	NS	NS	NS	NS	NS	*	*
Time point[b] (T)	***	***	***	***	***	***	***	***	**	*
Y×T	NS	NS	*	*	**	***	***	***	*	**
D×T	***	***	***	***	***	***	***	***	***	***
Y×D×T	NS	NS	*	NS	NS	**	NS	NS	***	***

[a] * = Signficant at P < 0.05; ** = significant at P < 0.01; *** = significant at P < 0.001; NS = nonsignificant.

[b] Growth stage for spinach and beets.

Table 4.2. Predicted values and standard errors of regression parameters for models describing the response of onion, broccoli, spinach, and beets to flame weeding treatments. Regression parameters were estimated using equation 4. Regressions for onion and broccoli included for flame treatments at the first time point only. Regressions included for spinach and beets flamed at the 4- and 6-leaf growth stages.

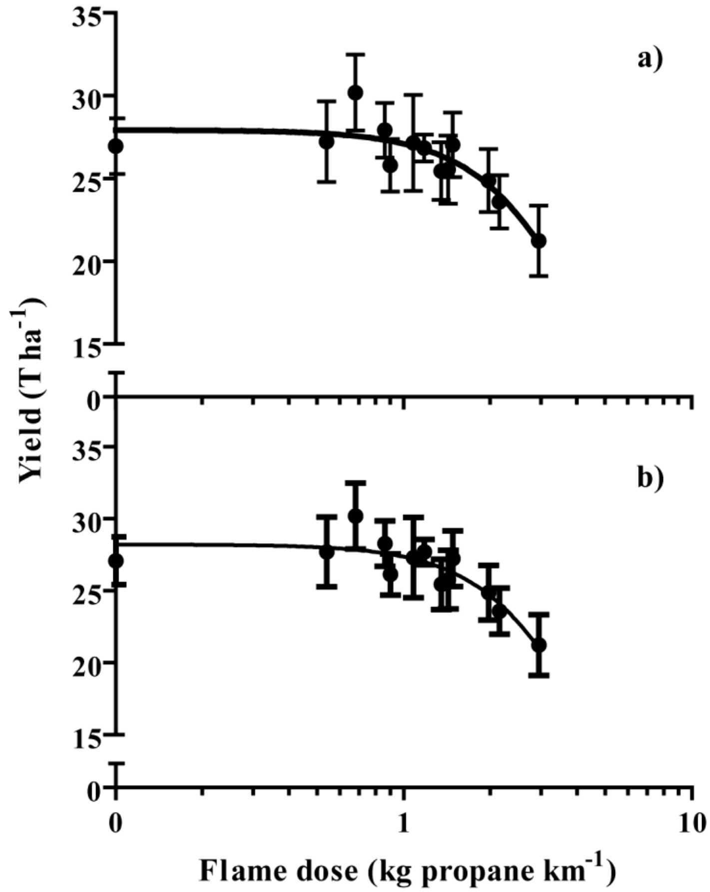
				Regression parameters (± SE)					
Year	Crop	Yield parameter	DAT[a]	D^{b}	b	ED_{50}	ED_{5}	ED_{10}	ED_{25}
2005-06	Onion	Marketable yield	15, 9	27.9 ± 1.26	2.18 ± 1.28	4.94[c] ± 1.79	1.28 ± 0.63	1.80 ± 0.56	2.98 ± 0.48
2005-06	Onion	Total yield	15, 9	28.2 ± 1.22	2.22 ± 1.21	4.77[c] ± 1.54	1.26 ± 0.58	1.77 ± 0.52	2.91 ± 0.43
2005-06	Broccoli	Marketable yield	14, 10	9.5 ± 1.06	2.41 ± 1.29	2.56 ± 0.48	0.75 ± 0.49	1.02 ± 0.51	1.62 ± 0.45
2005-06	Broccoli	Total yield	14, 10	9.7 ± 0.86	2.73 ± 1.34	2.65 ± 0.41	0.90 ± 0.46	1.19 ± 0.45	1.77 ± 0.38
2006	Broccoli	Head diameter	10	89.2 ± 5.35	2.73 ± 1.34	2.65 ± 0.41	0.90 ± 0.46	1.19 ± 0.45	1.77 ± 0.38
			Growth stage						
2005	Spinach	Plant number	4-leaf	16.2 ± 1.61	3.27 ± 1.11	1.66 ± 0.18	0.67 ± 0.23	0.85 ± 0.23	1.19 ± 0.21
2006	Spinach	Plant number	4-leaf	20.6 ± 1.63	4.07 ± 1.04	1.32 ± 0.09	0.64 ± 0.14	0.77 ± 0.13	1.00 ± 0.11
2005	Spinach	Plant number	6-leaf	15.2 ± 1.52	3.65 ± 1.20	1.46 ± 0.14	0.65 ± 0.20	0.80 ± 0.20	1.08 ± 0.17
2006	Spinach	Plant number	6-leaf	20.7 ± 1.14	1.88 ± 1.05	4.89[c] ± 1.72	1.02 ± 0.61	1.52 ± 0.59	2.73 ± 0.48
2005-06	Spinach	Marketable yield	4-leaf	2.8 ± 0.24	4.12 ± 1.14	1.28 ± 0.10	0.63 ± 0.15	0.75 ± 0.14	0.98 ± 0.12
2005-06	Spinach	Total yield	4-leaf	3.5 ± 0.31	3.82 ± 1.05	1.34 ± 0.11	0.62 ± 0.16	0.75 ± 0.15	1.00 ± 0.13
2005-06	Spinach	Marketable yield	6-leaf	3.0 ± 0.28	2.98 ± 0.78	1.34 ± 0.13	0.50 ± 0.16	0.64 ± 0.16	0.93 ± 0.15
2005-06	Spinach	Total yield	6-leaf	3.7 ± 0.33	2.96 ± 0.76	1.43 ± 0.14	0.53 ± 0.16	0.68 ± 0.17	0.99 ± 0.16
2005	Beet	Marketable yield	4-leaf	17.9 ± 1.77	3.60 ± 1.11	1.37 ± 0.13	0.61 ± 0.18	0.74 ± 0.18	1.01 ± 0.16
2006	Beet	Marketable yield	4-leaf	14.2 ± 2.24	1.44 ± 0.54	1.31 ± 0.35	0.17 ± 0.16	0.29 ± 0.21	0.61 ± 0.30
2005	Beet	Total yield	4-leaf	23.4 ± 1.59	4.27 ± 1.11	1.63 ± 0.11	0.82 ± 0.16	0.98 ± 0.15	1.26 ± 0.13
2006	Beet	Total yield	4-leaf	16.8 ± 2.51	1.69 ± 0.81	2.04 ± 0.49	0.36 ± 0.33	0.56 ± 0.40	1.07 ± 0.46
2005	Beet	Marketable yield	6-leaf	17.3 ± 1.59	2.66 ± 1.11	2.34 ± 0.33	0.77 ± 0.37	1.03 ± 0.37	1.55 ± 0.33
2006	Beet	Marketable yield	6-leaf	14.7 ± 2.16	1.44 ± 0.64	1.98 ± 0.50	0.26 ± 0.26	0.43 ± 0.34	0.92 ± 0.44
2005	Beet	Total yield	6-leaf	24.1 ± 2.38	1.71 ± 0.80	3.00[c] ± 0.64	0.54 ± 0.41	0.83 ± 0.47	1.58 ± 0.48
2006	Beet	Total yield	6-leaf	17.2 ± 2.07	2.37 ± 1.87	3.17[c] ± 0.92	0.91 ± 0.78	1.25 ± 0.78	1.99 ± 0.64

[a]Number of days after transplantation that flame treatment took place. Cells with two values indicate DAT in 2005 and 2006, respectively.

[b] *D*, the upper assymptote; *b* , the slope of the curve at the ED_{50} ; ED_{x} , the effective dose giving 50, 5, 10, and 25% response.

[c]Uncertain predicted value outside of range of observed data.

Figure 4.1. Marketable (a) and total (b) onion yield (T ha^{-1}) regressed over flame dose at the first time point (15 and 9 DAT in 2005 and 2006) and averaged over two years. Regressions were made using equation 4 and parameter values are found in Table 4.2. Points represent mean values ±SE.



a)
b)
Yield (T ha-1)
35
30
25
20
15
0
0
1
10
Flame dose (kg propane km-1)

Figure 4.2. Broccoli head diameter (mm) regressed over flame dose at the first time point (14 DAT) in 2005 (a) and 2006 (10 DAT) (b). Marketable (c) and total (d) broccoli yields regressed over flame dose at the first time point (14 and 10 DAT) in 2005-06. Note difference in scale of x-axis for broccoli head diameter in 2005 (a) compared to other regressions (b, c, and d). Regression equation for broccoli head diameter in 2005 (a) made using the linear equation: $y = A + Bx$. Parameter estimates ±SE for (a): A, 144 ± 5; B, -12 ± 4. Regressions for (b), (c), and (d) made using equation 4 and parameter values found in Table 4.2. Points represent mean values ±SE.

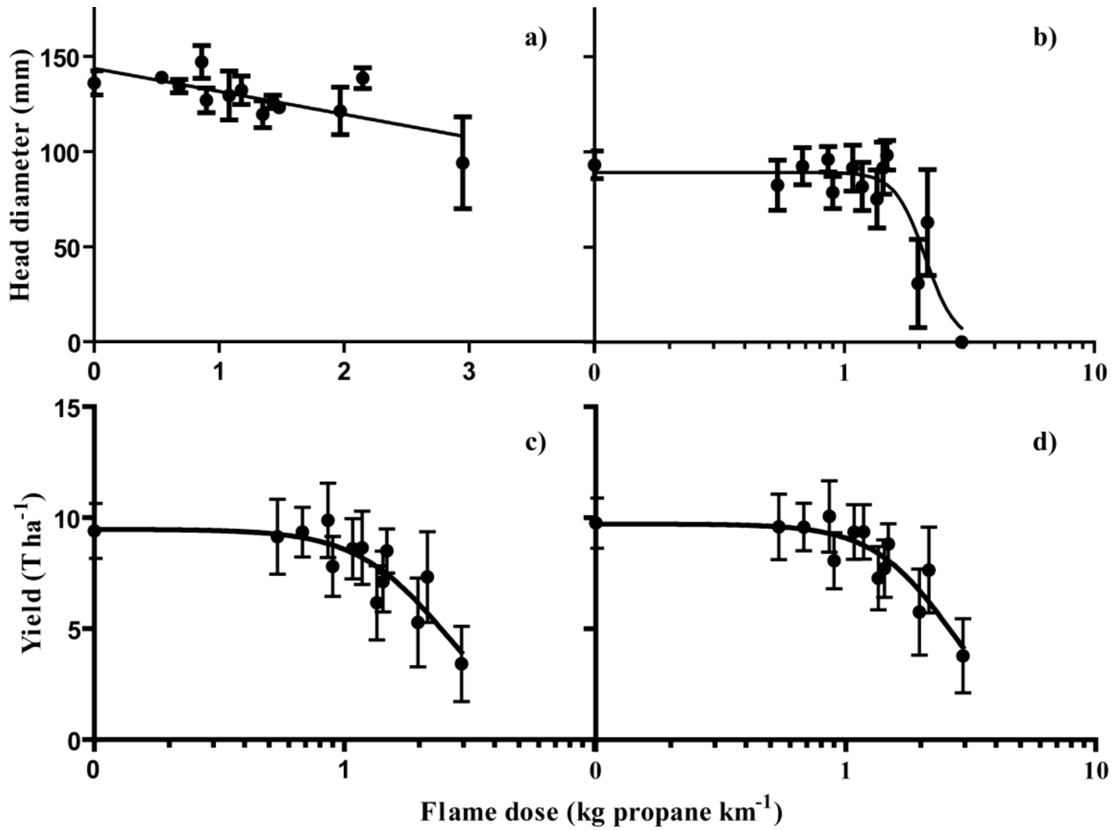
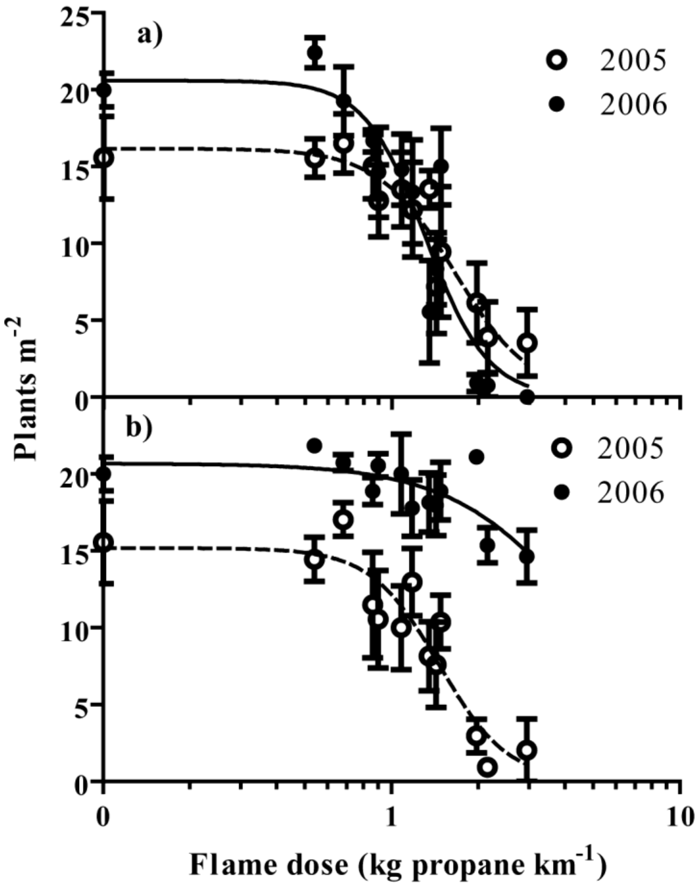

a)
b)
c)
d)
Head diameter (mm)
Yield (T ha^{-1})
Flame dose (kg propane km^{-1})

Figure 4.3. Number of spinach plants m^{-2} regressed over flame dose at the 4-leaf (a) and 6-leaf growth stage (b) in 2005 and 2006. Regressions were made using equation 4 and parameter values are found in Table 4.2. Points represent mean values ±SE.



a)
b)
2005
2006
Plants m-2
Flame dose (kg propane km-1)
0
5
10
15
20
25
1
10

Figure 4.4. Marketable spinach yield (T ha^{-1}) regressed over flame dose at the 4-leaf (a) and 6-leaf growth stage (b) in 2005-06. Total spinach yield (T ha^{-1}) regressed over flame dose at the 4-leaf (c) and 6-leaf growth stage (d) in 2005-06. Regressions were made using equation 4 and parameter values are found in Table 4.2. Points represent mean values ±SE.

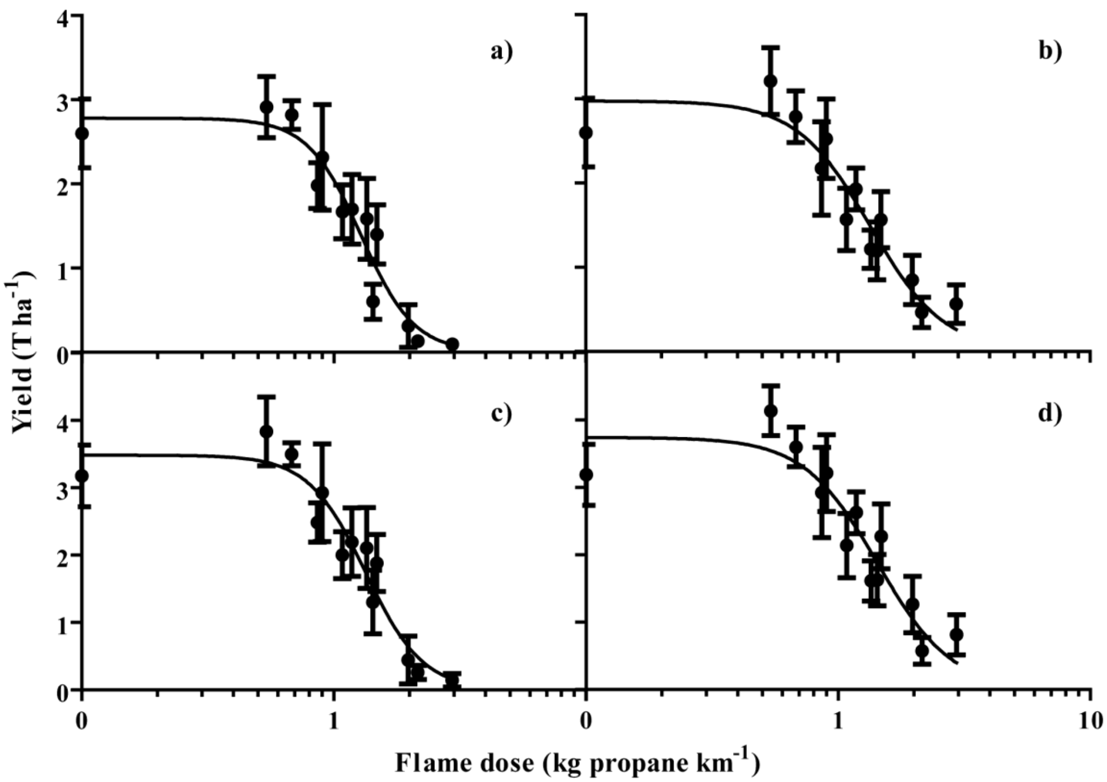
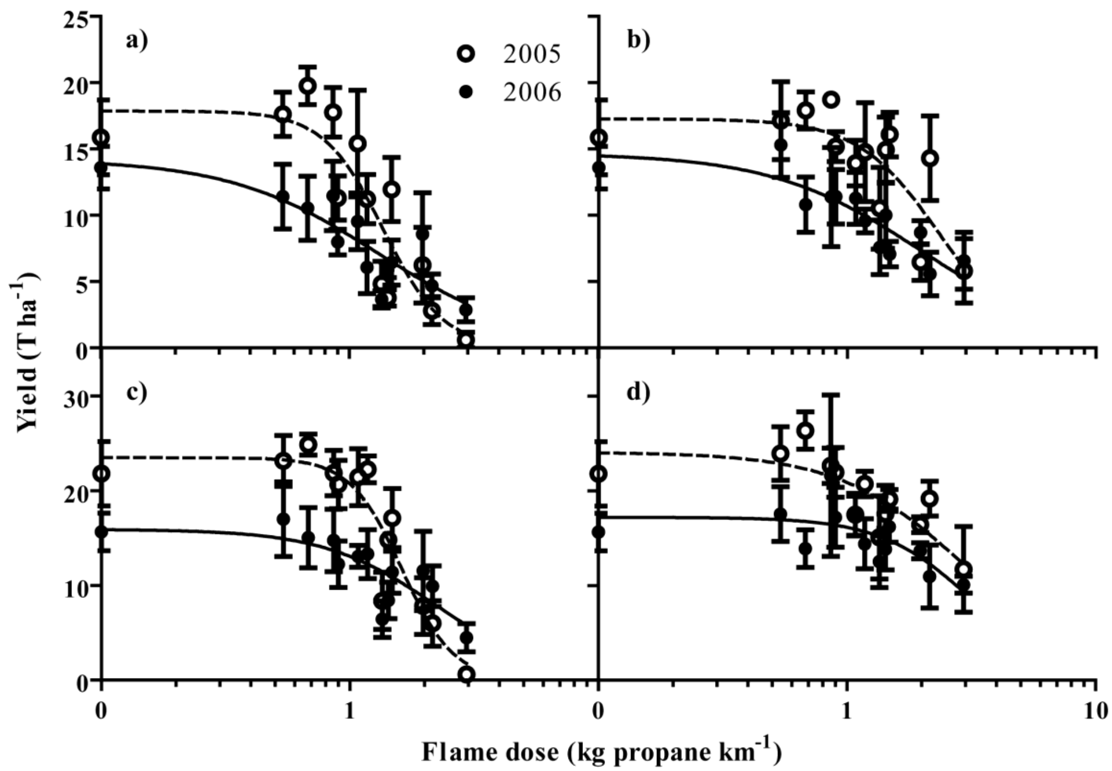

a)
b)
c)
d)
Yield (T ha-1)
Flame dose (kg propane km-1)
0
1
2
3
4
0
1
10

Figure 4.5. Marketable beet yield (T ha^{-1}) regressed over flame dose at the 4-leaf (a) and 6-leaf growth stage (b) in 2005-06. Total beet yield (T ha^{-1}) regressed over flame dose at the 4-leaf (c) and 6-leaf growth stage (d) in 2005-06. Regressions were made using equation 4 and parameter values are found in Table 4.2. Points represent mean values ±SE.



a)
b)
c)
d)
2005
2006
Yield (T ha^{-1})
Flame dose (kg propane km^{-1})
0
5
10
15
20
25
30
1
10

4.8 Connecting text

In the previous study, four vegetable crops were evaluated for tolerance of flame weeding. Yield losses in spinach and beet were observed from flame doses in the range necessary to control dicot weed species. Broccoli and onion were found to be highly resistant to flame weeding treatments at most time points evaluated. However, these treatments utilized a single flame treatment, whereas controlling weeds with flame weeding would take multiple flame treatments depending upon weed composition and crop competitiveness.

