Identifying the locations, movement and habitat of the European Fire Ant, *Myrmica rubra*; an invasive species in the urban/suburban environment of Halifax, Nova Scotia

by

Susan M. L. Horton

A Thesis Submitted to Saint Mary's University, Halifax, Nova Scotia,
in Partial Fulfillment of the Requirements for the
Degree of Master of Science in Applied Science

December 2011, Halifax, Nova Scotia

Approved:	Dr. Cathy Conrad Supervisor Geography Department
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Approved:	Dr. Doug Strongman Supervisory Committee Member Biology Department
Approved:	Dr. Cristian Suteanu Program Representative
Approved:	Dr. Kevin Vessey Dean of Graduate Studies

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Abstract

Myrmica rubra, also known as the European Fire Ant, is a pestiferous, invasive species in North America. It administers a painful sting and causes many residents to be unable to use their outdoor property. In this study, nest preferences in the urban/suburban environment were investigated. Habitat characteristics such as vegetation, substrate and edaphic factors were compared to nest densities. The species showed high adaptability to all nest features. However, the majority of nests were found in ground flora roots or against a hard substrate in a slightly acidic soil. This differs from nest sites in rural invaded areas where most nests are found in leaf litter or wood. A secondary study measured densities of ants using sucrose bait stations. The results were mapped using GIS and showed a correlation between locations of nests and densities in vials. An exponential increase of residents encountering this species indicates an irruption is occurring.

December 9th 2011

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This Master of Science degree has been quite an adventure for me. I first met my supervisor, Dr. Cathy Conrad at an environmental education day where she was a guest lecturer. I knew I had found a kindred spirit, as her enthusiasm for her subject and ability to engage the public immediately appealed to my love of education and public service. I would like to thank Cathy for believing in me and allowing me to choose my own path.

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1 Introduction

1.1 Introduction

Halifax, a coastal city on the eastern seaboard of Canada, has a large ice free harbour and consequently is an active port. The Halifax peninsula is the primary population centre of the Halifax Regional Municipality (HRM). Historically, Halifax has been an important hub for trade. This trade often involved the movement of live plants and animals, and water or soil from many areas of the world (much of which was used as ship ballast). Many of the species purposely or accidentally introduced, have integrated into the local ecosystem and are generally unnoticed. However, there are some that have become invasive. The change from introduced, or simply non-native to invasive depends on the detriment to native biodiversity. One such species which has become pestiferous, particularly in the last 10 years, is *Myrmica rubra* (Linnaeus). This species is also known as the European Fire Ant. In its native Palearctic habitat of northwestern Europe and western Asia, this ant is generally less-aggressive and the genus plays an important role in the conservation of the Blue Butterfly *Maculinea spp.* (Elmes et al. 1998). In its non-native environment however, it has very aggressive tendencies and a higher than average population density in the colony (Groden et al. 2005). There is currently a well established distribution of colonies in the State of Maine, USA, where this ant has become a focus of researcher and public attention. There, the problem has become so great that many people are unable to use their outdoor property for fear of being attacked and stung (Groden et al. 2005); a situation is which is now occurring in Halifax (Hebda, 2009 personal communication). It is unknown whether this species has invaded areas other than North America but Wetterer and Radchenko (2011) map locations in Greenland, India, Kashmir and Turkmenistan. They label these locations as unconfirmed and 'highly questionable'. There have also been erroneous locations previously recorded including one in Japan (Wetterer and Radchenko 2011, Czechowski et al. 2000).

This research focused on the ants' preferred urban habitat and how humans have contributed to the spread and success of the species.

1.2 Objectives

Myrmica rubra has been studied in its native ecosystems for decades but the investigations into its abundance and behaviour in introduced habitats is relatively new. In particular there are few focused studies in the urban/suburban environment. In Canada, at the time of the present study, there was almost no evidence of research into this species in any context and was therefore an important factor in determining the approach to the study. It was felt that information was needed about the species before manipulation of conditions for further scientific understanding and for management purposes could occur. Therefore the direction of the study was primarily observational. This involved various methodologies including the field, laboratory and presentations. The stages are broken into preliminary, macro studies, and a micro study.

The research question for this study was: "**What are the ecological parameters of variables that determine habitat choice for *Myrmica rubra* in an urban environment?**".

The objectives that supported the research question were divided into two categories: a scientific and sociological. Knowledge of this species as an invasive is still in its infancy and mostly focused on management techniques or in relation to more rural environments. This study focused on the urban/suburban so was unique in this respect. The scientific investigation studied the currently unknown ecological parameters by measuring habitat gradients such as the vegetation or substrate (hard or non-organic substances nests are found within or against); atmospheric factors such as light percentage and air temperature; and edaphic factors such as temperature, moisture, percentage of sand, and pH. Secondly, it was important from a sociological point of view. Residents were concerned about the devastating effect this species has had on their quality of life. Part of the study therefore was dedicated to public outreach, in particular, public education in order to attempt to slow down the rate of spread. The use of citizen scientists to observe and report sightings was important in locating infestations and can be used to track the rate of spread over time.

In summary, the objectives for this study were to:

- Determine which, if any, abiotic factors were preferred in habitat choice for *M. rubra* in an urban environment

- Determine the validity of a baiting method which may be used in the future to locate colonies in unknown areas

- Identify and map locations of known and potential infestations in the HRM area, therefore creating a baseline study by which populations can be tracked over time

- Determine methods by which humans have aided the introduction of the species into new areas, altered the landscape to create a hospitable environment and attempted to control the infestations

- Inform and educate the public in order to increase awareness and help reduce the rate of spread

These objectives were achieved by locating nests, measuring abiotic factors; calculating nest densities; measuring densities and engaging residents.

1.3 Identifying characteristics and Life Cycle of *Myrmica rubra*

The genus *Myrmica* is well represented in North America with between seventy five to one hundred different species. Correct identification of *M. rubra* is therefore important in locating colonies for monitoring. There has been a volatile history of this species beginning with Linneus referring to the species as *Formica rubra* in 1758. Santschi (1931) suggested this species was synonymous with *M. ruginodis* or *M. laevinodis* used by Nylander in 1867, as both of these species also have a powerful sting. *Myrmica rubra* has had a succession of synonyms including *M. laevinodis bruesi* Weber and *M. rubra r. champlaini* Forel among many others (Groden et. al 2005, Wetterer and Radchenko 2011).

Myrmica rubra workers have a body length of 3.5-5mm with a pair of propodeal spines. The antennae have twelve segments and an indistinct club. The base of the scape is not sharply angled as is found in native myrmicines. The angled clypeus is also well developed. Worker caste colour is usually a reddish brown but sometimes described as a yellowish brown (Collingwood 1979, Klotz 2008). Queens are similar in colour but the body is larger with a length of 5.5-7mm. Males are generally darker with lighter appendages but can have variation into near black. Their body length is 4.5-5.5mm (Collingwood 1979, Klotz et al. 2008).

There are several identifying features that help distinguish the subfamily Formicinae and Myrmicinae. The presence of a post petiole in myrmicinae is a distinctive characteristic differentiating it from formicines which have only a single petiole. Formicines also lack a stinger and instead spray formic acid from an acidopore. Further identifying features can be found in Klotz et al. (2008) and Fisher and Cover (2007).

Figures 1.1 - 1.3 show photographs of various angles of specimens found at York Redoubt, Halifax. These images are reproduced with the permission of of P.S. Ward and A. Nobile and were found on the website 'Antweb.org'.[1]

1 http://www.antweb.org/description.do?rank=species&genus=Myrmica&name=rubra&project

Figure 1.1: *M. rubra worker frontal view (Photo A. Nobile)*

Figure 1.2: *M. rubra worker side view (Photo A. Nobile)*

Figure 1.3: *M. rubra worker dorsal view (Photo A. Nobile)*

Myrmica rubra is highly polydomous and polygynous, meaning there are many nests of the same colony and there are many queens in each nest. *Myrmica* ant colonies grow logistically following the equation:

$$W_{t+1} = R * W_t / (1 + \Theta W_t)$$

W = number of workers in year *t*

R = rate of natural increase

Θ = the restriction on unlimited growth

with *M. rubra* forming the largest colonies; generally about 1000 workers (Brian, 1965; Elmes 1973; Hochberg et al. 1994; Elmes et al. 1998). In their native habitats the nuptial flights of virtually all *Myrmica* populations occur between mid-August and mid-September regardless of their geographical location. The only exception appears to be species living in the tundra of the far north (Alaska), which complete their cycle some weeks earlier (Elmes et al. 1998). Colonies of *M. rubra* have not been observed conducting nuptial flights in the invasive range. It is thought that the mode of expansion is through 'budding'. This occurs when the density of ants in a nest gets high enough to need more space or better resources. The method is by the movement of a queen or queens leaving the primary nest and starting up a new nest nearby with a selection of workers. There may be a winged dispersal method as winged specimens have been collected in Halifax, but no flying alates have been found in Nova Scotia to date; although, an observation by Dr. Barry Hicks in Carbonear, Newfoundland found one population of flying males (Hicks 2011). He observed the swarms flying at a height of 6-9 metres above the ground. This is consistent with native flights in England where the swarms fly to equally high places such as near the tops of tors (Groden 2011, personal communication). A similar phenomenon has been observed for the Argentine ant in citrus groves in California where Markin (1970) suggested that nuptial fights for this invasive species were also rare and heavily male biased, and new gynes frequently mate and remain within their nest. (Groden et al. 2005).

There are several generations during the yearly life cycle. The first generation is composed of sterile workers who take care of the queen and brood. During the summer two further broods may be laid down with a final brood produced in late Summer/early autumn. This brood overwinters as larvae in diapause, triggered by short day photoperiods and is ready to pupate in spring (Kipyatkov 2001). In Halifax the first emergence of active workers from their wintering nests in 2010 was on April 4th. In 2011, the emergence was one week earlier on March 31st.

Food sources are varied as the species is omnivorous. As colonization of satellite nests is high in June the workers seek sugar based carbohydrates such as honeydew. In July the workers seek increased protein as the brood develop then return to high sugar levels in the autumn. In its native range *M. rubra* is known to feed elaiosomes to larvae and are therefore myrmecochorous (Fisher et al. 2005).

1.4 *Myrmica* species- Global Locations

Myrmica rubra, part of the family Formicidae, sub family Myrmicinae (Bolton 2003), is a Palearctic north temperate ant species with a native geographic range that extends from Ireland and the UK, through northern Europe to western Siberia (Czechowski et al. 2000; Groden et al. 2005). In Europe, its range covers > 25°of latitude (Elmes et al. 1999) where populations can be found from as far north as the Arctic Circle and south to the Black Sea (Agosti and Collingwood 1987, Elmes et al. 1999, Groden et al. 2005). It is an adaptable species with a wide climatic tolerance that varies from extreme oceanic in the west, to continental in the east (Elmes et al. 1999). An occurrence of *M. rubra* had been reported in Japan in the 1980s (Onoyama 1989), but this report is thought to be a misidentification (Czechowski et al. 2000; Groden et al. 2005). In fact there are conflicting views on the areas this species has been found. A search on several Internet sites revealed a multitude of locations and omissions. Clearly, there is scattered information in the popular press. The newest definitive source of the world wide distribution of *M. rubra* was published in Jan 2011 by Wetterer and Radchenco. They compiled data from over 2000 sites from sources both published and unpublished. They have traced the earliest reports of the species in 71 geographic areas including countries, major islands, US states, Canadian provinces and Russian Federal Districts. They have reported three new areas with no previously published records including Prince Edward Island, Washington State and the Far Eastern Federal District of Russia. They include several tables explaining the distribution in the native areas and the newer distribution in the invasive range (Wetterer and Radchenko 2011).

New areas have recently been confirmed in other parts of Canada including British Columbia (Greater Vancouver, Burnaby) (Higgins 2011, personal communication) and Newfoundland (Saint John's, Corner Brook and Carbonear) (Hicks 2011). Several new locations in Nova Scotia and New Brunswick have been reported (to Horton, via email 2010 and 2011). Many locations are yet to be confirmed but all indications are that this problem is more widespread than previously thought.

In Maine, most infestations seem to be situated within 10 km of the coast with one colony inland about 70km (Groden et al. 2005). However, it was found that the property owners of the inland infestation had purchased potted plant material from a nursery in an infested coastal area (Groden et al. 2005). This coastal affinity appears to be confirmed in a report given to the USDA Research Forum on Invasive Species in 2009 where Lund et. al. (2009) attempted to further identify the locations of infestations of pest species of ants in Maine and New York. They mailed out 500 requests for samples to residents and businesses. Of the ninety samples returned in Maine, sixty seven percent were from coastal communities. This is inconclusive however, as the report does not clarify which of the two most frequent species of pestiferous ant, *Formica exsectoides* and *Myrmica rubra,* was found in which areas.

Groden et al. (2005) also found infestations ranging into New Brunswick, although there have been no reports in the public domain of this as a problem area in Canada. There are historical references to populations in Quebec and Ontario, with Ontario suffering an urban infestation similar to Halifax. The problem had become great enough in the Richmond Hill area, outside of Toronto, ON., that a management plan was created in 2006 by the Parks Recreation and Culture Department, Natural Heritage Section. Aside from the basic plan, which is modelled on the University of Maine factsheet [2], no further publications have been found. The amount of public complaints prompted the Town Council to release an information sheet to the public[3]. That factsheet is based on the University of Maine information. The urban reports have continued as *M. rubra* were confirmed in private yards in Cambridge, Massachusetts (greater Boston) during the summer of 2010.[4] Cambridge is an urban area and will undoubtedly have increasing interactions between the pest and humans.

1.5 Invasive Species

Invasive species are becoming a problem in ecosystems globally. Wilson (1992), said of invasives that the introduction of exotic species was second only to habitat destruction in its contribution to declines in native species abundance. Areas in the world where trade

2 http://www.umext.maine.edu/onlinepubs/htmpubs/2550.htm
3 http://www.richmondhill.ca/documents/prc_fireant_brochure.pdf
4 http://www.pctonline.com/European-fire-ant-Cambridge.aspx

has occurred over long periods of history are now realizing the effects of invasive species on the environment and economy.

1.5.1 Characteristics of Invasives

Invasive species are non-native species, aliens, which have been introduced to a new environment and have become detrimental to that environment. Not all introduced species however are invasive. There is much confusion in the terms used in invasive biology. Colautti and MacIsaac (2004) define the term invasives not in a single definition but in terms of stages and effects at each stage. These stages encompass detrimental effects to ecosystems and humans. They list no fewer than thirty two terms associated with invasive species and the stages they most closely resemble. They also take into consideration the mode of transport of these species and whether they are indigenous to an area or introduced. It should also be considered that often invasives have a detrimental effect on the economic health of an area particularly if the invasive species affects a major resource such as agriculture, fishing or forestry.

The likelihood of invasion is normally conceptualised as being determined by three traits: a) the number of attempts/introductions, b) the ability of first immigrants to find food, shelter and mates so they can survive and reproduce, and c) the potential of the initial population to grow and spread (Elton 1958; Usher 1986; di Castri 1989; Ebenhard 1991; Moller 1996). In all invasive biology it is worth considering the effect of the Allee Effect. This is defined as a positive relationship between any component of individual fitness and either numbers or densities of conspecifics (Stephens et al. 1999). It indicates that species in small populations benefit from an increase in density and works at the population level.

1.5.2 Effect of Invasives on Systems

In nature, there are many examples of species which co-exist and are neutral, or are perceived a minor irritation. Some species are considered beneficial and are introduced purposefully. The majority of purposeful introductions are specifically to aid in biological control of other species (native or non-native); those deemed a 'problem' by groups or individuals. For example, introduced species of Asian Lady Beetles are used by the horticulture industry to control aphids (Albright et al. 2006). The problem is that the effects of non-indigenous species may not be limited to the uses we expect (Simberloff and Stiling 1996; Albright et al. 2006).

Some invasive species completely decimate native populations and proliferate to the point where species diversity in an area is severely reduced. Interspecific competitions which have gained notoriety include European rabbits in Australia, which displaced the native Bilby (Read et al., 2008), or grey squirrels inhabiting the same niche as the smaller red squirrel (Bertolino et al., 2008). Both of these are examples of species which outcompete the indigenous fauna for habitat and food. 'Rules of Assembly' or 'Ecological Assembly Rules' assume that interspecific competition is greatest between species that are most similar in morphology and function (and thus resource use), and as a consequence, patterns of species co-occurrence, such as the invader being the victor for resources, are manifested (Ward and Beggs, 2007). In some instances the introduced species, even a domesticated species, causes the extinction of a native species, as was the case of the Dodo (Janoo, 2005). Janoo (2005) discusses the discovery of Dodo bones in highland areas not native to the bird. This suggests the extinction was caused by not only by the many introduced domesticated and wild animal species but included the extremely invasive *Homo sapiens sapiens*.

Invasives are found in the three main types of environments: terrestrial, aquatic and marine (with transitional environments bordering each. e.g. wetlands, riparian zones, estuaries). There is now strong evidence of large numbers of invasives along the east coast of the United States and Canada in the Gulf of Maine, the body of water on the eastern seaboard of North America (surrounded by Nova Scotia, New Brunswick, Maine, New Hampshire and Massachusetts) (Dijkstra, 2007). The most numerous of these aquatic invasive species (AIS) include crustaceans, bryozoans, algae and ascidians. These species have seriously affected marine transport, recreational boat users and the fishing industry, where they entangle fishing gear and foul boats. It took many years for the effects of invasives to be recognised by authorities and are now beginning to be considered a formidable threat.

In both Canada and the United States, government websites[5] provide information on policies relating to the movement, prevention and management of a selection of these invaders. Factsheets suggesting guidelines for reducing introductions are becoming more readily available for the public and commercial organizations. Despite this, there is very

5 http://www.ec.gc.ca/eee-ias/default.asp?lang=En&n=02101A38-1#ws02A8C8D1

little legislation currently in place that regulate this problem as a whole. In 2004 the Government of Canada and its Provincial/Territorial counterparts introduced *An Invasive Alien Species Strategy for Canada.* Previous to this, invasive alien species fell under a suite of at least fifteen existing federal legislation Acts. These are all now part of the Invasive Alien Species Partnership Program.[5]

In terrestrial ecosystems, the focus of invasive species is primarily aimed at the agricultural and forestry industries. Research money is more easily accessible if a direct link with one of these areas can be proven. In countries such as Canada, which relies heavily on a resource based economy, the introduction of a harmful species can cost billions.[5]

In the International Year of Biodiversity, 2010, many initiatives were launched to bring the various aspects of conservation to the global stage. Invasive alien species have, until now, been fairly low priority unless there has been a direct correlation to economic health in a major industry. This is now a concern being highlighted by conservationists around the globe as a major threat to biodiversity and in turn to ecosystem health. A report by McGeoch et al. (2010) investigates invasive alien species (IAS) and coordinated efforts to quantify patterns in the extent of biological invasion, its impact on biodiversity and policy responses. They declare that for this indicator of threat, the 2010 Biodiversity Target has thus not been achieved. The results nonetheless provide clear direction for bridging the current divide between information available on IAS and information needed for policy and management for the prevention and control of IAS. It further highlights the need for measures to ensure that policy is effectively implemented, such that it translates into reduced IAS pressure and impact on biodiversity beyond 2010.

Byers et al. (2002) also report on IAS but focus on directing research that reduce the impacts of non-indigenous species and predict detrimental impacts. They try to encourage new research aimed at the introduced species that are most likely to increase as a result of the increase in international trade of plants and animals. They also consider the possible effects of climate change as an added stressor and believe that management decisions are being made by failing to actively prevent new invasions and control those recently discovered or already established. To accomplish this goal, they outline key questions to help guide researchers and managers in their decisions on how best to spend time and money. In spite of these recommendations, in the eight years since publication, climate change has not, as of yet, been investigated in terms of its synergistic effect with introduced species at a national level.

1.5.3 *Myrmica rubra* as an Invasive Species

Research into the invasive form of *M. rubra* is still in its infancy compared to work conducted in Europe over the past 60 years where *Myrmica* is one of the most intensively studied of all ant genera with *M. rubra* especially so in the UK (Bourke and Franks 1995, Evans et al. 2010). Until 2010, the literature available regarding the invasiveness and history of *M. rubra* has been limited. Dr. E. Groden, University of Maine, recently collaborated on an article outlining work related to new fungal pathogens (Evans et. al. 2010) and their effect on *M. rubra*. The article explains how fungi were identified as a possible control mechanism in the indigenous version in England. However, Evans et al. (2010) report that the impact and dynamics of these fungal pathogens on *M. rubra* populations, and their relevance to invasion biology, still remain unknown.

To date there are few publications on this species as an urban invasive. The limited references refer mostly to colony locations, rather than study of the insect itself, as in the case of Wetterer and Radchenko (2011) and Lund et al. (2009). Almost all research conducted from or associated with the University of Maine has centred around rural areas such as Acadia National Park and the native habitats of England (Evans et al. 2010). Many investigations are also focused on management experiments in the laboratory, not urban environments where the human-ant interactions occur on a regular basis. It may be that the invasiveness of the species is different in an unnatural, anthropogenic setting than that of a rural environment. This idea has been hinted upon by Wheeler (1908. page 339) when he observed *M. rubra* in Massachusetts (incorrectly described as as *Myrmica laevinodis*). He wrote, "It is very fond of attending aphids and, unlike our timid native *Myrmica spp* which live in the retirement of woods, bogs, heaths and waste places generally, it prefers to nest in cultivated soil. Hence it may become a nuisance in lawns and dooryards, like the fire-ant (*Solenopsis geminata*) of the Southern States.".

