USE OF PERENNIAL LEGUMINOUS LIVING MULCHES FOR THE FERTILIZATION OF BROCCOLI IN AN ORGANIC AGRICULTURE SYSTEM

by

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A thesis submitted to McGill University
in partial fulfilment of the requirements
of the degree of

MASTER OF SCIENCE

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June 2008

CONTRIBUTION OF CO-AUTHORS TO MANUSCRIPTS

This thesis has been written in the form of manuscripts to be submitted to scientific journals. The content of chapters 3 and 4 corresponds to two manuscripts that will be submitted for publication in the International Journal of Vegetable Science. The candidate was responsible for designing and conducting the field and laboratory research experiments, and for preparing the thesis dissertation and manuscripts. Dr. Philippe Seguin and Katrine Stewart, Professors in the Department of Plant Science at McGill University, and co-authors of the manuscripts, provided supervisory guidance for the whole duration of the project and edited the manuscripts for publication.

ABSTRACT

Use of alfalfa and red clover living mulches and green manures as alternative, biologically-based, means of fertilizing organic broccoli was studied. Both green manures increased broccoli head weight, hollow stem incidence, SPAD readings, total N content of the plants, and soil available N. These green manures can supply N to a broccoli crop in excess of the recommended dosage and produce satisfactory yields in organic production systems. Living mulches reduced broccoli yields (head weight and diameter), reduced N uptake (SPAD readings), even though they increased soil N availability (especially alfalfa). Living mulches appear to have the potential to supply N to broccoli, but cropping practices should be modified to limit competition. When row covers and living mulches are used together, the creation of a sheltered microenvironment provides better conditions for imported cabbage worm (*Pieris rapae)* larvae, which, in this experiment, were more abundant in living mulch plots.

RESUME

L'utilisation du trèfle rouge et de la luzerne en paillis vivant et en engrais vert pour la fertilisation biologique du brocoli a été testée. Les deux engrais verts ont augmenté le poids du brocoli, la présence de cœur creux, l'absorption d'azote (SPAD), l'azote total du brocoli et l'azote disponible du sol. Ces engrais verts peuvent fournir plus d'azote que les doses recommandées et assurer des rendements satisfaisants en régie biologique. Les paillis vivants ont diminué les rendements et l'absorption d'azote (SPAD), mais ont augmenté l'azote disponible du sol (surtout la luzerne). Les paillis vivants ont le potentiel de fournir de l'azote au brocoli, mais la régie doit être modifiée pour limiter la compétition. Lorsque des bâches flottantes sont utilisées en combinaison avec les paillis vivants, un microenvironnement est créé. Nous avons observé une plus grande abondance de *Pieris rapae* dans les paillis vivants.

AKNOWLEDGEMENTS

I would like to give special thanks to my co-supervisors, Dr Philippe Seguin and Dr Katrine Stewart, without whom this project would not have been as successful. They provided necessary moral, technical and academic support and insightful comments throughout the project, it has been very appreciated.

Special thanks also to the whole team at the Horticulture Center. First ones first, Michael Bleho who provided technical and mechanical assistance, performed important operations for the project, and shared his workforce in times of crunches. Simon for his great mechanical skills, Laurence, Rudy, James, Laura, and Jen for their timely help.

Thanks to the seed farm team as well, where the fun drying and grinding work has happened. Thanks to Amélie for her amazing willingness to help and for the numerous times where she saved my life, or at least made it easier. Thanks to Jolianne for showing me how to work the forage harvester, to Mike for sharing his dryer, and to Jim for letting me do what I needed to do.

Thanks to Bruce and Dagobiet who were very kind to take some time to show me how to work with the lab and field equipment. Thanks to Evan, Ruben, Wucheng and to Bruce again and especially for companionship in the lab and the office.

Thanks to Carolyn, Lise, and Roslyn for their support going through the red tape.

Thanks to the Fond Québécois de la Recherche sur la Nature et les Technologies (FQRNT) for providing me with the financial means to sustain myself for two years, while doing research about a topic that I liked and I feel is useful. Thanks also to the McGill University for reminding me to apply for their graduate studies fellowship.

Thanks to Dubois Agrinovation for their generous material contribution (irrigation and row cover) who relieved Philippe's headaches about how expensive my project could have been.