In the following study, we sought a greater understanding of the effects of multiple selective flame weeding treatments in onion. Effects on onion yield, development, and crop quality were monitored. In addition, weed control in the different flame weeding treatments was evaluated.

The manuscript was co-authored by the candidate, Dr. Maryse Leblanc, Insitut de recherche et de développement en agroenvironnement, Dr. Daniel Cloutier, Institut de malherbologie, Dr. Philippe Seguin, Department of Plant Science, Macdonald Campus of McGill University, and Dr. Katrine Stewart, Department of Plant Science, Macdonald Campus of McGill University. The candidate designed the experiments, carried out the field experiments, performed the laboratory analyses, conducted the data analyses, and was the primary author of the manuscript. Drs. Leblanc and Cloutier provided funds and reviewed the manuscript. Drs. Seguin and Stewart assisted the candidate in the experimental design, provided funds and supervisory guidance, and reviewed the manuscript.

5.0 Impact of Selective Flame Weeding on Onion Yield, Pungency, Flavonoid Concentration, and Weeds

5.1 Abstract

Field experiments were conducted to evaluate the effects of multiple selective flame weeding treatments in onion. Onions were flamed between one and six times over the course of the season with a high (1.43 kg propane km^{-1}) or low (0.90 kg propane km^{-1}) flame dose. In each treatment, one subplot was maintained weed-free in addition to flame treatment to remove differential weed effects, while the other received only the prescribed flame weeding regimen as weed control. Control of dicot weeds was better than that of monocot species. Dicot weed density and shoot mass were reduced as flaming dose and the number of flame treatments increased. Monocot density was reduced by 50% in all flamed treatments compared to the non-treated control, but no differences between flamed treatments were observed. Effects of flame treatments on monocot shoot mass were minimal. Among weed-free treatments, onion was able to tolerate up to six flame treatments with either dose without a loss of yield. Treatments that received only flame weeding as weed control had total onion yields 37 and 80% of the weed-free flamed treatments in 2006 and 2007, respectively. Flame weeding treatments had little effect on the amount of time to reach maturity, leaf and bulb development, onion pungency, or quercetin concentration.

5.2 Introduction

Flame weeding is a weed control method that can be used as an alternative to synthetic herbicides. The use of flame weeding has become more common with the rise in organic crop production as well as concerns about the effects of herbicides on human health and the environment. Controlling weeds without the use of selective chemical herbicides can be difficult, and has been cited as the most difficult aspect involved in the transition from conventional to organic crop production (Walz, 1999).

Onion is a weakly competitive crop with a shallow root system and an open leaf canopy. Effective weed control is therefore required for the entire duration of the growing season. A number of herbicides are currently available for use in onion in Canada, including preemergence (chlorprophram and chlorthal dimethyl) and postemergence (e.g., fenoxyprop-p-ethyl, sethoxydim, pendimethalin, and oxyfluorfen) herbicides (OMAFRA, 2008). Under organic production, without the use of herbicides, weed control is difficult. Care must be taken if cultivating close to the crop, as the shallow onion roots can be easily damaged. Therefore large amounts of hand-weeding are often required at considerable expense (Ascard, 1989). Flame weeding is an alternative method that can decrease the amount of hand-weeding required. Flaming is used to control weeds that occur along the crop row, as inter-row weeds can be effectively controlled through mechanical cultivation (Melander, 1998). Weeds that grow close to crop plants within the row are more difficult to control as cultivation is either ineffective or causes unacceptable damage to crop plants. The advantage of flame weeding is that it provides effective weed control within the crop row where mechanical cultivation is difficult, and reduces or eliminates the amount of costly hand-weeding that is necessary.

Flaming has been historically used primarily as a preemergence treatment, either prior to planting or before crop emergence (Ascard, 1995). Burner(s) are set parallel to the direction of travel, often with a shield to improve fuel efficiency, and centered directly over the center of the crop row. With the weed population destroyed, the emerging crop is provided an early season advantage. Alternatively, flaming can be used after crop emergence or planting in tolerant species, a process referred to as selective flaming. Selective flaming requires a different system, where uncovered, angled burners are staggered and directed towards the center from both sides of the row.

A number of studies have been conducted investigating the effect of weed characteristics and flaming specifications on weed susceptibility to flaming, including species and growth stage (Ascard, 1995; Cisneros and Zandstra, 2008; Knezevic and Ulloa, 2007a; Sivesind et al., 2009), plant size and density (Ascard, 1994), and fuel pressure and burner setup (Ascard, 1997). In an earlier study (Sivesind et al., 2009, Chapter 3) we evaluated monocot and dicot weeds for susceptibility to flame weeding. Depending upon flaming dose and plant development, dicot species could be effectively controlled. For grass species, while survival was high, above ground biomass was substantially reduced in the short term. These results indicate that flame weeding is able to kill or severely damage many weed species; however, a single flaming does not impart extended control. The ability of flame weeding alone to control weeds for an entire season has not been well established. There is relatively little information available on the effects of flaming postemergence on many potential crop species. Ascard (1989) conducted a study comparing pre- and postemergence flaming to herbicide applications in set and seeded onion. Onion was reported to be reasonably tolerant of post-emergent flame weeding, with no yield reductions reported in seeded or hand planted sets, and a moderate reduction in yield in machine planted sets. However, there are a number of important considerations besides yield which need to be evaluated before an informed recommendation can be made concerning selective flaming in onion. For example, there are no reports in the literature regarding the effects of flaming on any of the many aspects of onion quality besides simple yield determinations. Onions are highly valued for their flavour. Onions accumulate large quantities of sulphur compounds, particularly the gamma glutamyl peptides and S-alkenyl cysteine sulfoxides (ACSOs). In intact tissue, ACSOs are found in cytoplasm while the enzyme alliinase is stored within the vacuole (Lancaster and Collin, 1981). When onions are eaten or chopped for cooking, ACSOs are hydrolyzed by alliinase, and form a number of volatile S-compounds responsible for the characteristic flavour and aroma associated with onions (Coolong and Randle, 2003). A number of factors are known to affect flavour intensity in onion, including temperature (Coolong and Randle, 2003) and sulfur fertilization levels (Randle et al., 2002) during plant growth. Therefore, it is important to determine if stresses associated with flame weeding affects onion flavour characteristics.

Onion has been reported to contain large quantities of flavonoids (Hertog et al., 1992; Hirota et al., 1998; Patil et al., 1995), phenolic compounds which are widely distributed in the plant kingdom (Sellappan and Akoh, 2002). Flavonoids function in plant defense response to a number of stimuli: as a deterrent to herbivory, as a response to attack by pathogens (Stafford, 1997) and in response to physical damage caused by chemicals or UV light (Olsson et al., 1998). The major flavonoid in onion is quercetin, which is found within the plant mostly as glucosidic conjugates (Price and Rhodes, 1997; Sellappan and Akoh, 2002). Flavonoid content in onion has been reported to vary with cultivar (Marotti and Piccaglia, 2002; Price and Rhodes, 1997), and is affected by environmental conditions (Patil et al., 1995). Quercetin has garnered much interest due to its antioxidant properties and potential benefits regarding cardiovascular disease (Hertog et al., 1993; Hertog et al., 1995) and cancer (Hosokawa et al., 1990; Scambia et al., 1990). Quercetin levels in onion therefore may be important in relation to their value as a functional food. Alternatively, in light of the role of many flavonoids in defence response to many biotic and abiotic stresses, increased quercetin levels may impart a protective effect on the plant. Given the number of biotic and abiotic stressors that can alter flavonoid levels and profiles, the response of quercetin to flame weeding in onion should be investigated.

Flame weeding has been used for weed control in onion, but normally in conjunction with other weed control strategies (Ascard 1989; Ascard and Fogelberg, 2008; Melander, 1998; Melander and Rasmussen, 2001); we are not aware of any studies that have examined the use of flame weeding alone to provide season-long weed control in onion. In this study we investigated the use of repeated flame weeding treatments to provide season long weed control. Secondly, previous studies (Chapter 4) demonstrated that onion is quite tolerant of flame treatment; 10% yield loss occurred when onion was flamed a single time with a dose 1.77 kg propane km^{-1} between 9 and 15 days after transplantation (DAT), while yields were unaffected when onions received a single flame treatment with doses up to 2.95 kg propane km^{-1} from 20 DAT onwards. Due to the lack of residual weed control from flaming, repeated treatments would be necessary. This study was intended to determine the effects of selective flame weeding on onions in terms of yield, development, and crop quality. To accomplish these goals, experiments were conducted to test different flaming doses and number of treatments on onion yield, weed control, and a number of factors relating to plant growth and crop quality, including pungency and flavonoid concentration.

5.3 Materials and methods

5.3.1 General field management

Experiments were conducted at the Institut de recherche et de développement en agroenvironnement (IRDA) in Saint-Hyacinthe, QC, Canada (45° 38' North, 72° 57' West) in 2006 and 2007. The soil type was a Duravin loam with 2.2% organic matter and a pH of 6.7. Fields were fertilized in accordance with local recommendations as indicated by soil tests (Centre de Référence en Agriculture et Agroalimentaire du Québec, 2003). In 2006, the field received a broadcast of 420 kg ha^{-1} of 13-12-23 NPK fertilizer on May 28. A banded application of 80 kg ha^{-1} of 27-0-0 NPK was applied 15 cm from the row at a depth of 2 cm on July 6. In 2007, 420 kg ha^{-1} of 13-12-23 NPK was broadcast on May 22. A banded application of 106 kg ha^{-1} of 27-0-0 NPK was applied 15 cm from the row at a depth of 2 cm on July 6. Sixty-day-old onion plants (cv. Vaquero) were transplanted into the field May 28, 2006 and May 24, 2007. Plots consisted of a single row 5 m long with plant spacing of 15 cm and 90 cm between rows.

5.3.2 Experimental design

Experiments were set up in randomized complete block design (RCBD) with split-plot restriction and four replications. Main plots were factorial combinations of flaming dose and number of flame treatments. There were two levels for flaming dose: a low flaming dose (0.90 kg propane km^{-1}) and a high flaming dose (1.43 kg propane km^{-1}). These rates were chosen based on results from previous experiments (Chapters 3 and 4) which indicated that these rates would be appropriate for use in onion and provide effective weed control. There were six levels for the number of flame treatments: plots were flamed 1, 2, 3, 4, 5, or 6 times over the course of the growing season. In addition one control plot was included that received no flame treatment. Plots were split into two subplots; one subplot was hand-weeded in addition to the appropriate flame treatment, the other received only the flame weeding treatment. In hand weeded splits, all weeds that were not controlled with flaming were removed following flame treatment in order to remove confounding effects of differing weed pressures based upon flame treatments. One subplot of the control therefore received no weed control treatment and served as the weedy check, the other subplot was maintained weed-free through hand-weeding and served as the non-flamed, weed-free control.

The experiment contained 52 main plots (4 repetitions of 2 flaming doses × 6 number of flame treatments + 1 control) each split into two subplots. Time points for flame treatments were at 10, 24, 34, 52, 61, and 73 days after transplantation (DAT) in 2006, and 15, 26, 36, 50, 62, and 77 DAT in 2007. Flame treatments were administered when weeds reached the cotyledon to 2-leaf stage, developmental stages determined in an earlier study (Chapter 3) to be susceptible to the flaming rates employed. In 2006 and 2007, due to a high number of escapes and uneven emergence later in the season, the third and fourth flame treatments were administered approximately every two weeks.

Onion leaf counts and bulb measurements were made on four plants in the middle of each split-plot five times in each year: 25, 38, 51, 64, 78 DAT in 2006, and 20, 33, 47, 63, and 75 DAT in 2007. Onion bulb and neck diameters were measured with a digital calliper in order to gauge bulb initiation and bulb development. In addition, the number of leaves produced were counted and recorded. At the conclusion of the experiment, weeds within a 20 × 100 cm quadrat placed along the center of the row in each split-plot were collected, sorted by species, counted, dried for four days in a 60 °C forced air oven, and weighed. Split-plots were harvested individually when 75% of tops had fallen, placed in onion bags, and the date recorded for use in determining the number of days between transplantation and harvest. Onions were cured for 3 days in a forced air oven at 35° C followed by two weeks indoors at ambient temperature until necks were closed and dry. Cured onions were then graded according to established guidelines (OMAFRA, 2008) and weighed.

Onions were then placed in cold storage at 4 °C for a short time (1 week) until pungency and flavonoid analyses.

5.3.3 Flaming specifications

Flame treatments were performed using a propane fuelled, tractor mounted unshielded Red Dragon two burner system equipped with two liquid phase Model LT 1 ½ × 6 Liquid Torches (Flame Engineering, Inc., LaCrosse, KS) directed perpendicularly to the crop row. Burners were set at an angle of 30° with respect to the horizontal 18 cm from the row measured along the angle. Burners were staggered to keep flames from intersecting and deflecting upwards and damaging crop canopy. To achieve a flaming dose of 0.90 kg propane km^{-1}, the fuel pressure was set at 117 kPa and a tractor speed of 3 km h^{-1} was used. A fuel pressure of 214 kPa and a driving speed of 3 km h^{-1} were utilized to achieve a flaming dose of 1.43 kg propane km^{-1}.

5.3.4 Flaming dose calculation

Flame weeding doses are often expressed in terms of fuel used per area of coverage (e.g. kg propane ha^{-1}). We have presented flaming doses used in this study as propane burned per unit row length (kg propane km^{-1}). We used this method as it describes the flaming dose effectively and would be simple to accurately compare dosages used in separate studies by authors using different equipment. To convert the rates used in this study to kg propane ha^{-1}, all that is required is to multiply by 10 and divide by the row width in meters. For example, in this study we used a row spacing of 0.90 meters, so a dose of 0.54 kg propane km^{-1} would be equal to 6 kg propane ha^{-1}. For a row spacing of 0.60 meters, this same example would be equal to 9 kg propane ha^{-1}. This approach simplifies the comparison of rates used by different parties, and makes it easy to calculate the actual amount of fuel that is required for any given field.

5.3.5 Flavonoid extraction

After cold storage (< 1 week), samples from five bulbs from each sub-plot were combined, frozen in liquid nitrogen, and stored at -80 °C until lyophilization. Freeze-dried onion tissue was ground to a fine powder using a mortar and pestle. Ground freeze-dried material was stored in tightly capped tubes at -20 °C until flavonoid extraction.

Flavonoid extraction was performed according to an established procedure (Hertog et al., 1992) with some modifications. Acid hydrolysis was used to convert flavonoid glycosides to the aglycon form for quantification. Aliquots (0.25 g) of freeze dried and ground material were extracted with 19.5 mL of 61.25% aqueous methanol with 2g L^{-1} tert-butylhydroquinone (TBHQ) added as an antioxidant and 0.5 mL of 500 μg ml^{-1} daidzein in methanol included as an internal standard. Samples were brought up to 25 mL by the addition of 5 mL 6 M HCl, yielding a 1.2 M HCl, 50% aqueous methanol solution. Samples were then placed in a water bath at 90 °C for 2 hours and mixed regularly. Samples were filtered through 0.45 μm PFTE filters and stored at -20 °C until HPLC analysis. All sample extractions were performed in duplicate.

5.3.6 HPLC analyses

Samples were analyzed using a Varian Polaris HPLC system employing two model 210 pumps, a model 410 autosampler, a PDA detector model 330, and Star Chromatography workstation System control software version 6.30 (Varian, Inc., Palo Alto, CA, USA). The column employed was a C_{18} Phenomenex Luna (250 × 4.5 mm, 5 μm). HPLC separation was achieved using a method presented by Price and Rhodes (1997) and Lombard et al. (2002) with some modifications. Flavonoids were separated using a linear gradient of water acidified with 0.05% trifluoroacetic acid (Pump A) and acetonitrile (Pump B) with a flow rate of 1.0 mL min^{-1} as follows: isocratic 20% B for 5 min; linear gradient to 55% B for 30 min; linear gradient to 95% B for 1 min; isocratic 95% B for 1 min; linear gradient to 20% B for 1 min; and isocratic 20% B for an additional 4 minutes to equilibrate the column in preparation for the next sample. Column temperature was maintained at a constant 30 °C during analysis. Peak areas were quantified by comparison to standard curves constructed from known quantities of purchased external standards of quercetin, kaempferol, and myrcetin (Sigma Chemical Co., St. Louis, MO).

5.3.7 Flavour characteristics

Five onions from each sub-plot were analyzed for overall pungency. Enzymatically produced pyruvic acid (EPY) is a good indicator of onion pungency (Schwimmer and Weston, 1961). Enzymatically produced pyruvic acid was determined by subtracting background levels of pyruvic acid from total pyruvic acid after sample preparation.

