Population Dynamics of Dandelion (*Taraxacum officinale*) in Turfgrass as Influenced by a Biological Control Agent, *Sclerotinia minor*

By

Mohammed H. Abu-Dieyeh

Department of Plant Science

McGill University

Montreal, Quebec, Canada

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To my wife and children

ABSTRACT

Control of *Taraxacum officinale* (dandelion) and other broadleaf weeds in turfgrass has been readily achieved with phenoxy herbicides, but the herbicide option has been revoked in many regions, necessitating alternative weed control strategies. One biological alternative is *Sclerotinia minor*, an Ascomycete fungus. The impact of *S. minor* on broadleaf weed dynamics and biotic interactions were studied in a turfgrass environment. The goal was to maximize effectiveness of a *S. minor* formulation as a biocontrol of dandelion using an ecological approach. *S. minor* efficacy was not affected by turf microenvironments and was similarly efficacious with spring or fall application. All accessions from a worldwide collection of dandelion and 32 turfgrass broadleaf species were susceptible to *S. minor*. Biocontrol efficacy was inversely correlated with dandelion age, but efficacy on all ages was enhanced in the presence of grass competition. When combined with regular mowing at 7-10 cm, the *S. minor* suppressive effect on dandelion was similar to the herbicide effect, particularly in the following season. Weed suppression was less with close mowing at 3-5 cm due to increased dandelion seedling recruitment. While spring herbicide application was effective to suppress dandelion population, the *S. minor* treatment has no residual activity, necessitating a second application to suppress seedling recruits. Root regrowth after *S. minor* infection was minimal and was further reduced in superior quality turf after season-long mowing, and after spring applications. *S. minor* infected dandelion seeds, reduced the dandelion seedbank, and reduced dandelion seedling emergence by 98%. *S. minor* did not affect the emergence or the total biomass of cool season temperate turfgrass species. Turfgrass quality was improved following *S. minor* application and populations of other broadleaf weeds were also controlled by *S. minor*. Understanding the biotic interactions within the turfgrass environment has rewardingly lead to successful integration of the *S. minor* biocontrol with the common management tools of mowing and over-seeding to achieve excellent control of dandelion and a healthy thriving turf.

RÉSUMÉ

Taraxacum officinale (pissenlit) et les autres mauvaises herbes à feuilles larges sont facilement contrôlés par les herbicides. Par contre, les herbicides étant interdits dans plusieurs régions, l'emploi de méthodes alternatives est nécessaire. *Sclerotinia minor*, un champignon de type Ascomycète, est une alternative biologique. L'impact de *S. minor* sur la dynamique des mauvaises herbes à feuilles larges et sur les interactions biotiques a été étudié dans un environnement de pelouse en plaque. Le but était de maximiser l'efficacité d'une formule contenant *S. minor* afin de l'utiliser contre les pissenlits. L'efficacité de *S. minor* n'a pas été affectée par les microenvironnements de la pelouse ni par la période d'application. Toutes les variétés de pissenlit sélectionnées ainsi que 32 espèces de mauvaises herbes étaient susceptibles à *S. minor*. L'efficacité du biocontrôle était inversement proportionnelle à l'âge des pissenlits et augmentait due à la compétition de l'herbe. Combiné avec une tonte longue et régulière, l'effet suppressif de *S. minor* sur les pissenlits était similaire à l'effet de l'herbicide. La suppression des mauvaises herbes était moindre lorsque la tonte était courte due à l'augmentation du taux de recrutement des semis de pissenlit. Deux applications de *S. minor* ont eu des effets similaires à une application d'herbicide. Le traitement de *S. minor* n'ayant pas d'activité résiduelle, deux applications sont nécessaires. La repousse des racines suite à l'infection par *S. minor* a été minimale, encore plus dans la pelouse de qualité. *S. minor* a infecté les graines, a réduit la banque de graines, et a réduit l'émergence des semis de pissenlit de 98%. *S. minor* n'a pas affecté l'émergence ou la biomasse des espèces de pelouse acclimatées aux saisons tempérées. La qualité de la pelouse en plaque était améliorée suite à l'application

de *S. minor* et les autres mauvaises herbes étaient aussi contrôlées par *S. minor*. La compréhension des interactions biotiques à l'intérieur de l'environnement de la pelouse en plaque a permit l'intégration réussie de *S. minor* comme agent de biocontrôle dans les pratiques courantes (la tonte et le sursemis) utilisées pour atteindre un excellent contrôle des pissenlits et une pelouse résistante et en santé.

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CONTRIBUTIONS OF AUTHORS

In this thesis there are six manuscripts (Chapters: 3, 5, 6, 7, 8, 9) either already published, accepted for publication, under revision or to be submitted to refereed journals.

In all manuscripts, I designed the experimental set-up, performed the experiments and the statistical analysis and wrote the manuscript myself. Professor Watson co-authored all manuscripts; he supervised the work, provided financial and technical resources and corrected all manuscripts.

Chapter 9 represents a manuscript that already published in *Biocontrol Science and Technology*, December 2005; 15(8):815-825. In this manuscript, Jerome Bernier is a co-author; he helped me in seed collection, in laboratory work related to seed germination experiment and isolating seed microflora. More details about contributions of authors are mentioned in the preface of each chapter.

A specific acknowledgment is mentioned at the end of each chapter describing the nature of work provided by certain people who helped me in one way or another related to the designated chapter.

CHAPTER 1

INTRODUCTION

Weeds are familiar plants in our environment, infesting a wide range of variable habitats and posing major threats to agriculture, biodiversity, ecosystem integrity and public health. They are commonly found in areas where the native vegetation has been replaced with a controlled system of cropping and management, or with other activities related to civilization (Anderson 1996). After the Second World War, the ecology of turfgrass systems has been changed due to massive turf monoculture industrialization (Robbins & Birkenholtz 2003). The monoculture turfgrass system replaces the ecology of a natural environment, and provides a favourable environment for better adapted organisms including weeds. Nearly every turfgrass environment has weed problems that require some degree of management to be usable and aesthetically pleasing (Monaco et al. 2002). McCarty et al. (2001) described 73 grass and grass-like and 145 broadleaf species as weeds in turfgrass environments. These weeds do not only compete with turfgrass for light, soil nutrients, soil moisture and physical space, but also are hosts for pests (McCarty et al. 2001).

A significant turfgrass industry has evolved to manage these new environments, and has become an important economic component in North America and Europe accompanied by increased inputs in petrochemicals, fertilizers and pesticides (Robbins & Birkenholtz 2003; Cisar 2004). In 1990, it was estimated that there were approximately 30 million acres of turfgrass in USA (Emmons 1995). This area is predicted to increase year after year. Monaco et al. (2002) mentioned an estimate of 46 million acres of turf in the United States. The annual expenditure on turf was estimated during 1982-1993 in USA as $45 billion (Beard & Greeen 1994). Turfgrass not only provides recreational and aesthetic benefits for human but also several functional benefits for the environment e.g. reduction of soil erosion (Beard & Green 1994).

Dandelion (T*araxacum officinale* G.H. Weber ex Wiggers) is a common world wide perennial weed occurring in a wide range of habitats including grassland areas. The high seed potential and dispersal abilities and the regenerative capacity of the strong tap root are major competitive features in dandelion leading to its prevalence in turfgrass environments and to the dilemma of its control (Stewart-Wade et al. 2002b). Current methods for dandelion control in turfgrass are proper management practices and chemical control with chlorophenoxy herbicides including 2,4-dichlorophenoxy acetic acid (2,4-D) (Riddle et al. 1991). However, chemical herbicides have received considerable negative publicity worldwide and nowadays various levels of governments are enacting legislation restricting or banning the use of pesticides in urban areas (Schnick et al. 2002; Cisar 2004). In response to this situation, alternative approaches including biological control are being researched.