Thanks to Dan, Emily, Reid and Renée from the Ferme Coopérative Tourne-Sol, who now have to deal with my weeds, and who were very understanding and supportive when my masters work was impeding on farm time.

And finally heartfelt thanks to my love Dana, and to my family for their love and support throughout the whole project.

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1. GENERAL INTRODUCTION

1.1. Statement of the problem

In recent years, organic agriculture has been one of the most rapidly expanding sectors in the agri-food business (Cameron, 2004). This trend, observed at the consumer level, is confirmed by a 10.6 % increase in the number of Canadian organic producers between 2003 and 2004 (Macey, 2005). The Community Supported Agriculture (CSA) model has emerged as an interesting commercialisation alternative for organic vegetable producers in North America, and is especially well supported in Québec (Équiterre, 2008). This model brings farmers and consumers together in a "partnership" that gives more security for the farmer, and that insures a supply of high quality local organic produce to the consumer (Hendersen and Van En, 2007).

CSA farms are generally highly diversified, with complex rotations, and their access to manure is sometimes limited. Furthermore, with newly adopted governmental regulations about water protection and the environment in Québec, on-farm composting will be severely limited or prohibited (Weil, 2005).

Broccoli is an important crop in Québec, and is often produced on CSA farms. It has high nutrient requirements and needs large doses of compost. Farmers with limited or restricted access to compost must look for alternatives. The use of cover crops and legumes for biological fixation of nitrogen is one of these alternative options, their soil benefits are important for agricultural producers (Vanek et al., 2005). The goal of the present master's thesis is to contribute to the development and better understanding of leguminous perennial living mulch techniques that could be used by organic farmers to provide nitrogen and enhance fertility in vegetable crops.

1.2. Objectives for the project

- Determine and quantify the effect of perennial legume living mulches and green manures on organic broccoli (*Brassica oleracea* var *italica*) yields (head weight and diameter), earliness, nitrogen uptake, hollow stem incidence, and soil available nitrogen.

- Determine the effect of a living mulch system design including irrigation, mowing of the living mulch, and tillage of a strip on the competition between crop and living mulch, and the repercussions on broccoli yields.
- Quantify living mulch and green manure productivity and calculate estimates of the nitrogen applied through their biomass.
- Determine if application of living mulch clippings to the crop row will affect broccoli yields, nitrogen uptake, and soil available nitrogen.
- Quantify the effect of living mulches on *P.rapae* larvae presence in broccoli.

1.3. Hypotheses for the project

- Use of leguminous green manures or living mulches will result in broccoli yields, hollow stem incidence, plant nitrogen uptake and soil available nitrogen similar to those of plants fertilized with compost.
- The nitrogen supplied from alfalfa (*Medicago sativa*) green manures and living mulches will be greater than that of red clover (*Trifolium pratense*) similar green manures and living mulches.
- Applying living mulch clippings to the row will improve broccoli yields, plant nitrogen status and soil available nitrogen compared to a standard living mulch design.
- Use of living mulches will reduce *P.rapae* pressure in the broccoli crop.

2. LITERATURE REVIEW

2.1. Organic farming industry and principles

Agricultural production has changed markedly over the past 100 years with factory-made implements replacing animal and human labour and the use of artificial fertilizers (Grigg, 1984). After World War II, the jump in productivity was even more spectacular due to increases in fertilizer use, breeding of highly responsive and high yielding cereal varieties, and the advances in chemical control of diseases and pests (Evenson and Gollin, 2003).

After the release of "Silent Spring" by Rachel Carson (Carson, 1962), it became increasingly obvious that the widespread use of pesticides, and their residues on foods represented risks both for human health, and the environment. Links were made between degradation of water bodies and rivers, and the excessive use of fertilizers. It is now widely recognized that nitrogen from cropland can leach into water bodies causing algal blooms, eutrophication and death of marine organisms (Emmett, 2007). Phosphorus adsorbed to soil particles, which are eroded from the land and move into the waters, can cause the same effects. Nitrates, herbicides and other pesticides are sometimes found in water sources of cities and villages representing a risk for people's health and the environment (Jordan, 2004).