1.5.4 Insect invasives

Social insects are among the most successful of animal invaders (Holway et al. 2002; Groden et al. 2005). *Apis mellifera scutellata* (African Honey Bee/Killer Bee);

Coptotermes formosanus (Formosan termite); *Paravespula germanica* (German Yellowjacket Wasp); and several ant species have have reached high population densities over broad geographic areas (Groden et al. 2005). In North America there are various well established insect invaders including *Linepithema humile* (Argentine Ant) and *Solenopsis invicta* (red imported fire ant or RIFA) (Gutrich et al. 2007). *Solenopsis invicta* is a native of South America and has successfully infested vast areas of the USA causing ecological and economic damage and detrimental effects to human health (Gutrich et al. 2007). In Louisiana it was found that this species had infiltrated all ecosystems except forested areas with 100% complete closed canopy (LaFleur et al. 2005). Hawaii is particularly concerned about the introduced *S. invicta*. Over 70% of endemic land bird and snail species were lost before its introduction (Steadman 1995; Loope 1998); the impacts of invasive species contributed greatly to these past declines and are now the predominant cause of biodiversity loss in Hawaii (Loope 1998; Gutrich et al. 2007).

Although *S. invicta* is presently confined to the United States, many areas in Canada must be observant. This species prefer open, disturbed habitat such as pastures, lawns, and roadways, but are generally capable of colonizing any area with temperatures greater than -12°C, about 25–50 cm of rainfall annually or other sources of water, and at least some exposure to sunlight (Wojcik 1983; Vinson 1997; Korzukhin et al., 2001; Gutrich et al. 2007), all of which can be found in coastal zones.

Most insects are solitary animals living to satisfy their own needs at a basic evolutionary level. Very few insect species communicate beyond the necessary fighting or mating: parental care is rare. Ants, along with bees, wasps, and sawflies belong to the order

Hymenoptera. Only members of this order and Isoptera (termites) are widely considered eusocial (social insects). Sociality occurs when overlapping generations of parents and offspring coexist in an integrated unit or colony and the population form a containment or nest (Elzinga 2004). Eusocial insects have a highly developed caste system where there is a division of labour and reproduction is not carried out by all members (Wilson 1971). Evolutionarily speaking, Hymenoptera are the most advanced insect group (Elzinga 2004). Wilson (1985) considers sociality to be one of the landmark events in the evolution of insects. Many thousands of arthropod species have become associated with social insects and have taken opportunities to either mimic, parasitize or become mutualistic (Elzinga 2004). In many ant species there are close relationships with Hemiptera (true bugs such as aphids). The ants provide the trophobionts with protection and in return they are "milked" by their hosts for their sugar solutions, a honeydew kept in their hind gut and released upon tactile stimuli (Dixon 1985, Agosti et al. 2000). *Myrmica rubra* exhibits these traits, but unlike some other ant species has not been associated with a particular hemiptera species; contrary to *S. invicta* (Helms and Vinson 2003, Groden et al. 2005).

Partial reasoning for the success of ants as invasives may be the colony structure itself. Many invasive ants, as well as some ants in their native habitat, have a unicolonial structure. This is often defined as the breakdown of colony boundaries and the reduction of intraspecific competition (Garnas et al. 2007). These are successful invaders partially because they do not need to expend energy in defending territory against other nests or colonies of the same species: energy is put into the common good, i.e. greater numbers of ants (Helantera et al. 2009). They are therefore less, if at all aggressive towards neighbouring nests. Unicolonial ants are genetically related, especially in species that have a single queen where the relatedness of workers is 75%. In polygynous ants this is less so, especially in polyandrous species. In many unicolonial or supercolonies of ants, multiple queens does not mean automatic aggressiveness despite fewer genetic ties. However, although *M. rubra* exhibits traits of a unicolonial species and has often been assumed to be this (van der Hammen et al. 2002), Garnas et al. (2007), using aggression assays, found the Maine populations of *M. rubra* to be multicolonial. Aggression at 10m intervals between colony areas had a high level of aggressiveness then increased as the distance between the nest site increased.

1.6 Habitat

Myrmica rubra appears to be an adaptable species using a variety of microhabitats for nesting. Groden et al. (2005) found that nests were found most frequently under or within downed woody debris or in the leaf litter where they place their brood in a curled leaf.

Within their native range, *M. rubra* has been reported to exploit a variety of habitats and to use different nesting substrates in different parts of its range. In a study by Brian (1956), all nests in the study area of Scotland were found on the south facing slopes of the sandstone hills between the River Clyde and Loch Lomond. In England, they are found most commonly under stones along pasture edges (Elmes 1975, Elmes and Wardlow 1981; Groden et al. 2005), but along the Black Sea, they primarily nest in rotten wood in wet, shady woodlands, and in Russia in moss tussocks in open pine forests (Groden et al. 2005). In Maine, *M. rubra* seems to not only exploit multiple nesting microhabitats but also multiple ecosystems. Infestations were found in lawns and gardens, old field habitats, scrub-shrub, wetlands, and deciduous forests but not within the dense spruce coniferous forest. This supports the findings of Brian (1956) in regards to the decrease in nests due to the closing of the canopy. They have also been observed nesting and foraging within the coastal zone at the edge of the spruce forest into the intertidal zone in parts of Mount Desert Island, Maine (Groden et al. 2005).

1.7 Citizen Science

"Citizen Science" is a term used to describe non-scientists working in the field of science, particularly in the environment, and generally for the purpose of data collection. Individuals or groups carry out activities such as observation, monitoring (i.e. taking samples- measuring and recording), and computation. History has many examples of citizens contributing a great deal to scientific knowledge, and with modern day technology, ease of sharing of information is taking science from the auspices of the select few to the masses. The future may see a much greater involvement by the public as governments cut science programmes and community groups fill the voids. In Nova Scotia there are many community monitoring groups taking stewardship of watersheds and other initiatives.

It is becoming more obvious that citizen science has a role to play in scientific data collection. Balmford et al. (2003) state that monitoring long-term trends in the abundance and distribution of species is important in conservation, and as a way of tracking anthropogenic impacts. The United Nations Environment Programme stresses public participation as an essential component of sustainability and recognizes that public participation has become an increasingly important part of environmental management (Au et al., 2000). In Nova Scotia the emphasis on citizen participation seems to be through the various community based ecological monitoring groups. Monitoring in North America is particularly prolific, especially in relation to the water quality of lakes and rivers (Griffin, 1999; Savan et al. 2003).

The use of citizens in gathering data in resource-poor countries can be a way to create a synergism between research and education. In an article by Braschler et al. (2010), a case study is presented on how participation in ant monitoring in South Africa can enhance the scholars' interest in science, introduce them to new career paths and mesh information on biodiversity with the collection of data. In this case study ants were chosen as a tool to monitor biodiversity because ants are diverse enough to enable meaningful studies are not usually so diverse that their study is impracticable (Agosti et al. 2000, Braschler et al. 2010).

In a study on ants in an urban environment, citizens can aid simply by observing ant behaviour or assisting in nest finding. In this study citizen scientists were also asked to locate nests in public areas and report to the Nova Scotia Museum of Natural History or HRM directly.

1.8 GIS Applications in Ecology

In the past decade, the uses of Geographic Information Systems (GIS) have soared. The applications are now being utilized in disciplines not traditionally associated directly with geography. One area which is benefiting enormously is biology, or more specifically ecology. Often, ecological studies have spatial data that need organizing and analysis, where there are multiple variables to consider and patterns to identify. This is now easier thanks to the visual aids of maps and the use of geodatabases. Resource managers are using GIS to map habitats and locate species, especially those in critical ecosystems.

One of the newer areas natural resource managers are now investigating is that of the control of non-indigenous, and in particular, invasive species. Considering Wilson (1992) identified this threat nearly 20 years ago, it has become imperative to find measures of control and where possible, eradication. GIS maps are being created to detect, predict and manage the spread of aquatic, marine and terrestrial species. Manrique et al. (2008) used maps to predict the generational development of the invasive Brazilian peppertree (*Schinus terebinthifolius Raddi*), a species which has established itself throughout central and southern Florida. The objectives of this study were to determine development rate and survival of *E. utilis*, a potential biocontrol agent of this invasive species, at seven constant temperatures and generate prediction maps of the number of generations per year this species may exhibit. From these results the next stage would be to implement a management plan and monitor using a similar mapping technique. The project is still in production but may set a precedent for future users.

Prediction is a powerful tool when applied to the management of (detrimentally) abundant or conversely, endangered species. There are many ecological analyses tools used to determine species diversity and ecosystem structure. Unfortunately, not all of these methods are as accurate as a natural resource manager would need. Menke et al. (2009) investigated the use of species distribution models (SDMs) or, more specifically, ecological niche models (ENMs) to characterize and predict the distribution of the invasive Argentinian Ant *(Linepithema humile)*, in California. These models are a useful and rapidly proliferating tool in ecology and global change biology. ENMs are used to understand associations between a species and its environment; to draw biological inferences in order to predict potential occurrences in unoccupied regions and forecast future distributions under environmental change (Menke et al. 2009). To sample the full habitat and climate heterogeneity of the southern region, sampling points were allocated to different land-cover types. This was based on their spatial extent and presumed probability of *L. humile* occurrence given pitfall trap results from the northern region. They used Hawth's analysis tools for GIS to randomly place sampling points in each vegetation category based on the proportion of the land-cover type occupied by that category. To determine whether or not Argentine ants were present at sites in the southern region, they used visual surveys and bait transects. Models were analysed on the multiple variables and maps generated.

China is also facing a problem with invasive ants. The South American species *Solenopsis invicta,* or red imported fire ant (RIFA), appears to be spreading at an alarming rate. This species, already a nuisance species in most parts of the United States, is difficult to control, and has proven nearly impossible to eradicate. Now, GIS is being used to help identify areas which could potentially be suitable for colonization. The aims of this study were threefold: to see how the weather conditions in the Jiangsu Province meet the requirements for development and colonization of RIFA; to evaluate the suitability of the province for RIFA; and to provide information for making quarantine decisions and adopting technical measures. An isotherm map, indicated that several regions of Jiangsu Province (Changzhou, Liyang, Wuxian Dongshan, Nanjing and Lvsi) are at high risk of *S. invicta* infestation, especially from late July and early August. Quarantine officials are using this information to be vigilant for any accidental introductions of this pest in the susceptible regions and time (Xiong et al. 2007).

There are many examples of the spread of alien species globally, and researchers are combining tradition with technology to combat this threat. Costs associated with non-native species in the United States is estimated at over $120 Billion/year alone (Pimentel *et al.* 2005). Graham et al. (2007) have developed a web-based system, called Global Organism Detection and Monitoring (GODM) to provide real-time data from a broad spectrum of users on the distribution and abundance of non-native species. Information includes attributes of their habitats for predictive spatial modelling of current and potential distributions.

2 Study Area – Halifax Regional Municipality

2.1 Location

Halifax, the capital city of Nova Scotia, is located half way between the equator and the North Pole on the eastern seaboard of North America. The province of Nova Scotia is divided into several counties with the newly amalgamated Halifax Regional Municipality found along the south to eastern shores of Nova Scotia (Figure 2.1).

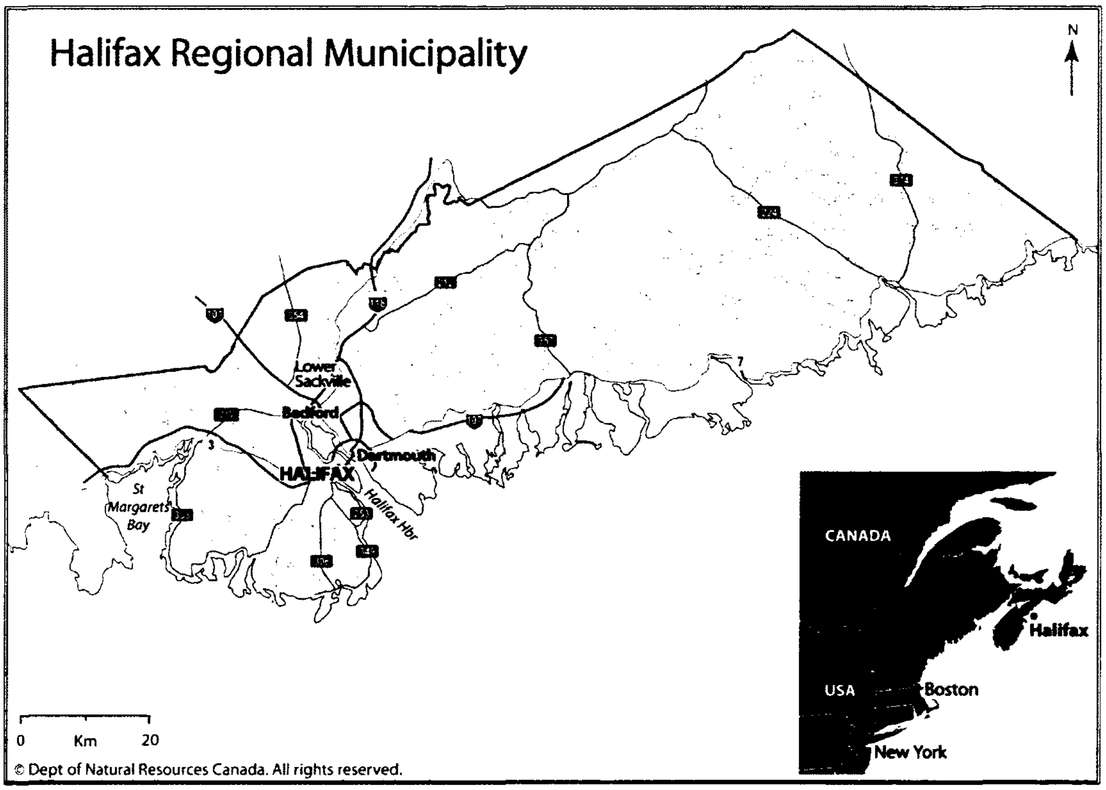

***Figure 2.1**: Location of Halifax, Nova Scotia (insert) and the Halifax Regional Municipality (HRM). (Courtesy of Will Flanagan. Saint Mary's University 2011)*

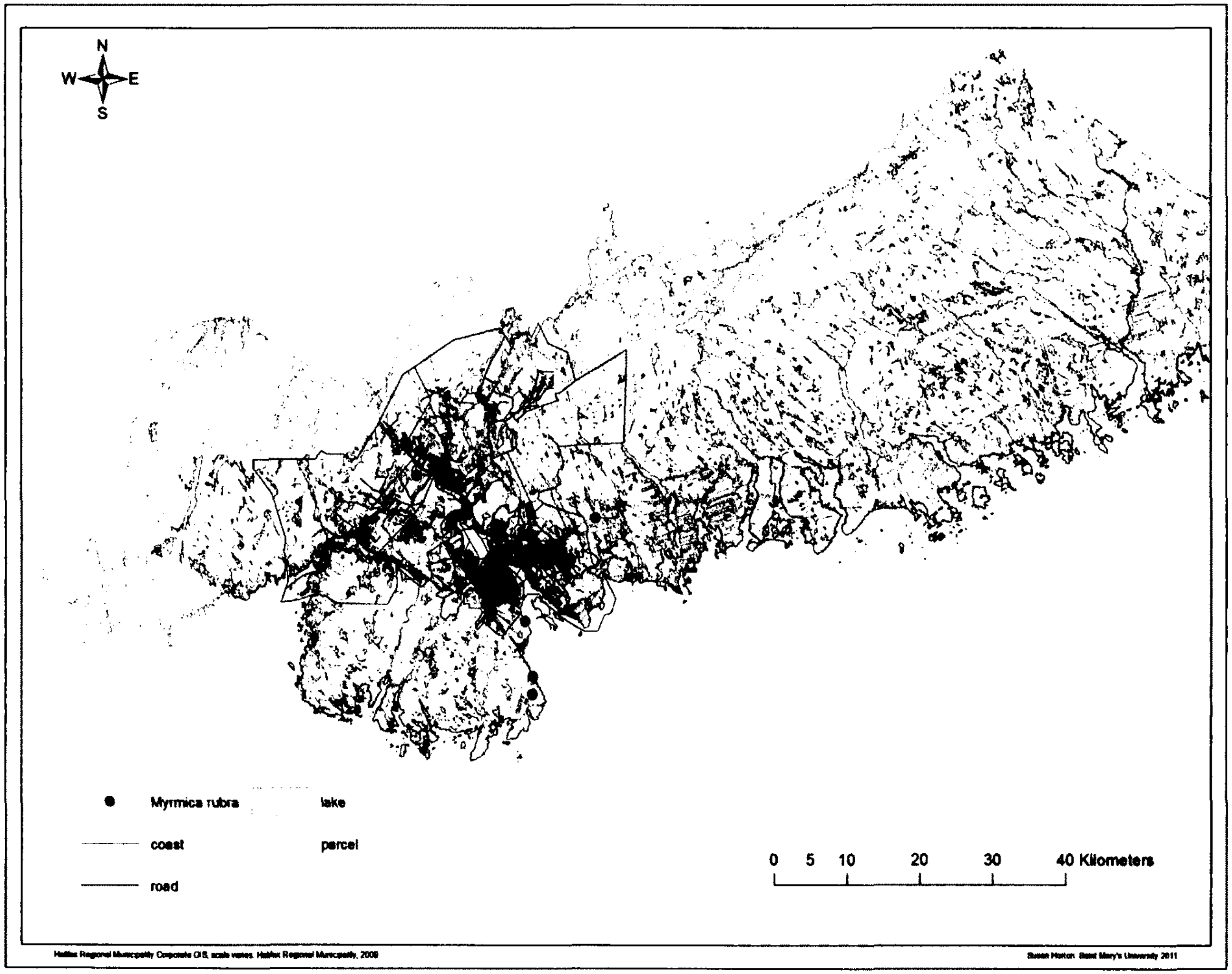

Figure 2.2: *Halifax Regional Municipality showing geocoded locations of reported Myrmica rubra locations. (Source: Author)*

The location of the European Fire Ants are concentrated in the urban core of the Halifax peninsula with reports lessening as the distance from the centre increases. The larger study, or macro-study, entailed recording the reported locations and conducting a property survey identifying nests sites. Figure 2.2 shows the geocoded reported infestations in the HRM area.

A smaller study, or micro-study, was also conducted in Halifax on an infested, rectangular city block (refer Chapter 3 for details). This block was considered a 'typical' city block for the peninsula and was situated in an area where many other similar blocks are also infested (Figure 3.10).

2.2 History

Halifax was established in 1749 as a British garrison. The Halifax Regional Municipality came into existence in 1996 by combining the cities of Halifax, Dartmouth, the Town of Bedford and the County of Halifax. This means the regional boundary includes a variety of urban, suburban and rural environments. The population of the 200 communities which comprise HRM, is around 400 000 (Halifax Regional Municipality 2011). The highest population density is centred around the downtown cores of the old cities of Halifax and Dartmouth. The majority of the municipal government is found on the Halifax peninsula, the primary focus area of the study. This area has a highly developed urban core and mature neighbourhoods. Due to the concentration of trade on the shores of the peninsula and evidence of other species being introduced throughout history (e.g. heather, Japanese knotweed and brown spruce longhorn beetle in Point Pleasant Park on the south end of the peninsula), it is possible that *M. rubra* may have been introduced in this areas initially as well.

2.3 Geography

Halifax has a large deep harbour, carved by rivers, enlarged by glacial action and finished by geologically recent relative sea-level rise. Metropolitan Halifax is located along a rugged, low relief fjord coastline, created as post-glacial sea-level rise flooded glacially modified river valleys. [6]

The Atlantic coast is highly irregular, with numerous embayments such as Bedford Basin in Halifax Harbour. The coast has many small bedrock islands, which reflect the complex bathymetry of the inner shelf. The rivers are incised into Paleozoic rocks of the Meguma Terrane. These rocks form a broad, upland region with a relatively flat surface which slopes from about 300m inland to sea level on the coast.

There are two types of glacial deposits and landforms in the Halifax area: drumlins and stony till plain with the rest of the area being made up of bedrock.[6]

Figure 2.3 shows a map of the surficial geology of the metro area.[7] The areas most studied during this research are comprised of either Till Veneer from Beaver River Till or

6 http://www.gov.ns.ca/natr/meb/field/stop1.asp
7 http://www.gov.ns.ca/natr/meb/data/mg/ofm/pdf/ofm_2011-001_d447_dp.pdf

bedrock. The Beaver River Till is a diamicton with sandy matrix and locally derived clasts. Sediments were deposited by ice and derived from subglacial erosion and have an average thickness estimated 0.5–5 m with some exposed bedrock and some areas measuring a locally derived till with a thickness of >5m. In Figure 2.3 the Beaver Till comprising most of the peninsula and downtown Dartmouth, can be seen in green; the mainland shows the dominance of bedrock in pink.