Sclerotinia minor Jagger is an ascomycete plant pathogen that has biocontrol potential for dandelion in turfgrass. Several formulations of *S. minor* have been shown to have a mycoherbicidal activity on dandelion in turfgrass systems (Ciotola et al. 1991; Riddle et al. 1991; Briere et al. 1992; Schnick et al. 2002; Stewart-Wade et al. 2002a). *S. minor* mycelium in sodium alginate granules (Briere et al. 1992) and mycelial-colonized barley grits (Ciotola et al. 1991) were found to be the most effective formulations on dandelion and plantain without causing damage to turfgrass species (Stewart-Wade et al. 2002a).

In the absence of ecological information, reliance on a single weed control strategy may result in weed management failure, increasing the weed problems or having negative environmental and economic consequences (Booth et al. 2003). The biocontrol process involves several biotic and abiotic interactions. Biocontrol is successful when the biotic components and the environment interact in such a manner that weed control or suppression occurs (Kennedy & Kremer 1996). Given the high costs of such programs, the success rate should be maximized and this task cannot be achieved without understanding the ecology of the components of the plant: pathogen system (Cousens & Croft 2000). As a consequence of weed control, species composition, abundance and/or distribution are usually changed. Therefore, investigating different biotic interactions and studying population dynamics of the involved weed species are necessary to evaluate the success of a biocontrol agent and to determine the impact of the biocontrol on the populations and on the environment (Burpee 1990; Deacon 1991; Kennedy & Kremer 1996; Radosevich et al. 1997; Headrick & Goeden 2001).

The ecology and population dynamics of dandelion and other broad-leaf weed species in turfgrass systems under the influence of *S. minor* have never been investigated. The major goals of this study were to understand the ecological interactions of the pathosystem and to evaluate the extent of pathogenesis between *S. minor* and dandelion and seek out an ecological approach that maximizes the biocontrol process.

1.1. Research Hypotheses

A research program was established to acquire knowledge concerning biotic and abiotic interactions that increase disease and damage to dandelion caused by ***Sclerotinia minor*.** The following hypotheses were tested:

1-Dandelions of different ages and growth forms (biotypes or accessions) will respond differently to *Sclerotinia minor*.

2-Different mowing heights of turfgrass have different effects on the biocontrol process of dandelion caused by *Sclerotinia minor*.

3-Turfgrass density and vigour are synergic with *Sclerotinia minor* to suppress dandelion through competitive interaction.

4-Different application times of *Sclerotinia minor* have different effects on the control efficacy and or the susceptibility of dandelion.

5-The application of *Sclerotinia minor* in turfgrass systems changes the plant populations, influencing species composition, abundance, and distribution.

6-No adverse effects are expected on turfgrass growth and establishment due to *Sclerotinia minor* successive applications

1.2. Research Objectives

Based on the above hypotheses, the following research objectives were defined:

1- To evaluate the effect of plant age, and dandelion biotypes on the performance of *S. minor* to cause mortality and population suppression of dandelion.

2- To evaluate the effect of grass competition on dandelion control by *S. minor*.

3- To determine the effect of grass mowing height on dandelion suppression with *S. minor*.

4- To evaluate the effect of seasonal application of *S. minor* on dandelion control.

5- To investigate the effect of *S. minor* on seed germination and seedling establishment of dandelion and turfgrass

6- To study the effect of *S. minor* on the population dynamics of dandelion and other broadleaf weed species in turfgrass systems.

CHAPTER 2

GENERAL LITERATURE REVIEW

2.1. Weeds and their implications

Weeds are characteristic of human activities and civilization, particularly in controlled cropping systems and managed lands. Usually, weeds thrive in disturbed sites, having strong competitiveness for the available resources and strong reproductive abilities (Monaco et al. 2002). In disturbed areas, weeds comprise the first stage of secondary plant succession and often have the capability to replace the original plant species (Anderson 1996). Since 1879, several definitions and descriptions were given for weeds. Baker (1974) listed twelve biological characteristics that describe weeds and suggested that a plant species might possess various combinations of these characteristics resulting in a range of weediness from minor to major or highly successful weeds. Zimmerman (1976) characterized weeds by four features: ability to colonize disturbed habitats, invasive, locally abundant, and of little economic value. Aldrich (1984) described weeds as plants that originated under a natural environment and in response to [human] imposed or natural conditions are now interfering associates of crops and human activities. However, The Weed Science Society of America, 1994 described a weed simply as any plant that is objectionable or interferes with the activities or welfare of man (Radosevich et al. 1997). All of these definitions imply that weeds have some common biological traits but also a level of relative undesirability as determined by humans. Radosevich et al. (1997) indicated that relative abundance of plants, their location, and the potential use of the land they occupy should be considered in weed definitions.

In Canada, the estimated average annual loss caused by weeds in 58 crops was $984 million (Swanton et al. 1993). The loss was distributed as $372 million in eastern Canada and $612 million in western Canada. In The United States, the estimated average annual loss caused by weeds in 46 crops grown in 1991 was $4.1 billion. This annual loss would have been $19.6 billion if herbicides had not been used (Bridges 1992). The successful competition of weeds results in crop quantity loss by being more aggressive in growth habit, obtaining and utilising the essentials of growth, development and reproduction at the expense of crop plants and in some cases, by secretion of chemicals that adversely affect the growth and development of crop plants (Monaco et al. 2002).

Crop quality is also reduced when green, moist vegetation and the reproductive parts of weeds are harvested along with the crop (Anderson 1996). Crop loss is not the only adverse effect of weeds. They can serve as hosts for plant pests and diseases and also weeds are the major cause of hay fever and dermatitis which infect millions of people in the United States, Canada and other parts of the world (Anderson 1996). The cost of impact due to weeds could be as great to ecosystem as land-use but is harder to estimate (Booth et al. 2003).

Weed competition is one of the critical factors limiting crop yield in agricultural systems. Synthetic organic pesticides are the primary method of control for weeds, insects and pathogens. In Canada, sales of these products exceeded $1.4 billion in 1998, primarily for herbicides applied to cereal and oil seed crops (Floate et al. 2002). Continuous development of new pesticides has been driven by firstly, a desire to replace existing products with more target specific, lower mammalian toxicity, and less environmental persistence products, and secondly, the need to find alternatives to products that become less effective due to the development of a certain degree of pesticide resistance (Floate et al. 2002). The introduce of herbicide resistant crops, using genetic engineering, reflects the continuous desire to secure weed control markets (Müller-Scharer et al. 2000).

Resistance to one or more herbicide classes has been reported for populations of seven broadleaf weed species on the Canadian prairies in the past decade (Beckie et al. 1999). Four populations of wild oat, *Avena fatua* L., in intensive, continuous cropping systems of wheat, barley and canola in Alberta were found to be completely resistant to all herbicides registered for use in wheat (Beckie et al. 1999). Indeed, the large-scale and repeated application of broad-spectrum herbicides raises other concerns like the transfer of resistance genes to wild and weedy relatives, the spread of resistant volunteer crops and weed shift toward more tolerant species.

Applications of herbicides also introduce chemical residues into the environment, with undetermined consequences. Herbicides such as 2,4-D (2,4-dicholorphenoxy acetic acid), bromoxynil (3,5-dibromo-4-hydrooxybenzonitrile) and dicamba (3,6-dichloro-methoxybenzoic acid) were frequently present in rainfall at concentrations that may have adverse effects on sensitive plant species and on the quality of surface water in Alberta (Hill et al. 1999). According to Floate et al. (2002), the Pest Management Regulatory Agency in Canada is reviewing all pesticides registered prior to 31 December, 1994 (74% of the 550 currently registered active ingredients) to stay current with the reassessment with a new standard: “**reasonable certainty that no harm will result**

from aggregate exposure to each pesticide from dietary and other sources". In addition to the negative effects of excess herbicide use on our ground water and environment, public awareness and concern have also increased resulting in the reduction in use or banning of chemical pesticides, especially in urban environments. Therefore searching for alternative means of weed management, such as biological control, have been encouraged.