Pyruvic acid levels were obtained from the combined tissues of five onions from each plot (Randle, 1992). Equal portions of each of five onions were combined and homogenized in a standard blender with an equal weight of water for 3 min. The puree was left to sit for 12 min. before filtering out particulates using a Whatman #1 filter. In order to determine background levels of pyruvic acid, equivalent sections from each onion were microwaved for 1.5 s g^{-1} in order to deactivate alliinase. Water was added to bring the total weight to twice that of the pre-microwave weight of the onion, and the mixture was homogenized in a blender for 3 min. Samples used to determine background pyruvic acid levels and those used for total pyruvic acid analyses were treated identically in all subsequent steps.

A 25 μL aliquote of the homogenate was brought up to 1 mL with deionized water, mixed with 1 ml of 0.025% 2, 4 dinitrophenylhydrazine (DNPH) in 1M HCl, vortexed, and heated for 10 min in a water bath set at 37 °C. The mixture was then added to 1 mL of 1.5 M NaOH and absorbance read in a spectrophotometer at a wavelength of 515 nm (Anthon, 2003). Background levels of pyruvic acid were subtracted from total levels in order to determine enzymatically produced pyruvic acid.

Soluble solids content is highly correlated with water-soluble carbohydrates in onion (Mann and Hoyle, 1945). A hand held refractometer was used to measure sugar levels using an aliquot of the aqueous extract that was prepared for pyruvic acid analysis.

5.3.8 Statistical analyses

All data were subjected to analysis of variance (ANOVA) using the MIXED procedure of the Statistical Analysis System to identify significant treatment effects and interactions (SAS, 2003). Homogeneity of variance was tested using the Brown-Forsythe test. Weed data was log transformed (log y+1) to homogenize variance between treatments when appropriate. Transformed data was back transformed for presentation within the text. Year, flame dose, number of flame treatments, and weeding treatment were considered fixed. Repetition and interactions with repetition were considered random. Leaf counts and bulb measurements were performed at five time points in each of 2006 and 2007. Treatments were not included in the analysis of leaf count and bulb measurement data for a time point if the full treatment regimen had not yet been completed (i.e. treatments to be flamed five times were not included in analysis at the second leaf count and bulb measurement in 2007, as only two flame treatments had been administered at this point in time). The control was not included in the initial ANOVA in order to allow for analysis of the factorial structure of the experiment. The control was included and the experiment analyzed as a one-way ANOVA using single degree of freedom contrasts to test for differences between groups of treatments and the control.

Differences between fixed effects means were determined using the LSMEANS statement and the PDIFF option, which declares differences using Fisher's protect least significant difference (LSD). When interactions between fixed effects were significant, simple effects were determined using the SLICE option of the LSMEANS statement (Littell et al., 2002). Significance was declared at $P \leq 0.05$ unless noted. Regression analyses were performed when analysis of variance indicated significant effects for quantitative factors. Linear regression was performed using SAS (SAS, 2003). Linear, quadratic, and cubic factors were tested for significance. Non-linear regressions were performed using GraphPad Prism v. 5.02 (GraphPad Software, Inc., La Jolla, CA). Different models were evaluated, and Akaike's information criterion (AIC) was used to determine the model which best fit the data (Motulsky and Christopoulos, 2004). Lack-of-fit *F*-tests were used to test the fit of the model to the data (Motulsky and Christopoulos, 2004). Models evaluated included the exponential model

$$y = (A - B)*\exp(k*x) + B \qquad [1]$$

where A is the value of y when $x = 0$, B is the lower plateau, and k is the rate of change of x. Other data was best fit by

$$y = Y_o*\exp(k*x) \qquad [2]$$

where Y_o is the value of y at $x = 0$ and k is the rate of change of x.

5.4 Results and discussion

5.4.1 Weed response

The weed population was a diverse mixture of dicot and monocot species (Figure 5.1). The most common broadleaf weeds included common lambsquarters (*Chenopodium album* L.), redroot pigweed (*Amaranthus retroflexus* L.), common ragweed (*Ambrosia artimisiifolia* L.), and shepherd's-purse [*Capsella bursa-pastoris* (L.) Medik]. Predominant grass species included barnyardgrass [*Echinochloa crus-galli* (L.) Beauv.], yellow foxtail [*Setaria pumila* (Poir.) Roemer and J.A. Schultes], green foxtail [*Setaria viridis* (L.) Beauv.], and witchgrass (*Panicum capillare* L.). For analysis, weeds were grouped into monocot and dicot species.

Main effects due to flame dose and the number of flame treatments were observed for dicot weed density; however, a three-way interaction between year, flame dose, and the number of flame treatments indicated that the effect of flame dose depended upon the number of treatments and varied year to year, and that the effect of the number of flame treatments depended upon dose and varied from one year to the next (Table 5.1). Overall, dicot weed density decreased as the number of flame treatments increased (Figure 5.2). In 2006, dicot weed density was similar between the low and high doses up to two flame treatments (Figure 5.2), after which dicot density in high dose treatments continued declining at the same rate while in low dose treatments further declines were diminished. By the sixth flame treatment, dicot weed density in low dose treatments was 4 plants m^{-2} and in high dose treatments 0 plants m^{-2}. In 2007, dicot weed density declined as the number of flame treatments increased, reaching 1 and 9 plants m^{-2} in high and low dose treatments flamed six times (Figure 5.1). Shoot mass of dicot weeds was affected by both flaming dose and the number of flame treatments (Table 5.1). Dicot shoot mass was reduced from 630 g DM m^{-2} in the non-treated control to 51 g DM m^{-2} in 0.90 kg propane km^{-1} flamed treatments and 11 g DM m^{-2} in 1.43 kg propane km^{-2} flamed treatments (Figure 5.3). Shoot mass of dicot weeds decreased as the number of flame treatments increased, regardless of dose (Figure 5.4). Dicot shoot mass was reduced from 190 g DM m^{-2} in treatments flamed once to 4 g DM m^{-2} in treatments flamed six times. The response of dicot shoot mass to increased number of flame treatments was largely similar to that of dicot weed density; both dicot weed density and shoot mass were reduced as the number of flame treatments increased.

Monocot density in flamed treatments was not affected by flame dose or the number of treatments (Table 5.1). Single degree of freedom contrasts revealed a difference between the non-treated control and all flamed plots (66 and 33 plants m^{-2}, respectively). A flame dose × number of flame treatments interaction was present for shoot mass of monocot weeds (Table 5.1). There was no difference in monocot shoot mass between the high (1.43 kg propane km^{-1}) and low (0.90 kg propane km^{-1}) flame doses in treatments flamed one to four times. Shoot mass was higher in the low dose than the high dose treatment in treatments flamed five times (158 and 19 g DM m^{-2}) and six times ($P = 0.1$) (93 and 31 g DM m^{-2}). These results suggest that a greater flame dose was effective at further reducing monocot shoot mass when the number of flame treatments is increased. There was no difference between the non-treated control and high or low dose treatments for shoot mass of monocot weeds. As monocot density was lower in flamed plots than in the non-treated control, but no difference was observed for monocot shoot mass, it appears that weeds that survived were able to grow larger under conditions of lower competition when weed density was reduced. Shoot mass therefore remained unchanged despite fewer individuals being present.

Total weed density was affected by both flaming dose and the number of flame treatments (Table 5.1). As flaming dose increased, total weed density declined (Figure 5.5). There was no interaction between flame dose and year in the factorial analysis; however, when the control was included in the one-way ANOVA, an interaction was present as the non-treated control had higher total weed densities in 2006 (539 plants m^{-2}) than in 2007 (277 plants m^{-2}) while the flamed treatments were similar year to year (Figure 5.5). Though total weed density declined as the flaming dose increased from 0.90 kg propane km^{-1} to 1.43 kg propane km^{-1}, the largest reduction in total weed density occurred as the flaming dose increased from 0 to 0.90 kg propane km^{-1}, reflecting the overall effect of flame weeding on total weed density at either flame weeding dose evaluated. In 2006, total weed density declined from 539 plants m^{-2} in the non-treated control to 91 plants m^{-2} in treatments flamed at 0.90 kg propane km^{-1} and 76 plants m^{-2} in treatments flamed at 1.43 kg propane km^{-1}. In 2007, total weed density declined from 277 plants m^{-2} in the non-treated control to 59 plants m^{-2} in treatments flamed at 0.90 kg propane km^{-1} and 24 plants m^{-2} in treatments flamed at 1.43 kg propane km^{-1}. Increasing the number of flame treatments caused a decrease in total weed density as well (Table 5.1). There was no interaction between the number of flame treatments and year in the factorial analysis; however, when the control was included in the one-way ANOVA, an interaction was present as the non-treated control had higher total weed densities in 2006 than in 2007 while the flame treatments were similar year to year (Figure 5.6). Similar to the effect of flaming dose on total weed density, the biggest reduction occurred between the non-treated control and treatments flamed once (Figure 5.6). A single flame treatment reduced total weed density to 31 and 28% of the non-treated control in 2006 and 2007, respectively; six flame treatments resulted in total weed density 12% of the control in each of 2006 and 2007. As was observed for total weed density, total weed shoot mass was affected by both flame dose and the number of treatments (Table 5.1). Total weed shoot mass declined as flaming dose increased from zero to 1.43 kg propane km^{-1} (Figure 5.7). Total weed shoot mass declined from 1187 g DM m^{-2} for the non-flamed control to 543 g DM m^{-2} for the 0.90 kg propane km^{-1} treatment and 342 g DM m^{-2} for the 1.43 kg propane km^{-1} treatment. A decline in total weed shoot mass was observed due to increasing the number of flame treatments (Figure 5.8). Total weed shoot mass declined 77% between the non-treated control and treatments flamed six times.

Flame weeding treatments had a greater effect on dicot weeds than monocot weeds in this study. These results are in line with results of dose response studies, which generally found dicot weeds to be much more susceptible to flame weeding than monocot species (Ascard, 1995; Cisneros and Zandstra, 2008; Knezevic et al., 2008; Knezevic and Ulloa, 2007a; Sivesind et al., 2009). However, there is little information in the literature regarding season-long effects of flame weeding on weed populations in the field. End of season dicot weed density declined as the number of flame treatments increased, and shoot mass of dicot weeds declined as both flame dose and the number of flame treatments increased. These results make sense in light of the results of earlier studies we conducted. Flame doses in the range used in this study 0.90 to 1.43 kg propane km^{-1} were effective in controlling dicot weeds at a number of growth stages, and weed mortality increased as flame dose increased

(Chapter 3). Monocot weed density was lower in flamed treatments than in the weedy, non-treated control, but did not decline as the number of flame treatments increased from one to six. In our earlier study, monocot weeds could not be effectively controlled by flaming with dose in the range used in these experiments.

5.4.2 Onion yield

Repeated flame weeding treatments had limited effects on onion yield. No main effects or interactions between flame dose and the number of flame treatments was observed for marketable or total onion yields (Table 5.2). Among flamed treatments, an effect due to hand-weeding treatment was observed; treatments that were hand-weeded in addition to flaming had higher marketable and total yields than plots that were flamed only (Table 5.2), though the magnitude of the difference varied between years. Flame-only treatments had marketable and total yields 37% of the hand-weeded and flamed treatments in 2006 and 80% of the hand-weeded and flamed treatments in 2007. Onion yields in flame-only plots in 2006 were considerably lower than yields in 2007. Single degree of freedom contrasts revealed no differences between the hand-weeded, non-flamed control and hand-weeded and flamed treatments in either 2006 or 2007 (data not shown). However, the weedy control had reduced marketable and total yields as compared to flame-only treatments in both 2006 and 2007. In 2006, the weedy control had marketable and total yields of 2.37 and 2.44 T ha^{-1}, while all flame-only treatments had marketable and total yields of 8.53 and 8.62 T ha^{-1} averaged over flame dose and number of treatments. In 2007, the weedy control had marketable and total yields of 4.80 and 4.89 T ha^{-1}, while all flame-only treatments had marketable and total yields of 31.04 and 33.3 T ha^{-1}.

There was higher disease incidence in 2007 than in 2006, which resulted in higher yields of unmarketable onions in that year (3.7 and 0.14 T ha^{-1}, respectively, averaged over all other factors). The most common diseases observed were slippery skin, sour skin, and soft rot. A year × weeding interaction was present for unmarketable yield. There was no difference in unmarketable yield between flame-only and hand-weeded and flamed plots in 2006; however, in 2007, hand-weeded plots had nearly twice the yield of unmarketable onions as the flame only plots (5.1 and 2.3 T ha^{-1}, respectively). This result was surprising and the cause unclear. If injury due to flame weeding were to blame, it could be expected that having greater numbers of potentially vector carrying weeds in close contact could increase disease prevalence. However, the bacterial pathogens that cause slippery skin, sour skin, and soft rot are often secondary invaders that gain entry into a plant through an existing injury (McDonald, 1994). If injuries to leaves due to flame treatments were present, perhaps having weeds present would reduce splashing of soil onto injured leaves by rain thus reducing infection by these secondary pathogens.

Flame weeding treatments did not reduce total or marketable onion yields in either year of this study regardless of flame dose or the number of flame treatments. Among weed-free treatments, no differences between the hand-weeded, non-flamed control and flamed treatments were observed. In this study, onion was able to withstand up to six flame weeding treatments without any measurable loss of yield. Flame-only treatments had reduced yields as compared to weed-free flamed treatments in both years of the study. Flame-only treatments had marketable and total yields 37 and 80% of the weed-free flamed treatments in 2006 and 2007, respectively. This indicates that the weed control provided by the flame treatments alone was not sufficient to avoid yield loss. The weed population that remained in the flame-only treatments was sufficient to cause yield losses of 63 and 20% in 2006 and 2007 as compared to the hand-weeded and flamed treatments. However, flame-only treatments did have yields considerably greater than the weedy control, indicating that flame treatments were able to control weeds enough to increase yields. This difference can be attributed to a single flame dose early in the season as there were no differences between flame-only treatments due to the flame dose or number of treatments; subsequent flame treatments did not further increase onion yields.

Ascard (1989) found that up to three selective flaming treatments did not reduce yields compared to mechanically and chemically treated onions in hand-planted sets, though yield reductions were observed in machine planted sets that received the same treatments. Flaming preemergence and once during growth resulted in no yield reduction in seeded onions. In all these treatments, hand-weeding was used in addition to mechanical, chemical, or thermal treatments. Yield reductions observed in mechanically planted sets were attributed to uneven development in the young plants. Employing onion transplants is a good way to ensure a minimum level of development in young plants in order to help minimize the chance of harming under developed individuals. The results from the current study are in general agreement with those presented by Ascard, in that onions appear to be reasonably tolerant of selective flame weeding.

5.4.3 Days to harvest

As flame weeding has the potential to cause high degrees of stress on growing plants, we wished to determine if flame weeding treatments delayed or otherwise affected onion maturity. Effects on the number of days between onion transplantation and harvest due to flame weeding were limited. The number of flame treatments had no measurable effect on the time to reach harvest maturity (Table 5.3). A main effect due to flaming dose was observed, with 1.43 kg propane km^{-1} flamed treatments taking longer to mature, averaging 104 days to harvest while 0.90 kg propane km^{-1} flamed treatments averaged 101 days between transplantation and harvest. However, when compared to the non-flamed control, no difference was seen between the control and flamed treatments among hand-weeded plots. Hand-weeding in addition to flame weeding treatment affected the number of days from transplant to harvest (Table 5.3). Among flamed plots, those that were hand-weeded in addition to flaming (108 d) took longer to reach maturity than the plots that were flamed only (97 d). Visual observation suggested that onions that grew with high weed densities were less robust, perhaps resulting in less resistance to having tops fall. Overall, the plants in the flame-only treatments were less robust, and onion yield in these treatments was diminished as was described above. Flame weeding may cause damage to the crop canopy. Previous studies have demonstrated that moderate and severe foliage loss can delay crop maturity in onion (Bartolo et al., 1994). However, delays in crop maturity due to flame weeding were not observed in our study. The results in our study suggest that though flame weeding may have some subtle effects on the time required for onions to reach maturity, these effects can be expected to be minimal.

5.4.4 Leaf development and maturity

Flame weeding treatments did not negatively affect onion leaf or bulb development in this study. Leaf count data in 2006 was unreliable due to a failure to account for natural leaf senescence. Therefore, only analyses of the 2007 leaf and maturity data are presented. No effects on either leaf number or bulb diameter were observed due to flame dose or number of flame treatments at any of the five sampling times during the season (data not shown). Beginning with the third sampling time (47 DAT), both the number of leaves and bulb diameter were reduced in flame-only plots as compared to the hand-weeded and flamed plots. At 47 DAT, hand-weeded treatments averaged 12.3 leaves per plant, while flame-only treatments averaged 11.9 (P=0.08). By 63 DAT, plants in hand-weeded treatments averaged 16.4 leaves, while those in flame-only plots had 15.8 leaves each, and by 75 DAT, onion plants in hand-weeded and flamed plots averaged 17.8 leaves, greater than the 17.1 leaves per plant for onions in the flame-only treatments. The effect of hand-weeding on bulb diameter was similar to leaf number. Onion diameters in hand-weeded and flamed plots were 10% larger than those in flame-only plots, averaged over flame dose and number of treatments, at 47, 63, and 75 DAT. The differences in leaf number and bulb diameter between hand-weeded, flamed plots and flame-only plots can be attributed to the increased competition for light, water, and nutrients due to the greater weed presence in flame-only plots (Aldrich and Kremer, 1997).

Flame weeding has the potential to cause damage to leaves in onion, especially if treatments occur early in the season. In addition, if burners are not positioned correctly, unnecessary flame damage to the crop canopy may result. Foliage damage has been shown to decrease yields and delay crop maturity in onion (Bartolo et al., 1994; Muro et al., 1998). In addition, final bulb size can be predicted based upon bulb size at bulbing and the number of leaves produced after bulbing has begun (Lancaster et al., 1996). No effects of flame weeding on leaf or bulb development in onion were detected in this study. Onion appears to be sufficiently tolerant of flame weeding to justify its use.

5.4.5 Flavour characteristics

Overall, onion pungency was greater in 2007 (3.45 μmoles g^{-1}) than in 2006 (2.40 μmoles g^{-1}), though the difference between the two years varied according to treatment (Table 5.3). A year × flaming dose interaction was present, as onion pungency was greater in both the high and low flame doses in 2007 than in 2006, but to differing degrees. However, there was no effect due to flaming dose in either year. A year × weeding interaction was present as well. Onion pungency in both the hand-weeded and flamed treatments and the flame-only treatments was greater in 2007 than in 2006, though the difference in pungency in the hand-weeded and flamed treatments was greater than in flame-only treatments. There was no difference in onion pungency between hand-weeded and flamed and flame-only treatments in either year. In addition, no difference was observed in onion pungency between the non-flamed control and the flamed treatments.