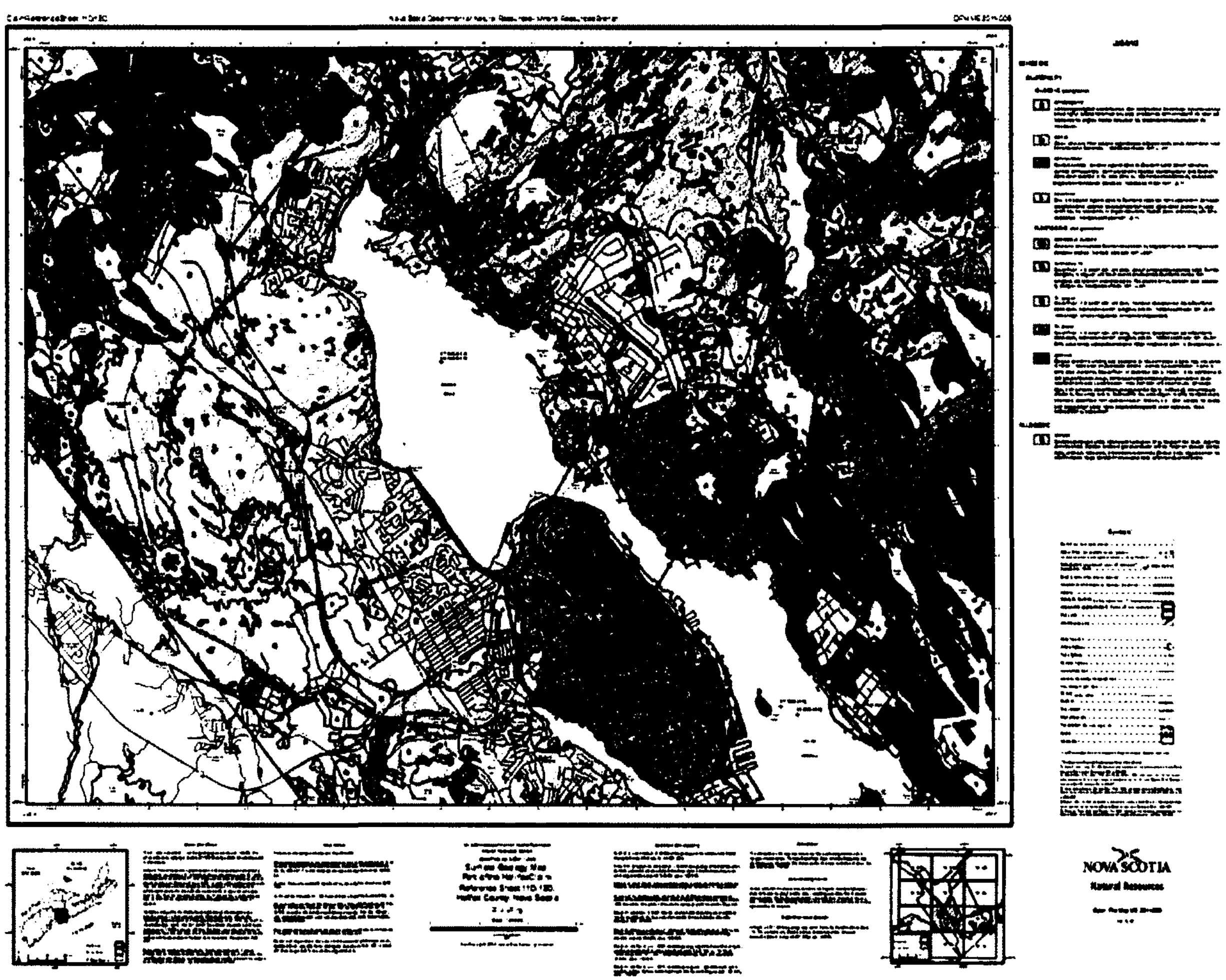

Figure 2.3: *Surficial geology of the Halifax, Dartmouth, Bedford area.*
(Source: Nova Scotia Department of Natural Resources. Open File Map 2011-009). Author: Utting 2011

2.4 Ecology

The terrestrial environment consists mostly of mixed deciduous and coniferous forests. Forested areas around the Gulf of Maine make up some 237,600 square kilometres and cover about 75% of land. According to the World Wildlife Fund and National Geographic ecological boundaries, Nova Scotia falls under the eco-region of the New England-Acadian forests. This region is described as a transition zone between the boreal spruce-fir forest to the north and the deciduous forest to the south[8]. The forest shown in Figure 2.1 is similar to the invasive habitat of *M. rubra* in Acadian National Park in Maine.

The climate of Nova Scotia is characterized by warm, moist summers and cold, snowy winters. Maritime air masses moderate the extremes and bring precipitation to the area year round. This precipitation is evenly distributed in the primary forms of either rain or snow. Climatic averages for the HRM area, taken at Halifax Stanfield International Airport over the period of 1971-2000[9] show an average precipitation of 1452.2mm and an average temperature of 6.3°C. This temperate environment is similar to the climatic conditions found in the native ranges of the *M. rubra* (Czechowski et al. 2000, Groden et al., 2005).

8 http://www.nationalgeographic.com/wildworld/profiles/terrestrial/na/na0410.html
9 http://climate.weatheroffice.gc.ca/climate_normals/results_e.html?stnID=6358&autofwd=1

Figure 2.4: *Acadian mixed forest in eastern HRM. (Source: Author)*

2.5 Urban Habitats

The surficial geology is undoubtedly the greatest influence on the composition of the soil but the majority of the studied areas are urban and therefore have been manipulated to suit the individual's needs; this in turn influences the composition of biota. The majority of properties visited were primarily grass lawns with varying degrees of trees, ornamental plants or vegetable gardens interspersed throughout. There was a limited variety of trees with maples being the dominant species on the peninsula and oak, beech, birch, chestnut, ash, yew and some alder also being found: very few coniferous species were in evidence. Bedding plants were varied but some species were favoured such as hostas, ornamental grasses, ground cover plants and rhododendrons. Figure 2.5 shows a typical peninsula property with ornamental plants and invasive plants (e.g. gout weed, hawk weed). The flags show locations of ground flora nests. There are also poison (boric acid) bait stations placed throughout the lawn (these were not directly part of the study).

***Figure 2.5**: Halifax peninsula property showing locations of M. rubra nests (flagged) and feeding stations. (Source: Author)*

The majority of properties had regular additions of soil improvers in the planting areas (fertilizers, mulch etc.) although very little was done to change the condition of the lawns except moving. Some residents had occasionally put lime down. Many residents used some form of chemical insecticide.

The properties found in the suburban/rural boundaries had distinctive areas of manicured lawns with defined edges leading into native dense scrub-shrub and forest. These areas were often impenetrable and therefore were not able to be studied. The infestations were in the managed areas of the property.

3 Study Design

3.1 Introduction

The investigation was broken into three stages: the pre-collection of habitat data known as the 'Preliminary study'; the collection of data of habitat variables know as the 'Macro-study'; and the more focused ant density count known as the 'Micro-study'.

3.1.1 Preliminary study

The preliminary study focused primarily on the recruitment of the residents of HRM who believed they had an infestation. As part of public service, educational presentations were given on how ants may be inadvertently spread by movement of infested soil or organic material. As this research involved working with people, Research Ethics Board approval was sought (File #10-104). Once approval was granted potential participants were contacted and invited to participate in the study.

Interested residents were given information and subsequently, if they wanted to fully participate, then a more detailed arrangement was made for a visit to the property to conduct the physical survey. In order to help reduce bias, the residents were contacted on a first come, first serve basis. If nests were found and the species confirmed, then a macro study would take place. The potential participants were given the questionnaire (Appendix A) and information about the study including all Ethics Board forms.

The purpose of the questionnaire was to help determine a time line or history of the species in the area, trace possible methods of infestation (e.g. transport of infected plant material or soil), and gather information on the use of pest control.

Another important factor in public education was the use of the media. The attendance at presentations was limited but the coverage of the problem by the media was more extensive. Over the course of the summer of 2010 several interviews took place in a variety of forms (TV, radio and newspaper). The media was focused on showing the impact this species has on the lives of local residents and to ask questions on how to manage them. As this study does not include management techniques, a link to the university of Maine[10] was given. Figure 3.1 shows choice chambers where biological controls are being tested.

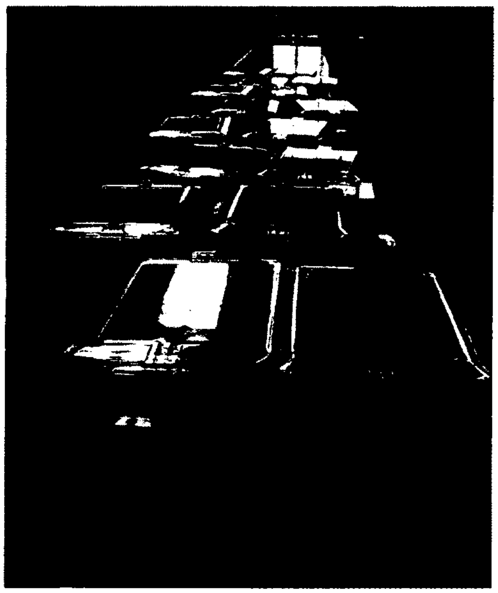

***Figure 3.1**: University of Maine research lab testing biological control agents. (Source:author)*

10 http://sbe.umaine.edu/fireant/

3.2 Field work

The scientific portion of the research was the collection and analysis of data gathered in the field. The focus of this work was to measure nest characteristics to determine the preferred habitat. There were also a lot of observational notes made on aspects of ant behaviour and habitat descriptions. The fieldwork was conducted in two parts, the whole property surveys (macro-study) and the use of baited vials for counting ant densities (micro-study).

3.2.1 Macro-study

The purpose of the macro-study was to measure the ecological parameters of nest sites and determine nest density. Each site location was either an individual property based on a civic address or public property. Permission to inspect HRM land was obtained from HRM's Energy and Environment Department (Sustainability and Environment Management Office or SEMO at the time of the study). Several site inspections took place and were mapped, but not all have data associated with them due to lack of available equipment. The statistical analysis of data is based on the surveys with measurements but some maps reflect all inspections and reports. The studies were conducted in 2010 during the months of July and August with two in early September and one preliminary study at the end of May (which was re-sampled in early July and used for comparison for the effects of pesticide applications). All surveys were on dry days, almost uniformly with no cloud cover, and between the hours of 10am-5pm (exception: one between 5-8pm). If rain had occurred prior to the appointment, at least 12 hours was allowed to pass between the end of the rain and the start of the survey to allow for drainage of soil.

Each inspection took on average 2-4 hours but longer if the property was large. All residents were invited to participate in the physical inspection. If the property owner assisted, then confirmation of nest composition was done by the researcher before recording values. A nest was defined as an aggregation of ants where brood and/or reproductives (queens, gynes or males) were present, as was the method used in Groden et al. 2005.

Before beginning the nest search, an air temperature was recorded. A complete systematic ground level search was then performed. The search was conducted in a grid pattern starting with the furthest corner on left hand side of the property boundary (when facing the house). This was deemed the origin and later used as the origin of the micro-study for the purposes of spatial data to be used in GIS maps. The search involved turning over all movable hard substrates such as stones, wood, paving slabs and furniture. The edges of all grass or soil against a hard surface such as a sidewalk or foundation were pulled back and disturbed by hand, trowel and foot. If a nest was present the ants would be agitated and pour out of underground holes. If the nest was under a flat hard substrate then the nest was easily seen against the surface, as the ants nest in the upper layers of soil (Figures 3.2 and 3.3). In all observed cases of a flat substrate, the ants present were considered to be of one nest. If the paving slab (or equivalent) was large, for example 50cm in diameter then more than one nest was possible. Nests would be considered separate if there were no tunnel connections between them. This would involve carefully excavating the visible nest at the edges. In a ground flora nest there were generally one to three entry holes which were found close together (i.e. 3-5 cm).

Figure 3.2: *Nest under paving slab. The white particles are brood, mostly larvae. The workers are reddish brown and can be seen amongst the brood and on the upturned paving slab. (Source:author)*

Figure 3.3: *Shallow nest under a paving slab. Worker ants and queens can be clearly seen.(Source:author)*

If the property had a grass lawn, a walking sweep was conducted and percussion was used. This was achieved by sweeping the ground with a foot or tapping with a trowel. Ground flora nests were confirmed by an excavation of soil on the surface of the grass (Figure 3.4). Ground nests were opened by pulling the vegetation thatch off by hand or by using a trowel (Figure 3.5). Dandelion nests were more cryptic and required a lot of percussion around the base of the stem. If *M. rubra* were present they would pour out in defence, thus determining whether the nest was in use. Occasionally an abandoned nest was found and often there were several active nests close by.

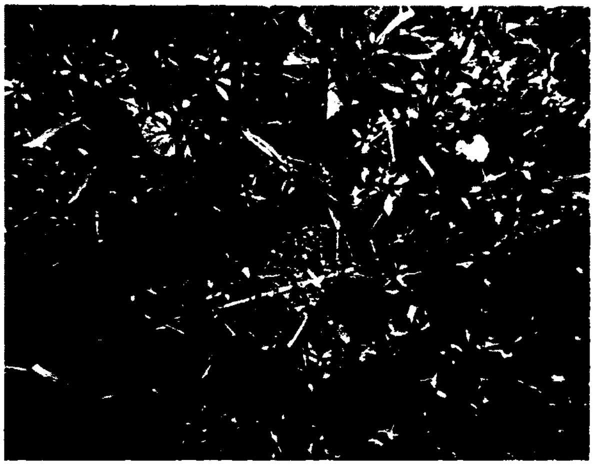

Figure 3.4: Undisturbed ground flora nest. (Source:author)

Figure 3.5: Ground flora nest after disturbance with boot. Workers and brood visible close to soil surface. (Source:author)

If a nest was found in soil under a paving slab, then both the soil and slab were tallied. This allowed areas to be identified where nests are most likely to be found on a property. Nest were categorized as: grass roots (nests in matrix of grass roots); weed roots (nests in matrix of small ground flora roots not grass); tree roots (under or against); shrub roots (under or against); moss (under thatch of moss); soil (in soil of any kind); under or against hard substrates/surfaces such as paving slabs, stones, wood, plastic, or foundations/sidewalks; rotten wood (nests inside decaying wood such as stumps, logs or branches); weed suppressant (under fabric); mulch (specifically mulch used in urban gardens); and compost (in compost bins or piles). Examples can be seen in Figures 3.6 to 3.8. For example of tally sheet for vegetation and substrate please refer to Appendix E.

***Figure 3.6**: Nest sites under logs in soil and beside logs in grass. (Source:author)*

***Figure 3.7**: Nest sites under weed suppressant fabric, in root matrix of ornamental plants and in mulch. (Source:author)*

***Figure 3.8**: Nest sites under hard substrates such as stones and logs. (Source:author)*

A Hydrosense(™) soil moisture probe and Oakton(™) soil temperature probe were then inserted into the nest chambers and measurements taken (Figure 3.9). Not all nests could be probed, if, for example the nest was contained in leaf litter or in highly stony ground. In these cases a 'no data' entry was registered.

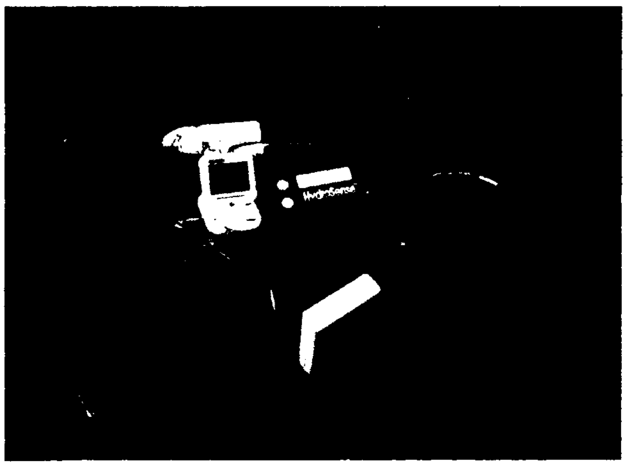

Figure 3.9: *Soil moisture probe and soil temperature probe measuring data in a ground flora nest. (Source:author)*

Finally, a soil sample was taken for a representative nest for the property. This was selected to reflect the many different micro-habitats the nests were found within. Two flat scoops of soil from the confines of the nest were taken with a trowel and were placed into a plastic food bag. This was sealed and labelled. The first bag was placed in another then a third. The triple bagging was necessary to prevent escape of ants which were inadvertently caught. Occasionally the first bag would be chewed through. Storage of soil is dependent on the type of soil and location of sampling. There are no absolute rules (Sheppard and Addison 2008) and tests such as particle size are unaffected by storage considerations. As there were no biota or sensitive nutrients being measured it was acceptable to place the sample in conditions similar to those of a Maritime winter. Therefore, the bags were stored in a dark, cool environment and placed in a freezer within three days.

Light percentage was recorded, based on the amount of sun the nest received during the duration of the day between 9am to 5pm. Factors determining the amount of light included aspect, vegetation cover and surrounding buildings. Observations were made by the researcher during the hours on the property and this was confirmed by the resident(s). This method was based on one used by the Groden team on Mount Desert Island, Maine (Groden, personal communication).

In order to understand whether there was a preference of conditions between nest sites and non-nest sites a comparative study of soil temperature was conducted on the property of CaseStudy 201001 during the summer of 2011. Nest sites and uninfested ground areas within 50cm of each nest were probed using an electronic thermometer. In total twenty seven nests were found. Environmental variables were kept the same for each set of measurements (light percentage, aspect, vegetation cover, moisture levels, soil type and depth of probe). A two tailed T-test was used to test the hypothesis.

Other data were graphed and statistically analysed in R (means, medians, modes, standard deviation, histograms and scatterplots).

3.2.2 Micro-Study

The micro-study was a focused study based on one city block. The purpose of this study was to measure the density of ants using a bait sampling method and compare this to known nest locations. This allowed for validation of the trapping method and for comparison of density to anecdotal evidence of introductions.

Bait sampling is a technique used in areas where it may be difficult to place pitfall traps due to surface composition or practicality of the traps being left undisturbed. Interactions between species and changes in abundance and species of ants or to determine dominance (Lach et al. 2010, Andersen 1992).

The city block chosen as a study site is depicted in Figure 3.10.

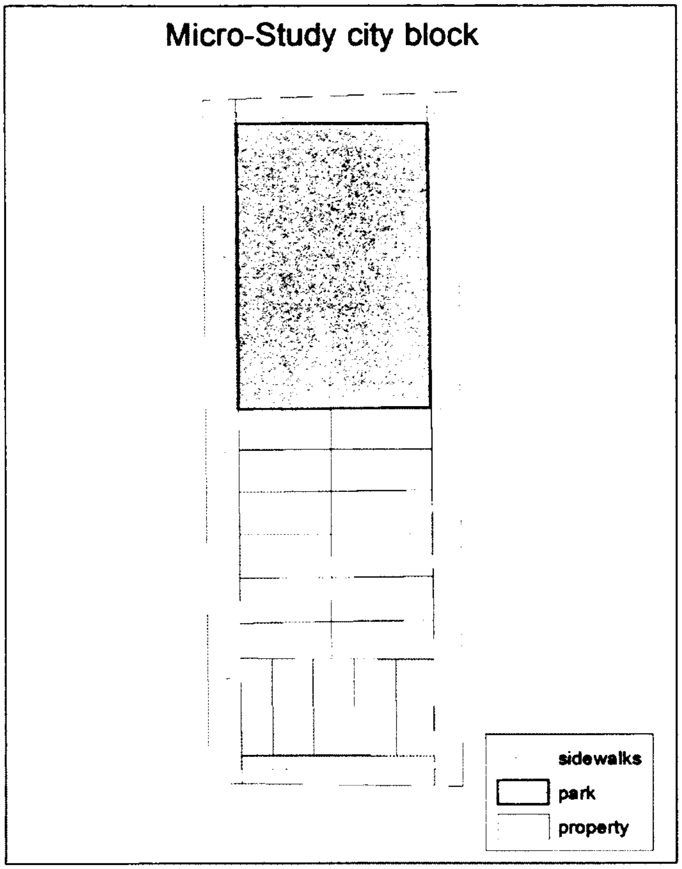

Figure 3.10: *Micro-study block with identifying features altered or removed (Source:author)*

This block was selected for several reasons:

- it had a known infestation
- there is a mix of private properties and a small public park and playground therefore representing a mixed use area
- the size and shape of the block is typical for the peninsula area thus representing the urban area
- there are two lane roads surrounding the rectangular block.

This block was chosen as a generic representative of many other similar areas in the city known to be infested. The uniqueness of this location compared to other similar blocks is the addition of the public park. This represents a variety of ecosystems, uses and management strategies. It is thought that although the introduction patterns may be different for each continuous habitat in the city, this block is a valid location to determine the use of this baiting technique.

It is hypothesized that infestations can occur on an unbroken habitat but cannot cross large barriers. The exact width of a barrier is not known but preliminary investigation concluded that the surrounding city blocks were not infested and therefore all populations were contained to the one area. This was based on preliminary observations on surrounding blocks and Groden 2010 (personal communication).

A study conducted by Groden et al. (2008 unpublished) in Orono, Maine focused on a residential block of mixed land use in a suburban environment. The purpose of the experiment was to establish the densities of *M. rubra* then test a variety of poisoned baits and monitor the resulting changes in populations. In the Orono study, the ant densities were measured using a 25% sucrose bait in scintillation tubes across transect lines at approximately 2m intervals. Each transect line was approximately between 2-10 metres apart.

In this study, gaining permission to access individual properties limited the freedom to run transect lines across the block, therefore, each parcel (as was outlined using civic addresses on the HRM GIS datapack. Source: Halifax Regional Municipality Corporate

GIS 2009) was taken as an isolated study and merged later using GIS. This method covered the block in a continuous manner as if it is a continuous habitat. This mimicked studies done with transect lines such as those by Groden et al.(2008 unpublished), and Andersen (1992), who used a 6 metre spacing whilst baiting ants using tuna in the tropics of Australia.