2.2. Biological control

Biocontrol uses natural enemies to control insect, pathogen and weed pests. Recently, the definition of biocontrol has been broaden to the use of natural or modified organisms, gene or gene products to reduce the effect of undesirables organisms (pests) and to favour desirable organisms such as crops, trees, animals and beneficial insects and microorganisms (Cook 1987). Biocontrol involves one or more natural processes (e.g. antibiosis, parasitism, competition, predation and induced host resistance) that are influenced by abiotic and biotic factors from the surrounding environment. These factors often limit the interactions between plant pathogens and their antagonists resulting in less than acceptable suppression of disease or reduction in pathogen populations (Cook & Baker 1983).

The origin of research on biocontrol of soil-borne plant pathogens was traced to the 1920s when, in experimental conditions, saprophytic microorganisms were co-inoculated with pathogens into previously sterilized soils and shown to exert control (Garret 1965: cited in Deacon 1991). The first recorded example of biological weed control was the control of *Opuntia vulgaris* by the intentional introduction of a cochineal insect, *Dactylopius ceylonicus* to northern India from Brazil in 1795 (Goeden 1988: cited in Watson 1993). This insect was also introduced to Sri Lanka (prior to 1865) and resulted in successful biological control of *O. vulgaris* throughout that country (Watson 1993).

In Canada, notable biological control successes over the past 20 years were reviewed by Mason & Huber (2002). Some of these projects have resulted in successful pest control including the control of *Sphaerotheca* and *Erysiphe* powdery mildews using Sporodex® (Belanger et al. 2002), the successful establishment of exotic agents, e.g. European apple sawfly (Vincent et al. 2002), and development and registration of *Chondrostereum purpureum* under the trade names Chontrol® or Myco-Tech® for control of stump sprouting and regrowth of alder, birch and poplar in utility rights-of-way and forest vegetation management (cited in Mason & Huber (2002) as: Dr W.E. Hintz, Mycologic Inc. personal communication).

2.2.1. Biological control of weeds

Weeds are the most significant pests in economic and environmental terms as measured by effort spent on their control and the herbicide share of global pesticide sales (Bridges 1992; Powell & Jutsum 1993). In Canada, the herbicides comprised 85% of total pesticide sales in 1998 (Floate et al. 2002). The literature on biological control of weeds is relatively compact, and the United States, Australia, South Africa, Canada and New Zealand use biocontrol the most (McFadyen 1998). Several reviews have documented the progress and limitations of biological weed control (Watson 1993; Kennedy and Kremer 1996; McFadyen 1998; Müller-Scharer et al. 2000; Charudattan & Dinoor 2000; Mason & Huber 2002; Hallett 2005).

Three main strategies for biological control are available for weed suppression: classical, inundative and integrated management approaches. The classical approach involves the importation of exotic natural enemies for release, dissemination and self-perpetuation on an introduced target weed. Inundative or bioherbicide approach is the augmentation or addition of a virulent pathogen to suppress weeds. In this approach the biocontrol agent is not self-sustaining and must be applied to the target host in repeated manner. Integrated approach is a broad approach which involves technologies, ecological interactions and management practices to conserve or enhance native enemies of weeds.

Classical biological control is the only weed control method able to provide a long term, often permanent solution to a serious weed problem with limited or no further inputs after the agent is introduced (Watson 1993). Insects and rust fungi have been successful classical approaches to weed control (Kennedy & Kremer 1996; Charudattan & Dinoor 2000). This approach is the predominant method in weed biocontrol and there is little actual use of augmentation as commercial or practical methods in the field (McFadyen 1998). One of the most successful examples is the introduction of a rust fungus *Puccinia chondrillina* along with three insects to control rush skeleton weed (*Chondrilla juncea*) in Australia. This fungus disseminated rapidly and widely and was able to control the most predominant biotype of rush skeleton weed (Charudattan & Dinoor 2000).

Augmentation using pathogens, almost entirely with fungi, has had limited commercial or practical success in the field. Currently, five fungi and one bacterium are registered and formulated as bioherbicides (Charudattan & Dinoor 2000). DeVine® (*Phytophthora palmivora*) is used to control *Morrenia odorata* (milkweed vine) in citrus fields in Florida. Collego® (*Colletotrichum gloeosporioides* f.sp. *aeschynomene*) is used to control *Aeschynomene virginica* (northern jointvetch) in Arkansas, Mississippi and Louisiana. BioMal® (*Colletotrichum gloeosporioides* f.sp. *malvae*) was registered in Canada for control of *Malva pusilla* (round-leaved mallow), but has not been commercialized due to production problems. Dr. BioSedge® was formulated based on the rust fungus *Puccinia canliculata* and registered in the United States to control *Cyperus esculentus* (yellow nutsedge). Stumpout® is a stump-treatment product based on the wood-infecting basidiomycetes, *Cylindrobasidium laeve* is registered in South Africa to control resprouting of cut trees in natural and trees plantation areas. CAMPERICO® an isolate of a wilt-inducing bacterium, *Xanthomonas campestris* pv. *poae*, is registered in Japan for the control of annual bluegrass in golf courses (Charudattan & Dinoor 2000). Although research is continuous on many potential bioherbicides, problems with mass-production, formulation and commercialization continue to prevent their implementation (Cook 1996; McFadyen 1998; Charudattan & Dinoor 2000).

The requirements for an effective augmentation program must include a comprehensive understanding of the natural enemies involved, the biology and population dynamics of the target weed(s), the optimum requirements for delivery of the natural enemy, the optimum conditions for subsequent infestation of the target weed population, and the complex interactions within the host-parasite or host-pathogen system (Watson 1993). The integrative approach is not yet well-defined, but usually includes practices and applications to enhance or conserve the biocontrol agent (Kennedy and Kremer 1996). Biological control organisms are most often host specific and usually control only one weed species, and this is one of their commercial limitations. Different strategies may be used to overcome this limitation by enhancing the effectiveness and acceptability of the bioherbicide. The combination with other methods including hand weeding, mechanical habitat management or low rates of chemical herbicides are required to obtain a wide spectrum control of common weed species associated with most crop production systems (Watson 1993). The use of low rates of the chemical herbicide 2,4-D with *Sclerotinia minor* as a mycoherbicide was found to be synergic and effective to control dandelion (Schnick et al. 2002). In Europe, successful control of *Chenopodium album* in maize and sugar beet crops was achieved by combining the pathogen *Ascochyta caulina* with a phytotoxin produced by the same fungus (Müller-Schärer et al. 2000). The one-weed-one-bioherbicide can be replaced by a multiple-pathogen strategy using a mixture of host specific pathogens, each one controlling a specific group of weeds (Chandramohan et al. 2002; Chandramohan & Charudattan 2003; Hallett 2005).

Biological weed control takes advantage of biotic factors that influence the distribution, abundance, and competitive abilities of plant species. It is successful when the weed, the biocontrol agent and the environment interact in such a manner that weed control or suppression occurs (Kennedy & Kremer 1996). Many authors (Burpee 1990; Daecon 1991; Watson 1993; Kennedy & Kremer 1996; Cousens & Croft 2000; Headrick & Goeden 2001) focused on the understanding of the environment around the biocontrol agent and the target host to promote successful biocontrol processes.

Recently problems associated with classical biological control have been raised as the biocontrol agent may move from its target plant to attack closely related native plant species. The musk thistle weevil has been introduced across the United States to control musk thistle (*Carduus nutans*), a serious weed of pastures, rangeland and other non cultivated areas. The weevil has been successful in suppressing musk thistle infestation but also exploited five native thistle species in the U.S. national parks and nature conservancy preserves (Louda et al. 1997). The authors concluded that greater caution with regards to environmental costs must be considered when evaluating biocontrol agents. The breadth of diet, potential host range and ecological effects need to be investigated and then carefully weighed against the environmental costs of the pest and of alternative management options. Non-target effects of fungal pathogens used as biocontrol agents were also reported, e.g. *Rhizobium* spp. caused reduction in mycorrhizal root colonization of plants and disordered commercial mushrooms (Brimner & Boland 2003). However McFadyen (1998) indicated that biocontrol offers the only safe economic and environmentally sustainable solution. Moreover Headrick & Goeden (2001) proposed that biological control could be a useful tool for restoration and maintenance of ecosystems that are in ecological decline due to destruction by pests.