Flame weeding had little effect on soluble solids concentration in onion in this study. Analysis of variance detected differences in soluble solids concentration due to the number of flame treatments and whether plots were hand-weeded in addition to being flamed (Table 5.3). As the number of flame treatments increased, soluble solids concentration trended downwards, but no significant regression was found (Figure 5.9). Soluble solids content in treatments flamed once or twice was higher than in treatments flamed four and six times. A main effect due to hand-weeding treatment was observed as well; hand-weeded and flamed treatments had lower soluble solids concentrations (7.2%) than flame-only treatments (7.6%). This result was consistent for the weedy check (8.0%) and the hand-weeded control (6.7%) as well, suggesting that the difference is caused by the greater weed flora present in flame-only plots as compared to hand-weeded plots rather than the flame treatments themselves. Soluble solids concentration in onion is has been demonstrated to be affected by cultivar and length of time in storage (Kopsell and Randle, 1997). No effect of flame weeding on soluble solids concentration was observed in this study.

Onion flavour is affected by fertilization levels, especially sulphur availability (Lancaster and Boland, 1990; Randle et al., 1995; Randle et al., 2002). Onion pungency increases as growing temperature increases (Coolong and Randle, 2003), and may be negatively correlated with bulb size (Lee et al., 2009. However, flame weeding had little effect on onion pungency or soluble solids concentration in our study.

5.4.6 Flavonoid content

The only flavonoid found in onion in detectable quantities after acid hydrolosis was quercetin. Kaempferol and myrcetin have been detected in onion in minor amounts in some studies (Leighton et al., 1992; Sellappan and Akoh, 2002;), but not in others (Hertog et al., 1992). Neither kaempferol nor myrcetin were detected in our study. Quercetin and its glycosides have been routinely found to be the major flavonoids present in onion (e.g., Hertog et al., 1992; Lombard et al., 2002; Price and Rhodes, 1997).

A year × weeding treatment interaction was observed for quercetin content (Table 5.3). In 2006, flame-only treatments had greater quercetin concentration (4032 $\mu g\ g^{-1}$ DM) than hand-weeded and flamed treatments (2238 $\mu g\ g^{-1}$ DM), while in 2007, quercetin concentration in flame-only treatments was similar to hand-weeded and flamed treatments. In addition, flame-only plots had higher quercetin concentration in 2006 (4032 $\mu g\ g^{-1}$ DM) than in 2007 (2107 $\mu g\ g^{-1}$ DM), while hand-weeded and flamed plots were similar between years. Though we could not find any reports in the literature concerning the effect of weeds on flavonoid levels in onion, weed pressure has been reported to increase flavonoids in soybean (Al-Tawaha and Seguin, 2006). A flame dose × number of flame treatments interaction was observed for quercetin content (Table 5.3). This interaction was caused by a difference between the 1.43 kg propane km^{-1} (3272 $\mu g\ g^{-1}$ DM) and 0.90 kg propane km^{-1} (2382 $\mu g\ g^{-1}$ DM) treatments only among plots flamed three times (data not shown). A difference between the high and low flaming doses was not observed in plots flamed fewer or greater numbers of times; the reason for this observation was not determined.

Flavonoid concentration in onion has been reported to vary according to cultivar (Marotti and Piccaglia, 2002; Mogren et al., 2006; Price and Rhodes, 1997) and environmental growing conditions (Patil et al., 1995). In other species, flavonoids have been reported to be affected by natural elicitors (Al-Tawaha et al., 2006; Gagnon and Ibrahim, 1997; Sivesind and Seguin, 2006). Flavonoids are also produced as a response to attack by pathogens (Stafford 1997) or physical damage caused by chemicals or UV light (Olsson et al. 1998). Flame weeding had minimal effect on quercetin concentration in onion in this study. Data from other studies did not find any temporary increases in quercetin concentration due to flame weeding that would not be detected at harvest maturity (Sivesind, unpublished data).

5.5 Summary and conclusions

This is the first study to report the effects of a season-long flame weeding regimen on a weed population in onion. Weed control efficacy differed for dicot and monocot species. Dicot weed density and shoot mass declined steadily as flame dose and the number of treatments increased. However, monocot density did not differ due to flame dose or the number of treatments in plots flamed between one and six times. Averaged across flame doses and the number of treatments, flamed treatments had a 50% reduction in monocot density as compared to the non-treated control. Effects of flame treatments on monocot shoot mass were limited. Monocot weeds that survived flame treatments therefore grew larger and made up for the reduced weed population, resulting in no reduction of monocot shoot mass. Total weed density was reduced as the number of flame treatments and flaming dose increased, though the largest declines were between the non-treated control and the first flame treatment and lowest flame dose. Total weed shoot mass was reduced as flame dose and the number of treatments increased. These results suggest that multiple flame weeding treatments are an effective method for reducing weed density and biomass in onion. However, total weed pressure in this study remained unacceptably high for onion production no matter the number of flame treatments or flame dose. Weeds that survived a flame treatment, whether through avoidance (e. g. hidden behind a clod of dirt) or tolerance, quickly grew beyond the size range at which flame weeding is effective. Additional weed control measures would be required to reduce weeds to acceptable levels. The number of flame weeding treatments necessary would therefore depend upon the frequency and efficacy of additional weed control measures. Targeted hand-weeding treatments could be used periodically to remove weeds that survived flame treatments. In-row mechanical weeding, such as the use of a torsion weeder or finger weeder, could be used in conjunction with flame weeding as well. Further research in this area is necessary.

Onion was able to withstand up to six flame weeding treatments at a dose of 0.90 or 1.43 kg propane km^{-1} with little effect on yield of marketable or total onions. No differences between the weed-free, non-flamed control and weed-free flamed treatments were observed. Flame-only treatments had reduced yields as compared to weed-free flamed treatments in both years of the study. Flame treatments alone were not able to control weeds sufficiently to avoid yield loss in this study. A combination of flame weeding and hand-weeding or mechanical measures is therefore recommended to reduce weed populations to levels acceptable for onion production.

The results of these experiments suggest that flame weeding has little effect on time to maturity, leaf and bulb development, pungency, and soluble solids content in onion. No effects on quercetin concentration due to flame treatments were detected. We were not able to find any previous reports on the effects of flame weeding on any of these factors in onion. Onion is quite tolerant of multiple flame weeding treatments, with few negative effects observed. However, as onion is a weak competitor with weeds, additional methods to satisfactorily control weeds may be necessary in order to ensure yields. The results of this study suggest that multiple flame weeding treatments would be an acceptable and valuable addition to in-season weed control in onion as long as additional weed control measures were used to increase the level of control. The overall efficacy of flame weeding treatments would also be dependent upon the composition of the weed population, with flame weeding more effective for populations where the ratio of dicot to monocot weeds is high.

5.6 Acknowledgements

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5.7 Tables and figures

Table 5.1. Significant main effects and interactions of flame dose and number of treatments on monocotyledon, dicotyledon, and total weed density (plants m^{-2}) and shoot mass (g DM m^{-2}) in onion in 2006-07. Data was log transformed to stabilize variance when necessary.

	Monocots		Dicots		Total[a]	
Effect	Density	Mass	Density	Mass	Density	Mass
Year (Y)	NS	**[b]	NS	NS	NS	NS
Flame Dose (D)	NS	NS	***	***	*	***
Y×D	NS	NS	NS	NS	NS	NS
Flame Number (N)	NS	NS	***	***	***	***
Y×N	NS	NS	NS	NS	NS	NS
D×N	NS	*	NS	NS	NS	NS
Y×D×N	NS	NS	*	NS	NS	NS

[a]Total represents the sum of monocots and dicots.

[b]* = Signficant at $P \leq 0.05$; ** = significant at $P \leq 0.01$; *** = significant at $P \leq 0.001$; NS = nonsignificant.

Table 5.2. Significant main effects and interactions of flame dose, number of flame treatments, and weeding treatment on marketable, unmarketable, and total onion yields in 2006-07. Plots were flamed with a dose of 0.90 or 1.43 kg propane km^{-1} between one and six times over the course of the growing season.

Effect	Marketable	Unmarketable	Total[a]
Year (Y)	***[b]	**	***
Flame Dose (D)	NS	NS	NS
Y×D	NS	NS	NS
Flame Number (N)	NS	NS	NS
D×N	NS	NS	NS
Y×N	NS	NS	NS
Y×D×N	NS	NS	NS
Weeding (W)	***	**	***
W×D	NS	NS	NS
W×N	NS	NS	NS
W×D×N	NS	NS	NS
Y×W	***	**	*
Y×W×D	NS	NS	NS
Y×W×N	NS	NS	NS
Y×W×D×N	NS	NS	NS

[a]Total is sum of marketable and unmarketable onion.

[b]* = Signficant at $P \leq 0.05$; ** = significant at $P \leq 0.01$; *** = significant at $P \leq 0.001$; NS = nonsignificant.

Table 5.3. Significant main effects and interactions of year, flame dose, number of flame treatments, and weeding treatment on pungency, soluble solids concentration, and quercetin concentration in onion in 2006-07. Plots were flamed with a dose of 0.90 or 1.43 kg propane km^{-1} between one and six times over the course of the growing season. Plots either received flame treatment alone or were hand-weeded in addition to flame treatment.

Effect	Days to harvest	Pungency	SSC[a]	Quercetin
Year (Y)	***[b]	**	NS	**
Flame Dose (D)	*	NS	NS	NS
Y×D	NS	*	NS	NS
Flame Number (N)	NS	NS	**	NS
D×N	NS	NS	NS	*
Y×N	NS	NS	NS	NS
Y×D×N	NS	NS	NS	NS
Weeding (W)	***	NS	***	***
W×D	NS	NS	NS	NS
W×N	NS	NS	NS	NS
W×D×N	NS	NS	NS	NS
Y×W	NS	*	NS	***
Y×W×D	NS	NS	NS	NS
Y×W×N	NS	NS	NS	NS
Y×W×D×N	NS	NS	NS	NS

[a]Soluble solids concentration

[b]* = Signficant at $P \leq 0.05$; ** = significant at $P \leq 0.01$; *** = significant at $P \leq 0.001$; NS = nonsignificant.

Figure 5.1. Density of weed species (plants m^{-2}) in end of season collection averaged across all treatments in 2006 (a) and 2007 (b). Bars represent means ±SE. Weeds are designated by their official Bayer codes; for plants without Bayer codes, US codes are used. Weed species abbreviations: AMAPO, Powell amaranth (*Amaranthus powellii* S. Wats.); AMARE, redroot pigweed (*Amaranthus retroflexus* L.); AMBAR, common ragweed (*Ambrosia artemisiifolia* L.); BROIN, smooth brome (*Bromus inermis* Leyss.); CAPBP, shepherd's-purse (*Capsella bursa-pastoris* (L.) Medik.); CHEAL, common lambsquarters (*Chenopodium album* L.); ECHCG, barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.); GASCI, hairy galinsoga, (*Galinsoga quadriradiata* Cav.); PANCA, witchgrass (*Panicum capillare* L.); POLPE, ladysthumb (*Polygonum persicaria* L.); POLSC, pale smartweed (*Polygonum lapathifolium* L.); SECE, cereal rye (*Secale cereale* L.); SETLU, yellow foxtail (*Setaria pumila* (Poir.) Roemer & J.A. Schultes); SETVI, green foxtail (*Setaria viridis* (L.) Beauv.); SINAR, wild mustard (*Sinapis arvensis* L.); SOLSA, hairy nightshade (*Solanum physalifolium* Rusby); SONAR, perennial sowthistle (*Sonchus arvensis* L.); SONOL, annual sowthistle (*Sonchus oleraceus* L.); TAROF, dandelion (*Taraxacum officinale* G.H. Weber ex Wiggers); TRIRE, white clover (*Trifolium repens* L.).

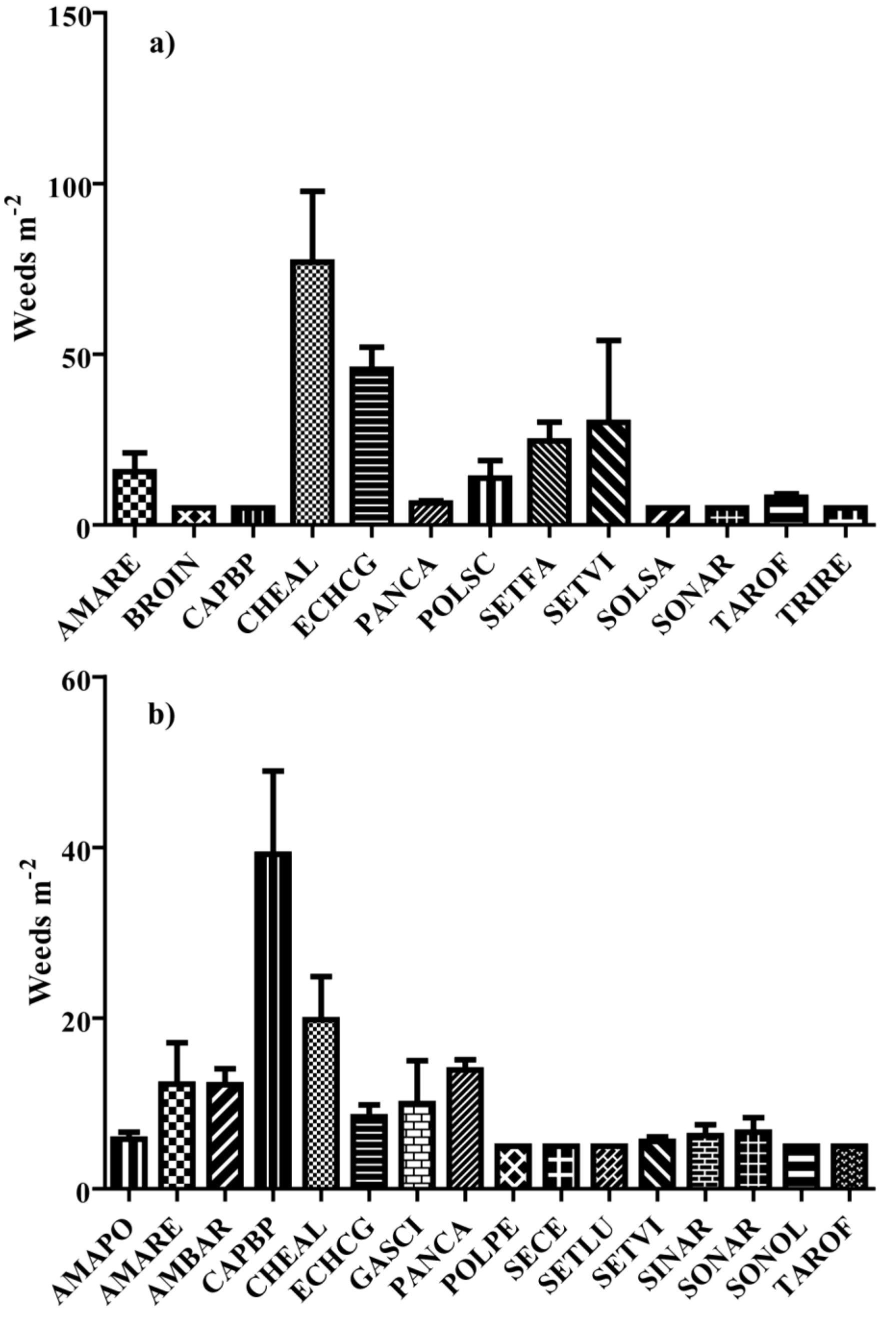
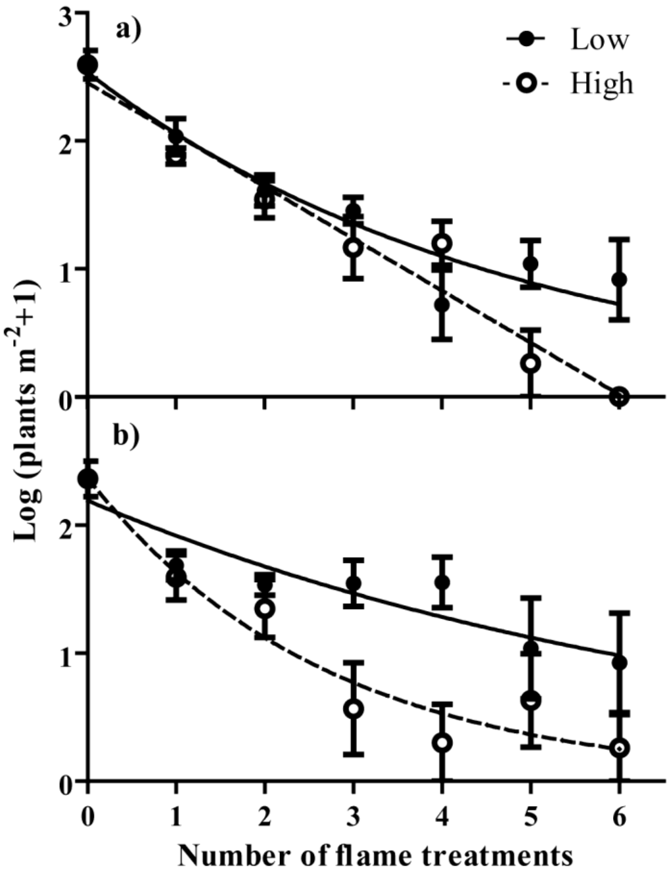

a)
Weeds m-2
150
100
50
0
AMARE
BROIN
CAPBP
CHEAL
ECHCG
PANCA
POLSC
SETFA
SETVI
SOLSA
SONAR
TAROF
TRIRE
b)
Weeds m-2
60
40
20
0
AMAPO
AMARE
AMBAR
CAPBP
CHEAL
ECHCG
GASCI
PANCA
POLPE
SECE
SETLU
SETVI
SINAR
SONAR
SONOL
TAROF

Figure 5.2. Dicot weed density (plants m^{-2}) regressed over the number of flame treatments in 2006 (a) and 2007 (b). Plots were flamed with either the low (0.90 kg propane km^{-1}) or high (1.43 kg propane km^{-1}) flaming dose between zero and six times over the course of the growing season. Points represent mean value of replicates ±SE. Regressions for low dose in 2006 and high and low doses in 2007 made using equation 2: $y = Y_o * \exp(k * x)$. Regression for high dose in 2006 made using a linear equation: $y = A + Bx$. Parameter estimates ±SE: 2006 Low (0.90 kg propane km^{-1}), Y_o, 2.54 ± 0.16; k, -0.21 ± 0.03. 2006 High (1.43 kg propane km^{-1}), A, 2.45 ± 0.12; B, -0.41 ± 0.03. 2007 Low (0.90 kg propane km^{-1}), Y_o, 2.19 ± 0.0.18; k, -0.13 ± 0.03. 2007 High (1.43 kg propane km^{-1}), Y_o, 2.37 ± 0.23; k, -0.38 ± 0.07.



a)
b)
Low
High
Log (plants m-2+1)
Number of flame treatments

Figure 5.3. Dicot shoot mass (g DM m^{-2}) regressed over flame dose, averaged over years and the number of flame treatments. Treatments were flamed between zero and six times over the course of the season. Points represent mean value of replicates ±SE. Regression made using a linear equation: $y = A + Bx$. Parameter estimates ±SE: A, 2.80 ± 0.28; B, -1.20 ± 0.25.