In this urban study where lot sizes are relatively small, each parcel was divided into a grid of 5 metres by 5 metres with the origin located at the lower left hand corner (while facing the main structure). Location flags were placed at five metre intervals in both horizontal and vertical axes unless there was a structure or a hard surface (sidewalk, driveway). Once flags were in place, scintillation vials containing a 2cm x 2cm gauze square saturated in a 25% sucrose solution, were laid on the ground and left for 1.5 hours. The vials were then capped and the species identified as *M. rubra* or native (all *Lasius spp.*) and density recorded on the scale set by Groden et al. (2008 unpublished) for standardization (see Table 3.1).

Table 3.1: *Key for densities of ants in scintillation vials.*

Density	**Number of ants**
Zero (Z)	0
Low (L)	1-25
Medium (M)	26-50
High (H)	51-100
Very High (VH)	100+

Table 3.2: Example of recording sheet for micro-study

Flag	X (m)	Y(m)	rubra density	other density
1a	0	0	L	Z
1b	5	0	H	Z
2d	15	5	Z	M

Although all private properties were measured at five metre intervals, the public park was measured at 12m x 15m. This larger grid was chosen due to the safety concerns of placing vials in a well used public space during peak times and of practicality of gathering data in one session in such a large area. To prepare the public for disruption, public notices were placed on fencing for a period of a week prior with information and a contact email address. No enquiries resulted.

3.3 Laboratory

The laboratory provided the means to analyze the soil component more thoroughly than in the field. Frozen soil samples were thawed as and when needed to conduct the various stages of soil tests. Measurements of edaphic factors included moisture, organic matter, pH and texture (particle size). Wherever possible, guidelines in Carter and Gregorich (2008) were followed.

Soil is an important medium in which ants can nest. Although many nests are found in leaf litter, especially in a forested environment (Groden et al. 2005), it appears most colonies in the urban environment have overwintering nests in protected soil. In summer they expand the territory with satellite nests in more exposed locations (ground flora). Soil samples were taken from most sites to identify if soil characteristics play a role in the choice and success of the species. The samples taken were chosen to represent a wide variety of micro-habitats. Each sample was chosen to represent a micro-habitat of that site (such as ground flora, under stone, against substrate, under log etc.).

In the field, two readings were taken for each nest: moisture and temperature. These were averaged for each location so as to be comparable to soil sample variables measured in the lab. The bagged soil was taken back to the lab and left in the freezer for two to three months. All *M. rubra* perished, but as a safety precaution, samples were thawed at room temperature in a small tray immersed in a larger tray filled with soapy water. If any ants were able to escape, they would then have had a moat of surfactant to cross and perished. The soil was then analysed to determine the following attributes: moisture content, organic content, texture and pH.

3.3.1 Moisture

Although percentage of moisture was read in the field, the soil samples were also dried in the laboratory oven to measure the gravimetric soil moisture.

The mass of aluminum pie plates was taken before adding thawed soil sample. The mass was retaken and recorded. Samples were heated at 105°C for 24 hours. Samples were allowed to cool in a desiccating chamber and mass was retaken until no change in mass occurred (Kroetsch and Wang 2008, VanProosdij 2010 personal communication).

***Figure 3.11**: Taking the mass of soil samples for moisture content. (Source:author)*

***Figure 3.12**: Drying soil samples in the oven.(Source:author)*

3.3.2 Organic Matter

The dried samples were divided and a sub-sample taken and put in a measured crucible. The crucibles were placed in a muffle furnace at 550°C for two hours. Sub-samples were removed and allowed to cool in a desiccating chamber then re-weighed. The difference was calculated and converted to percentage.

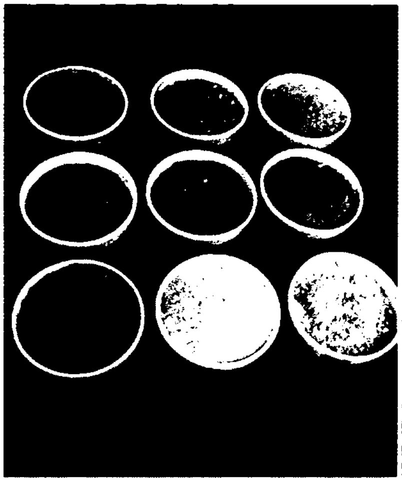

Figure 3.13: *Muffle oven with sub samples testing organic matter component. (Source:author)*

Figure 3.14: *sub samples of soil after burning. Note the grey ash in one crucible where the soil had a very high organic percentage. (Source:author)*

Figure 3.15: *Desiccation chamber with samples and sub samples. (Source:author)*

3.3.3 Texture

Mechanical separation was for grain size analysis. Dried samples were ground in a mortar and pestle until hardened soil particles were reduced to dust. The samples were then placed in a Tyler Ro-Tap mechanical sieve shaker using standard Canadian Soil standards brass sieves (Table 3.3). Dried soil was placed in the top pan and shaken for 1 minute. Each sieve was removed, the soil was brushed out onto a measured pie plate on a balance using a soft haired brush (to avoid damaging the mesh), and weighed. Occasionally soil clods were found in the large mesh sieves. If this occurred, all soil was removed from sieve, re-crushed in the mortar and returned to the shaker and re-shaken (Kroetsch and Wang 2008). Each mass was recorded and categorized according to the CSSC guidelines.

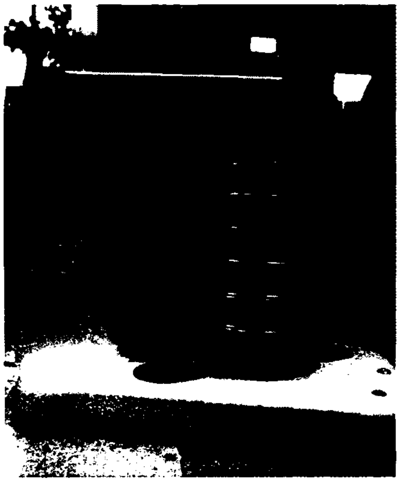

Figure 3.16: Sieve shaker with stacks of brass sieves.(Source:author)

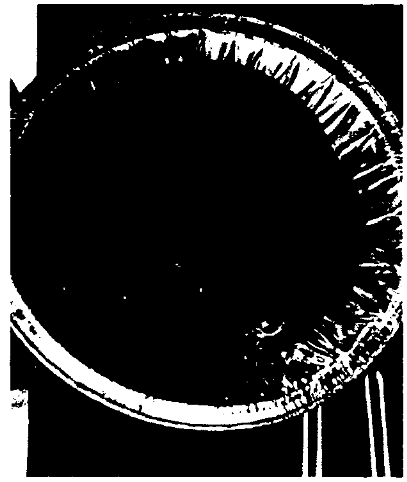

Figure 3.17: Various particle sizes displayed after sieving and masses taken. (Source:author)

Table 3.3. Brass sieve sizes in various measurements.

Size (mm)	Sieve Number	Phi (ϕ)
4.000	No.5	-2
2.000	No.10	-1
1.000	No.18	0
0.500	No.35	1
0.250	No.60	2
0.125*	No.120	3
0.630*	No.230	4

*sieve sizes correspond with World Reference Base (WRB) guidelines.(IUSS Working group WRB 2006)

3.3.4 pH

Measurements of pH are generally conducted on air dried samples but due to the limited amount of soil it was necessary to use the oven dried soil for the pH tests. This was discussed with a soil scientist (Burton, personal correspondence 2011) at the Nova Scotia Agricultural College who cautioned but said there should be no difference as the samples are from Nova Scotia and are not high in inorganic carbonates (as is the case in Western Canada).

The test used to measure the pH was 'Soil in Water'. This method is adequate for the detail of analysis needed in this investigation.

***Figure 3.18**: pH probe in supernatant solution. (Source: author)*

According to Hendershot et al. (2008) the main concern of the measurement of soil pH in water is that the increase in the amount of water will cause an increase in the pH; and therefore it is important to keep the ratio constant and as low as possible. 10 grams of soil was placed in a 50ml beaker. 20ml of 'pure' or double deionized water was added and stirred with a glass rod. For the organic samples, 2g of soil was used in 20ml water. If more soil or water was needed, the ratio was kept the same. The solution was stirred on low on a stirring plate for 30min. A magnetic stirring bar ensured a homogeneous solution. The suspension was allowed to settle for one hour to separate sediments and supernatant.

Before immersion, the electronic probe was calibrated with buffers pH4 and pH7 using Logger Pro software. The voltage reading was 2.48V. The probe was rinsed with deionized water between readings. Once calibrated, the probe was immersed in the supernatant ensuring the glass membrane and porous salt bridges were submerged and a reading was taken once the measurement was constant.

3.4 Geographic Information Systems

GIS maps are useful tools whenever spatial data is presented. The purpose of the maps used in this investigation was to show the locations of reports across the HRM, to show the locations of nest sites on individual properties and to display the densities from the bait vials on the micro study block. The software used was ESRI ArcGIS, ArcMap v9.3 and ArcMap v10.

3.4.1 Data Management

Data integrity was crucial to the project. Due to the fact that homeowners were consulted and addresses used to create a database, it was essential to protect the identity of residents according to the agreement laid out in the Research Ethics Board approval. Therefore a database spreadsheet was used and identities and collected data, were separated. The commonality used for all data manipulation was the case study number. All data used for purposes other than a map were referred to only by the case study number. All data with spatial references were further separated from the names of the homeowners.

3.4.2 Maps

There were several maps created to show a variety of aspects of the data. The primary map was a geocoded map of infestation sites in the HRM area using reported findings from the public. This was made using ArcMap v10 accessing information stored centrally at ESRI in the coordinate system of GCS_WGS_1984. The coordinate systems used for the rest of the map data was: was ATS_1977_MTM_5_Nova_Scotia (projected coordinate system) and GCS_ATS_1977 (geographic coordinate system). This could cause problems at the street level with coordinates not matching by several metres, but as this map is at a large scale and only meant to show general locations, it does not affect the quality of the displayed data. The geocoded map was based on civic addresses as opposed to GPS coordinates. This was chosen due to standardize the locations. If more detailed locations of nest sites in a parcel were needed, for example in large public parks, then longitude and latitude coordinates were used. The addresses were processed using the geocoding tool under the 'tools' drop down menu. Any addresses not matching were checked, corrected and rematched. There were some discrepancies due to the change of name of some roads but these were resolved using data from HRM.[11] A large scale map and a smaller scale of the concentrated infested areas which can be viewed by the public are included. Detailed maps showing streets and roads are found in Appendix B.

The micro-study maps were based on the data collected from the sucrose bait stations on each grid point of the studied property. There was no latitude and longitude information needed as the property was based on a civic address. Therefore, the grid of X and Y

11 http://www.halifax.ca/giss/

coordinates had an origin of 0,0. This data was exported from the spreadsheet (OpenOffice Calc) to a .csv file (text) for insertion into the ArcGIS program. ArcGis ArcMap 9.3 was used and a base map of the HRM area created. The micro-study .csv data was added and converted into a shapefile. The shapefile had to be dragged from the global origin of the equator and dropped into Halifax. Using the rotate tool the grid was moved into position to match the measurements taken in the field. The edits were saved and repeated for all nine sites. Once all sites were complete the merge tool found in the Spatial Analyst extension was used to make one one shapefile from the separate studies. This vector map was customized to show the densities of the fire ants as low, medium and high in a colour coded hierarchy. This process was repeated for the densities of native ants.

The vector maps were then interpolated to raster maps using the vector to raster interpolation tool. Krigging, Spline and IDW (inverse distance weighting) maps were made but it was decided, based on the uses of each in Heywood et al. (2006) the best interpolation of the data was IDW. This method most closely represents the type and amount of data available for the study area.

Please note that the microstudy maps have been reorientated and the north arrow removed. Also, some features were altered or disguised. The purpose of this was to make the maps less identifiable to protect personal property.

4 Results

4.1 Preliminary Study

The primary goal of the preliminary study was to recruit participants. Public education was a secondary goal. Residents who were unsure of the species on their property took samples to Andrew Hebda (NSNHM) or to Saint Mary's for identification. Mr. Hebda has been receiving samples of *M. rubra* since the late 1990s and identified this species in Nova Scotia. Formal acknowledgement of the species at a federal level was given by the Canadian Food Inspection Association (CFIA) in 2008 (Hebda, personal correspondence, Wetterer and Radchenko 2011).

The emails from the public to the researcher totalled close to one hundred and fifty, the majority of which were from the HRM area and elsewhere in Nova Scotia. There was interest from other parts of Canada as well due to a story in the Canadian Press. There has now been a positive identification of the species in parts of Vancouver by an entomologist (Robert Higgins, personal correspondence 2011) and a recent article written about flying male *M. rubra* in Newfoundland (Hicks, 2011). These researchers give evidence of continuation of spread of the species throughout Canada.

4.1.1 Questionnaire results

Based on the feedback from residents a timeline was created to show when *M. rubra* were first perceived as a problem. This is shown to have increased during the past decade (see Figure 4.1). Other results of the questionnaire are found in Appendix C.

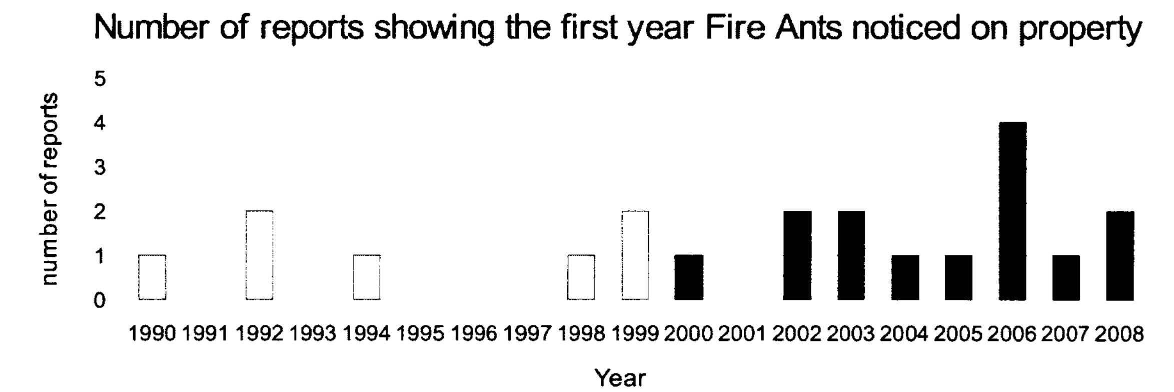

***Figure 4.1**: Graph showing the year M. rubra became a problem for homeowners.*

The years 1990 - 2008 are inclusive as this is the period of time people were certain of the presence of the infestations. There have been reports dating back to thirty five years via members of the public but these have not been included in this graph as confirmation was not attained directly from the questionnaire.

In Figure 4.2, the total number of reports of first infestation show an exponential growth. This indicates there may be an irruption of the species occurring. If the total number of complaints were also graphed, these would be expected to also show an exponential increase. There were also increases of reports in periods after media exposure as was the case after media releases during 2010. Therefore such data may have a bias.

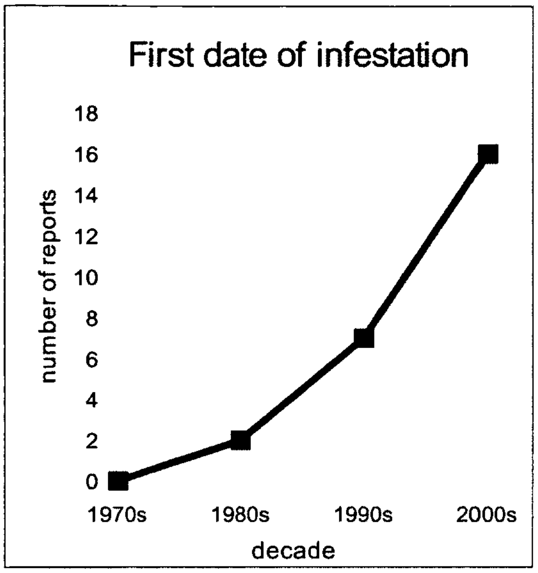

Figure 4.2: *Date of the first notice of the infestation on the property of residents over the past four decades.*

One question was regarding the possible origin of infested material. It was found that residents bought plants and material from a variety of sources (Figure 4.3). There appears to be no difference of choices of one origin of organic material (or soil) over another. It is inconclusive that there were greater reports showing Superstore as a source of plants as it is not known whether people shop more frequently at one business over another. There is no measurable evidence to show that any of these businesses was a source of ant infestations. However, anecdotal evidence on two properties shows that there was no infestation before a particular type of organic material was introduced and within five years, the infestations were at problem levels. This time frame of five years occurs in another report where a resident had transplanted infested plants from her Halifax property to her cottage, and within five years the cottage property was also infested to problem levels.

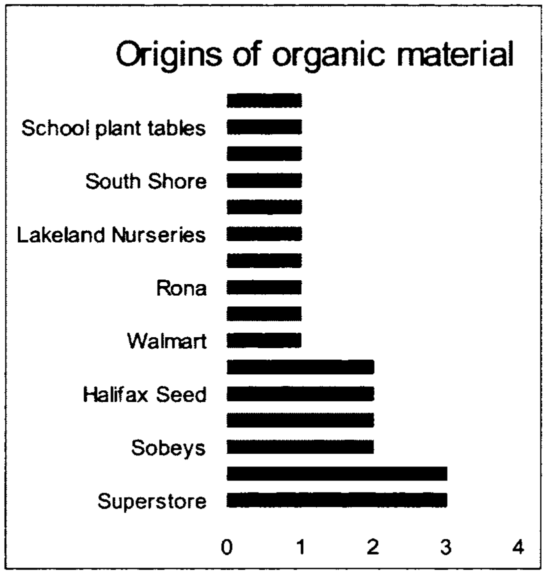

***Figure 4.3**: Graph showing sources of organic material for properties known to have an infestation.*

When asked whether there has been a noticeable increase or decrease in other species, residents reported both greater and fewer earwigs and spiders. However, all reported a decrease in other ant species. Almost all residents reported using some type of chemical control. There was no noticeable reduction in *M. rubra*. The only success reported in reducing numbers of ants, was a family who dug up their lawn, laid new sods and paving stones, and started aggressively baiting with a home-made 1-2% borax solution. This evidence is based on two summers (case 201016). It is unknown what the long term effects will be. In 2011 case 201014 began aggressively removing nests and using chemical controls in the soil left behind. It is reported that the numbers of ant nests and stings has significantly reduced. This method is also being applied to case 201001 but has yet to be complete enough for a significant result.

4.2 Macro-Study

The macro study focused on recording characteristics of nests and establishing the micro habitat each nest was found in. In total, thirty five locations, including portions of five city parks were searched for nests. Of that number, the species was confirmed and nests found in twenty three properties. Measuring equipment was available for use for seventeen of the infested sites. Measurements and soil samples were only taken at these sites (one large, diverse site had three soil samples taken).

4.2.1 Nest Characteristics

This investigation was primarily based on observations of nest conditions. Several variables were measured, some at the site level and others at the nest level. Table 4.1 shows the summary of data analysed both in the field and in the laboratory. Raw data was taken from measurements of three hundred and sixty six nests. This was then summarized for standardization and comparison purposes (n=19).

Table 4.1: Summary of site characteristics for macro-study cases.

CaseStudy	Organic%	pH	Sand%	Soil temp °C	Soil moisture%	Light %	Nest density m^{-2}
201001	21.90	6.23	73.58	20.5	11.93	70	0.11
201002	13.02	5.87	79.71	20.64	18.7	50	0.1
201002	9.02	6.40	79.50	20.64	18.8	50	0.01
201002	2.57	6.99	56.21	20.64	18.18	50	0.01
201003	11.20	5.36	71.79	22.13	17.27	80	0.04
201004	9.42	5.77	66.86	19	17	30	0.04
201005	5.84	6.17	80.10	19.13	16.45	56	0.09
201006	23.28	4.31	74.14	20.5	15.82	10	0.01
201007	52.13	5.90	86.13	18.55	15.75	60	0.1
201009	23.06	6.49	93.72	19.91	15.08	59	0.03
201010	34.79	5.56	80.21	19.52	13.83	87	0.12
201011	2.12	4.47	81.41	21.63	13.33	77	0.02
201012	74.41	3.47	93.74	21.63	12.67	48	0.03
201013	14.90	6.57	74.07	21	12.23	50	0.16
201014	8.00	5.94	62.66	21.71	11.23	32	0.18
201015	13.85	6.45	81.72	20.14	10.69	79	0.03
201016	10.06	7.47	62.51	17.42	10.23	40	0.01
201017	15.48	6.41	78.32	13.8	10.92	42	0.03
201019				17.06	11.92	90	0.13

The soil temperature, moisture and light were based on measurements taken in the field. The organic component, pH and percentage of sand (SumSand) were all calculated from soil samples analysed in the laboratory. The nest density was calculated taking the number of nests and dividing by the total available habitat of each parcel of land in metres squared.

The organic component had a wide range of values starting as low as 2.57% and as high as 74.41%. The mean was 19.17, a median of 13.43 and a mode of 9 (Table 4.2). The standard deviation is high at 18.36. Therefore the preferred organic component appears to be around one fifth of the composition of the soil but is not essential to the nest choice.