While the classical biocontrol method is the most safe, practical, economically feasible, and sustainable in the long term (Watson 1993; McFadyen 1998), the current popularity of inundative biological control may result in problems, as application activities will be executed by untrained persons (van Lenteren et al. 2003). Therefore a comprehensive methodology for environmental risk assessment of exotic natural enemies used in inundative biological control has been proposed (van Lenteren et al. 2003).

2.2.2. Ecological interactions and weed biological control

Ecology and weed science have developed separately but looking for applied answers from ecologically based questions about weed invasion and control is the major interest of weed ecologists (Booth et al. 2003). Ecological principles and concepts are necessary to understand the nature of weediness. Once this understanding is established, it is possible to investigate the relationships and interactions that exist among environment, weeds, and crops in agro-ecosystems. However, without considering the ecological foundation, weed management may fail, becomes worse and/or instigates economical and environmental impacts (Booth et al. 2003).

All populations have intrinsic potential for exponential growth when environmental regulation is lacking (Begon & Mortimer 1986). A weed infestation is a plant population lacking negative feedback control to compensate for the positive response of reproduction and growth (Radosevich et al. 1997). Negative regulation could be related to food webs, nutrient cycling, individual response to density and so on, as an example, if the soil is fertilized, the negative response to nutrient deficiency is removed; therefore a plant population outbreak occurs because of the of positive feed pack of the fertilizer (Radosevich et al. 1997). The lack of negative regulation in a system also may occur if a new exotic species is incorporated into the system, this is why many invasive species become troublesome weeds (Booth et al. 2003).

Weed infestation in a field is defined by four parameters: the number of species present, the genetic diversity of each species, the density of each species, and the distribution of the species across the field (Radosevich et al. 1997). While the number of species in a field remains relatively constant from year to year, the other factors fluctuate widely in response to environment, pathogens, cultural practices and weed management tactics (Blackshaw et al. 2001; Cousens & Croft 2000; Busey 2003). It is the continual changes in weed population that make successful weed control such a difficult task to consistently achieve (Hartzler 2000).

Within a weed population, the individuals are found in various functional stages, interacting with each other, with populations of other species and with the environment (Radosevich et al. 1997). Moreover a weed may affect other organisms by changing their survival and growth or changing one or more of ecosystem processes like nutrient cycling (Booth et al. 2003) Plant demography is the study of how plant populations change in size and structure during various stages of their life cycle. It is possible, using demographic principles, to assess how weed populations might change through time or respond to perturbation in their habitat or environment (Radosevich et al. 1997).

Seasonal and long term population dynamics have been studied under different weed management tactics including tillage and crop rotation (Johnson and Mullinix 1997; Felix & Owen 1999; Blackshaw et al. 2001; N'Zala et al. 2002), crop spacing and spatial distribution of weeds (Darwent & Elliott 1979), grazing intensity (Harker et al. 2000), and herbicide application methods (Johnson and Mullinix 1997; Felix & Owen 1999; Fernandez-Quintanilla et al. 1987). Results of these studies suggested that the development of integrated weed management systems is a complex task and must be supported by thorough understanding of the dynamics of weed populations through changing weed coverage, density, species composition and distribution in response to agronomic practices. Therefore, in order to develop effective and economical weed control measures, it is important to study the weed flora and their dynamics during the crop cycle (Fernandez-Quintanilla 1988).

Several current publications (Burpee 1990; Deacon 1991; Kennedy & Kremer 1996; Radosevich et al. 1997; Cousens & Croft 2000) considered the importance of studying ecology and population dynamics of weeds to enhance weed biocontrol programs, but to my knowledge, no specific study has been published. Burpee (1990) reviewed the impact of abiotic factors on interactions among soil-borne plant pathogenic fungi, and microbial antagonists and he emphasized the effect of edaphic factors on disease or pathogen-suppressive activities of microbial antagonists, rather than population dynamics of these organisms.

Fernandez-Quintanilla (1988) discussed the different approaches available to analyze the population dynamics of weeds depending on levels of complexity. The method of long-term studies is the simplest level and involves monitoring a single component of the population throughout several seasons, determining population trends and rates of changes. This approach was recommended by the author to describe the effects of certain management practice on a weed population (Fernandez-Quintanilla et al. 1987).

2.3. Turfgrass

2.3.1. Turfgrass industry

Turfgrasses are grasses that act as a vegetative ground cover with numerous recreational and aesthetic benefits for human and serve a functional environmental purpose by preventing soil erosion (Beard & Green 1994). Turfgrass offers more advantages to life, it releases significant amount of oxygen into the air, causes about 50% cooling effects of sun's heat through transpiration and it helps to remove air pollutants and dust particles from the atmosphere (Emmons 1995).

Turfgrasses have been utilized by humans for more than ten centuries but a revolution of turf industry has been evolved primarily during the past three decades in parallel with modern civilization and urbanization (Walsh et al. 1999). Turfgrass is the major vegetative ground cover in the American landscape with an estimate of more than 46 million acres (~18.6 ha) of turf represented by 93 million dwellings including home lawns, commercial lawns, golf courses, athletic fields, parks, campuses, recreational areas and roadsides (Monaco et al. 2003). While most of the households participate in do-it-yourself lawn care, about 9.3 million lawns are maintained by professional lawn care operators (Monaco et al. 2003). The annual expenditure for maintaining turfgrss in the USA was conservatively estimated to be $45 billion (Beard & Green 1994). Robbins & Birkenholtz (2003) estimated the lawn coverage of Franklin County, OH as 23% of land cover and discussed the implications of this expansive turf coverage on replacement of natural and agricultural lands. They concluded significant impacts regarding chemical exposure, water and energy demands and wildlife conservation.

Turfgrass species grown in North America are categorized as either cool-or warm-season grasses. Cool-season grasses have a C-3 photosynthetic pathway and are more common to Canada and the Northern United States (Anderson 1996). Cool-season grasses include Kentucky blue grass, fine fescues, tall fescues, creeping bentgrass and perennial ryegrass. The most important and widely used in North America is Kentucky blue grass (*Poa pratensis*), it was introduced to North America from Europe in the

1600's (Emmons 1995). The turfgrass seed market is the second largest market after hybrid corn seeds as there are 14000 golf courses in the US alone and 300 new golf courses are constructed annually (Lee 1996).

2.3.2. Turfgrass weeds and management

As a man-made or man–interfered environment, weeds invade turfgrass. Weeds often are symptoms of a weakened turf, not the cause of it (McCarty et al. 2001). In general a dense healthy grown turfgrass is the best defence against weed colonization (Monaco et al. 2002; Busey 2003). Therefore weeds are mostly found where soil has been exposed or disturbed by compaction (e.g. side walk edging) or in a weakened turf because of adverse environmental conditions, pests or improper selection of not adapted grass species to a local environment (McCarty et al. 2001). However weed seeds could be introduced to the turf any time by various ways of seed dispersal and nearly every lawn has weed problems and needs some degree of management (Monaco et al. 2002). Weed species common to turfgrass vary with geographic regions, but many are common to more than one region (Anderson 1996). McCarty et al. (2001) described 73 grass and grass-like and 145 broad-leaf species as weeds in turfgrass environments.