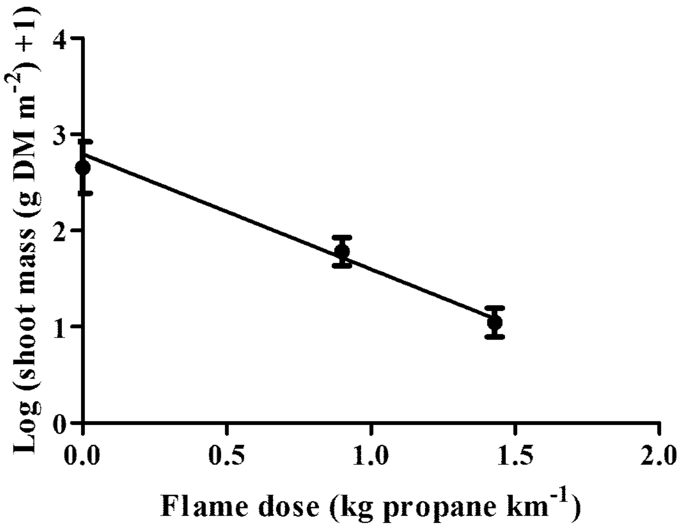

Log (shoot mass (g DM m-2) +1)
4
3
2
1
0
0.0
0.5
1.0
1.5
2.0
Flame dose (kg propane km-1)

Figure 5.4. Dicot shoot mass (g DM m^{-2}) regressed over the number of flame treatments, averaged over years and flaming doses. Points represent mean value of replicates ±SE. Regression made using equation 2: $y = Y_o*\exp(k*x)$. Parameter estimates ±SE: Y_o, 2.87 ± 0.23; k, -0.23 ± 0.03.

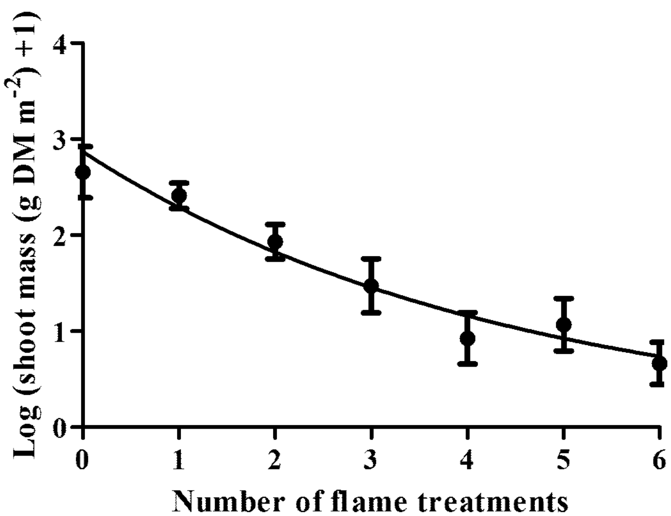

Log (shoot mass (g DM m-2) +1)
4
3
2
1
0
0
1
2
3
4
5
6
Number of flame treatments

Figure 5.5. Total weed density (plants m^{-2}) in 2006 and 2007 regressed over flaming dose and averaged over number of flame treatments. Treatments were flamed between zero and six times over the course of the season. Points represent mean value of replicates ±SE. Regression for 2006 made using equation 1: $y = (A - B)*\exp(k*x) + B$. Regression for 2007 made using equation 2: $y = Y_o*\exp(k*x)$. Parameter estimates ±SE: 2006, *A*, 538.8 ± 44.6 ; *B*, 73.9 ± 24.0 ; *k*, 3.69 ± 2.15. 2007, Y_o, 276.6 ± 20.7; *k*, -1.72 ± 0.15.

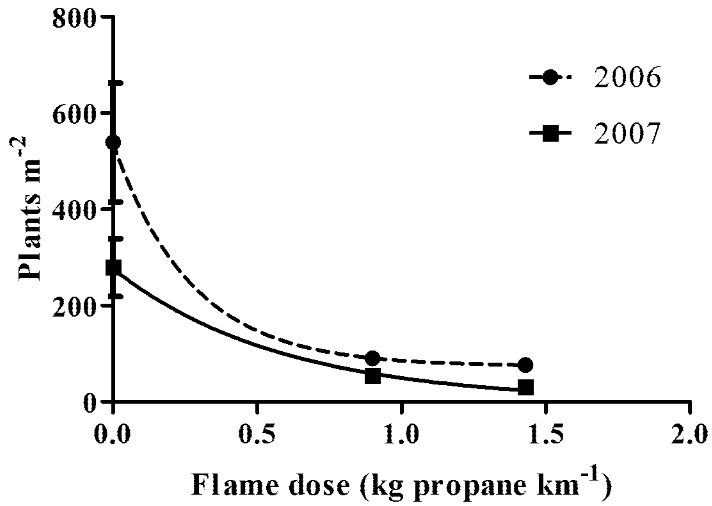

800
600
400
200
0
Plants m-2
0.0
0.5
1.0
1.5
2.0
Flame dose (kg propane km-1)
2006
2007

Figure 5.6. Total weed density (plants m^{-2}) in 2006 and 2007 regressed over the number of flame treatments and averaged over flaming dose. Points represent mean value of replicates ±SE. Regressions made using equation 1: $y = (A - B)*\exp(k*x) + B$. Parameter estimates ±SE: 2006, *A*, 537.7 ± 41.0; *B*, 61.7 ± 14.7; *k*, -1.53 ± 0.32. 2007, *A*, 278.2 ± 20.3; *B*, 33.6 ± 7.1; *k*, -1.73 ± 0.37.

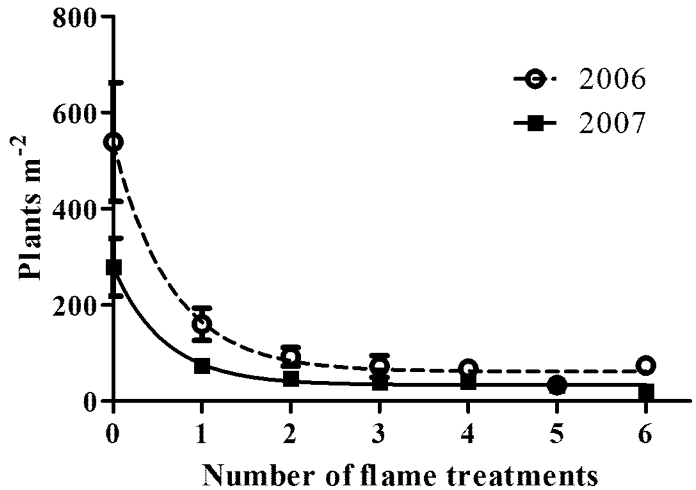

800
600
400
200
0
Plants m-2
2006
2007
0
1
2
3
4
5
6
Number of flame treatments

Figure 5.7. Total weed shoot mass (g DM m^{-2}) in 2006-07 regressed over flaming dose and averaged over number of flame treatments. Treatments were flamed between zero and six times over the course of the season. Points represent mean value of replicates ±SE. Regression made using equation 2: $y = Y_o * \exp(k * x)$. Parameter estimates ±SE: Y_o, 1187.0 ± 133.9; k, -0.87 ± 0.13.

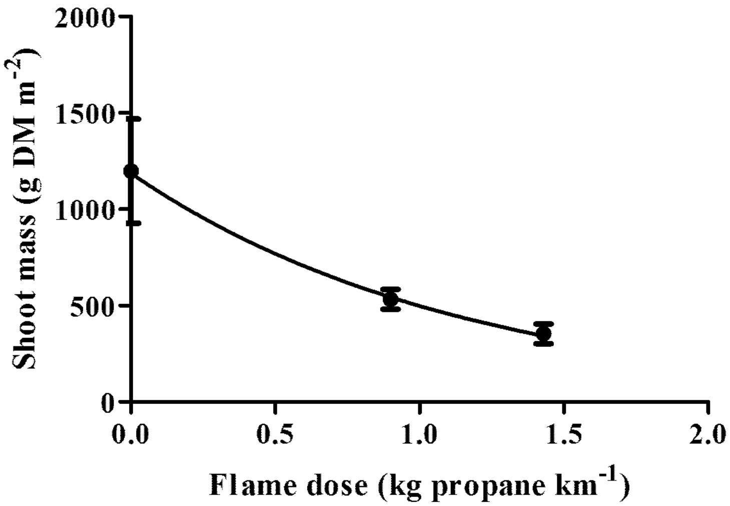

Shoot mass (g DM m-2)
2000
1500
1000
500
0
0.0
0.5
1.0
1.5
2.0
Flame dose (kg propane km-1)

Figure 5.8. Total weed shoot mass (g DM m^{-2}) in 2006-07 regressed over the number of flame treatments and averaged over flaming dose. Points represent mean value of replicates ±SE. Regression made using equation 1: $y = (A - B)*\exp(k*x) + B$. Parameter estimates ±SE: *A*, 1160.0 ± 123.0; *B*, 218.5 ± 112.6; *k*, -0.50 ± 0.19.

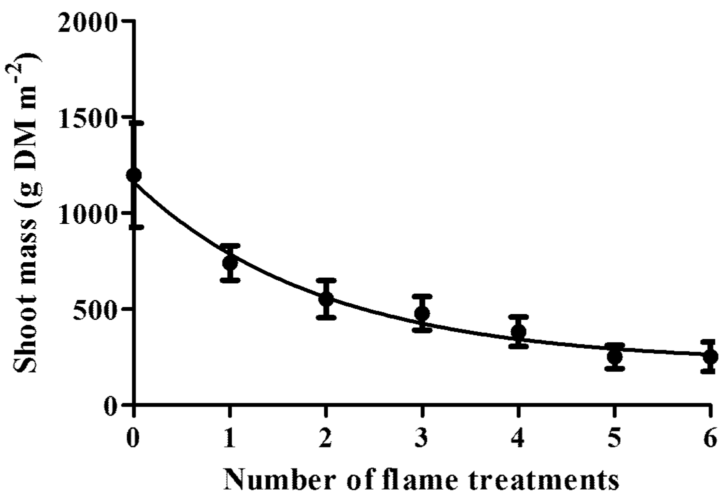
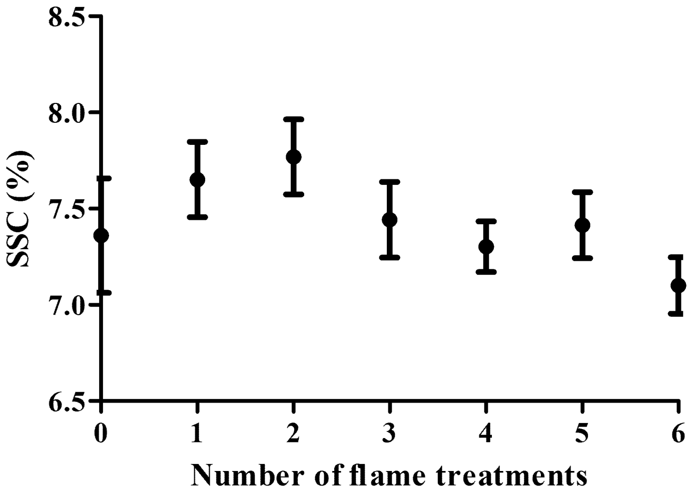

Shoot mass (g DM m-2)
2000
1500
1000
500
0
0
1
2
3
4
5
6
Number of flame treatments

Figure 5.9. Soluble solids content (SSC) in 2006-07 in treatments flamed zero to six times over the course of the season and averaged over years, weeding treatment, and flaming dose. Points represent mean value of replicates ±SE.


8.5
8.0
7.5
7.0
6.5
SSC (%)
0
1
2
3
4
5
6
Number of flame treatments

5.8 Connecting text

In Chapter 4, we evaluated four vegetable crops in their ability to withstand flame weeding. In that study, onion and broccoli proved to be the most flame tolerant. In the following study, we evaluated the effects of multiple flame weeding treatments in broccoli on yield, development, and crop quality parameters, as well as efficacy as weed control. This approach provides a more complete picture of the effects that multiple flame treatments would have on broccoli production.

The following manuscript was co-authored by the candidate, Dr. Maryse Leblanc, Insitut de recherche et de développement en agroenvironnement, Dr. Daniel Cloutier, Institut de malherbologie, Dr. Philippe Seguin, Department of Plant Science, Macdonald Campus of McGill University, and Dr. Katrine Stewart, Department of Plant Science, Macdonald Campus of McGill University. The candidate designed the experiments, carried out the field experiments, performed the laboratory analyses, conducted the data analyses, and was the primary author of the manuscript. Drs. Leblanc and Cloutier provided funds and reviewed the manuscript. Drs. Seguin and Stewart assisted the candidate in the experimental design, provided funds and supervisory guidance, and reviewed the manuscript.

6.0 Selective Flame Weeding in Broccoli: Effects on Productivity, Development, Glucoraphanin Concentration, and Weeds

6.1 Abstract

Field experiments were conducted to evaluate the effects of multiple selective flame weeding treatments in broccoli. Broccoli was flamed from one to four times over the course of the growing season with either a high (1.43 kg propane km^{-1}) or low (0.90 kg propane km^{-1}) flame dose. Broccoli yields in treatments flamed with either the high or low flame dose were not different than the non-flamed, weed-free control. Treatments that were hand-weeded in addition to flame treatment had greater yields than treatments that were only flame weeded in one of two years; in the other hand-weeded and flame treatments had lower yields. Broccoli yield was not affected by the number of flame treatments. Density and shoot mass of monocot weeds decreased as the number of flame treatments increased in one of two years. Dicot weed density declined as the number of flame treatments increased. Dicot shoot mass was not affected by the number of flame treatments; though shoot mass did not differ between flame doses, the non-treated control had greater shoot mass than all flame treatments. Flame weeding treatments did not affect the number of days to maturity. Low dose treatments had greater glucoraphanin concentrations than high dose treatments, but overall effects of flame weeding on glucoraphanin concentration were limited. Results of these experiments suggest that broccoli is reasonably tolerant of flame weeding and can withstand multiple selective flame weeding treatments.

6.2 Introduction

Flame weeding is increasingly being utilized for weed control as an alternative to herbicides. It is often used in organic agriculture and other situations where herbicides are unavailable or undesirable. Flaming can be quite valuable as a means to reduce the expensive and labor intensive hand-weeding that would otherwise be required in the absence of chemical herbicides (Ascard, 1989; Mojžiš, 2002; Nemming, 1993). Flame weeding is commonly used before the crop is present, either as a stale seedbed technique prior to seeding or planting, or before crop emergence. This is often the case in small seeded, slowly germinating crops such as onion and carrot (Ascard, 1995). Because there is no differentiation between targeted and non-targeted plants in these cases, this type of flaming can be referred to as non-selective. An alternative to non-selective flaming is selective flaming, when crop plants are present. Selectivity occurs by flaming at doses high enough to control weeds but low enough not to cause undue harm to the crop plants.

A number of studies have been carried out investigating the use of flaming, including technical aspects (Ascard, 1997), and weed response (Ascard, 1994; Ascard, 1995; Cisneros and Zandstra, 2008; Knezevic and Ulloa, 2007a; Knezevic et al., 2008; Sivesind et al., 2009). Fewer studies have investigated the impact of flame weeding on crop species. Ascard (1989) compared the use of selective flaming to herbicides and mechanical cultivation in onions. In hand-planted sets, no reduction in yield was reported after one flame treatment at emergence and two selective flame weeding treatments. Knezevic and Ulloa (2007c) investigated the use of flame weeding in six crops; they reported field corn and sorghum the most tolerant, while red clover and alfalfa were both susceptible to flame damage. Vester (1988) flamed curly kale (*Brassica oleracea* Acephala group) at the 4-5 leaf stage and observed considerable damage when flamed at 66 kg ha^{-1}, but only minor damage when the flame dose was reduced to 44 kg ha^{-1}. Wszelaki et al. (2007) investigated the effects of selective flaming on weeds, crop quality, and yield of tomato (*Solanum lycopersicum* L.) and cabbage [*Brassica oleracea* L. (capitata group)]. This study reported that flaming slowed cabbage growth by two weeks and delayed harvest. Yield parameters were reduced in all flamed plots as compared to a hand-weeded control. The latter report is one of the few published investigations of the effects of flame weeding on crop quality, where the incidence of blossom end rot was reduced by flaming in tomato. However, flaming frequency was not evaluated as plots were only flamed a single time.

Broccoli (*Brassica oleracea* L. var. *italica*) is a hardy, popular cool season vegetable which can be grown as a spring or fall crop. In the spring, transplants can be used in order to shorten the time required to maturity. Some early investigations into the use of flame weeding in cole crops have been carried out (Wilson and Ilnicki, 1966). However, there is little current information available in the literature regarding the use of selective flame weeding in broccoli.

Glucosinolates comprise a class of thioglucosides that are found in appreciable quantities in a number of plant species, but primarily in plants belonging to the *Brassicaceae*. Glucosinolates and their derivatives are thought to be part of the plant defense response to herbivory and fungal infection (Kiddle et al., 2001). Glucosinolates may also play a role in protecting plants from bacterial pathogens

(Charron et al., 2002). Glucosinolates are hydrolyzed by mirosinase into a number of volatile and non-volatile compounds, among these the isothiocyanates. The isothiocyanate sulphoraphane, a hydrolyzed product of glucoraphanin, has been shown to be an effective inducer of phase II detoxification enzymes and may provide anti-cancer benefits (Fahey et al., 1997; Vallejo, 2003; Zhang et al., 1992). In addition, glucosinolates and their derivatives are important components of the flavour profile of edible cruciferous crops (MacLeod, 1976). Glucosinolate concentrations in cruciferous species have been shown to be affected by both genetic and environmental factors (Ciska, 2000; Shelp et al., 1993). Fertilization (Aires et al., 2006; Zhao, 1994), developmental stage (Vallejo, 2003), insect predation (Bodnaryk, 1992), and fungal infection (Ludwig-Müller et al., 1999) have all been cited as affecting glucosinolate concentrations in various *Brassicaceae* species.

Due to their role in plant defence, flavour characteristics, and potential health benefits, it is important to understand all factors which could affect glucosinolate concentration in cruciferous crops. There have been no reports in the literature concerning the effect of flame weeding on glucosinolate content in any *Brassicaceae* species. As a number of other stresses have been shown to alter glucosinolate content in cruciferous crops, there is a realistic possibility that flame weeding may as well. Flaming can impose a significant stress on crop species and any effect on glucosinolate concentration needs to be understood in order to determine the impact of management on crop quality, which ultimately could affect its value.