Table 4.2: Characteristics of soils sampled in the laboratory: simple statistical analysis

	Mean	Median	Mode	StdDev
Soil Organic Content	19.17	13.43	9	18.36
Soil pH	5.88	6.06	6	0.98
Sand percentage in soil	76.46	78.91		10.06

The pH showed a small variability having only a 0.98 standard deviation. The mean was 5.88, median 6.06 and the mode clearly a 6. The lowest pH was 3.47 and the highest 7.47. Generally in Halifax the soil is slightly acidic due to the geology of the area. It is unknown whether there is a preference for slightly acidic soil or if the ants have adapted to the acidity. In England, *M. rubra* are found in the heathland where soils are ericaceous, therefore this may be a factor in their success. Most plants prefer a range of pH between 6.5-7 where most nutrients are readily available. *M. rubra* are most frequently found in the roots of plants (Figure 4.7) followed by soil against a hard substrate. This shows acidic conditions are not a disincentive for habitation. It is unknown whether the same is true for high pH soils, as no nests were found above a neutral pH.

It was hypothesized that sand would be a small component of the mineral make up of the soil as silt and clay are better retainers of moisture. This however, was proved incorrect as the percentage of sand started at a high percentage (56.21%) and had a range up to the ninetieth percentiles (93.74%). The mean was 76.46 and the median 78.91. The standard deviation was 10.06 showing a wider range of preference than is clear from the average and median.

Based on averages for each site, soil moisture ranged from 10.23 to 18.80%. This gives an overall average of 14.32%, a median of 13.23 and a standard deviation of 2.89. This was accumulated data from the total of the three hundred and sixty six nests. However, if statistics are calculated from the raw data (Table 4.3) the products are more varied. This can be accounted for by the wide range of in situ nest measurements from 3% to 39%.

***Table 4.3**: Characteristics of soils sampled in the field*

	Mean	Median	Mode	StdDev
Soil Moisture %	11.71	10	7	6.61
Soil Temp °C	19.68	20	21	2.07

The temperature of the air was measured at each site in the shade. The average air temperature was 25°C. Therefore the nests were kept on average about 5°C cooler than the ambient temperature. This is in keeping with other social insects such as bees (Drummond 2011, personal communication). Ambient light had a wide range with a mean of 59.72, median of 23 and mode of 40. Standard deviation was high at 27.36. The range of light varied from 10% to 100%. Therefore the amount of light on the nest area has little effect on determining the position of the nest.

Nest density, the dependent variable, involved the calculations of the possible habitat nesting areas of each property. Where the property had breaks in habitat areas such as buildings or paving, each section was individually measured and added together.

The average density of nests found in the city was 0.12 nests m^{-2} with a median of 0.1 nests m^{-2}. The standard deviation was 0.11. This compares to the densities found in the native habitat of Britain where the range was from 0.02 to 0.13 nests m^{-2}(Groden et al. 2005). There were areas of high infestation where nests density was 6 nests m^{-2}. This compares with the findings of Groden et al. (2005) who measured up to 4 nests m^{-2} in Maine. However, the average nest density was greater at 1.24 nests m^{-2}.

The statistical program R was used to analyse the data to look for relationships between the variables. For the purposes of R the title of each variable was altered slightly. Data from the spreadsheet was converted into a .csv file and read into the program. Histograms were plotted to determine frequency distribution (Figure 4.4).

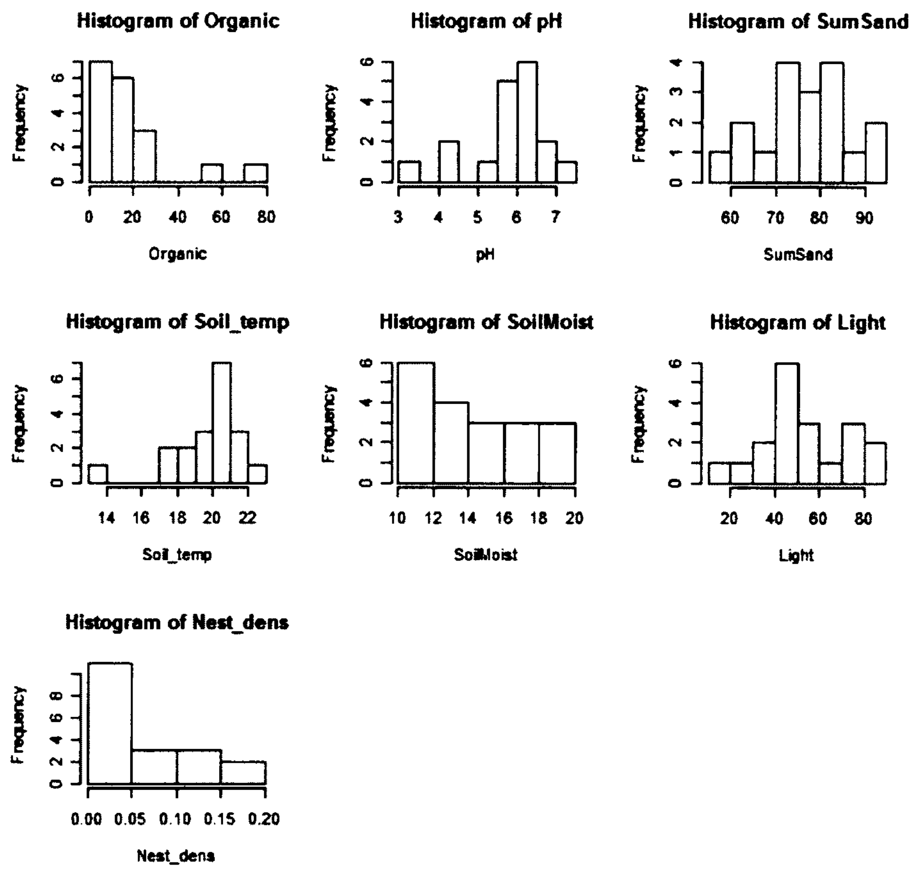

***Figure 4.4**: Histograms of M. rubra nest characteristics and nest density.*

The histograms in Figure 4.4 show a variety of distribution curves. The percentage of organic matter have a positively skewed distribution. The higher values of organic material are due to soil samples taken from a compost heap which was rich in humus and an area in which organic matter had been added to the soil by the resident. The pH shows a negatively skewed distribution. The soils in the HRM area have a slightly acidic pH due to the slate bedrock and glacial till. However, in the urban environment some home owners add an alkaline substance such as calcium in an attempt to raise the pH. In this study two residents reported adding lime to their soil. The highest pH, 7.47 was found at one of these properties. The other resident who had reported increasing their pH had a reading of 5.36. There appears a normal distribution of the percentage of sand but only against the axis spanning the samples. If sand percentage was plotted against a zero origin, there would be a negatively skewed distribution as all of the values are above 50% and most are between 70-80%. It was expected that the percentage of sand would be closer to that of a loamy soil, i.e. 40% sand (40% silt and 20% clay). In Maine and England the ants have shown a preference for moist environments so this was unexpected. The soil temperature appears to have a negative skew but if the outlier of 13.8°C is removed, the curve is a normal distribution bell shape. Soil moisture shows an even distribution across a wide range of percentages with a slight positive skew. Light percentage has a normal bell shape distribution. The dependent variable of nest density has a definite positive skew. The highest frequency of values are found in the 0.00-0.05 range.

Scatterplots in R, which compare all variables against each other in a matrix, were used to look for trends between nest variables and help determine the correct statistical test to apply (Figure 4.5). There appears no strong correlations between variables. Most plots show an evenly distributed collection of points. There may be some concentration of data points between organic material and pH. The organic matter versus percentage of sand also may show some slight linear trend. Organic matter against the percentage of sand may also show some concentration. All have many outliers which may indicate a further trend or may show a more random distribution. Organics versus soil moisture, percentage of light and nest density show a mostly random distribution with some possible linear pattern. All trends are based on observation of points on the graphs but would need a larger sample size to do a regression analysis and determine if these were true.

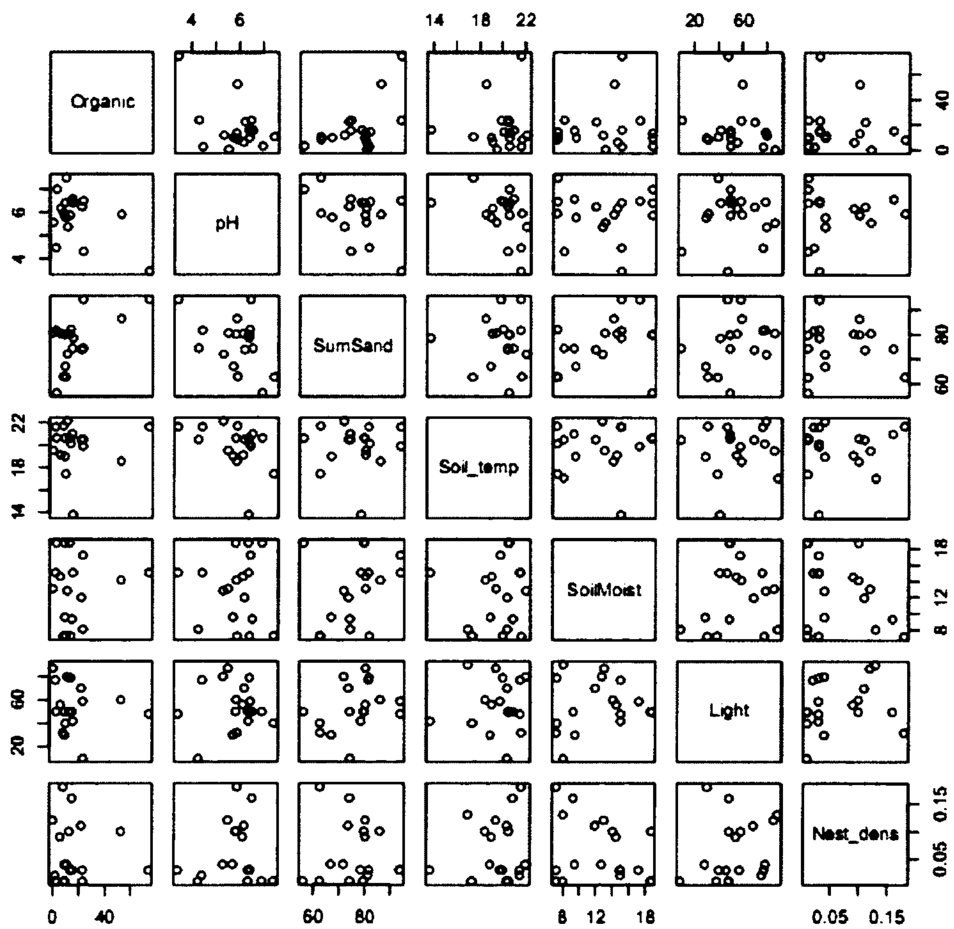

Figure 4.5: Scatter plots of M. rubra nest characteristics and nest density.

pH has a randomly distributed pattern on most graphs when plotted against other nest variables. There may be some vague trend with soil temperature. Soil temperature too shows very little possible trends in any graph except organics and pH. Soil moisture also has little correlation with any variable except the possibility of a linear trend with organics and soil temperature. Light shows an almost uniform distribution on all graphs except possibly organics. Nest density, the dependent variable shows no strong trends against any nest characteristic.

4.2.2 Habitat: Vegetation and Substrate preference

The urban environment in Halifax was recorded as two categories: nests amongst vegetation and nests against or within a hard surface (substrate). There is a large variety of habitat possibilities under each category, as people personalize their outdoor environment. However, some patterns have emerged. Figure 4.6 shows the full array of urban habitats recorded in the field. The number of nests found in each micro-habitat is on the y-axis and the micro-habitat types are found on the x-axis. Appendix E has a sample of the full spreadsheet of tallies.

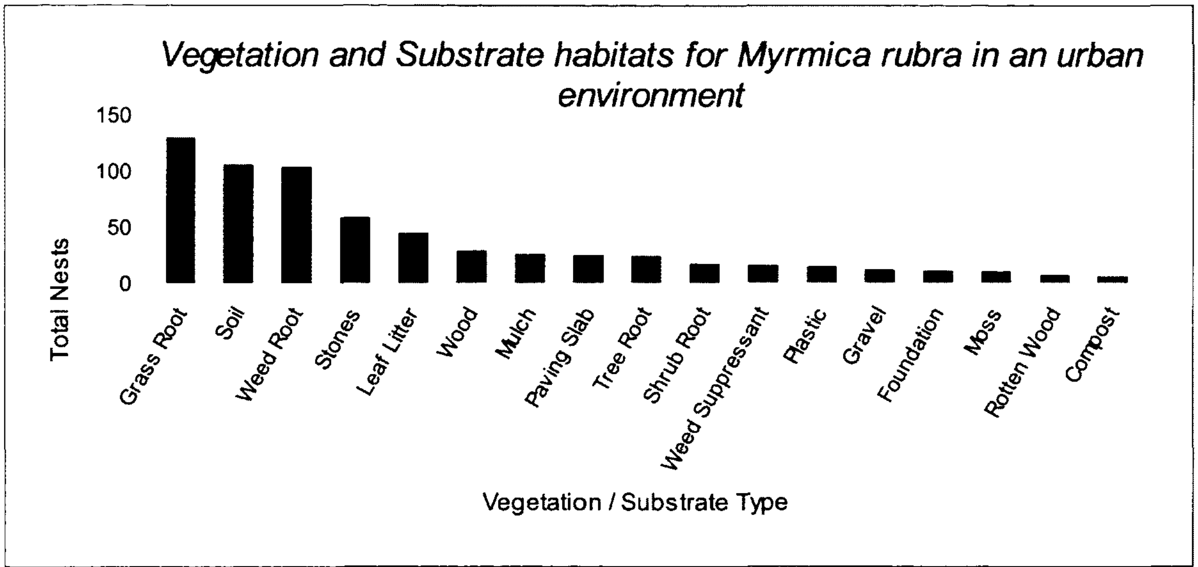

Figure 4.6*: Graph showing the various micro-habitats of Myrmica rubra nests.*

Grass nests were in the greatest abundance. These nests were closely followed by nests in soil. The soil nests were found almost entirely against or under a substrate, as opposed to nests openly exposed in soil or soil mounds, such as those built by other species. This can be seen more clearly in Figure 4.7 where soil is considered uncovered. Weed roots, predominantly dandelion roots, was only slightly fewer than grass. It was observed by a botanist studying an infested area that many nests on a particular property were found within the roots of dandelions (Lundholm 2011, personal communication). On a similar property (case201001) an observational comparative study was conducted and found that in areas where both grass and dandelion plants are in equal abundance, the nests were primarily found in dandelion roots. On most properties surveyed however, dandelions had previously been removed or prevented and therefore grass was the dominant plant.

The micro-habitat categories were amalgamated to form fewer categories to be comparable with the habitat graph for Maine. The summary can be seen in Table 4.4.

Table 4.4: *Reduction of tallied categories of vegetation and substrate micro-habitats to simplified categories for comparison with other graphs showing invaded environments.*

Simplified Habitat Category	Original Habitat Category	Tally
Soil under tree roots =	tree root	23
	shrub root	16
		39
Soil mound nest in herbaceous roots=	grass roots	129
	weed roots	103
	moss	9
		241
Soil=	soil (only)	3
		3
Leaf litter=	leaf litter	44
	compost	5
	mulch	25
		74
Downed woody debris=	Rot Wood	6
		6
In soil against a substrate=	gravel	11
	paving slab	24
	stones	58
	foundation	10
	plastic	14
	weed supressent	15
	wood	28
		160

Summary	number of nests	percentage	%whole
Soil mound nest in herbaceous roots	241	46.08	**46**
In soil against a substrate	160	30.59	**31**
Leaf litter	74	14.15	**14**
Soil under tree roots	39	7.46	**7**
Downed woody debris	6	1.15	**1**
Soil	3	0.57	**1**
Total	523	100	100

***Table 4.5**: Summary of table 4.4 showing absolute number of nests converted into percentages and rounded.*

The absolute number of nests was changed to percentages for standardization and ease of comparison. It was found that the ground flora nests were most abundant in the urban environment at 46%, followed closely by soil against a hard substrate 31%. Leaf litter was present at 14%. Soil under tree roots less than 10% at only 7%. Downed woody debris and exposed soil were very low at only 1% and less than 1 respectively (Figure 4.7).

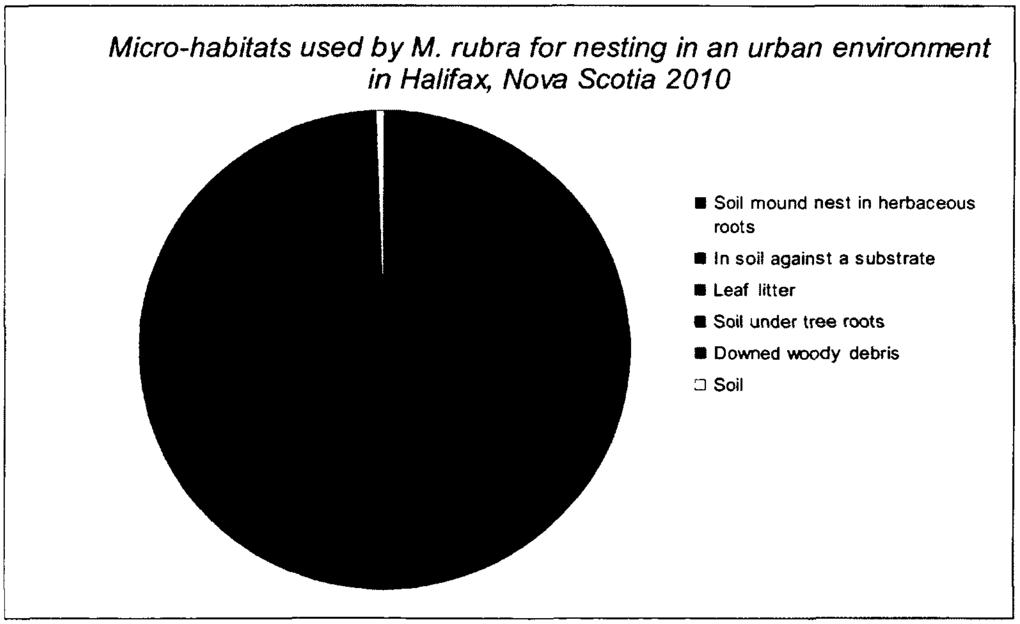

***Figure 4.7**: Graph showing micro-habitat choices in invaded urban environments.*

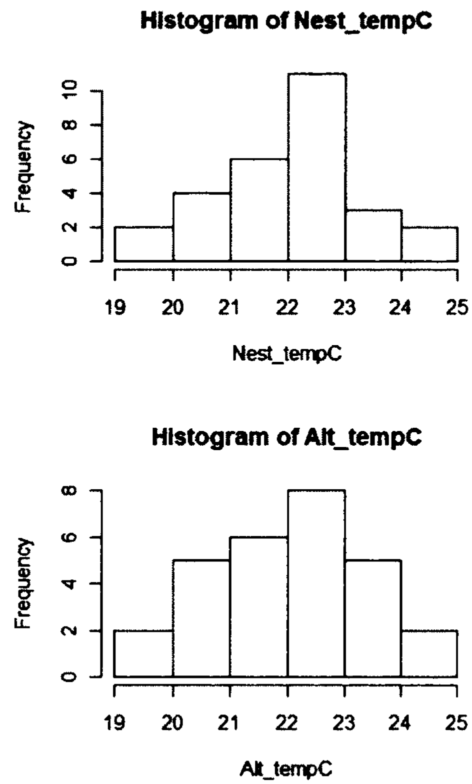

Figure 4.8: *Histograms of nest temperatures and alternative habitat temperatures.*

The comparison of soil temperatures of nest sites compared to non-nest sites showed a close relationship. In R, the measurements were graphed using a histogram (Figure 4.8) and a T-test conducted.

Both histograms show a normal distribution bell shaped curve with the range the same of 19°C to 25°C. A paired T-test was chosen as the data is paired, normally distributed, has less than 30 values and is interval. This was calculated using R.

It was found that there was no difference in soil temperature between the nest sites and the uninfested sites (Paired t-test, $t = -1.1485$, $df = 27$, $p = 0.2608$).

4.3 Micro-Study

The number of ants counted in the scintillation vials were categorized in ranges of zero (0 ants); low (1-25 ants); medium (26-50 ants); and high (51-100 ants). The results are detailed in Appendix F. Figure 4.9 shows the density of both native ants and *M. rubra* on a GIS map of the micro-study block.