The most obvious impact of weeds on turf areas is the competition for light, soil nutrients, soil moisture and physical space which may lead to replacement of turf by weeds and disturbs the visual turf uniformity due to different growth habits, different leaf shape and size or color contrast (McCarty et al. 2001). Some common weeds are poisonous if consumed (e.g. black nightshade); cause inflammation when touched (e.g. stinging nettle, poison ivy) or cause allergic reaction (e.g. common ragweed). Turfgrass weeds also can harbour pests such as plant pathogens, arthropods and rodents (Anderson 1996; McCarty et al. 2001).

Cultural management involves turfgrass selection and planting, irrigation, cultivation, fertilization and mowing and hand weeding. The proper use and time of each of these cultural practices depend on the turf species and the environment. Although cultural practices do not eliminate weeds, they are a necessity to reduce dependence on synthetic herbicides (Busey 2003). Hand weeding is the oldest practice of weed control but the strength and/or depth of the vegetative reproductive parts of perennial weeds make them difficult to be removed and remaining parts can resprout (Anderson 1996).

Chemical management of weeds in turfgrass originated about 1895 (Hansen 1921: cited in Busey 2003) but substantial use of chemicals began after the discovery of 2,4-D in 1944. Herbicide application could be before planting or after turf establishment (Monaco et al. 2002). 2,4-D is widely used and if applied properly, it controls a number of broadleaf weeds without adverse effects on the grass (Emmons 1995). Mixtures of at least two of 2,4-D, mecoprop (MCPP) [(±)-2-(4-chloro-2-methylphenoxy) propioinic acid] , dicamba, triclopyr {[(3,5,6-trichloro-2-pyridinyl)oxyl]acetic acid} and clopyralid (3,6-dichloro-2-pyridinecarboxylic acid) are commonly used and ensure better broad–spectrum weed control (Emmons 1995). Recently there has been increased public concern raised concerning the possible adverse environmental effects of lawn pesticides (see section 2.4.4.2).

Recent studies have shown selected microorganisms to be promising bioherbicides to control certain weeds in turfgrass systems. Examples include *Xanthomonas campestris* pv. *poannua* to control annual blue grass (Johnson et al. 1996); *Bipolaris setaria* and

Pyricularia grisea to control goosegrass (Figliola et al. 1998); *Sclerotinia minor* (Ciotola et al. 1991; Riddle et al.1991) and *Phoma herbarum* (Neumann & Boland 1999) to control dandelion. Corn gluten hydrosylate has been shown to have herbicidal activity on crabgrasses and germinating seeds of other species (Christians 1991).

A multiple management approach to control weeds has evolved, integrated weed management (IWM). This approach combines available control measures (cultural, chemical, biological and ecological) to attain successful weed control with minimum environmental impact. Synergic efficacy was recorded using reduced herbicide rates combined with a pathogen (Schnick et al. 2002; Schnick & Boland 2004) or with mowing (Lowdey & Marrs 1992). Integrating a pathogen with interspecific competition from a crop or a cover crop (Guntli et al. 1999; Story et al. 2000) or combining a pathogen with mechanical cutting or defoliation (Green et al. 1998; Kluth et al. 2003) exerted more suppressive effect on weeds than the pathogen alone.

2.4. Dandelion (*Taraxacum officinale* Weber ex wigger)

2.4.1. Distribution and habitat preference

Dandelion is a herbaceous perennial plant belongs to Asteraceae. It is successful in colonizing a broad range of climatic conditions and found in almost every cold, subtemperate, temperate, and subtropical regions of the world (Solbrig & Simpson 1974; Holm et al. 1997). Dandelion has been reported in over 60 countries worldwide (Holm et al. 1997). The species is native to Eurasia and was introduced to North America by European settlers probably in the 17 century (Solbrig & Simpson 1974).

The first Canadian collection of *Taraxacum officinale* was made in Montreal, QC in 1821 (Rousseau 1968). Dandelion has been reported throughout Canada including almost all isolated regions (Stewart-Wade et al. 2002b).

Dandelion is commonly found on disturbed soils and has become a common weed infesting home lawns, turfgrass swards, pastures, forages, golf courses, athletic fields, wood areas, and roadside verges (Stewart-Wade et al. 2002b). Dandelion is somewhat unique in colonizing habitats with widely variable environments. It can grow in a wide range of soil types (Simon et al. 1996), wide range of soil pH (Von Hofsten 1954), resists drought (Von Hofsten 1954), and adapts to a wide range of light and shade intensity (Longyear 1918).

2.4.2. Description, competitiveness and intraspecific variations of dandelion.

Dandelion was described by Bouchard and Neron (1999) as follows:

> "Leaves are elongated, margins wavy-toothed to deeply divided, irregular segments, margins usually toothed. The leaves are variable in shape from one plant to another, usually hairless but may also be hairy. Leaves are all arranged in a rosette at the base of the plant. No stems appear but hollow floral scapes, 5 to 30 cm high arise from the center of the rosette. Plants have very strong tap roots and yellow single flower heads. Seeds have a white long beaked pappus and are wind-dispersed".

Phenotypic plasticity in dandelion increases its ability to colonize a wide range of habitats (Stewart-Wade et al. 2002b). The variable growth habits of dandelion rosettes, the strong deep taproot, and the high seed potential are responsible for the strong competitive abilities of dandelion. The rosette growth habit of dandelion can spread flat on the ground surface or be more or less erect tufts enabling dandelion to survive weather conditions, grazing, mowing and competition with grasses (Longear 1918; Baker 1974; Lovell & Rowan 1991).

The thick, branched taproot can be 2-3 cm in diameter, grows up to 1-2 m in length (Von Hofsten 1954; Solbrig 1971), and extends below the level of grass roots, making it difficult to remove manually (Lovell & Rowan 1991). It is surmounted by a highly divided crown, which can produce up to 22 branches depending on plant age and crowding factor (Roberts 1936). The root is highly regenerative, producing shoots and roots from small segments (Emmons 1995), and has the ability to overwinter, as the crown is contractile at the end of the growing season, drawing the crown below the soil surface, providing protection against the harsh winter (Longyear 1918). The parenchymaous cells of the secondary phloem and xylem in the roots are able to develop into new shoots and roots (Higashimura 1986). Bioassay of root extracts showed that relatively high auxin and cytokinin activities were present compared with gibberellin (Booth & Satchuthananthavale 1974).

From the basal rosette of a dandelion plant, one to numerous glabrous, hollow cylindrical scapes (5-50 cm tall) are able to rise (Holm et al. 1997). The scape bears a single terminal capitulum of 2-5 cm diameter and composed of up to 250 ligulate, perfect yellow florets (Holm et al. 1997). In dandelion, there is no distinction between ray and disc-florets either in appearance or function with all florets being ligulate and equally fertile (Roberts 1936). Most pollen grains of dandelion are abortive, sterile and cannot form pollen tubes (Solbrig 1971) so the seeds are develop without fertilization (Roberts

1936). Ford (1981) found that the number of inflorescence/plant, number of seeds/inflorescence and consequently the number of seeds/plant vary with the habitat of this agamospecies. In a heavily infested area in Canada, 48-146 inflorescence/plant (average 93) and 130-412 seeds/inflorescence (average 252) were recorded, resulting in an average of 23,436 seeds/plant produced in a growing season (Roberts 1936). Bostock & Benton (1979) reported lower fecundity in the United Kingdom with an average of 12.2 inflorescence/ plant and 2,170 seeds/ plant.

Dandelion scapes elongate significantly to enhance dispersal and the seeds have pappi that further aid in wind dispersal (Radosevich et al. 1995). The papus serves as a parachute and causes the relatively low falling velocity of about 0.38 m s^{-1} (Tackenberg 2003). Sheldon and Burrows (1973) correlated dandelion seed dispersal with wind speed while Tackenberg et al. (2003) concluded that the long-distance (more than 100 m) dispersal of dandelion seeds is mainly caused by convective updrafts rather than wind speed. Dandelion seeds are also dispersed in the excreta of animals like horses, cattle, and birds and by water via irrigation ditches (Radosevich et al. 1995).