This study was conducted to investigate the use of multiple post-emergent flame weeding treatments in broccoli. Previous studies (Chapter 4) indicated that broccoli was tolerant to a single flame weeding treatment occurring at a variety of different points during the season. However, a single flame treatment will not be sufficient to control weeds for an entire growing season. Producers who invest in flame weeding equipment will be interested in utilizing this method at multiple points during the season to control weeds. There is not any information currently available concerning the effects of multiple flaming weeding treatments on broccoli, or on the efficacy of such treatments for weed control. Therefore, we conducted field experiments to investigate the effects of multiple flame weeding treatments on broccoli yield, development and maturity, and glucosinolate concentration, and weed control efficacy.

6.3 Materials and methods

6.3.1 General field management

Experiments were conducted at the Institut de recherche et de développement en agroenvironnement (IRDA) in Saint-Hyacinthe, QC, Canada (45° 38' North, 72° 57' West) in 2006 and 2007. The soil type for all experiments was a Duravin loam with 2.2% organic matter and a pH of 6.7. Fields were fertilized according to local recommendations as indicated by soil tests (Centre de Référence en Agriculture et Agroalimentaire du Québec, 2003). In 2006, during soil preparation fields received a broadcast application of 685 kg ha^{-1} of 14-21-21 NPK and 1.5 kg ha^{-1} boron on May 29. A banded application of 80 kg ha^{-1} of 27-0-0 NPK was applied 15 cm from the row at a depth of 2 cm on July 6. In 2007, 420 kg ha^{-1} of 13-12-23 NPK fertilizer was broadcast on May 22, with a banded application of 106 kg ha^{-1} of 27-0-0 NPK on July

6. Sixty-day-old broccoli seedlings (cv. Everest) were transplanted into the field on May 30, 2006, and May 23, 2007. Plots were 5 m long with plant spacing of 30 cm and 90 cm between rows, resulting in a plant density of 30,000 plants ha^{-1}.

6.3.2 Experimental design

Experiments were set up in randomized complete block design (RCBD) with four replications and split-plot restriction. Main plots were factorial combinations of flaming dose and the number of flame treatments. There were two levels for flaming dose: a low flaming dose (0.90 kg propane km^{-1}) and a high flaming dose (1.43 kg propane km^{-1}). These rates were chosen based on results from previous experiments (Chapters 3 and 4) which indicated that these rates would be appropriate for use in broccoli and provide effective weed control. There were four levels for the number of flame treatments: plots were flamed 1, 2, 3, or 4 times over the course of the growing season. In addition one control plot was included that received no flame treatment. Plots were split into two subplots; one subplot was hand-weeded in addition to the appropriate flame treatment, the other received only the flame weeding treatment. In hand weeded splits, all weeds that were not controlled with flaming were removed following flame treatment in order to remove confounding effects of differing weed pressures resulting from different flame treatments. One subplot of the control therefore received no weed control treatment and served as the weedy check, the other subplot was maintained weed-free through hand-weeding and served as the non-flamed, weed-free control. The experiment therefore contained 36 main plots (4 repetitions of 2 flaming doses × 4 number of flame treatments + 1 control) each split into two subplots. Time points for the flame treatments were 10, 22, 39, and 50 days after transplantation (DAT) for the 1, 2, 3, and 4 times flamed treatments, respectively, in 2006, and 16, 27, 37, and 48 DAT in 2007. Flame treatments were administered when weeds reached the cotyledon to 2-leaf stage, developmental stages determined in a previous study (Chapter 3) to be susceptible to the flaming rates employed. Due to a high number of escapes and uneven emergence later in the season, the third and fourth flame treatments were administered two weeks following the previous treatment, weather permitting.

Four broccoli plants in each split-plot were monitored for leaf production at three points over the course of the season: 27, 41, and 51 DAT in 2006, and 21, 36, and 50 DAT in 2007. Leaf counts were based upon the total number of leaves produced, including those that had already senesced. To gauge the efficacy of the different flame weeding regimens, a 20 ×100 cm quadrat was placed along the center of the row in each weedy split-plot, and at broccoli harvest all weeds within were separated by species, counted, dried at 60 °C until constant mass, and weighed. Seven broccoli plants were randomly selected per plot and individual terminal heads were harvested upon reaching maturity. Stalks were cut to give an overall length of 20 cm and the date of harvest recorded. Head diameter in millimeters was measured using a digital caliper and heads weighed individually. Samples of florets from each head were removed and frozen in liquid nitrogen, placed in resealable polyethylene bags and stored at -25 °C until lyophilization for glucosinolate analysis.

6.3.3 Flaming specifications

Flame treatments were performed using a propane fuelled, tractor mounted unshielded Red Dragon two burner system (Flame Engineering, Inc., LaCrosse, KS), directed perpendicularly to the crop row. Burners were set at an angle of 30° with respect to the horizontal 18 cm from the row measured along the angle. Burners were staggered to keep flames from intersecting and deflecting upwards and damaging crop canopy. To achieve a flaming dose of 0.90 kg propane km^{-1}, the fuel pressure was set at 117 kPa and with a driving speed of 3 km h^{-1}. A fuel pressure of 214 kPa and a driving speed of 3 km h^{-1} were utilized to achieve a flaming dose of 1.43 kg propane km^{-1}.

6.3.4 Flaming dose calculation

Flame weeding doses are often expressed in terms of fuel used per area of coverage (e.g. kg propane ha^{-1}). We have presented flaming doses used in this study as propane burned per unit of row length (kg propane km^{-1}). We used this method as it describes the flaming dose effectively and it would be simple to accurately compare dosages used in separate studies by authors using different equipment. To convert the rates used in this study to kg propane ha^{-1}, all that is required is to multiply by 10 and divide by the row width in meters. For example, in this study we used a row spacing of 0.90 meters, so a dose of 0.54 kg propane km^{-1} would be equal to 6 kg propane ha^{-1}. For a row spacing of 0.60 meters, this same example would be equal to 9 kg propane ha^{-1}. This approach simplifies the comparison of rates used by different parties, and makes it easy to calculate the actual amount of fuel that is required for any given field.

6.3.5 Glucosinolate analyses

Glucosinolate content was determined using a procedure modeled after the EU official method (ISO, 1992). Florets from broccoli heads previously frozen were mixed, lyophilized, and ground to a fine powder using a mortar and pestle. Samples of 500 mg were extracted by adding 5 mL of 70% methanol heated to 75° C with 200 µL of 5 mM sinigrin added as internal standard (no sinigrin was detected in test samples). After centrifugation, supernatants were set aside and residues extracted a second time with an additional 5 mL of 75° C methanol (70%). Supernatants were mixed and added to 1 mL 0.5 M barium acetate and centrifuged at 3000 g for 10 min (Schreiner et al., 2006). Two mL of this extract was applied to columns containing 250 µl of DEAE A-25 Sephadex resin (ISO, 1992). Glucosinolates were desulfated with the addition of 250 µL of purified *Helix pomatia* sulfatase and left to sit for 18 h. Desulfo-glucosinolates were then eluted with 2 mL of d_2H_2O and subjected to HPLC analysis.

Glucosinolate quantification was conducted on a Varian Polaris HPLC system (Varian, Inc., Palo Alto, CA) consisting of two model 210 pumps, a model 410 autosampler, a PDA detector model 330, and Star Chromatography workstation system control software version 6.30. Glucosinolates were separated on a Phenomenex (Phenomenex, Torrance, CA) Luna C18 column (5 µm, 250 × 4.6 mm). Desulfo-glucosinolates were separated using a linear gradient of water (Pump A) and acetonitrile (Pump B) with a flow rate of 1.0 mL min^{-1} as follows: 0 min: isocratic 100% A for 2 min, from 2 to 25 min a linear gradient from 0 to 30% B, isocratic 30% B for 2 min, linear gradient from 30 to 0% B over 1 min, isocratic 100% A for 4 min. Glucosinolates were detected at a wavelength of 229 nm (ISO, 1992). Because of difficulties acquiring authentic standards of glucosinolates it was decided to limit quantification to glucoraphanin. Glucoraphanin has been identified as being the most prevalent glucosinolate in broccoli, representing between 50 (Tian et al., 2005) and 56% (Kushad et al., 1999; Schreiner et al., 2006) of total glucosinolates, and 68 (Kushad et al., 1999) and 87% (Schreiner et al., 2006) of aliphatic glucosinolates. In addition, sulphoraphane, the isothiocyanate derived from glucoraphanin, is known to be an inducer of mammalian detoxification and antioxidant enzyme activity and has been the subject of many studies investigating the health benefits of these compounds (Farnham et al., 2004). Identification and quantification of desulfo-glucoraphanin was determined by comparison to standard curves of authentic standards of desulfo-glucoraphanin (C_2 Bioengineering, Karslunde, DK) and to the internal standard.

6.3.6 Statistical analyses

All data were subjected to analysis of variance (ANOVA) using the MIXED procedure of the Statistical Analysis System to identify significant treatment effects and interactions (SAS, 2003). Homogeneity of variances was tested using the Brown-Forsythe test. Weed data was log transformed to homogenize variance between treatments. Transformed data were back transformed for presentation in the text. Year, flame dose, number of flame treatments, and weeding treatment were considered fixed. Repetition and interactions with repetition were considered random. Leaf counts occurred at varying times during the growing season; treatments were not included in an analysis if the full treatment regimen had not yet been implemented (i.e. plots receiving 4 flame treatments were not included in analyses of the first leaf count, which occurred prior to the fourth flaming). Because leaf counts occurred at different points in relation to the flame treatments, leaf counts were examined in years separately.

Differences between fixed effects means were determined using the LSMEANS statement and the PDIFF option, which declares differences using Fisher's protect least significant difference (LSD). When interactions between fixed effects were significant, simple effects were determined using the SLICE option of the LSMEANS statement (Littell et al., 2002). Significance was declared at $P \leq 0.05$ unless noted. Control plots were not included in the initial ANOVA when testing for treatment effects. Single degree of freedom contrasts were then used to test for differences between experimental treatments and the control. Regression analyses were performed when analysis of variance indicated significant effects for quantitative factors. Linear regression was performed using SAS (SAS, 2003). Linear, quadratic, and cubic factors were tested for significance. Non-linear regressions were performed using GraphPad Prism v. 5.02 (GraphPad Software, Inc., La Jolla, CA). Different models were evaluated, and Akaike's information criterion (AIC) was used to determine the model which best fit the data (Motulsky and Christopoulos, 2004). Lack-of-fit *F*-tests was used to test the fit of the model to the data (Motulsky and Christopoulos, 2004). For data best fit by a nonlinear model, a number of equations were tried. Models evaluated included the exponential model

$$y = (A - B)*\exp(k*x) + B \qquad [1]$$

where A is the value of y when X=0, B is the lower plateau, and k is the rate of change of x. Other data was best fit by

$$y = Y_o*\exp(k*x) \qquad [2]$$

where Y_o is the value of y at x=0 and k is the rate of change of x.

6.4 Results and discussion

6.4.1 Yield

A flame dose main effect was present for both marketable and total yield (Table 6.1), with treatments receiving the low flaming dose producing higher yields (12.9 and 13.1 T ha^{-1} for marketable and total yields, respectively) than treatments flamed with the high dose (11.7 and 11.9 T ha^{-1}), averaged over all other factors. However, neither the high nor low dose had yields different than the non-flamed control (12.1 and 12.3 T ha^{-1} for marketable and total yields, respectively). Regressions of marketable or total yields against flame dose were not significant. An interaction between year and weeding treatment was present for marketable and total yields. In 2006, the flame-only treatments produced lower marketable yields (7.2 T ha^{-1}) than treatments that were hand-weeded in addition to being flamed (8.2 T ha^{-1}). Total yields in flame-only treatments (7.7 T ha^{-1}) were lower than hand-weeded and flamed treatments (8.5 T ha^{-1}) as well ($P = 0.07$). However, in 2007, flame-only treatments out produced hand-weeded and flamed treatments for marketable (17.8 and 15.9 T ha^{-1}) and total yields (17.9 and 16.0 T ha^{-1}). Weed pressure was much greater in 2006 than in 2007, resulting in the reduced yields observed in flame-only plots in 2006. The lower level of weed pressure in 2007 was clearly not enough to adversely affect yields. One possible explanation for this unexpected result is that treatments were hand-weeded too aggressively early in the 2007 season, causing damage to the young plants. A year × weeding interaction was present for the diameter of marketable broccoli heads, which mirrored what was observed for marketable yields. In 2006, hand-weeded and flamed treatments had greater marketable diameters than flame-only plots (114 and 110 mm, respectively), while flame-only treatments had greater marketable diameters than hand-weeded and flamed plots in 2007 (133 and 126 mm). A year × flame number interaction was observed for marketable diameter. Broccoli diameters were greater in 2007 for treatments flamed 1, 2, and 4 times; treatments flamed 3 times were greater statistically similar year to year. Regressions were not significant in either year for number of flame treatments. A year × weeding interaction was present for total broccoli head diameter. In 2006, no weeding effect was observed. In 2007, the flame-only treatments had greater total diameters than the hand-weeded and flamed treatments. No main effects of or interactions with flaming dose, number of flame treatments, or weeding status were observed for yield of unmarketable broccoli or unmarketable broccoli diameter. A main year effect for yield and diameter of unmarketable broccoli was present, with 2006 having greater yields (0.4 vs. 0.1 T ha^{-1}) and greater broccoli head diameters (28 vs. 6 mm) than 2007.

Overall, broccoli proved quite tolerant of multiple flame weeding treatments. No negative yield effects were observed due to the number of flame treatments. High dose treatments had decreased yields compared to low dose treatments, though neither were significant from the non-flamed control. Under greater weed pressure in 2006, hand-weeded and flamed treatments produced greater yields than flame-only treatments. It appears flame weeding alone was not sufficient to reduce weeds below thresholds which cause yield loss in 2006. Under lesser weed pressure, flame-only plots actually produced higher yields than plots that were hand-weeded in addition to flame treatments. The reason for this latter observation was not determined, but we hypothesise that it could be due to damage caused by the hand-weeding treatment.

There is little information available in the literature concerning the effect of flame weeding on broccoli. Wilson and Ilnicki (1966) investigated the use of flame weeding in a number of cole crops, including broccoli. No yield loss in broccoli was observed from one or two flame weeding treatments in their study. Other cole crops, such as cabbage (Netland et al., 1994; Wselaki et al., 2007) and kale (Vester, 1988) have been evaluated for flame weeding tolerance with mixed results.

6.4.2 Weed effects

The weed flora was a mixture of primarily monocot and dicot annual weeds (Figure 6.1). In 2006, the dominant species was barnyardgrass [*Echinochloa crus-galli* (L.)

Beauv.], with common lambsquarters (*Chenopodium album* L.) the most common dicot species. The weed flora was less dominated by a single weed species in 2007, with shepherd's-purse [*Capsella bursa-pastoris* (L.) Medik.] the most common species followed by common lambsquarters and several grass species. Weeds collected at the end of the season were harvested and separated according to species. For analysis, weeds were divided into monocots, dicots, and combined for total weeds (sum of monocots and dicots). As indicated by the non-treated control, total weed pressure was higher in 2006 (519 plants m^{-2}) than in 2007 (299 plants m^{-2}). A majority of this difference was a result of the greater density of monocot weeds in 2006 than 2007 (382 and 37 plants m^{-2}, respectively).

A year × flame number interaction was observed for monocot density (Table 6.2). In 2006, there was no difference between flamed treatments; however the non-treated control (382 plants m^{-2}) had greater monocot weed density than all flamed treatments (188 plants m^{-2}, averaged across all flamed treatments). In 2007, monocot weed density was reduced as the number of flame treatments increased (Figure 6.2). Monocot density decreased from 51 plants m^{-2} in treatments flamed once to less than 1 plant m^{-2} in treatments flamed four times (Figure 6.2). Monocot weed density was higher in 2006 than in 2007 for all flame treatments.

Response of shoot mass of monocots was similar to that of monocot density (Table 6.2). A year × flame number interaction was observed, as monocot shoot mass decreased as the number of flame treatments increased (Figure 6.3) in 2007. Plant shoot mass decreased from 8 g DM m^{-2} in the non-treated control to nearly 0 g DM m^{-2} when flamed three times. In 2006, there was no difference between flamed treatments; however the non-treated control had greater monocot weed density (154 plants m^{-2}) than all flamed treatments (61 plants m^{-2}).

A main effect of the number of flame treatments was present for dicot weed density (Table 6.2). Averaged over flame dose and years, dicot weed density decreased as the number of flame treatments increased (Figure 6.4). Weed reductions were most pronounced between the non-treated control and first flame treatment, and grew progressively smaller as the number of flame treatments increased. Dicot weed density was reduced from 165 plants m^{-2} in the non-treated control to 3 plants m^{-2} in the plots flamed four times. No differences were observed between flamed treatments for dicot shoot mass due to flame dose or number of flame treatments (Table 6.2). However, the non-treated control had greater dicot shoot mass (30 g DM m^{-2}) than all flamed treatments (2 g DM m^{-2}). A single early season flame treatment was sufficient to reduce dicot shoot mass by 93%; additional flame treatments were unable to reduce mass of dicot weeds further.

Total weed density was affected by both flaming dose and number of treatments (Table 6.2). Total weed density decreased from 346 plants m^{-2} in the non-treated control to 49 plants m^{-2} in the 1.43 kg propane km^{-1} flame dose (Figure 6.5). Similar to what was observed for monocot density, a year by number of flame treatment interaction was present. In 2006, no differences due to the number of flame treatments were observed in plots flamed one to four times. Still, the non-treated control had greater total weed density (519 plants m^{-2}) than all flamed treatments (201 plants m^{-2}). This effect can largely be attributed to differences in monocot weed density. In 2007, total weed density in decreased as number of flame treatments increased (Figure 6.6). Total density decreased from 294 plants m^{-2} in treatments flamed once to less than 4 plants m^{-2} in treatments flamed four times.

A flame dose × number of flame treatments interaction was present for total weed shoot mass (Table 6.2). When flamed with a dose of 1.43 kg propane km^{-1}, total shoot mass decreased as the number of flame treatments increased (Figure 6.7). However, shoot mass in plots flamed with the 0.90 kg propane km^{-1} dose were best described by a quadratic function; shoot mass decreased as the number of flame treatments increased from zero to three, but increased at the fourth flame treatment (Figure 6.7). This observation is unexpected, and the reason for its occurrence is unknown.

Weed density and shoot mass was affected by both flame number and number of treatments in this study. Monocot weed density and shoot mass was affected by the number of treatments; in both years of the experiment, monocot density and mass were reduced in flamed treatments compared to the non-treated control. Increasing the number of flame treatments further decreased density and shoot mass of monocot weeds in 2007 but not in 2006. It is notable that the non-treated control in 2006 had approximately 10 times as many monocot weeds per square meter as the non-treated control in 2007. Dicot weed density was negatively correlated with the number of flame treatments over the two years of the experiment. Shoot mass of dicot weeds was greater in the non-treated control than all flamed treatments; no further differences were observed. Total weed density was reduced as flame dose increased. Similar to monocot density, total weed density was reduced with each additional flame treatment in 2007 but not in 2006. This is largely a result of the response of monocot weeds to flame treatments. Total weed shoot mass was reduced as the number of flame treatments increased from zero to three for both high (1.43 kg propane km^{-1}) and low (0.90 kg propane km^{-1}) flame doses. Total weed shoot mass increased from three to four flame treatments in the low flame dose only.