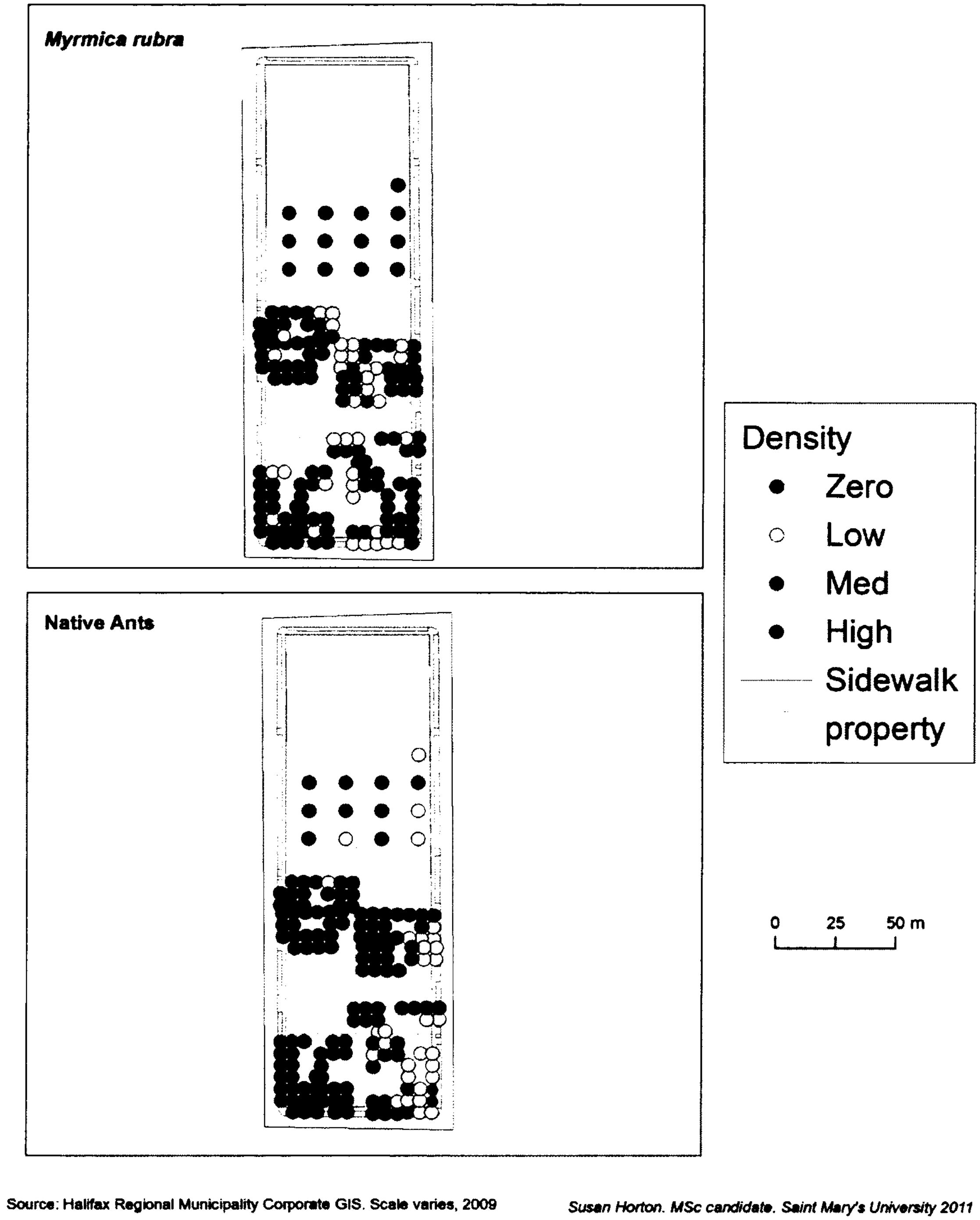

Figure 4.9: *Vector map showing ant densities on micro-study block.(Source:author)*

The coloured dots represent the number of ants found in each vial. It can be seen that there are many vials empty of native ants and a mix of densities of *M. rubra.* The vials contained either *M. rubra* or native ants, except for one vial which contained one native amongst a number of *M. rubra.* It is thought that given a food source, such as the sucrose solution, the *M. rubra* will out compete the native species and therefore be detected Groden et al. (unpublished). They found no mixing of species.

The southern most property, found on the corner lot had no nests of *M. rubra* in the macro study but did have scouts appear in vials along the sidewalk edge. The main native species found in many areas of the block are *Lasius spp.* In 2011, ant specimens were identified and it was found that there were five *Lasius spp.* and one *Camponotus sp.*, as well as *M. rubra.*

In Figure 4.10 the nest sites from the macro study have been added to the density maps. It can be seen that there is a correlation between location of nest sites and ants found in vials. Areas of white are where there are no standardized measurements, although *M. rubra* have been confirmed in all locations on the block, except the park area where there was confirmation of no *M. rubra.* The only properties where *M. rubra* were not present were the two closest to the park. These properties had scouts close to the southern neighbour's fence but did not have nests. In the upper park area no nests were found and no individual *M. rubra* were seen. A follow-up search confirmed this. *Lasius spp.* dominated the park. These discrepancies however are minimized by the use of the raster maps which interpolate the missing data.

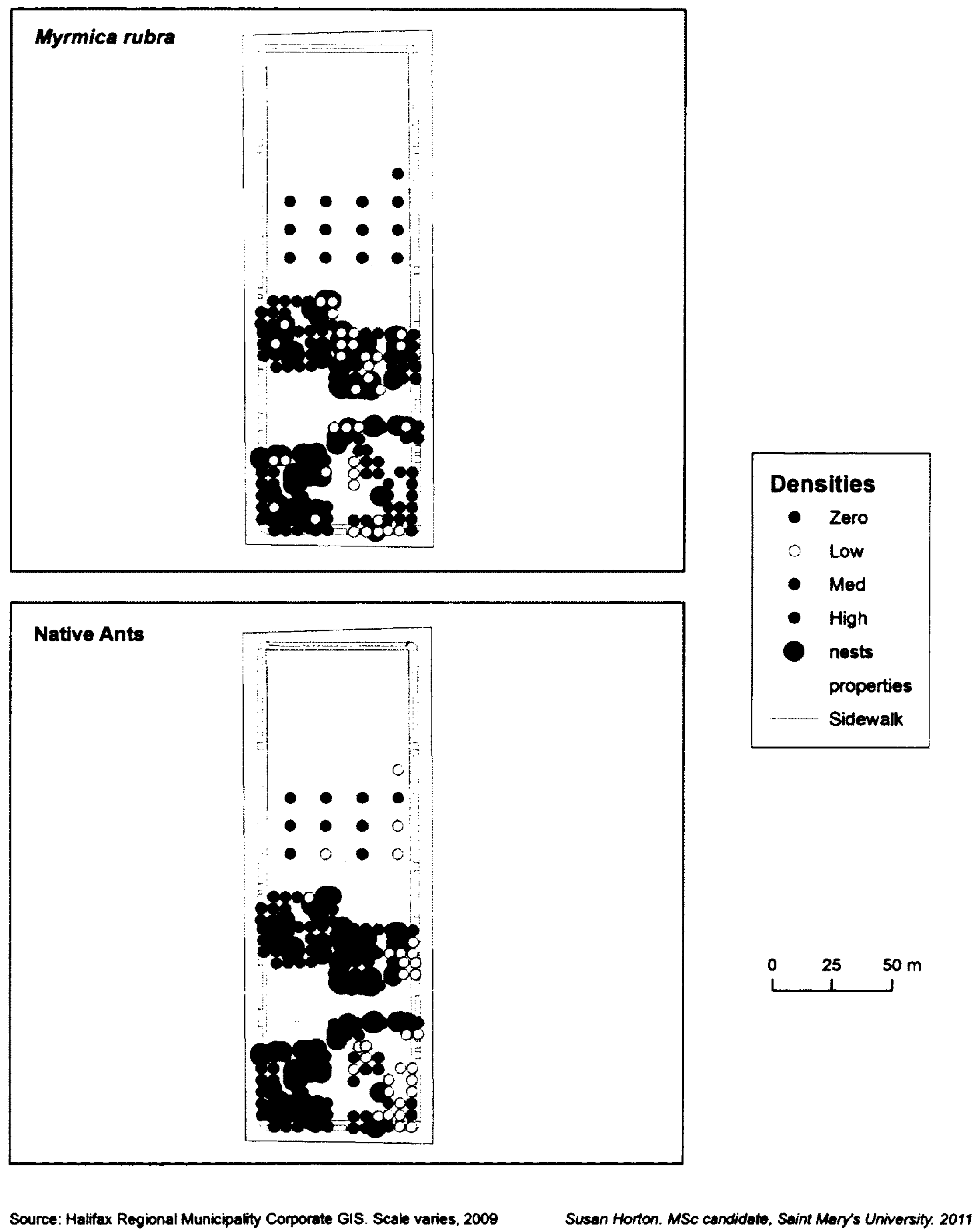

Figure 4.10: *Map of the location of the macronests of M. rubra superimposed on the M. rubra and native ant density from Figure 4.9 (Source:author)*

The vector maps were interpolated to Kriging, Spline and IDW raster and compared with the locations of the baited ants and the nests. It was found that the IDW maps most closely resembled the actual data.

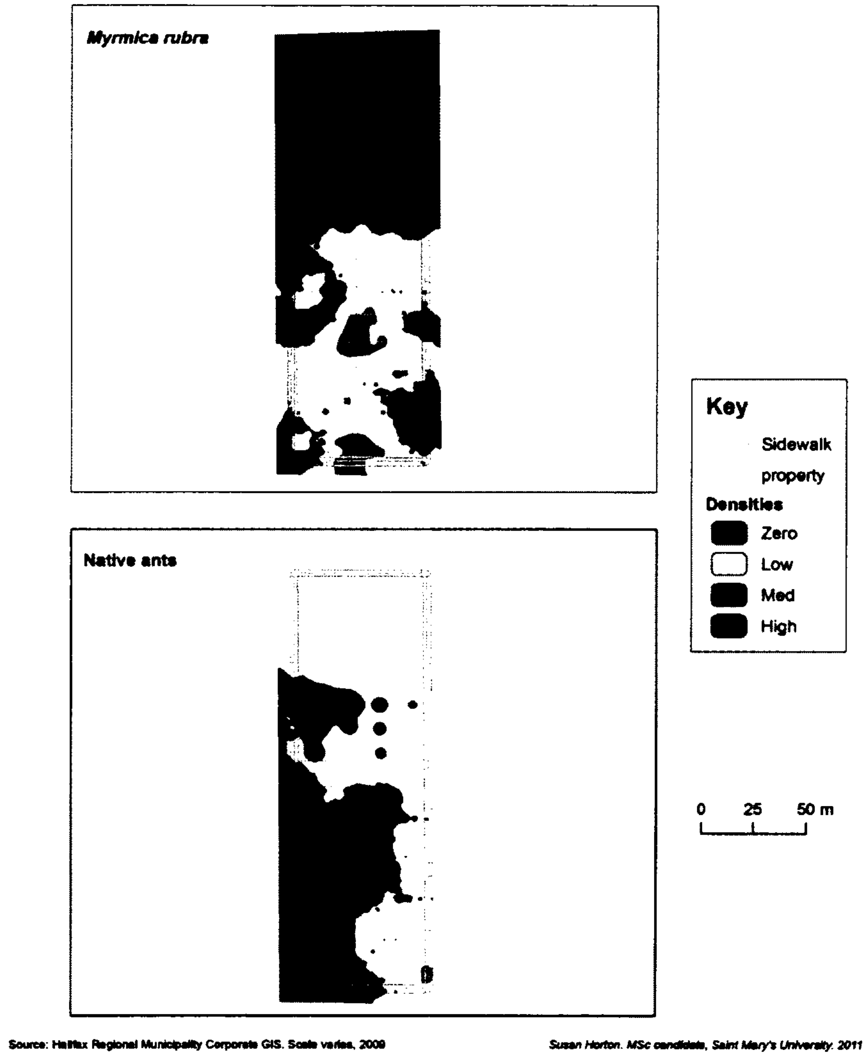

***Figure 4.11**: Inverse Distance Weighting map for M. rubra and native ant species on the micro-study block. (Source: author)*

Figure 4.11 shows the IDW maps for both native and *M. rubra* presence. There are two areas of the block which had a medium to high density of ants. This fits with anecdotal evidence by residents who thought there were introductions onto the block in two locations. The property with the highest density (in the south west part of the block) had a pest control company come in to exterminate the ants. The measurements on their property were made after three applications of pesticide by a professional company. The results may shown greater densities if the baiting was conducted the same time as the pre-application macro-study (Figure 4.14). The other 'hot spot' of high density was found near the centre of the block. Residents complained about a tree that had fallen during a hurricane (Juan in 2003) that had become heavily infested thus creating a source of ants which spread to adjacent properties. The ants would have been present before this but perhaps they multiplied to a problem level as a consequence. The circular pattern seen on the map appears to confirm this.

Slightly north of that high density area is an area of green almost surrounded by yellow. The residents here had removed their topsoil and have maintained a campaign of getting rid of nests and baiting continuously. They had a low density. It is worth noting however that the bait traps were set after a hurricane in September 2010 when the temperature cooled significantly and rapidly. Despite the presence of nests, the ants were not often found in vials. This may have been a behavioural response to the temperature change and affected the results.

A comparison of nest sites and densities Figure 4.12 overlays the nest sites on the IDW map. Nests are found in many of the same locations as areas where *M. rubra* were found in vials. The inverse is true for the native species. Although it has been observed that some native species, *Lasius* species in particular, do share habitat and even nests with *M. rubra* Figure 5.2, it is clear that where fire ant nests were found there was very little evidence of the presence of native species.

***Figure 4.12**: IDW density map with M. rubra nests overlaid in blue(Source:author)*

4.3.1 Additional studies

In one case study (201001) the number of nests was monitored for three summers. Figure 4.13 shows the increase in growth during that time. The property owner also engaged a campaign of using poisoned bait stations during the 2011 period. Any results from these should be seen in the 2012 summer season.

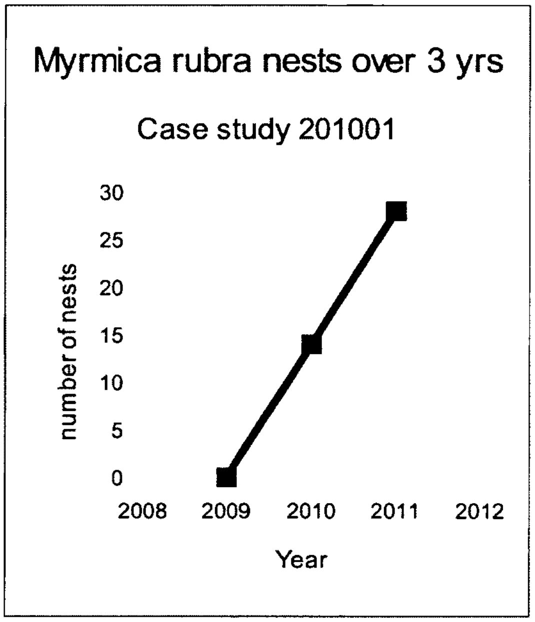

Figure 4.13:*Increased growth in number of nests on one city property over a three season period.*

In the first year (2009), the number of nests is listed as zero. A pest control company injected pesticide and the owners reported the nests may have been eradicated. There may have been nests still present but none were found at the beginning of the 2010 season and therefore concluded that few, if any were present the previous season, although it is difficult to tell until satellite nests are produced. However, this indicates there was a movement of ants from adjoining properties in the two years following as well as the creation of new nests on the property itself once the colony became re-established. Due to the location and configuration of the property it is possible that the majority of the nests came from one adjacent property as at least two other adjoining properties did not have nests present. The third may have had been the source of two of the nests but the spread was distinctive from the direction of the first.

A second additional study focused on the effectiveness of pest control. In one case study (case 201013) before and after treatment maps showed a reduction in nests but not an eradication (Figure 4.14).

It is known that *M. rubra* move nests occasionally but the movement of nests from the initial locations (green) to the new locations (purple) is significant. The pesticide used is known as a repellant as well as an insecticide (Lambda-Cyhalothrin 100g/L. Also known as "Demand"). This may account for the clear delineation of locations on the property. It may also be that the 'purple' are new nests due to colony budding from the neighbouring properties. It is interesting to note the patterns of nests. The green line of nests on the north boundary of the property was located under stones, logs and weed suppressant fabric. The nests in a green line in the centre north were under logs lining a walkway. The nests on the east side in purple were found against the wooden structure of a raised garden. No nests were found in open soil. The purple random distribution in the south west section were grass nests. This area had been sprayed but did not appear to repel ants. It would be interesting to test whether a pest control substance could be used as a barrier preventing further spread if it was used in a similar manner to the 'fire break' idea of the flat surface.

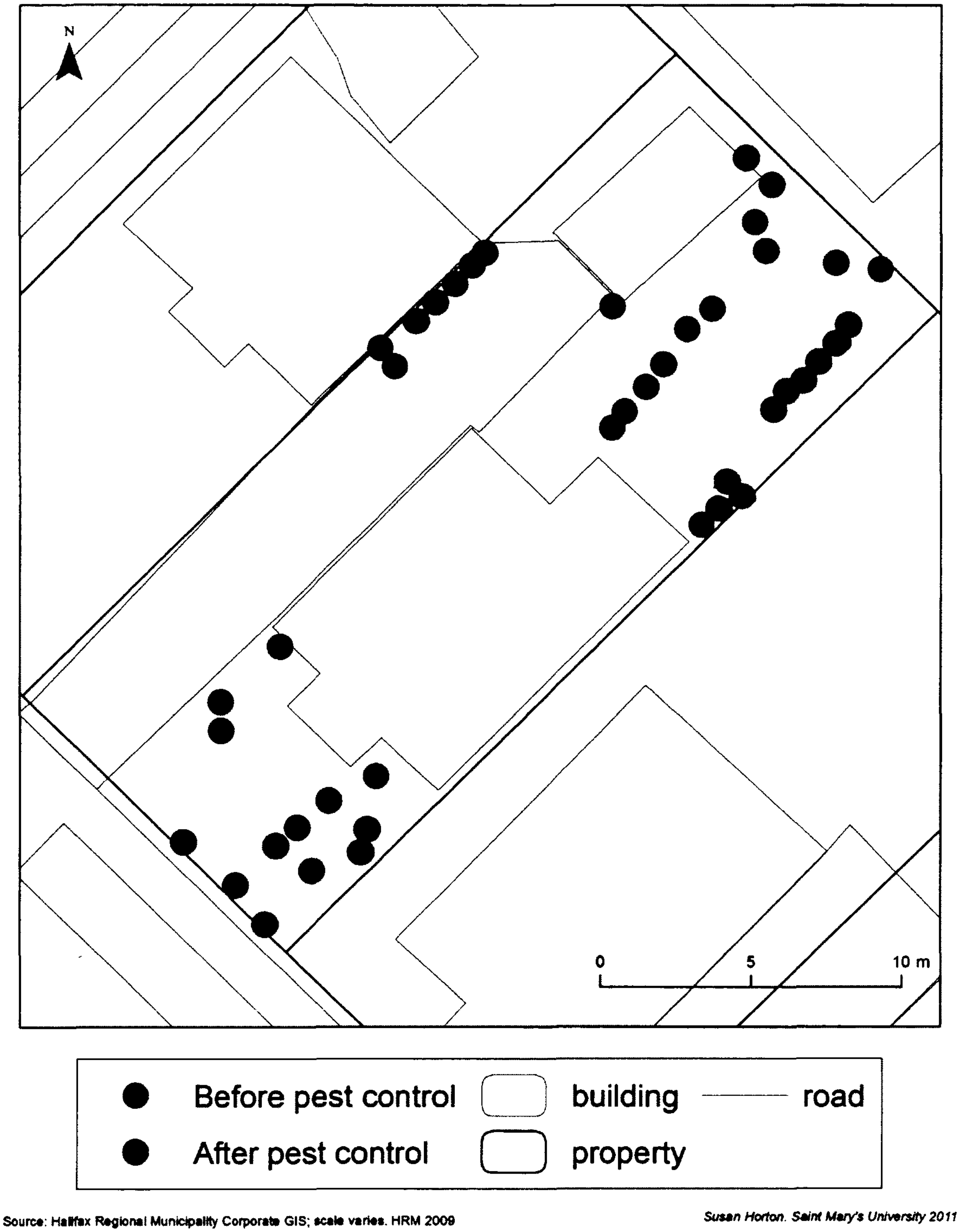

Figure 4.14*: M. rubra nest locations before and after three applications of pesticide. (Source: author)*

5 Discussion and Conclusion

5.1 Introduction

The presence of the ant in the urban/suburban environment has impacted the lives of ordinary citizens by interfering in their enjoyment and use of the outdoor environment. The source of introduction of these ants is still an unknown, although research is being done in Newfoundland and Helsinki to trace the genetic routes of these ants in North America and Europe (Hicks, 2011 personal communication; Higgins, 2011 personal communication). It appears the species may have been in the Halifax area for many decades, perhaps longer, but without concrete evidence this is difficult to confirm. There is however, anecdotal evidence linking childhood encounters over thirty five years ago and stories of "biting ants" from further back. The largest problem is misidentification of species; people assuming they were attacked by an indigenous species when in fact they might have encountered *M. rubra*. The recent increase in reports of this species becoming a problem confirms that this is no longer a case of isolated incidences but a widespread problem that will need attention from researchers and perhaps government officials. The past four decades alone show exponential growth in first encounter reports. If misidentification has been a problem in the past it is no longer the situation as the enduring presence of *M. rubra* ensure proper identification in the present.

The questionnaire was helpful in determining the counter measures residents have taken. Most home owners have used some form of chemical control. Very few have used a professional company, choosing instead to buy over the counter remedies commonly used for other species of ants. The residents were frustrated as these agents proved to be ineffective against the colonies as a whole. All residents reported an increase in the species with the small exception those who had major landscaping renovations before the study.

5.2 Locations and Movement

In Nova Scotia, affinity for coastal locations appears to hold true; most known colonies are within the Halifax Regional Municipality but others are found in coastal communities in many parts of the mainland. These coastal locations may be due to a preference for the milder maritime climate or its similarity to the UK climate. This assumes that the origin of all North American colonies is from western Europe which is possible given the first recorded infestation was Massachusetts in 1906 and attributed to potted plants brought over from England (Wheeler 1908). Given the history of Halifax as a port city having had primarily British colonization, it is not unreasonable to assume that early settlers may have brought plant materials with them as early as the 1700s. If so the species may have been present but in sufficiently small numbers to remained undetected until it reached a population density or location in the city by which to be noticed.