A physiological competitive feature of dandelion is its capability to manipulate the root carbohydrate reserve according to seasonal fluctuation and so the plant can adapt to temporal environmental stresses over other plants in the community (Wilson et al. 2001). According to the authors, understanding this feature may allow better timing of a control method.

The genus *Taraxacum* consists of 200 closely related species, 90% of which are polyploids and reproduce asexually by obligate agamospermy while the majority of the 10% are diploids reproduce sexually as obligate outcrossers, the remainder are primitive self-fertilized species (Hughes & Richards 1985). Indications of co-existence of sexual and agamospermous reproduction within the same agamospecies are also present (Hughes & Richards 1985). Both sexually and asexually Eurasian lineages are subjected to be introduced in the New World but only the asexual triploids (3n= 24) have successfully been able to colonize and reproduce via agamospermy (King 1993). Apomictic offspring are genetically identical and not expected to exchange genes but extensive intraspecific variation in North American dandelion populations is well documented in the literature (Stewart-Wade et al. 2002b). Significant differences in floral stage timings among different clones of North American dandelions were found when grown under a constant set of environmental conditions (Collier & Rogstad 2004).

Dandelions are very plastic, but very difficult to determine morphologically (Solbirg & Simpson 1974), therefore studies focused on allozymes to identify different biotypes of dandelion. In a local Michigan study, four genotypes were identified using isozymes analysis (Solbrig 1970), whereas 21 genotypes were identified in a comprehensive examination of allozyme diversity in North American dandelions (Lyman & Ellstrand 1984). van Oostrum et al. (1985) considered North American dandelion populations as composed of a mixture of clones, only a few of them being widespread. DNA analysis techniques have the potential to discriminate among clonal individuals that are not detected to be different with allozyme assays (Falque et al. 1998; Rogstad et al. 2001; van Dijk et al. 2003).

The morphological and genotypic variations among dandelion lead to its complex taxonomy (Small & Catling 1999). The genus is dealt with as many micro-species in Europe while as one species exhibiting considerable phenotypic plasticity in North

America (Richards 1973). The cause of phenotypic plasticity or genetic diversity in dandelion could be due to multiple introductions of European microspecies (Taylor 1987). King (1993) used restriction enzyme analysis of ribosomal DNA and chloroplast DNA to assess the source of genetic variation and concluded that multiple hybridization among microspecies populations, prior to their introduction to North America, was of greater importance than mutations in populations. In contrast, Rogstad et al. (2001) used genetic markers to examine the population structure of the central North American dandelions and suggested evidences that this diversification is more likely to be due to the steady accumulation of mutations rather than occasional sexual exchange.

2.4.3. Dandelion as a beneficial plant

This topic is beyond the subject of this research, however dealing with the biocontrol from an ecological point of view, the beneficial role of dandelion in our society and environment must be noted. Dandelion is one of the oldest medicinal plants with increasing significance of therapeutic, nutrition and beverage industries. Recently scientific reports have confirmed the traditional application of dandelion root extracts for therapeutic and nutritive uses and so a new program has been initiated at Laval University, Quebec, to introduce organic production of dandelion for commercial purposes (Letchamo & Gosselin 1995). Dandelion is also known as diuretic (Racz-kotilla et al. 1974); eliminates liver toxins, lowers cholesterol and blood pressure (Mattern 1994), and decreases body weight in obese patients (Dalby 1999). A potentially valuable source of antioxidant and bioactive materials were found in dandelion flower (Hu & Kitts

2003). In Canada, dandelion is one of 14 plant extracts in a health tonic sold under the brand name of MATOL (Michaud et al. 1993).

The nutritive value of dandelion leaves and flowers have received the attention of scientists regarding human consumption (Kuusi et al. 1984a & b). In Toronto alone, 155 tonnes of dandelion leaves were marketed as a salad green in 1988 and 1989 (Letchamo & Gosselin 1995). Extracts from dandelion have been used in cheese preparation (Akuzawa & Yokoyama 1988) and also in soups, beverages and as a coffee substitute (Stewart-Wade et al. 2002b). Dandelion has enough nutritive materials to exceed the established requirement for cattle (Bergen et al. 1990); it has a good digestibility and mineral availability for sheep (Derrick et al. 1993) and it's an excellent source of nectar for honey bee foraging (Mayer and Lunden 1991). The infusion of dandelion roots could be used as a source of carbon and energy for bifidobacteria due to its contents of oligofructans, glucose and fructose (Trojanova et al. 2004).

The global distribution of the common dandelion along with its ability to tolerate a wide range of environmental conditions and the analysis of heavy metals contents in the leaves make this species a particularly attractive candidate to evaluate its value as a biological monitor of environmental metal pollution since the accumulation of heavy metals corresponds to extent environmental pollution (Kulev & Dzhingova 1984; Rogstad et al. 2000; and Keane et al. 2001). Polychlorinated biphenyls were found to be accumulated in dandelion in the conditions of soil contamination with oil derivatives (Malgorzata & Boguslaw 2001). Polycyclic aromatic hydrocarbons were also monitored using dandelion (Malawska & Wilkomirski 2001). Trace metals like Cd, Cu, Mn, Pb, and Zn were also detected in dandelion to assess the pollution for Montreal Urban soils (Marr et al. 1999). The beneficial values of dandelion recommend the use of biocontrol or any other natural approach to suppress dandelion population rather than to eradicate it.

2.4.4. Dandelion as a problematic weed

Dandelion is a common weed that infests terrestrial habitats with widely variable environments. It is a noxious weed in pastures, forages, orchards, lawns, golf courses, municipal parks, and road sides (Holm et al. 1997). The biological traits that favours survival in these habitats are often contradictory, thus a trait that favours survival in an agricultural field could be unfavourable for survival in turfgrass (Solbrig & Simpson 1974; 1977). Genetic variability within dandelion populations was found to be an effective tool in regulating the population through different degrees of competitiveness and survival in response to pressures from interspecific competition (Vavrek 1998) and disturbances (Solbrig & Simpson 1974).

As an agricultural weed, it's known to reduce the yields of several crops as corn (Hartwig 1990), wheat (Ahmad 1993), alfalfa (Waddington, 1980), and spring canola (Froese & van Acker, 2003). Dandelion described as a prevalent weed in annual cereal and oil seed crops in Western Canada (Derksen & Thomas 1997). It is among the toughest weeds to control in reduced and no tillage fields and this was explained by the possibility of being trapped by the high amount of crop residues present in these types of fields (Frick & Thomas 1992). The high water content in dandelion tissues, especially the stems and ribs, causes the slow drying of hay and reduces its quality (Moyer et al. 1990).

Dandelion acts as an alternative hosts for several pests (as reviewed by Stewart-Wade et al. 2002b) and as an allergen causing allergic contact dermatitis (Goulden &

Wilknson 1998). In turfgrass systems, dandelion interrupts uniformity and limits the grass density (Riddle et al. 1991) and it is undesirable weed causing aesthetic problems during flowering and seed production period (Holm et al. 1979).

2.4.4.1. Cultural management practices to control dandelion

Cultural managements include fertilization, mowing, irrigation, cultivation and selection of proper turfgrass species have been practiced to control weeds in turfgrass (Busey 2003). Although extension recommendations often indicated that, a dense healthy turfgrass accompanied by proper mowing, watering and fertilization is the best defence against weed colonization but this often stated in generalities or based on a scant published research (Busey 2003).