Dicot weed density was reduced as the number of flame treatments increased in both years of the study. Monocot weed density and shoot mass were reduced as the number of flame treatments increased in one of two years of the study. Monocot density and shoot mass were affected by the number of flame treatments in 2007, which had significantly lower monocot weed density than 2006 (37 and 382 plants m^{-2}, respectively). Previous studies have found flame weeding to provide better control of dicot than monocot species (Knezevic et al., 2008; Knezevic and Ulloa, 2007a; Sivesind et al., 2009). However, these previous studies only examined short term effects of a single flame treatment, not season long control by multiple flame treatments.

6.4.3 Time to Maturity

Effects of flame weeding on the time for broccoli plants to produce a mature head were limited. A year × weeding × dose interaction was observed; when sliced for year a weeding × dose interaction was present in 2006 but not in 2007 (data not shown). In 2006 among plots flamed with the high dose (1.43 kg propane km^{-1}), the time for heads to reach maturity was greater in flame only plots (63 days) than in hand-weeded and flamed plots (61 days). There were no other effects due to weeding treatment or flame dose. The time to maturity was not affected by the number of flame treatments in this study. Single degree of freedom contrasts did not detect any differences between flamed plots and the non-flamed control. Results of this study indicate that flame weeding should not be expected to alter the time for broccoli to reach harvest maturity.

6.4.4 Developmental response

Due to variation in the timing of leaf counts with respect to flame treatments between the first and second year of the experiment, leaf count data were analyzed separately by count and year. In 2006, flame dose and number of flame treatments main effects were observed in count 1. Plants flamed at 1.43 kg propane km^{-1} had fewer leaves (8.1) than those flamed at 0.90 kg propane km^{-1} (8.4). However, regression analyses were nonsignificant. In addition, plants flamed twice had fewer leaves (8.1) than non-flamed plants and plants flamed a single time (8.4) (Figure 6.8). No differences were seen between treatments at the second or third leaf counts in 2006. In 2007, no difference in leaf number was observed between treatments at the first leaf count and the non-flamed control. At count 2 in 2007 (21 DAT), flame dose × flame number and weeding × flame dose interactions were observed. However, no significant regressions were found for either effect. No treatment effects on leaf number were observed in count 3 (36 DAT) in 2007.

Effect of flame dose and the number of flame treatments on the number of broccoli leaves were limited. In 2006, leaf number was reduced in plants flamed with the 1.43 kg propane km^{-1} dose and those flamed twice at the first count (10 DAT). No treatment effects due to flame treatments were detected at the second or third leaf counts. In 2007, effects of flame treatments on leaf count occurred only at the second leaf count (21 DAT). However, as no differences in leaf number were detected at the last leaf count conducted in either year, it appears as though any negative effects on leaf number due to flaming at the rates used in this study were short lived, and plants able to recover.

6.4.5 Glucoraphanin concentration

Glucoraphanin concentration was affected by flaming dose and weeding treatment (Table 6.3). A main effect due to flaming dose was observed, as plots flamed with the low dose (0.90 kg propane km^{-1}) contained greater glucoraphanin concentrations than those flamed with the high dose (1.43 kg propane km^{-1}) (2.72 and 2.31 μmoles g^{-1} DM, respectively). The high dose treatment had glucoraphanin concentrations 81% of the non-flamed control, which was similar in glucoraphanin concentration to the low dose treatment. Regression analyses were not significant. A weeding × year interaction was observed (Table 6.3), as the effect of weeding treatment on glucoraphanin concentration differed in the two years of the study. In 2006, flame only plots had higher glucoraphanin concentrations than hand-weeded and flamed plots (2.03 and 1.57 μmoles g^{-1} DM, respectively), while in 2007 the opposite was observed, with hand-weeded and flamed plots having higher glucoraphanin concentrations than plots that only received flame treatment as weed control (3.40 and 3.06 μmoles g^{-1} DM). There was much higher weed pressure in 2006 than 2007, especially for monocot species (383 and 34 plants m^{-2}, respectively), which could have induced an increase in glucoraphanin concentration in the flame-only treatments in 2006. However, it is unknown what would cause hand-weeded and flamed plots to have greater glucoraphanin concentration in 2007. Data from other studies did not find any temporary increases in glucoraphanin concentration due to flame weeding that would not be detected at harvest maturity (Sivesind, unpublished data).

The number of flame treatments had no effect on glucoraphanin concentration in broccoli florets in this study (Table 6.3). High dose treatments had lower glucoraphanin concentration than low dose flamed treatments or the non-flamed control. The response of glucoraphanin to confounding effects of flame treatment and weed pressure varied from year to year in this study. Glucosinolates may play an important role in the plant defence response, as glucosinolate derivatives have been demonstrated to suppress a number of disease causing pathogens (Charron et al., 2002; Rose et al., 1997). We are not aware of any other studies that have investigated the effect of flame treatments on glucosinolate concentrations in broccoli or any other plant species. However, flame damage has been demonstrated to initiate plant defence response; Vian et al. (1999) reported an increase in protein transcripts implicated in plant defence response after flame wounding in tomato. Glucosinolates concentrations are affected by a number of factors, both genetic and environmental (Brown et al., 2002; Farnham et al., 2004; Kushad, 1999; Shelp et al., 1993). The results of this study suggest that flame weeding has limited effects on glucoraphanin concentration in broccoli. However, as differences in glucoraphanin concentration were observed between the high and low flaming doses, further study of this effect is warranted.

6.5 Summary and conclusions

Results demonstrate that broccoli is quite tolerant of flame weeding and is a good candidate crop for use in selective flame weeding programs. These experiments tested the effect of flame weeding on numerous characteristics in broccoli and found few negative consequences. A flaming regimen utilizing a dose of 1.43 kg propane km^{-1} reduced yields compared to a regimen utilizing a dose of 0.90 kg propane km^{-1}, averaged across the number of flame treatments. In 2006, flame weeding did not suppress weeds sufficiently to avoid a yield loss compared to plots that were hand-weeded in addition to flame weeding. However, in 2007, flame-only plots out produced plots that were hand-weeded in addition to flaming, suggesting that weeds were suppressed sufficiently to eliminate yield loss. The reason for this increase in yield is unclear. Unmarketable broccoli yield and diameter were not affected by flaming dose, number of flame treatments or presence or absence of hand-weeding in addition to flame treatment. The number of flame treatments had little effect on broccoli yield. Monocot weeds decreased in both density and shoot mass with additional flame treatments in one of two years of this study. Dicot weed density, but not shoot mass, decreased with additional flame treatments. Effects of flame weeding on the time for broccoli heads to reach maturity and on the number of leaves produced were limited. High dose flame treatments (1.43 kg propane km^{-1}) caused a decrease in glucoraphanin concentration as compared to plots flamed with a lower dose (0.90 kg propane km^{-1}). These data suggest that broccoli is reasonably tolerant of flame weeding and would be able to withstand multiple flame weeding treatments of moderate intensity. In order to guarantee yields, flame weeding treatments may have to be supplemented with further weed control measures depending upon weed pressure.

6.6 Acknowledgements

The authors would like to thank Germain Moreau, field technicians, and summer students who assisted with field operations. We would also like to thank Ting-Li Lin of the College of Agricultural and Life Sciences Statistical Consulting Service at the University of Wisconsin-Madison for assistance with the statistical analyses. This research was supported by a grant from the Conseil des Recherches en Pêche et Agroalimentaire du Québec (CORPAQ).

6.7 Tables and Figures

Table 6.1. Significant main effects and interactions of year, flame dose, number of flame treatments, and weeding treatment on yield and head diameter of marketable, unmarketable, and total broccoli in 2006-07.

	Marketable		Unmarketable		Total	
Effect	Yield	Diameter	Yield	Diameter	Yield	Diameter
Year (Y)	***[a]	*	*	*	***	*
Flame Dose (D)	**	NS	NS	NS	**	NS
Y×D	NS	NS	NS	NS	NS	NS
Flame Number (N)	NS	NS	NS	NS	NS	NS
D×N	NS	NS	NS	NS	NS	NS
Y×N	NS	*	NS	NS	NS	NS
Y×D×N	NS	NS	NS	NS	NS	NS
Weeding (W)	NS	NS	NS	NS	NS	NS
W×D	NS	NS	NS	NS	NS	NS
W×N	NS	NS	NS	NS	NS	NS
W×D×N	NS	NS	NS	NS	NS	NS
Y×W	***	***	NS	NS	***	**
Y×W×D	NS	NS	NS	NS	NS	NS
Y×W×N	NS	NS	NS	NS	NS	NS
Y×W×D×N	NS	NS	NS	NS	NS	NS

[a] * = Significant at $P \leq 0.05$; ** = Significant at $P \leq 0.01$; *** = Significant at $P \leq 0.001$; NS = Nonsignificant

Table 6.2. Significant main effects and interactions of year, flame dose, and number of flame treatments on monocotyledon, dicotyledon, and total weed density (plants m^{-2}) and shoot mass (g DM m^{-2}) in broccoli in 2006-07. Data were log transformed prior to ANOVA to stabilize variance.

	Monocots		Dicots		Total[a]	
Effect	Density	Mass	Density	Mass	Density	Mass
Year (Y)	***[b]	***	NS	NS	***	***
Flame Dose (D)	NS	NS	NS	NS	**	NS
Y*D	NS	NS	NS	NS	NS	NS
Flame Number (N)	***	***	***	NS	***	***
Y*N	***	*	NS	NS	***	NS
D*N	NS	NS	NS	NS	NS	*
Y*D*N	NS	NS	NS	NS	NS	NS

[a]Total represents the sum of monocots and dicots.

[b] * = Significant at $P \leq 0.05$; ** = Significant at $P \leq 0.01$; *** = Significant at $P \leq 0.001$; NS = Nonsignificant

Table 6.3. Significant main effects and interactions of flame dose, number of flame treatments, weeding treatment (flame only or hand-weeding + flame), and year on glucoraphanin concentration in broccoli florets (µmoles g^{-1} DM) in 2006-07.

Effect	Glucoraphanin concentration
Year (Y)	***[a]
Flame Dose (D)	***
Y*D	NS[b]
Flame Number (N)	NS
D*N	NS
Y*N	NS
Y*D*N	NS
Weeding (W)	NS
W*D	NS
W*N	NS
W*D*N	NS
Y*W	***
Y*W*D	NS
Y*W*N	NS
Y*W*D*N	NS

[a] *** = Significant at $P \leq 0.001$

[b] NS = Nonsignificant result

Figure 6.1. Density of weed species (plants m^{-2}) in end of season collection averaged across all treatments in 2006 (a) and 2007 (b). Bars represent means ±SE. Weeds are designated by their official Bayer codes; for plants without Bayer codes, US codes are used. Weed species abbreviations: AGRRE, quackgrass (*Elymus repens* (L.) Gould); AMAPO, Powell amaranth (*Amaranthus powellii* S. Wats.); AMARE, redroot pigweed (*Amaranthus retroflexus* L.); AMBAR, common ragweed (*Ambrosia artemisiifolia* L.); BROIN, smooth brome (*Bromus inermis* Leyss.); CAPBP, shepherd's-purse (*Capsella bursa-pastoris* (L.) Medik.); CHEAL, common lambsquarters (*Chenopodium album* L.); DIGIS, smooth crabgrass (*Digitaria ischaemum* (Schreb.) Schreb. ex Muhl.); ECHCG, barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.); PANCA, witchgrass (*Panicum capillare* L.); POLPE, ladysthumb (*Polygonum persicaria* L.); SETFA, giant foxtail (*Setaria faberi* Herrm); SETLU, yellow foxtail (*Setaria pumila* (Poir.) Roemer & J.A. Schultes); SETVI, green foxtail (*Setaria viridis* (L.) Beauv.); SINAR, wild mustard (*Sinapis arvensis* L.); SONAR, perennial sowthistle (*Sonchus arvensis* L.); TAROF, dandelion (*Taraxacum officinale* G.H. Weber ex Wiggers).

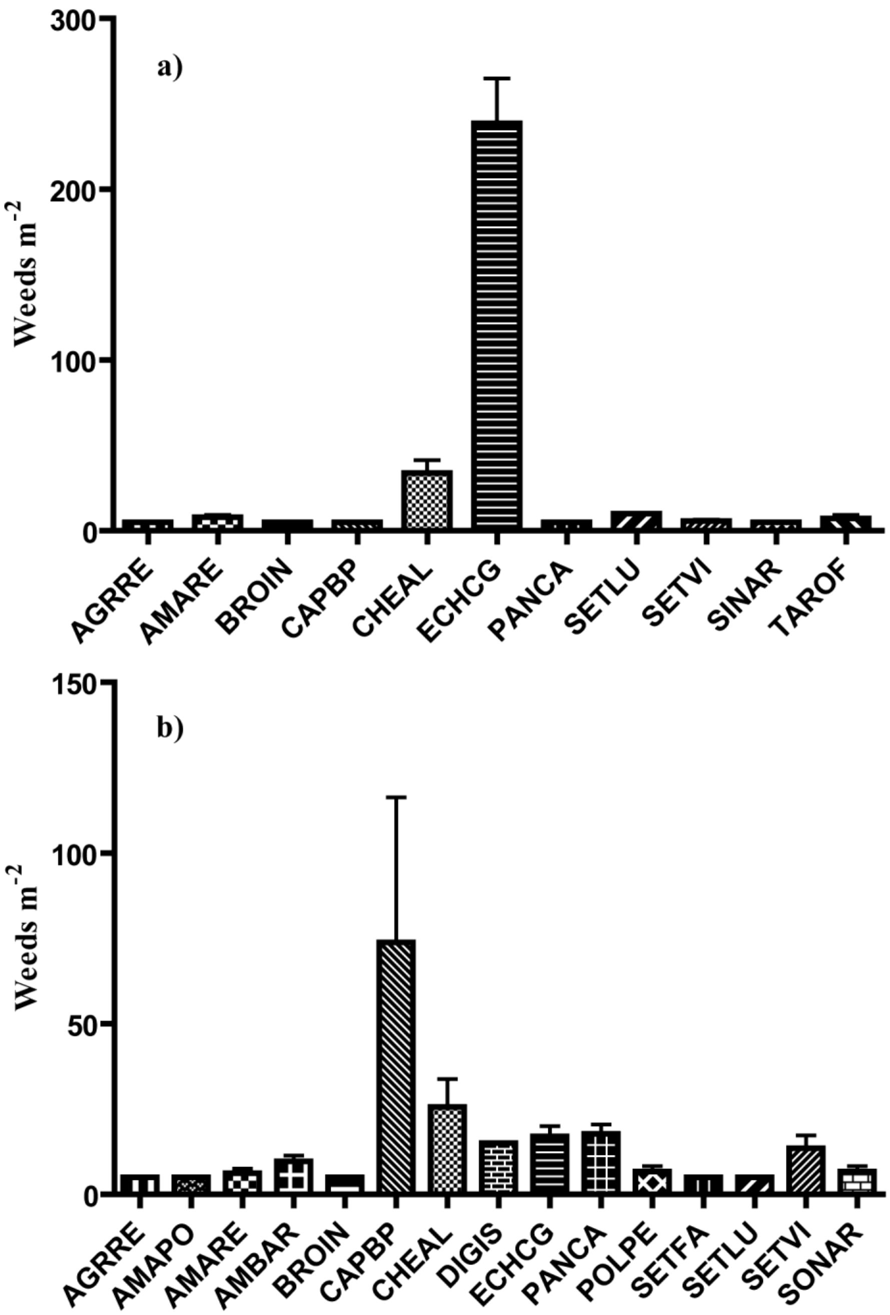

300
a)
200
Weeds m-2
100
0
AGRRE
AMARE
BROIN
CAPBP
CHEAL
ECHCG
PANCA
SETLU
SETVI
SINAR
TAROF
150
b)
100
Weeds m-2
50
0
AGRRE
AMAPO
AMARE
AMBAR
BROIN
CAPBP
CHEAL
DIGIS
ECHCG
PANCA
POLPE
SETFA
SETLU
SETVI
SONAR

Figure 6.2. Monocot weed density (plants m^{-2}) in broccoli in 2007 regressed over the number of flame treatments, averaged over two flaming doses. Points represent mean value of replicates ±SE. Regression made using a quadratic equation: $y = A + Bx + Cx^2$. Parameter estimates ±SE: *A*, 1.53 ± 0.43; *B*, 0.35 ± 0.05; *C*, -0.17 ± 0.05.

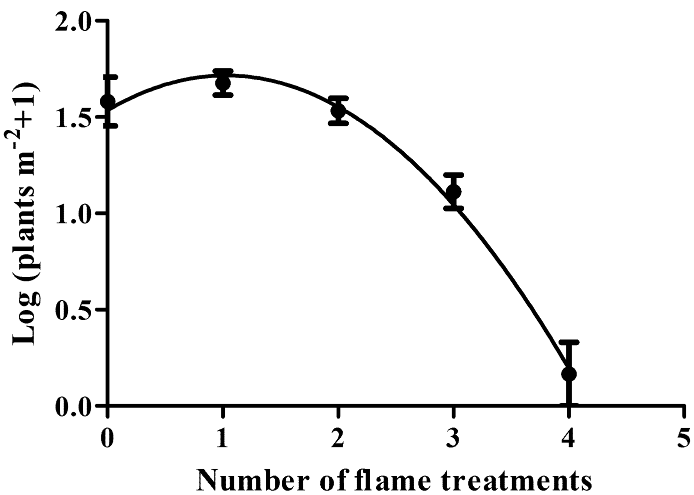

2.0
1.5
1.0
0.5
0.0
Log (plants m-2+1)
0
1
2
3
4
5
Number of flame treatments

Figure 6.3. Monocot shoot mass (g m^{-2}) in broccoli in 2007 as a function of number of flame treatments, averaged over two flaming doses. Points represent mean value of replicates ±SE. Regression made using a cubic equation: $y = A + Bx + Cx^2 + Dx^3$. Parameter estimates ±SE: *A*, 0.95 ± 0.11; *B*, -0.05 ± 0.22; *C*, -0.21 ± 0.13; *D*, 0.04 ± 0.02.