The locations of most of the ant infestations were concentrated in the urban core of the peninsula and appeared to radiate to other areas of HRM. This may be due to the fact that the greatest human population in the province is also found in HRM and therefore there is a greater chance of encounters. There is also the possibility that the greater media attention is focused in HRM therefore prompting more reports from the public.

The tracking of the spread of *M. rubra* has only begun during the last decade. This should be continued over time to measure the rate of spread and severity of the infestation. It is known that within five years of an introduction of infested plants there was an established colony at one resident's cottage. Undoubtedly new nests and colonies will radiate from that property and may spread unchecked for years due to the nature of the continuous habitat and land use. If the rate of spread in Nova Scotia is similar to a measured case in Maine, then there will be an area of around 100m x 70m in another 5 years. Case study 201011 also shows a rapid rate of spread. The owner believes the introduction of the species was six years previous to the study, in a shipment of firewood. On this property and the adjacent one (Case study 201010), the spread is already several hundred metres wide. It is unknown how many nests were transported in that shipment but there may have been multiple introductions onto the property. The leaf litter area next to the wood pile had an estimate of over one hundred nests and was too infested to accurately take measurements in. The ease at which this species appears to be spread may account for the increase in areas reported to have infestations.

Myrmica rubra increase the colony by budding and nuptial flights. Although only male swarms have been observed in North America (in Newfoundland only) (Hicks 2011), no spreading via nuptial flights have been confirmed. Therefore, the movement over ground is the considered the method of spread. The lack of nuptial flights is an advantage to management researchers. Budded groups have been observed moving nest sites in the grass and across open flat surfaces. However, it is thought that the movement across an open area has a risk of exposure and therefore there may be a limit of the distance a colony bud will travel with the queens. This is unknown at present, but based on observations, it is thought that the ants will cross a barrier as wide as a single driveway (or car width) but do not cross a double car width. If the ants were able to cross the physical barrier of a city street, this would show as a radiating pattern from an initial source of introduction. The evidence for this lack of radiating effect occurs frequently in the urban core. Here a grid system of primarily rectangular blocks have been used to show a distinct pattern. Blocks with known infestations were found across the street to blocks that were not infested. There was an almost chequered pattern in areas of peninsula of Halifax where severely infested blocks surrounded were surrounded by uninfested blocks. In some areas another infested block was found two streets over. This evidence suggested that the method of introduction was anthropogenic. This pattern has been shown in continuous habitat in Maine (Groden, personal communication 2011) and in Halifax (Case study 201026). Further evidence of this phenomenon occurred in areas near two case studies (201010 and 201011) which had very high numbers of nests. The residents live along a long continuous stretch of suburban/country road. There was a significant infestation on the case study side where both properties (neighbours) have continuous habitat. Across the two lane road from the properties, there was no evidence of *M. rubra.* A suggestion for further study would be to conduct and experiment testing what barrier width was needed to prevent colony budding across a flat exposed space.

Myrmica rubra may therefore partially depend on human transport. Species that use human vectors are considered "tramp" species. Characteristics of tramp species are their presence in significant numbers in urban habitats, unicolonial structure (or act similarly), are polydomous, and they may have lost the nuptile flight method of reproduction. They are opportunistic omnivores and often tend homopterans (Silverman 2005). *Myrmica rubra* could therefore be considered a tramp species.

5.2.1 Maps

In the micro-study, a comparison was made between the location of the nests and the ant densities. The results showed that the greatest densities were found near the greatest densities of nests. However, there were still areas where no ants were found in vials but the vials were located close to the nest sites. This may be due to the change in feeding pattern over the summer. *Myrmica rubra* feed more on sugar at the beginning and end of the summer and concentrate more on protein in July. It is also possible that the heat of the day was too great and created a greenhouse effect in the vials, thus preventing the ants from feeding. It is also possible that they were engaged in activities other than foraging or were staying close to the nests and the vials were beyond their movement range.

GIS maps were an important component to the understanding of the extent to which Halifax is infested. The area maps locating the reported sites were created using data collected from a variety of sources and based on reported incidents. This does leave an uncertainty as there will undoubtedly be a few areas that have had misidentification of species. However, in all the reported cases, only one resident had been incorrect. It appears that by the time people notice the problem and have sought information, they are quite accurate in identifying the species. The areas of the city where it was difficult to confirm the species were the large parks. In these cases a specific location would be needed to pinpoint the area to start a search. For example, the species was found in the stomachs of amphibians in a very large city park (Russell, 2011 personal communication). The salamanders' pond was located and a search conducted in a circular pattern from the edge of the water. After two hours and a hundred metre radius from the pond, no nests had been found. It is known for salamanders to forage several hundred metres away from their nesting site so it is possible the ant nests were quite far. This may have taken many working hours to find and is not feasible as a method of locating the presence of *M. rubra.* This would be an example of a use for the baiting technique used in the micro-study.

5.3 Habitat

In Halifax the majority of nests were found in ground flora but that may be more an indication of the availability of ground flora as opposed to other habitat types. Most properties had mostly grass as an available habitat. These results Figure 4.7 were clearly in contrast to the micro-habitats found in the rural environment in Maine where leaf litter accounted for 21%; downed woody debris 17%; soil under tree roots 13%; soil mound nests in herbaceous roots 6%; and soil nests 2% (Groden et al. 2005). In Halifax, only one case study had a high number of leaf litter nests. This property bordered a mixed forest similar to those in Acadia National Park.

Downed woody debris was not often found in the urban environment where lawns are more manicured and loose nesting material cleared. One agreement between the data is the lack of open soil nests. Other species are known to build open soil nests by burrowing tunnels and excavating to the surface (e.g. *Lasius spp.)* but *Myrmica rubra* prefer to build against a surface.

It would be a useful exercise to focus on which specific plant types *M. rubra* are associated with. As was possibly shown in the ground flora nests, there may be a preference for certain plants such as dandelions which have a large tap root and several secondary roots. This may be for ease of movement through the soil or as a source of moisture or protection. Some plants are also known to attract ants due to their high sugar content or the associated homopterans.

For the purposes of further work in management of the species, it was hoped that a clear preference of habitat characteristic would be observed. As can be seen in the analyses of variables there were no strong correlations. It was found however that the soil type had a slightly acidic pH and that some of the soils were very low in pH. If *M. rubra* prefer and can tolerate an acidic soil then perhaps a more alkaline soil would deter them. An alternative investigation could be to investigate the preferred pH range of soil for nesting.

5.4 Species Diversity

According to residents there was a decrease in other ant species over the years as the *M. rubra* became more numerous. There was conflicting evidence of the abundance of other species. It was observed in case study 201001 that there was an increase in the native population of ants in the second season (2011) despite the increase in the presence of *M. rubra* nests. On identifying species found in the same area as the *Myrmica*, all five were a species of *Lasius*. The previous month, newly dealated *Camponotus* queens were observed searching for nest sites (Figure 5.1). There were none present during the collection of the species on that day the micro-study was conducted.

***Figure 5.1**: Camponotus sp. queen.(Source: author)*

It was observed at case study 201006 that *M. rubra* was sharing a nest with a *Lasius sp.* The nest was under a shallow stone and both ants had brood filling the chambers. The brood lay side by side and the ants came into contact but otherwise ignored each other. Unfortunately no camera was available at the time but a similar photo of *M. rubra* and brood sharing a nest with *Lasius flavus* shows a similar situation (Figure 5.2)[12].

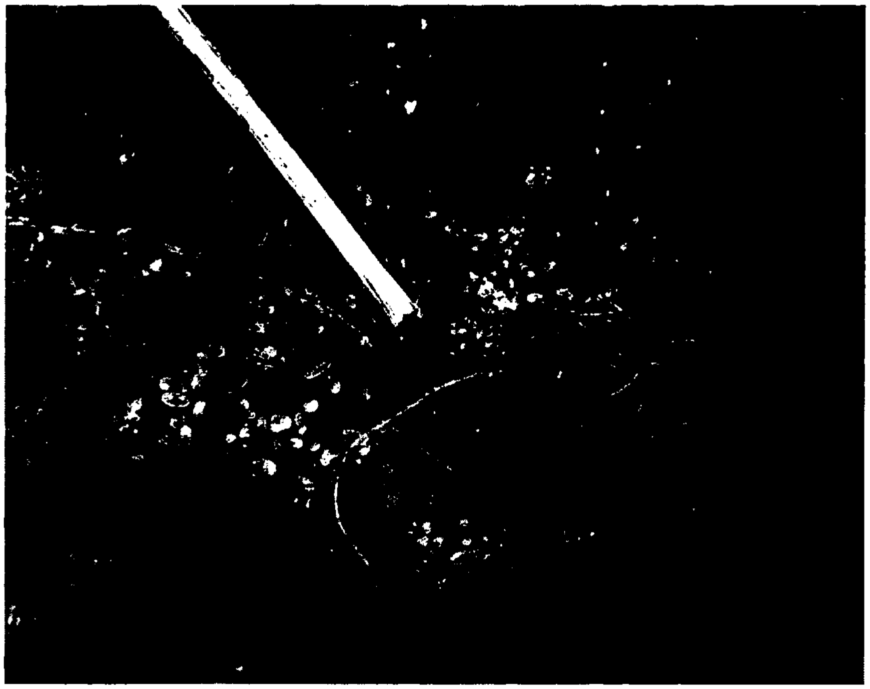

***Figure 5.2**: Myrmica rubra and Lasius flavus in the same nest. M. rubra can be seen on the left. The brood is M. rubra larvae. This nest was found in the native habitat in England. (Source: www.antwoodhill.co.uk)*

Another observation of *M. rubra* interacting with *Lasius* species occurred at case study 201001 where the *Lasius* had excavated an open soil tunnel and was creating chambers and the *Myrmica* appeared to be investigating. The tunnel was new that day and this was the only time the two species were seen together Figure 5.3. On closer inspection it appears the *Myrmica* were depositing cadavers close to the tunnel. For what purpose is unknown.

12 http://www.anthillwood.co.uk/anthunting.htm

Figure 5.3: *Lasius sp. (black garden ants) excavating nest in open soil with Myrmica rubra workers and cadavers on edge.(Source: author)*

5.5 Invasive Species

Myrmica rubra has been transformed from an introduced species in North America into an invasive. In all reported locations the presence of this species caused problems for people and the environment. Methods of introduction of non-native species vary depending on the ecosystem the introduced plant or animals inhabit and the characteristics of the species itself. Although the movement to and from many countries is now regulated, this legislation generally does not extend to the movement within countries. The sale of exotic species in the horticulture and aquaculture industries is still mostly unregulated. This has led to a number of inadvertent introductions where people share specimens or dispose indiscriminately of unwanted products. During presentations with the public and various horticulture groups in Halifax it was discovered that very few people are aware of the possibility of spreading non native species, and fewer still about the ease at which *M. rubra* may be spread.

In their 2002 article, Byers et al. also produced a thorough and detailed table identifying gaps in general research of invasive species. They suggest directions for studies and indicate data needed to complete these missing puzzles. Their suggestions can be adapted to help direct investigations of *M. rubra* by determining the gaps in general knowledge, of which there are many in it's invasive form. A selection of questions considered for research may be: 'Why do many invasions fail or have minimal effects but this one succeed?'; 'Is timing important?'; and 'Is success contingent on historical factors?'. Another category in the table, 'Types of studies or data required', could lead researchers to investigate whether established non-indigenous species are increasing in spatial extent or density, or whether indirect effects of non-indigenous species alter ecosystem properties.

5.5.1 Pest Control

In case study 201001 a poisoned bait trap, containing a 1% Boric acid solution was set near a disturbed nest where many ants were killed. Due to the movement of the trap some streaks of liquid poured out. After two hours, cadavers of *M. rubra* were found along the liquid runs (Figures 5.4 and 5.5). At first it was assumed the ants had fed and must have been highly susceptible to the poison, and therefore died at the source. This would have been unusual given the number of ants which regularly feed on this mixture elsewhere in the yard with no instant mortality. The feeding was watched for five minutes and it was noticed that the healthy workers were carrying the cadavers to the trap and placing the dead on the sticky liquid, using it as a makeshift midden pile (Figure 5.4).

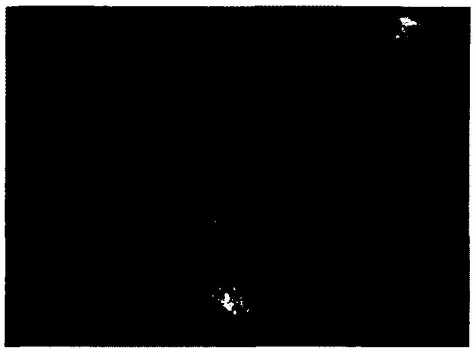

Figure 5.4*: Midden piles on the sweet solution leaking from a bait trap.(Source: author)*

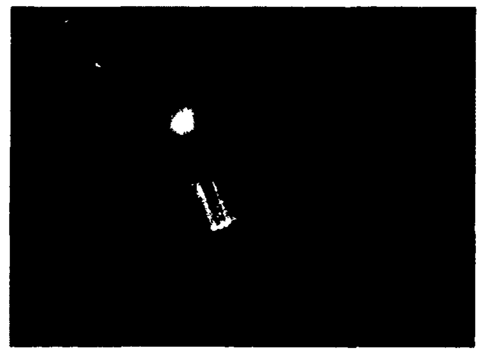

Figure 5.5*: Bait station beside a disturbed nest. Cadavers can be seen lining the streaks of solution.(Source: author)*

Midden piles have been observed at many locations. At one pumping station there were thousands of ant cadavers amongst the gravel lining the driveway. The infestation was not great if using the nest density as an indicator but the resulting number dead was large. There may have been nests on adjacent properties and the unoccupied station was used as a depository. It has been observed that *M. rubra* generally leave cadavers on open plots more than shaded plots (Graham et al. 2007). Although many cadavers were found on rocks, leaves, paving slabs and open ground, it was also found that during periods of heavy rain, the ants would come into a cool, relatively drier structure, and deposit their midden there. This phenomenon occurred at another pumping station where many cadavers were found inside the door of the cool, moist building. It was also observed on many occasions at case 201001 that the ants had gone into a garage and deposited the dead before returning outside. It is unexplained. A suggestion for an investigation could be to create a choice between a wet outdoor environment and a cool moist indoor environment and record whether the ants choose this or whether this was purely coincidence.

5.6 Observations

During the period of investigation, many observations were made that were not directly measurable. As information on this species is needed, behavioural observations may help identify topics for further research.

- One member of the public reported that several 'thousand' ants would regularly cover her electrical cords (for garden lights) in the plant pots. Other ants such as *Solenopsis invicta* are known to be attracted to electricity (Vinson and McKay 1990).

- *Myrmica rubra* carrying petals of vetch to the nest. This may be for the sugar content. They have also been observed feeding on peonies, also a sweet flower.

- One participant's house had approximately twenty *M. rubra* ants in the carpet scouting for sweet food left by the children. The ants entered the house by climbing up the foundation and accessing the floor through cracks. The encounters resulted in stings when the children unwittingly stepped or sat on the ants. Ants invading the house is the fear many people have. This appeared to be an isolated incident.

- *M. rubra* do not tend to follow linear trails of scent trails like many Formicines

- When searching for nests in ground flora, it was easier to find evidence of excavated nest chambers in June as there were small piles of dried soil throughout the grass. The soil mounds were present in July but not as common. More disturbance was needed to identify the ground nests. This may mean the establishment of satellite nests mostly occurs in June.

- The average size of nests in ground flora was: L x W x D= 20cm X 25cm x 15cm. Nest sizes under hard substrates such as stones or paving slabs would vary slightly depending on the size of the object the nest was under. One nest found under a stone was found to be 35cm x 20 cm x 20cm (case 202010)

- An interaction with another invertebrate was observed many times. Under substrates that had woodlice in high numbers there were no *M. rubra* found.

- Large numbers of ants did not always indicate a nest. High densities of ants were found found in and around a food source such as a caterpillar or worm or on detritus such as animal faeces or rotting dog bones

- They were frequently observed farming aphids or moving vertically on surfaces. The foraging was not confined to the ground.
- Many abandoned ground flora nests were found during searches. This increased in September after the hurricane and subsequent temperature drop.
- male and female alates were recorded in nests between mid-July and September

5.7 Further Work

Throughout the discussion, recommendations for further work have been suggested. The main focus of this investigation was to gather baseline data which can be used to monitor changes over time and to assist in focusing further investigations. The most important work that the public would like researched, would be an eradication method or management technique. There is continuing research on various pathogens at the University of Maine but alternative studies could take place on more traditional methods such as:

- Using chickens to naturally control population growth. It is unknown as to whether chickens would eat this species of ant and whether this would make any difference in the population if they do predate upon them
- Digging up nests as soon as they are located both in conjunction with the use of chemical control and without

- Further investigate the use of professional companies. It would be useful to take the micro-study city block and using a professional company treat all properties up to the edge of the park. This would hopefully show whether reinfestation is occurring from within or from an outside source.

- Continued monitoring of the results from properties that have had pest control treatments.

- Investigating the best use of the AntPro borax bait stations including what distance and density of stations is needed for effective control. The recommended number and distance on the information leaflet does not seem to be adequate for this species.

- Scientific testing of other labour intensive but easily accessible remedies such as boiling water, various surfactants, and home made borax trap designs.

The public want to know what they can do. It is also important to conduct a full survey of HRM (and perhaps Nova Scotia) to determine the extent of this infestation and create predictive tools. The idea of understanding the various characteristics of habitat choice was ultimately to be able to predict whether a property was susceptible to an invasion. Further understanding of the specific characteristics of the micro-habitat, obtained by comparing infested and non-infested areas would yield greater detail.

This species has been shown to be extremely adaptive to the maritime climate and is tolerant of extremes of many variables. The spread is relatively rapid and there is no known eradication. This is primarily an urban problem in Nova Scotia but the species has already been confirmed in the agricultural region. The consequences of this species infesting farms dependent on human pickers could be economically devastating. It is also a well known forest species as can be seen on the properties of those participants bordering wooded areas and more importantly in the forests of Acadia National Park. It is imperative that further research be conducted based on knowledge gained in this and other investigations.

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Appendix A Resident Questionnaire

SAINT MARY'S UNIVERSITY SINCE 1802
One University. One World. Yours.

Case # ____________

REB File # 10-104

European Fire Ant Questionnaire 2010

Questions are divided into tick box answers and written answers. Please print. Feel free to ask for clarification at any time. Answer to the best of your knowledge.

Name: ______________________________

Address: ______________________________

____

email address: ______________________________

Phone: ______________________________

Section 1. The House and Neighbourhood

1. **How long have you lived in this house?**
   <1 year ____ 1-5 yrs ____ 6-10____ 11-15____ 16-20____ 21-25____ 26-30____ >30___ if so how many?______
2. **When did you first notice these ants?** ______________**or** were they present when you first moved in yes/no. If so, did the previous owners or neighbours tell you how long the ants have been present? yes/no If yes when? ____________________
3. **When did you start having problems** (eg getting stung, pets getting attacked)? __________________
4. **How large is your property?** ____________________

5. **Estimate what percentage of your property you think ants are present.** ________%
6. **Have any of your neighbours complained about ant problems?** yes/no
7. **Could you mark an x on the image of where you think ants are present in your area** (attach a sketch of house and grounds)

Section 2. <u>The Yard/Garden</u>

8. **Have you (or a previous owner/tenant) planted or transplanted any vegetation?** yes/no If yes, **where have you bought plants from?**

   ____

   ____

9. **Where have you transplanted plants from?**

   ____

   ____

10. **Have you (or a previous owner/tenant) altered the landscape by...** - tick all that apply:
    adding rocks ____ paving slabs ____ weed suppressant ____ mulch ____ wood chips ____ gravel ____ logs ____ garden furniture ____ other decorative structure (eg statues, planters etc),- if so list items

    ____

11. **Have you removed any of the above items?** yes/no. If yes which?

    ____

12. **Have you altered the soil by...**- tick all that apply:
    adding fertilizer ____ compost ____ sand ____ silt ____ clay ____ a pH buffer such as lime(stone) ____ other?

    ____

13. **Have you transplanted plants or materials to other parts of the city or country** (i.e. to a cottage)? yes/no **Are the ants present in this new location?** yes/no/unsure

Section 3. <u>The colony</u>

14. **Have you found any nests (nests =many ants with eggs and/or larvae)?** yes/no/unsure
15. **How many did you find?** _____
16. **Where did you find them?** Tick all that apply:

    under stones ____ under dry wood ____ under logs ____ under paving slabs ____ under brick ____under plant pots ____ under garden furniture (indicate wood, plastic, metal , glass) ____ under/in leaf litter ____ under/in tree roots ____ under/in plant roots ____ under/in exposed soil____ in plant pots ____in grass____ other ___________________
17. **In areas where you find nests what plants are present?** please list

    ____

    ____

    ____

    ____
18. **Have you found any changes in the numbers of ants from year to year?** yes/no
19. **Have you found any changes in the numbers of ants from year to year?** yes/no

    If yes, did you notice any changes in the environment such as:

    weather - wetter ____ drier ____ warmer ____ cooler_____

    plants - more ____ less ____ change in types

    ____

    soils/ground - mulch, chippings etc

    ____
20. **Other species -** have you noticed more or less of other species such as other ants, woodlice, earwigs, spiders, etc?