A high rate of nitrogen fertilization (100-300 kg N $ha^{-1}yr^{-1}$) reduces population of dandelion and other broadleaf weeds in cool-season turfgrass (Murray et al. 1983). Mowing every two weeks did not prevent dandelion colonization in Kansas and dandelion was able to develop stands of 32 to 63 plants m^{-2} and buffalograss was the most competitive with dandelion compared to bentgrass and bermudagrass (Timmons 1950). In Ontario, Kentucky blue grass was the least and perennial ryegrass was the most competitive with dandelion among six turf species studied (Hall et al. 1992). Manual removal of dandelions using a special tool has been of limited value since a remained piece of root, covered by 5-10 cm of soil readily propagates a new plant (Falkowski et al. 1989). Interspecific competition also may exert a biological suppression on weed species. Density and abundance of dandelions were positively correlated with potassium level in its tissues and the use of potassium-free lawn fertilizer decreased dandelions because of increased competition from grasses (Tilman et al. 1999). In a competitive environment, the growth of all five *T. officinale* genotypes reduced to the least with *Poa pratensis* Kentucky bluegrass relative to other competitors *Plantago major* and *Trifolium pratense* (Vavrek 1998).

2.4.4.2. Chemical control of dandelion

The generally accepted practical current method of dandelion control has been the use of chemical herbicides. Repeated applications of dicamba or phenoxy herbicides such as 2,4-D and mecoprop or a combination product such as "KILLEX™" are extensively used for dandelion control (Anonymous 1997). Combination products are normally recommended to control a wide spectrum of broadleaf weeds (Emmons 1995). The major herbicidal effects are achieved by 2,4-D which causes epinasty, cell elongation, chloroplast damage, ethylene evolution, and increased biosynthesis of ATPase, nucleic acids and proteins (Ashton & Crafts 1981).

2,4-D represents the most common herbicide used for domestic purposes (non-agricultural sectors) in Canada and USA (Watson 2003). According to the United States Environmental Protection Agency (EPA) (2004) the quantities of yearly active ingredients of 2,4-D used for home and garden purposes were ranged as 3.6-5 Million kg based on 1999-2001 estimates. In Canada one tenth of these amounts are expected to be applied yearly (A. K. Watson personal communication). In Alberta alone 44 tonnes of herbicides were applied on home lawns and about 3.5 tonnes of herbicides were applied in Calgary national parks in 1998 (Alberta Environmental Protection 2001).

Generally speaking, the intensive use of synthetic chemical pesticides has resulted in increased environmental sustainability concerns (Robbins & Birkenholtz 2003), but more public awareness and concerns have been developed specifically on the use of 2,4-D and other lawn pesticides (Riddle et al. 1991). This primarily due to the proximity of the site of application to the site of human occupation, this leads to chronic exposure and persistence indoor contamination (Robbins & Birkenholtz 2003). The effect of these toxins upon human and specifically children is still not well investigated but surveys showed that children living in homes where pesticides were used have seven times more likely to develop childhood leukemia (Meyer & Allen 1994). The toxicity of 2,4-D was ranged as slight to high for birds, fishes and insects (USEPA 2001) and is known to cause mutations, birth defects, some damage to liver, kidneys and central nervous system as well as it is suspected to be carcinogenic (Meyer & Allen 1994). Moreover the carcinogen “dioxin” was detected in certain formulations of 2,4-D (Cochrane et al. 1981). On the other hand, under frequent lawn irrigation, 2,4-D, dicamba and MCPP were detected in the leachate and this can have impact on the movement of these herbicides through soil profiles causing water contamination (Starrett et al. 2000). A United States survey revealed the detection of one or more pesticides in 99% of urban stream samples (US Geological survey 1999 cited in: Robbins and Birkenholtz 2003).

All of the above mentioned health and environmental concerns imposed some municipal governments in Canada, USA and other countries to restrict or ban the use of 2,4-D and related herbicides in residential and public properties (Riddle et al. 1991; Tompkins et al. 2004; Cox 1999). This new situation has brought a new type of research emphasis to find natural substitutes for the turf chemical herbicides.

2.4.4.3. Biologically based and biological control of dandelion

Corn gluten meal is being marketed in the United States as a naturally occurring pre-emergent herbicide and a nitrogen source (Christians 1991). In Canada, this product has recently obtained a temporarily registration as TurfMaize® and reported to be effective in controlling crabgrasses and some control of certain broad leaf weeds (Tompkins et al. 2004). When applied, under glasshouse conditions, as a minimum as 324 g m^{-2}, corn gluten meal reduced survival of dandelion emergence by 75% of the control (Bingaman & Christians 1995). The active ingredients in this product are the dipeptides glycinyl-glycine and alaninyl-alanine which found to inhibit the root formation of susceptible species (Liu & Christian 1996).

The limitations of corn gluten meal as a natural herbicide to control dandelion are: 1) it inhibits root growth of germinating seeds but doesn't damage plants that have formed a mature root system (Christians 1991). 2) While published data indicated an accepted control for crabgrasses and perennial ryegrasses, the performance of this product on broadleaf species under field conditions is not clearly defined (Liu et al. 1994; Christians 1991; Bingaman & Christians 1995; Liu & Christian 1996). 3) The cost of using corn gluten meal is very expensive $418/acre, compared to synthetic herbicides which can be as low as $30/acre (Wilen & Shaw 2000). 4) Finally it's insoluble in water rendering it difficult to apply as an herbicide, so that the development of "gluten hydrolysate" (corn gluten meal hydrolyzed by proteinases) caused greater inhibition of germinating seeds and root growth (Liu et al. 1994).

Vertebrate herbivores which feed on dandelion could be used as biocontrol agents in certain agro-ecosystems. Müller et al. (1999) found that sheep and geese are good biocontrol agents, with sheep being superior in suppressing dandelion in Christmas tree plantations. Several insects have been reported to survive and feed on dandelion that may have biological control potential (Stewart-Wade et al. 2002a). These include the weevil *Ceutorhynchus punctiger* Gyllenhall (McAvoy et al. 1983), the potato leafhopper, *Empoasca fabae* (Harris) (Lamp et al. 1984), root-feeding larvae of the Japanese beetle, *Popillia japonica* Newman, the southern masked chafer, *Cyclocephala lurida* Bland (Crutchfield & Potter 1995), and the cynipid wasp, *Phanacis taraxaci* (Ashmead) (Bagatto et al. 1996). According to my knowledge neither the evaluation of suppression level nor the host specificity and key mortality factors has been studied for any of the above mentioned insects.

Phoma herbarum is a fungus has been reported as a potential bioherbicide for dandelion in turf environment (Neumann & Boland 1999). Four-week-old dandelions were significantly more susceptible to *P. herbarum* than older life stages and the use of mycelial suspension cause more severe disease than conidia (Neuman & Boland 2002). Variation in disease intensity and efficacy of *P. herbarum* under field conditions demonstrate the need to characterize optimal application conditions and formulations (Neumann & Boland 1999; Stewart-Wade et al. 2002a). *Phoma macrostoma* was also reported as a potential bioherbicide for dandelion and Canada thistle in turfgrass (Bailey et al. 2003). Interestingly Schnick & Boland (2004) found that sequential treatments of sub lethal rates of 2,4-D and *Phoma herbarum* were synergic for dandelion control.

Riddle (1989) reported that 8-week-old dandelions were highly susceptible to infection by strains of *Sclerotinia sclerotiorum* and *S. minor* isolated from various host species. Diseases caused by *S. sclerotiorum* and *S. minor* were most severe for dandelion under cool wet conditions (Riddle et al. 1991). Positive correlations between isolate virulence and reduction in the number of dandelion plants in inoculated turfgrass swards were observed. Another isolate, virulent on dandelion and plantain (*Plantago major* L.), was also evaluated for broadleaf weed control in turfgrass (Ciotola et al. 1991).

Burpee (1992) tested the efficacy of nine granular formulations of a bioherbicide containing the fungus *S. sclerotiorum* on vegetative growth from dandelion tap roots after incubation in a controlled environment. Two granular formulations inhibited the production of new petioles and leaves from treated tap roots after 96 h when grown at 23^{o}C and provided 100% relative humidity.