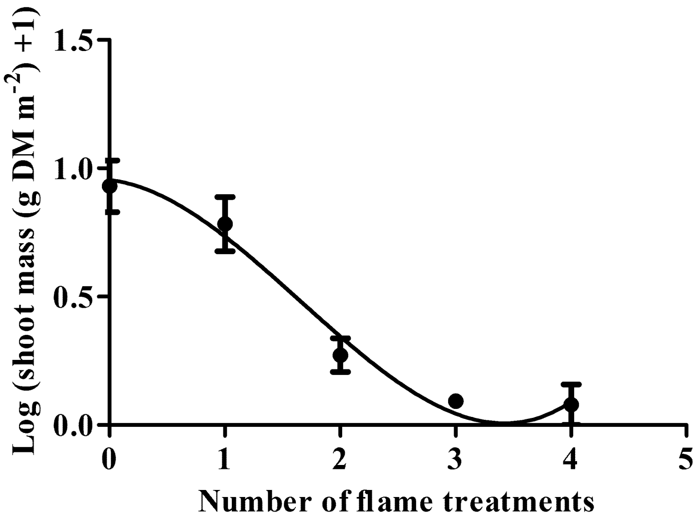

1.5
1.0
0.5
0.0
0
1
2
3
4
5
Log (shoot mass (g DM m-2) +1)
Number of flame treatments

Figure 6.4. Dicot weed density (plants m^{-2}) in broccoli regressed over the number of flame treatments, averaged over two flaming doses and two years. Points represent mean value of replicates ±SE. Regression made using equation 1: $y = (A - B)*\exp(k*x) + B$. Parameter estimates ±SE: *A*, 2.22.0 ± 0.17; *B*, 0.22 ± 0.31; *k*, -0.51 ± 0.20.

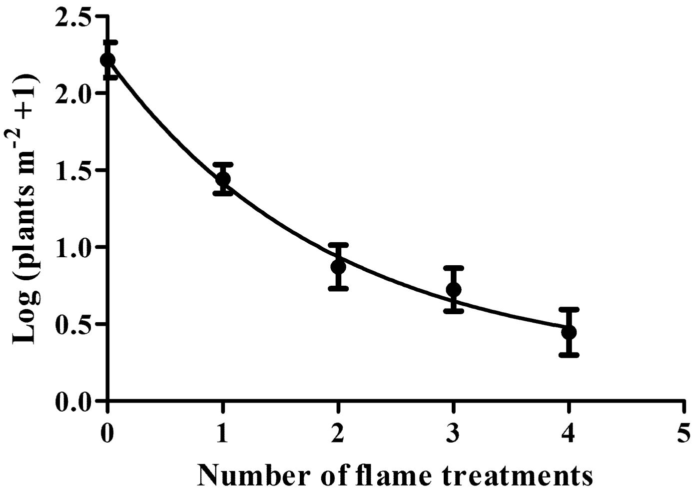
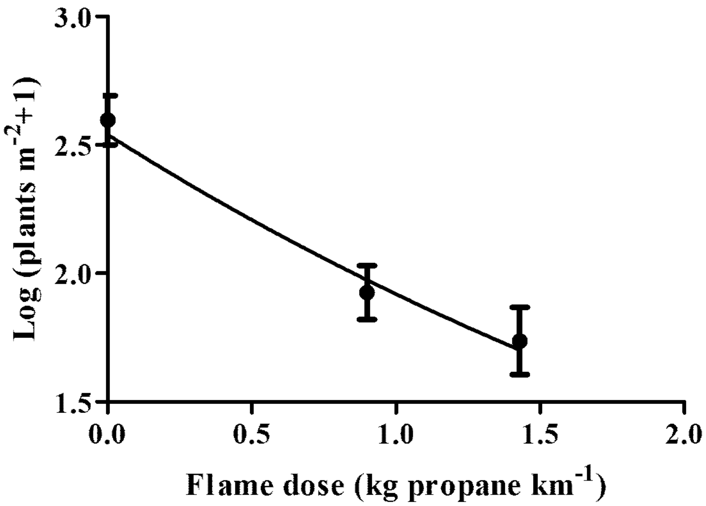
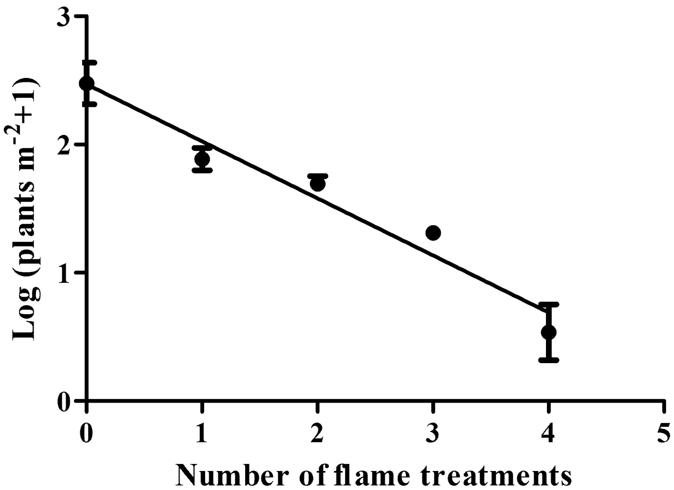

2.5
2.0
1.5
1.0
0.5
0.0
0
1
2
3
4
5
Log (plants m-2 +1)
Number of flame treatments

Figure 6.5. Total weed density (plants m^{-2}) in broccoli regressed over flame dose, averaged over the number of flame treatments and two years. Points represent mean value of replicates ±SE. Regression made using equation 2: $y = Y_o*\exp(k*x)$. Parameter estimates ±SE: Y_o, 2.54 ± 0.20; k, -0.28 ± 0.08.



3.0
2.5
2.0
1.5
Log (plants m-2+1)
0.0
0.5
1.0
1.5
2.0
Flame dose (kg propane km-1)

Figure 6.6. Total weed density (plants m^{-2}) in broccoli in 2007 regressed over the number of flame treatments and averaged over two flaming doses. Points represent mean value of replicates ±SE. Regression made using a linear equation: $y = A + Bx$. Parameter estimates ±SE: *A*, 2.47 ± 0.12; *B*, -0.45 ± 0.06.



Log (plants m-2+1)
3
2
1
0
0
1
2
3
4
5
Number of flame treatments

Figure 6.7. Total weed shoot mass (g DM m^{-2}) in broccoli plots flamed at 0.90 and 1.43 kg propane km^{-1} as a function of the number of flame treatments, averaged over two years. Points represent mean value of replicates ±SE. Regression for the high dose made using equation 2: $y = Y_o * \exp(k * x)$. Regression for the low dose made using a quadratic equation: $y = A + Bx + Cx^2$. Parameter estimates ± SE: High dose (1.43 kg propane km^{-1}), Y_o, 1.91 ± 0.22; k, -0.21 ± 0.06. Low dose (0.90 kg propane km^{-1}), A, 2.07 ± 0.21; B, -0.70 ± 0.25; C, 0.12 ± 0.06.

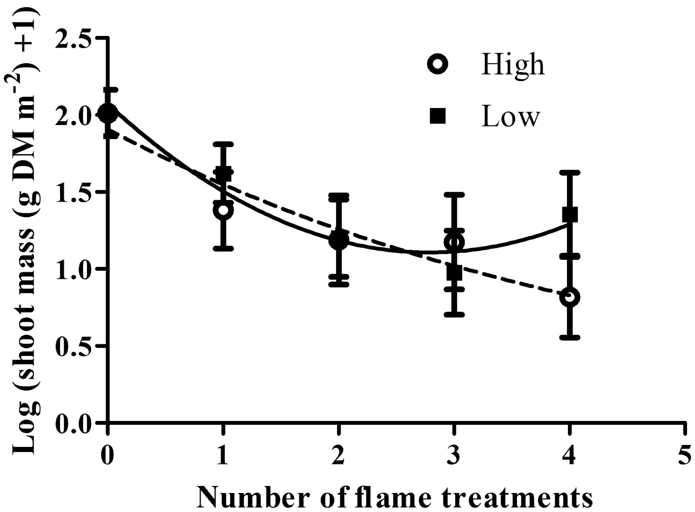
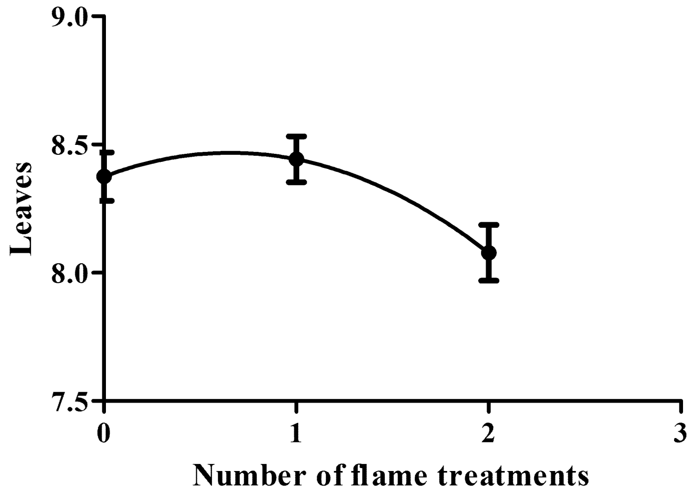

High
Low
Log (shoot mass (g DM m^{-2}) +1)
2.5
2.0
1.5
1.0
0.5
0.0
0
1
2
3
4
5
Number of flame treatments

Figure 6.8. Number of leaves per plant in broccoli in 2006 at count 1 (27 DAT) in plants flamed zero (non-flamed control), one, or two times, averaged over weeding treatment (flame-only or flame plus hand-weeding) and flame dose. Points represent mean value of replicates ±SE. Regression made using a quadratic equation: $y = A + Bx + Cx^2$. Parameter estimates ± SE: *A*, 8.38 ± 0.13; *B*, 0.28 ± 0.28; *C*, -0.22 ± 0.12.


9.0
8.5
8.0
7.5
Leaves
0
1
2
3
Number of flame treatments

7.0 General Discussion and Conclusion

This project was conducted to evaluate the use of selective flame weeding in vegetable crops. Response of a number of weeds to flame weeding was modeled using log-logistic models. Overall, dicot weeds were found to be more susceptible to flaming than monocot weeds, a similar conclusion reached by studies investigating non-selective flaming (Ascard, 1995; Cisneros and Zandstra, 2008; Knezevic and Ulloa, 2007a; Knezevic et al., 2008). For common lambsquarters, flaming doses required for 50% control ranged from 0.37 to 0.77 kg propane km^{-1} for weeds at the cotyledon and 6-leaf stage, respectively, while flame doses resulting in 95% control ranged from 0.83 to 2.85 kg propane km^{-1}. Response of redroot pigweed was similar to that of common lambsquarters, with LD_{50} values increasing from 0.32 to 0.97 kg propane km^{-1} for weeds at the cotyledon and 6-leaf stage, respectively. LD_{95} values for redroot pigweed ranged from 1.19 to 2.41 kg propane km^{-1} for the cotyledon and 6-leaf stage. Shepherd's-purse was effectively controlled at doses similar to that required for common lambsquarters and redroot pigweed (LD_{50} values of 0.58 and 0.85 at the cotyledon and 2-5 leaf stage; LD_{95} values of 1.15 and 2.78 at the cotyledon and 2-5 leaf stage). Flame doses of 1.50 kg propane km^{-1} should be sufficient to provide high levels of control for many dicot weeds at early growth stages (cotyledon to 2 leaves). As weeds develop, they become more difficult to control with flame weeding, and required flame doses increase; in this study flame doses of ~2.75 kg propane km^{-1} were required to provide 95% control of dicot weeds with 5-6 leaves, depending on the species. Neither barnyardgrass nor yellow foxtail were able to be effectively controlled; levels of control were inadequate with survival > 50% in all instances. Monocot weeds were often severely damaged with observable decrease in above ground shoot mass; however, regrowth allowed most weeds to survive.

When onion received a single flame dose, yield was affected by flame dose at the first flaming time point only (9-15 DAT), with a 10% yield reduction occurring at a flame dose of 1.78 kg propane km^{-1} (Table 4.2). No yield reductions were observed in onion beginning 20 DAT. Broccoli yields were only affected by a single flame treatment at the first time point (10-14 DAT), as yields were reduced 10% by a flame dose of 1.19 kg propane km^{-1}. Neither beets nor spinach were affected by preemergence flame weeding. However, yields were reduced at both the 4- and 6-leaf growth stages for both beets and spinach. In spinach, total yields were reduced 10% by flame doses of 0.75 and 0.68 kg propane km^{-1} at the 4- and 6-leaf growth stages. Total beet yields were reduced 10% at the 4-leaf stage by flame doses of 0.98 and 0.56 kg propane km^{-1} in 2005 and 2006, and at the 6-leaf stage by flame doses of 0.83 and 1.25 kg propane km^{-1} in 2005 and 2006, respectively. Based on the results of this study, postemergence flaming cannot be recommended for spinach or beets due to a high risk of yield loss. Preemergence flame weeding, however, has been used successfully by producers of many vegetable crops to reduce labor costs and caused no adverse effects in either spinach or beets in our study.

Onion was able to tolerate up to six flame treatments at a dose of 1.43 kg propane km^{-1} without any observed loss of yield. However, flame treatments alone were not able to control weeds sufficiently to avoid yield losses in onion. Therefore, additional weed control measures would be required in order to sufficiently control weeds. Flame weeding had minimal effects on time to reach maturity, leaf and bulb development, pungency or quercetin concentration in onion. Flame weeding approximately two weeks after transplantation in onion should not harm yields to a great degree as long as flame doses are kept to a minimum; flaming with a dose of 1.25 to 1.50 kg propane km^{-1} should be able to control dicot weeds at the cotyledon to 2-leaf growth stage, depending upon the species while minimizing yield losses. Beginning 20 DAT, flame doses up to 2.95 kg propane km^{-1} were safely used in onion without reducing yields; higher flame doses could therefore be used if necessary to control more advanced weeds. Up to six flame treatments were used in this study without reducing yield; however, we only tested multiple flame treatments with flame doses up to 1.43 kg propane km^{-1}. It is possible that multiple flame treatments with higher flame doses would have adverse effects on onion yields. Flame treatments should be able to reduce weed density and shoot mass, but additional measures may be necessary to ensure desired yields. This is would be especially important for areas with high weed pressure or a high degree of monocot weeds.

Broccoli was able to tolerate up to four flame treatments at a dose of 1.43 kg propane km^{-1} without any yield reduction. Flame-only treatments had lower yields than the flamed, weed-free treatments in one of two years; in the other year, the flame-only treatments had greater yields than the weed-free, flamed treatments. Density of monocot weeds declined as the number of flame treatments increased in one of two years, while dicot weed density declined as the number of flame treatments increased in both years. The effects of flame treatments on the number of days to maturity, leaf development, and glucoraphanin concentration were limited.

Broccoli was quite tolerant of flame weeding in this study. Flame weeding can safely be used in broccoli, and could serve as a method to reduce costly hand-weeding in organic production or to otherwise reduce the use of herbicides. If broccoli is flamed prior to three weeks after transplantation, care should be used to minimize the flame dose in order to reduce the risk of yield loss. In these first few weeks, flame doses should not exceed approximately 1.00 kg propane km^{-1}. If possible, it may be desirable to postpone flame weeding treatments in broccoli as long as possible by using a stale seedbed, early hand-weeding or other measures. Beginning 20 DAT, flame doses up to 2.95 kg propane km^{-1} were safely used in broccoli without reducing yields. Up to four flame treatments were used in this study without reducing yield; however, we only tested multiple flame treatments with flame doses up to 1.43 kg propane km^{-1}. The effects of multiple flame treatments utilizing doses above 1.43 kg propane km^{-1} are unknown.

Flame weeding is a valuable weed control option available to producers. As weed tolerance to flame weeding increases as plants develop, it is important to treat while plants are still young and more easily controlled. Understanding of its strengths and limitations is vital for effective results. Selective flaming has the potential to reduce the amount of costly hand-weeding employed in organic production for many flame-tolerant crops.

8.0 Hypotheses Conclusions

This project evaluated the use of flame weeding in vegetable crops. The overall research hypothesis was as follows:

Flame weeding has the potential to be a valuable weed control tool for producers of horticultural crops. However, both crop and weed species can be expected to vary in tolerance to flame weeding treatments. Furthermore, flame weeding causes significant stress on plants, which may result in morphological, developmental, and physiological changes.

This overall hypothesis was sub-divided into three sub-hypotheses which were tested in the experiments conducted for this project. The sub-hypotheses and their conclusions are as follows:

1. Weed species differ in their susceptibility to flame weeding. Within species, response to flaming will be dependent upon flaming intensity and plant development. Dose response curves can be constructed to describe the response of plants to flame treatment.

Conclusion: We accept this hypothesis. Overall, dicot species were able to be effectively controlled with flame weeding, while control of monocot species was generally poor. Further, doses required for equivalent responses varied between species and within species according to the growth stage at treatment.

2. Crop species will vary in their ability to tolerate flame weeding. Some species will be able to tolerate flame weeding in the intensity range required for weed control, while others receive unacceptable levels of damage.

Conclusion: We accept this hypothesis. Crop species differed in their ability to withstand flame weeding treatment. Broccoli and onion proved sufficiently tolerant to warrant further study, while postemergence flame weeding in spinach and beets posed too great a risk for yield loss to be able to recommend use.

3. Beyond yield considerations, flame damage is an extreme and acute stress and will cause other detectable developmental and physiological changes within crop plants.

Conclusion: We found little evidence to support this hypothesis for the parameters measured. We observed few effects from flame weeding on developmental and physiological parameters evaluated, though the exact effect varied according to each specific parameter. However, we only studied a small subset of possible parameters; no conclusion can be drawn for any parameters not specifically studied in this research.

9.0 Contribution to the Knowledge

This project contributes much information to the body of knowledge that was previously unavailable. To our knowledge, this is the first report of dose-response curves generated for weed species using the type of cross flaming system used for postemergence selective flame weeding in row crops. Second, this project substantially increases the information available concerning the use of postemergence flame weeding in onion, broccoli, spinach, and beets. This is the first report of the use of postemergence flame weeding in spinach and beets. To our knowledge, the information provided here concerning the use of postemergence flame weeding in broccoli is the first available since the 1960's. Selective flame weeding has been previously studied in onion, but this is the most complete study of the use of flame weeding at a wide range of flame doses and time points during the season.

The use of postemergence, selective flame weeding has been evaluated in crops by other researchers, but almost always in a dose-response manner testing only a single flame treatment at a particular growth stage. This is the first report of the use of multiple flame weeding treatments in broccoli. In addition, no other reports have previously evaluated the effects of flame weeding treatments on broccoli leaf development, time to maturity, or glucoraphanin concentration. Although some research has been published concerning the use of postemergence flame weeding in onion, this study evaluated a greater number of flame treatments and response parameters. No information is currently available on the effect of flame weeding on onion leaf development, time to maturity, or flavonoid concentration. In addition, these reports are the first quantifying the effects of a season-long flame weeding regimen on a weed population. This project significantly increases the breadth of knowledge concerning the use of postemergence, selective flame weeding in vegetable crops.

10.0 Recommendations for Future Research

The results of this project suggest several areas that warrant further research in flame weeding. Future research could include:

1. Model response of weeds not available in literature, such as giant ragweed (*Ambrosia trifida* L.) and many perennials.
2. Continue evaluation of crops for flame weeding tolerance.
3. Study flame weeding in concert with mechanical or other methods that would complement well.
4. Investigate ways to improve the control of monocot species by flame weeding.
5. Investigate any relationship between flame weeding and disease incidence, notably of secondary invaders.

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