    ____

21. **Have you noticed an increase or decrease of the areas affected by the spread of fire ants** (year to year or season to season)

____

Section 4. <u>Pest Control</u>

22. **Have you (or previous resident) used any store bought pesticides?** yes/no. If yes what were they

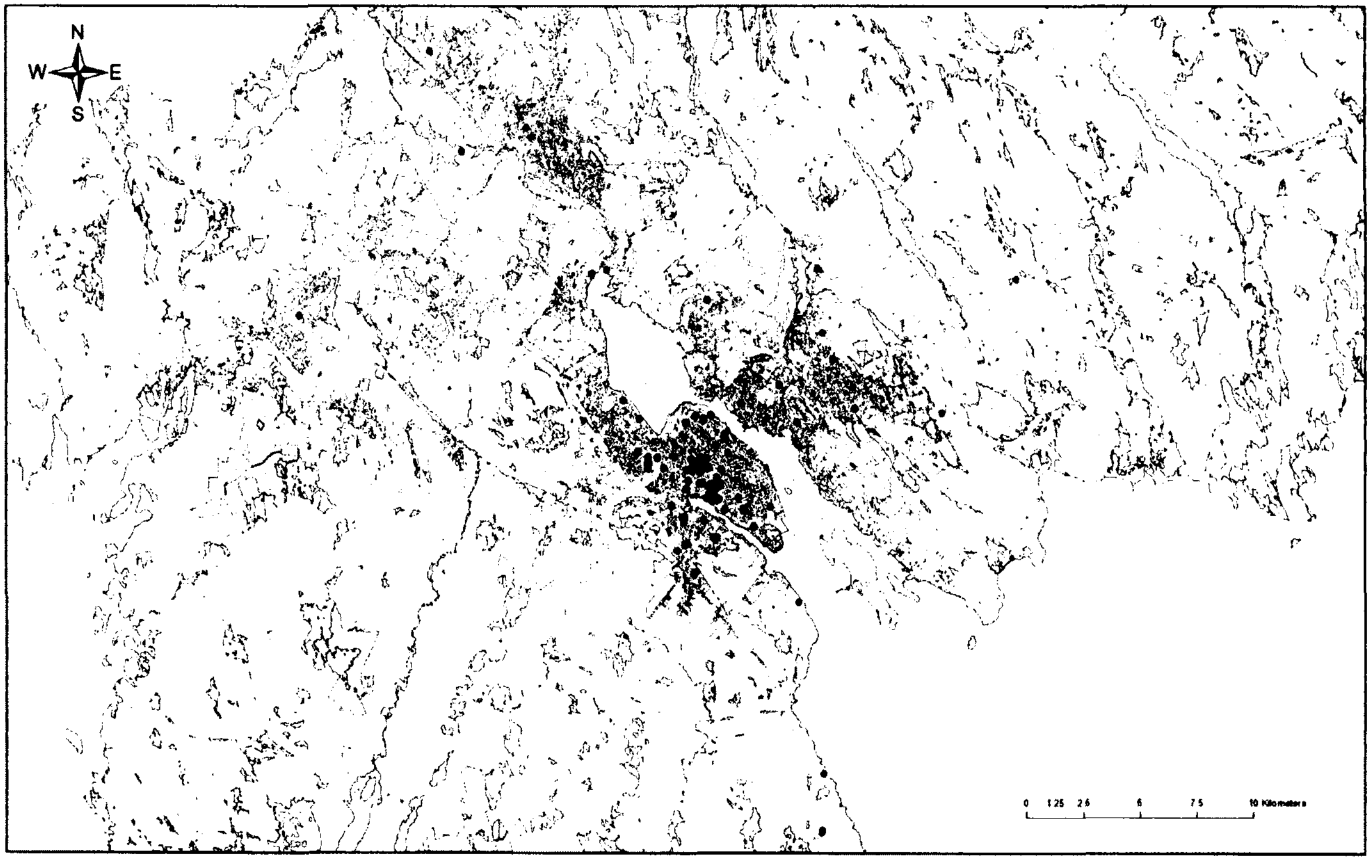
    ____

23. **Have you ever used a pest control company?** yes/no. If yes how long ago? __________
24. **How many applications did you have?** _____________
25. **Have you noticed any changes in the ant numbers since the application(s)?** large decrease/small decrease/no change/small increase/large increase
26. **Have you noticed any changes in the number of any other species since the application(s)?** Y/N/unsure

Any other comments:

Appendix B Geocoded HRM Detailed Map

Myrmica rubra locations in the Halifax Regional Municipality 2010



• Myrmica rubra	street		lake
coast	road		park

Halifax Regional Municipality. The red symbols show the locations of *M. rubra* as reported by the public. Note the concentration in the Halifax-Dartmouth metro area.

Halifax Regional Municipality Corporate GIS, scale varies. Halifax Regional Municipality, 2009

Susan Horton. Saint Mary's University 2011

Appendix C Questionnaire Results

CaseNr Q1 Q2 Q3 Q4 Q5 Q6 Q8 Q9 Q10 Q11 Q12 Q16 Q17/19 Q18 Q20
Q21 Q22 Q23 Q24 Notes
201001 <1 2010 2008 ~4000 100% Y N N N N grass, under stones Y/In N

201002 2000 2000 ~5000 100%

201003

201004 1-5 2005 2005 ~5000 65% Y Sobeys, Superstore, Walmart, Clems, South Shore, Valley WS, M, planters
N F, ComN perennials Y/In In-spiders, earwigs Y-ant traps N NA NA NA
201005 ? 2003 2003 ~4000 100% N Glace Bay R, WS, M N F, L N Y/In De-
spiders, earwigs, ants Y-ant traps N NA NA NA
201006 16-20 didn't notice ~4000 ? Y N N N N N N N
NA NA NA doesn't use yard
201007

201008 21-25 didn't notice no problems ~4000 ? Y Green house on property PS, G N F, L N
no change ants, earwigs Y- snail & earwig Y- 2008 2 no change Unsureno nests found on property but
ants found in sucrose vials
201009 16-20 2003 2003 ~10000 100% Y Garden centres, school plant sales, yard sales R, M N C
N Pennenials -hostas, spirea, irises, grass, lilacs Y/In De-native species N Y- 2010 2 increase
Unsuretwo months since pesticide application
201010 57 2006 2006 ~4500 90% Y Superstore PS, G Y-gravel F N Y/In
Sevin- cholinesterase inhibitor N
201011

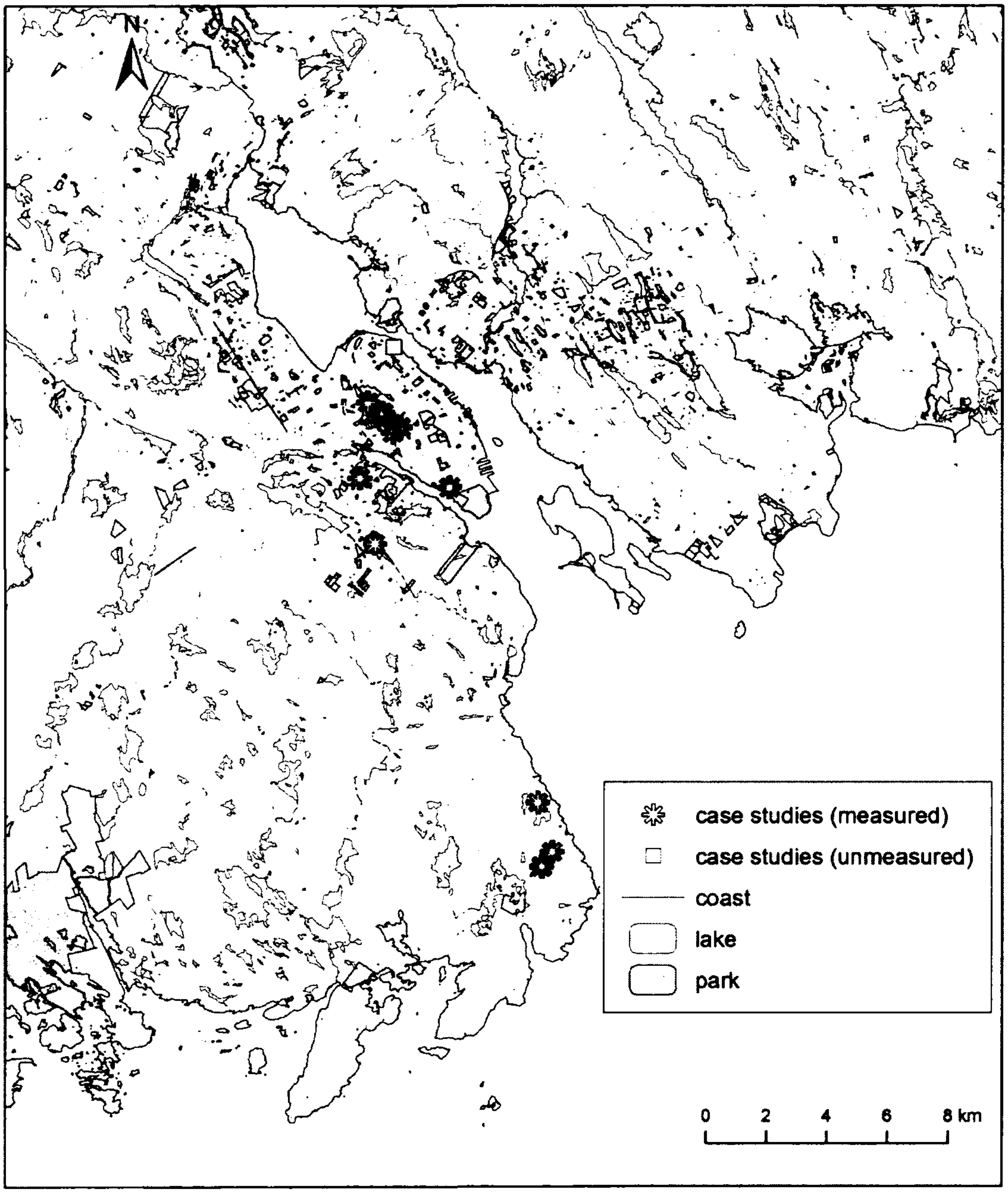
201012 6-10 2002 2002 ~45000 65% Y Bark mulch from Truro G N N no change few other species Y- Ortho Home Defense Max N
201013 1-5 2008 2008 ~4000 75% Y Halifax Seed WS, M, GF WS, MF, ComN Pennenials -hostas, poppies, lilies Y/In no other species Y-ant powder Y-20103 unsure Unsure Pesticide application one month before study
201014 6-10 2006 2006 ~6000 100% Y GF, herbs Y- old rotten wood beams N grass, herbs Y/In Y-borax, other home made remedies N very high infestation
201015 11-15 1999 1999 ~550000 20% Y Sobeys, Superstore, Blomidon Nurseries G N N apple tree, grass weeds Y/In More aphids, less spiders, less black ants except where fewer fire ants then more carpenter ants Y- basic ant control N Fewer black ants & carpenter ants in house over past two years
201016 6-10 2007 2007 ~4000 50% Y Farmer Clems, Kent, Rona, Home Depot, Halifax Seed R, PS, M, G, sand, stones Y- old concrete slabs F, S, L, soil, new sod in backyard N perennials- peonies, dahlias, lilacs, hostas, azaleas, daisies, crocuses, lilies Y/decrease In-spiders, earwigs. black ants Y- borax (3-4 times each summer 2010 & 2009) N decrease less fire ants & more black Pesticide app Qs relate to home made borax solution
201017

201019 16-20 1999 1999 ~4000 100% Y grass, under stones Y/In

201022 21-25 1985 1994 ~5000 100% Y Transplanted plants from previous address 1985. Plum trees & cherry tree bought 1990 from Lakeland Nurseries. Blueberry bushes from Brickton (Valley). Leaves from multiple sources, wood from multiple sources. PS, WS, M, WC, G, L deck 1990, raised beds Y- rotten wood Com, Sand, L, LL Y/Y all plants Y/In fewer other ant species Y-Borax 5%, Diatomaceous earth N Heard 30-35 years from other residents. Has transported contaminated leaf litter to cottage in last 5 years and now has ant problem there.
anon 1992 1992 ~5000 100% Y-Bug be gone spray temporary decrease Fewer black ants & carpenter ants in house over past two years

Appendix D Case Studies Map

Studied nest sites of *Myrmica rubra* in HRM



Source:Halifax Regional Municipality Corporate GIS; scale varies. Halifax Regional Municipality, 2009 *Susan Horton. Saint Mary's University 2011*

Appendix E Vegetation and Substrate Tally Table

CaseNr	StudyNr	NestNr	V_Moss	V_GrassRt	V_WeedRt	V_TreeRt	V_RotWood	V_ShrubRt	V_LeafLit	V_Compost	V_Mulch	S_Soil	S_Gravel	S_WeedSupp	S_PaveSlab	S_Stones	S_Foundatn	S_Plastic	S_Wood
201001	1	1		1														1	
201001	1	2		1														1	
201001	1	3		1	1														
201001	1	4		1	1														
201001	1	5		1	1														
201001	1	6		1	1														
201001	1	7		1	1														
201001	1	8							1										
201001	1	9							1							1			
201001	1	10							1			1							
201001	1	11							1							1			
201001	1	12		1	1														
201001	1	13		1	1											1			
201001	1	14		1	1														
201002	1	1		1	1														
201002	1	2						1											
201002	1	3		1															
201002	1	4		1															
201002	1	5		1															
201002	1	6		1															
201002	1	7						1								1			
201002	1	8						1								1			
201002	1	9						1											
201002	1	10													1				
201002	1	11													1				
201002	1	12													1				
201002	1	13													1				
201003	1	1	1																
201003	1	2		1	1														
201003	1	3		1	1														

201003	1	4		1	1							
201003	1	5		1	1							
201003	1	6		1	1							
201003	1	7										1
			1									
201003	1	8										1
				1								
201004	1	1									1	
201004	1	2									1	
201004	1	3										1
			1									
201004	1	4										1
			1									
201005	1	1			1							
201005	1	2			1						1	
201005	1	3		1		1						
201005	1	4			1							
				1								
201005	1	5			1							
				1								
201005	1	6										1
						1						
201005	1	7		1	1							1
201005	1	8		1			1					1
201005	1	9					1					
201005	1	10									1	
201005	1	11		1								
201005	1	12										1
				1								
201005	1	13				1						
				1								
201005	1	14		1								1
201005	1	15		1								1
201005	1	16										1
		1										
201006	1	1	1	1		1						
201006	1	2		1								
					1							

Continued for all case studies

	Totals	9	134	104	23	6	20	44	5	25
105	11	15	28	60	10	14	28			

	Full Names		Moss	Grass Root		Weed Root		Tree Root	Rotten Wood	
Shrub Root		Leaf Litter	Compost	Mulch	Soil	Gravel	Weed Suppressant		Paving Slab	
Stones	Foundation		Plastic	Wood						
		9	134	104	23	6	20	44	5	25
105	11	15	28	60	10	14	28			

Appendix F Microstudy Density Table

Densities of *M. rubra* in grid form for each case study on the micro-study city block.

CaseNr	Date	Row	Column	X	Y	Rubra	Native
201001	Aug 16, 10	1	B	5	0	Z	L
		1	C	10	0	Z	Z
		2	B	5	5	Z	L
		2	C	10	5	L	Z
		3	C	10	10	Z	Z
		4	C	10	15	Z	Z
		6	A	0	25	Z	L
		6	B	5	25	Z	Z
		6	C	10	25	L	Z
		7	B	5	30	Z	Z
		7	C	10	30	L	Z
		8	B	5	35	Z	Z
		8	C	10	35	L	Z
201006	Aug 6, 10	1	A	0	0	L	Z
		1	B	5	0	L	Z
		1	C	10	0	L	Z
		2	A	0	5	M	Z
		2	B	5	5	M	Z
		2	C	10	5	L	L
		3	A	0	20	L	Z
		4	A	0	25	L	L
		4	B	5	25	Z	Z
		4	C	10	25	Z	Z
		5	A	0	30	L	Z
		5	B	5	30	Z	L
		5	C	10	30	Z	Z
		6	B	5	35	Z	L
201007	Aug 9, 10	1	A	0	0	Z	L
		1	B	5	0	Z	L
		1	C	10	0	Z	Z
		2	A	0	5	Z	L
		2	B	5	5	L	Z
		2	C	10	5	L	Z
		3	A	0	10	Z	L
		3	C	10	10	Z	Z
		4	A	0	15	L	Z
		4	C	10	15	Z	Z
		5	A	0	20	L	Z

CaseNr	Date	Row	Column	X	Y	Rubra	Native
		5	B	5	20	Z	Z
		5	C	10	20	Z	Z
		6	A	0	25	M	Z
		6	B	5	25	L	Z
		6	C	10	25	L	Z
		7	A	0	30	L	Z
		7	B	5	30	L	Z
		7	C	10	30	L	Z
201008		1	A	0	0	L	Z
		1	B	5	0	L	L
		1	C	10	0	Z	L
		2	A	0	5	Z	L
		2	B	5	5	Z	L
		2	C	10	5	Z	M
		3	A	0	10	Z	Z
		3	B	5	10	Z	L
		3	C	10	10	Z	M
		4	A	0	15	Z	L
		4	C	10	15	Z	L
		5	A	0	20	Z	L
		5	C	10	20	Z	L
		6	B	5	25	Z	L
		6	C	10	25	Z	L
201010	Aug 13, 10	1	B	5	0	Z	L
		1	C	10	0	Z	L
		2	B	5	5	Z	L
		2	C	10	5	Z	L
		3	B	5	10	Z	Z
		3	C	10	10	Z	Z
		4	A	0	15	L	Z
		5	A	0	20	M	Z
		5	B	5	20	L	Z
		5	C	10	20	L	Z
		6	A	0	25	L	Z
		6	B	5	25	Z	Z
		6	C	10	25	M	Z
		7	A	0	30	H	Z
		7	B	5	30	H	Z
		7	C	10	30	H	Z
201013	Aug 20, 10	1	B	5	0	M	Z
		1	C	10	0	H	Z
		2	A	0	5	Z	Z
		2	B	5	5	L	Z
		2	C	10	5	M	Z

CaseNr	Date	Row	Column	X	Y	Rubra	Native
		3	A	0	10	Z	Z
		3	B	5	10	M	Z
		3	C	10	10	M	Z
		4	A	0	15	Z	Z
		5	A	0	20	Z	Z
		6	A	0	25	M	Z
		6	B	5	25	Z	Z
		6	C	10	25	L	Z
		7	B	5	30	Z	Z
		7	C	10	30	M	Z
201016	Sep 15, 10	1	A	0	0	Z	Z
		1	B	5	0	Z	Z
		1	C	10	0	Z	Z
		2	A	0	5	Z	Z
		2	B	5	5	L	Z
		2	C	10	5	Z	Z
		2	D	15	5	Z	Z
		3	A	0	10	Z	Z
		3	C	10	10	Z	Z
		3	D	15	10	Z	Z
		4	A	0	15	Z	Z
		4	C	10	15	Z	Z
		4	D	15	15	Z	Z
		5	A	0	20	Z	Z
		5	B	5	20	Z	Z
		5	C	10	20	Z	Z
		5	D	15	20	Z	Z
		6	A	0	25	Z	Z
		6	B	5	25	Z	Z
201017	Sep 21, 10	1	B	5	0	Z	Z
		1	C	10	0	Z	Z
		2	A	0	5	Z	Z
		2	B	5	5	Z	Z
		2	C	10	5	Z	Z
		3	A	0	10	Z	Z
		3	B	5	10	Z	Z
		3	C	10	10	L	Z
		4	A	0	15	Z	Z
		5	A	0	20	Z	L
		5	B	5	20	Z	Z
		6	A	0	25	L	Z
		6	B	5	25	Z	Z
		6	C	10	25	Z	Z
		7	A	0	30	L	Z

CaseNr	Date	Row	Column	X	Y	Rubra	Native
		7	B	5	30	L	Z
		7	C	10	30	Z	Z
201018	Aug 1, 10	1	A	0	0	Z	Z
		1	B	15	0	Z	L
		1	C	30	0	Z	Z
		1	D	45	0	Z	L
		2	A	0	12	Z	Z
		2	B	15	12	Z	Z
		2	C	30	12	Z	Z
		2	D	45	12	Z	L
		3	A	0	24	Z	Z
		3	B	15	24	Z	Z
		3	C	30	24	Z	Z
		3	D	45	24	Z	Z
		4	D	45	36	Z	L
201019	Sep 25, 10	1	B	5	0	Z	Z
		1	C	10	0	Z	Z
		1	D	15	0	Z	Z
		2	A	0	5	Z	Z
		2	B	5	5	Z	Z
		2	C	10	5	Z	Z
		2	D	15	5	Z	Z
		3	A	0	10	Z	Z
		3	B	5	10	L	Z
		3	C	10	10	Z	Z
		3	D	15	10	Z	Z
		4	A	0	15	Z	Z
		4	B	5	15	Z	Z
		4	D	15	15	Z	Z
		5	A	0	20	Z	Z
		5	B	5	20	Z	Z
		6	A	0	25	Z	Z
		6	B	5	25	Z	Z
		7	A	0	30	Z	Z
		7	B	5	30	L	Z
		7	C	10	30	L	Z

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