A collaborative project in Canada involving three academic institutions (University of Guelph, McGill University, and Nova Scotia Agricultural College) and three industrial partners (Dow AgroScience Inc., BioProducts Centre Inc., and Saskatchewan Wheat Pool) was established to develop a bioherbicide targeting dandelion in home lawns (Stewart-Wade et al. 2002a). After screening the fungal pathogens of dandelion, the organisms with the highest potential were chosen, formulated and tested under controlled and three different field environments (Ontario, Quebec and Nova Scotia) (Stewart-Wade et al. 2002a). Two solid formulations of *Sclerotinia minor* MAC1 (refers to Macdonald Campus, McGill University) containing mycelium in sodium alginate granules (Brière et al. 1992) and mycelial-colonized barley grits (Ciotola et al. 1991) were found to be the most effective formulations. In addition, MAC1 did not infect any of the turfgrass species tested, the transfer of infection to lettuce (a common highly susceptible host) requires direct contact, and the potential for infection of common garden plants was minimal (Stewart-Wade et al. 2002a). Sequential treatments of sub lethal rates of 2,4-D and *S. minor* were synergic for dandelion control (Schnick et al. 2002). An attempt to enhance the virulence of the granular *S. minor* formulation by maximizing the available amount of oxalic acid was conducted by Brière et al. (2000). A 330% increase in oxalic acid was obtained using modified Richard's solution (MRS) plus sodium succinate as compared to MRS alone, and a concomitant increase in virulence of 218% was expressed as increased lesion diameter. Currently, ongoing studies are investigating product costs, storage conditions, application timing, and health and environmental safety of this potential bioherbicide (A.K. Watson personal communication).

2.5. The fungus: *Sclerotinia minor* Jagger

As mentioned above, this fungus is a promising bioherbicide candidate for controlling dandelion in turfgrass systems. Of the five plant pathogenic species of the genus *Sclerotinia*, only *S. sclerotiorum* and *S. minor* have been reported in Canada (Bardin & Huang 2001). *S. sclerotiorum, S. trifoliorum* and *S. minor*, three closely related species, differ in mycelial, sclerotial, apothecial, morphogenetic, cytological, and electrophoretic characteristics, host ranges, and main mode of field infection (Willetts & Wong 1980).

S. minor Jagger (class: Discomycetes, order: Helotiales, Family: Sclerotiniaceae) is a soil-borne plant pathogen that can cause substantial losses in several crops particularly lettuce, *Lactuca sativa* L. (Melzer et al. 1997). Losses of 10-50% in lettuce are common (Melzer & Boland 1994) but losses up to 75% have been reported (Beach 1921: cited in Melzer et al. 1997). *S. minor* also causes economic losses in other crops, including peanut, chicory, green bean and sunflower (Abawi & Grogen 1979; Wadsworth &

Melouk 1985; Melzer et al. 1997). In Canada, S. *minor* has only been reported as a pathogen on lettuce crop in Quebec and Ontario (Bardin and Haung 2001). Surveys conducted from 1989 to 1991 in Holland Marsh, Ontario, indicated that *S. minor* was the most prevalent pathogen causing lettuce drop (Melzer et al. 1993). Disease caused by *S. minor* is characterized by rapidly expanding of watery, soft-rot lesions on the leaves and crown area of infected plants followed by the appearance of small (0.5-2 mm diameter) black sclerotia (Jagger 1920: cited in Melzer & Boland 1994). The crop canopy microclimate has no effect on the epidemiology of lettuce drop which is normally developed under moist soil conditions and temperatures of 5-25^{o}C after lettuce had developed to head stage (Melzer & Boland 1994). Plant to plant spread occurs occasionally by mycelial contact and sclerotia located within 2 cm of the taproot and 8 cm of the soil surface which can produce masses of the hyphae that infect nearby roots, stems, and senescent leaves (Subbarao 1998). Sclerotial formation occurs between 12-24^{o}C with more sclerotia produced at 12^{o}C but larger sclerotia produced at the higher temperature (Imolehin et al. 1980).

Recorded hosts of *S. minor* include 21 families, 66 genera and 94 plant species (Melzer et al. 1997). All hosts belong to Angiospermea of the plant division Spermatophyta. Three plant hosts occurred in the subclass Monocotyledonae (tulip and asparagus: Liliaceae; banana: Musaceae), while the other 19 families are all Dicotyledonae. The families Asteraceae, Fabaceae, Brassicaceae, Apiaceae and Caryophyllaceae had the greatest number of hosts, in decreasing frequency (Melzer et al. 1997). Recently, Meador & Melouk (2002) mentioned the host range of *S. minor* to be 222 plant species. Although *S. minor* has a broad host range that includes many economically important plants it is not considered to be a serious pathogen on most plants (Mezler et al. 1997). The host range of *S. sclerotiorum* is considerably greater and includes 75 plant families, 278 genera and 408 plant species as hosts for *S. sclerotiorum* (Boland & Hall 1994).

S. sclerotiorum infects hosts through dissemination of ascospores while *S. minor* infects through eruptive and myceliogenic germination of sclerotia (Abawi & Grogan 1979). The production of apothecia in *S. minor* in nature is extremely rare, has not been recorded in North America, and thus is considered to be of minor importance in the disease cycle (Abawi & Grogan, 1979). Infection of lettuce with *S. sclerotiorum* most often occurs at the ground level because it usually originates from ascospore infection of senescent lower leaves. In contrast, infection with *S. minor* can occur either at the soil line through senescent lower leaves or below ground as deep as 10 cm through root and stem tissue (Abawi & Grogan 1979).

The sclerotia of *Sclerotinia* species have been reported to survive in the soil for three to five years under natural conditions (Adams & Ayers 1979). In New Zealand, sclerotial numbers of *S. minor* rapidly declined in a horticultural soil box trial over the first three months and lower recoveries (9-11%) were observed in the field after six months (Alexander & Stewart 1994). Survival of MAC1 sclerotia (produced on barley grits formulation) in the field decreased significantly after four months (65% recovered and 17% viable), 20% recovered and 7% viable after seven months, and only sclerotia rinds were retrieved after 11 months (McGill Combined Research Report 1996-1998).

Mycelial germination of *S. minor* sclerotia was the predominant germination mode and occurred at 5-25^{o}C with an optimum of 15^{o}C, with no effect of soil type (Hao et al.

2003). In another study, germination was reported to occur from 6-30°C with an optimum of 18°C (Imolehin et al. 1980).

Species of *Sclerotinia* are able to spread locally or regionally by several means, including wind, farm equipment, animals, or human (Abawi & Grogan 1979). Viable sclerotia of *S. minor* may pass through the digestive tract of a ruminant and spread the pathogen from infested to noninfested areas (Melouk et al. 1989). Irrigation also has been shown to be involved in the spread of *Sclerotinia spp*. from field to field (Steadman et al. 1975). Long distance dissemination of *Sclerotinia spp*. probably occurs via seeds infected with mycelia or contaminated with sclerotia (Alexander & Stewart 1994).

Peanut seeds processed by hand only or by hand and machine had infection levels of 25.4 and 8.9% respectively, while seeds processed solely by machine had 1.4% infection (Wadsworth & Melouk 1985). Incidence of *S. minor* in seeds of four susceptible peanut genotypes in infested field plots ranged from 6.8% to 12.3% while the disease incidence value averaged 0.0 to 3.5% under a controlled greenhouse environment, therefore the source of seed before planting in disease-free fields should be considered (Akem & Melouk 1990). Peanut genotypes with a bunch growth habit exhibited a lower disease incidence than with a prostrate growth habit (Akem et al. 1992).

In addition to the research presented in this thesis, other unpublished research work have been conducted about *S. minor* IMI 344141. Appendix-1 contains unpublished results about screening of different fungal isolates and *S. minor* pathotypes for aggressiveness on dandelion and the characterization of the selected isolate while appendix-2 presents unpublished data about the role and fate of *S. minor* after field application.