For many people, all these problems are a clear indication that the time has come to change our philosophy, and our food production methods. Modern organic agriculture is one of the many approaches that have emerged as a response to the concerns about the agricultural model that stemmed from the industrial revolution. The US Department of Agriculture defines organic agriculture as follows:

> *" Production systems which avoid or largely exclude the use of synthetically compounded fertilizers, pesticides, growth regulators and (...) rely upon crop rotations crop residues, animal manures, green manures, off-farm organic wastes, mechanical cultivation, mineral bearing rocks, and aspects of biological pest control to maintain soil productivity and tilth, to supply plant nutrients and to control insects, weeds, and other pests." (USDA, 1980)*

Organic agriculture is based on a number of basic principles, the most central of which is the maintenance of soil health (Lampkin, 1990). The concept that a soil rich in organic matter is healthier because of its physical and chemical properties and improved capacity to sustain microbial life and diversity is central to organic farming.

The organic philosophy values self-reliance on one's own knowledge and resources while limiting the use of off-farm inputs (Jordan, 2004). Organic farmers aim to increase on-farm biodiversity, and to prevent problems rather than curing them by becoming more integrated in natural cycles.

Practices have been developed which have become the basis for successful organic land stewardship and good vegetable production. Crop rotation is one of the principal means to prevent insects, diseases and weed proliferation. Use of long rotations and the inclusion of sod and legume crops to allow the soil to rest, and ultimately to benefit from the soil improving effects of these crops is widely practised (Grubinger, 1992).

Adequate fertilization is another challenge faced by organic producers. Plant nutrition in organic production is mainly based on the adequate supply of organic residues from crops and animal manures. Farmers often make their own composts, a laborious process, by mixing high nitrogen content materials such as animal manures or vegetable wastes with low-nitrogen components such as leaves, straw, or woodchips (Jordan, 2004).

In regions where it is available, manure is sometimes acquired by farmers at considerable cost (Grubinger, 1992), but in some regions availability is limited. Furthermore, organic certification agencies are becoming stricter in their requirements relative to the sourcing of manure used for composting (Garantie-Bio, 2006). For many plant-based operations, the availability of manures and composts in the future is a topic of concern.

Many of these plant-based operations are vegetable farms that use alternate fertilization strategies such as green manuring with legumes and other crops. They will sometimes combine cover crops with the addition of rock phosphates, other mineral bearing rocks, powders, and other natural substances like blood meal (Arcand and Lynch, 2006). Whichever their situation, organic vegetable growers are generally eager to experiment with new management strategies that can improve nutrient cycling, recycling and addition on their farms (Grubinger, 1992). The development of such new management strategies for soil husbandry and fertility improvement without the use of manures will be the focus of this research document.

2.1.1. Legumes used as green manures

A traditional and simple way to use legume crops to provide fixed atmospheric nitrogen (N) for organic vegetable production is as green manures. The crop is plowed down and its fixed N and nutrients are released by decomposition of the residues by the soil microbial flora, and are made available to the following crop (Lampkin, 1990). Guldan et al (1997) reported that alfalfa (*Medicago sativa*) and hairy vetch (*Viccia villosa* Roth) had fertilizer replacement values ranging between 78 and 140 kg N/ha when used as green manures. Triplett et al. (1979) noted that in two years out of three, adding fertilizer to a plowed down alfalfa stand did not increase grain corn (*Zea mays*) yields, and that the alfalfa green manure alone was able to provide adequate nutrition for the crop. When different grain legumes [soybean (*Glycine max*), cowpea (*Vigna unguiculata)*, pigeon pea (*Cajanus cajan)* and groundnut (*Arachis hypogaea*)] were grown with corn and the residues were plowed down, Nair et al (1979) found intercrops left enough residual fertility to significantly increase the yields of a following wheat (*Triticum aestivum*) crop.

2.1.2. Inclusion of soil improving crops in vegetable production

Soil improving crops (SIC) such as legumes can be used as a source of biologically fixed N, but also as a means of reducing erosion, of increasing soil organic matter, and of increasing availability of certain elements. They are also used to control weeds, and to reduce insect pressure and soil compaction. (Sarrantonio, 1992). The opportunities for the use of SIC in vegetable cropping systems are presented in Figure 2.1.

In a northern climate such as Montréal with a frost free period of 140 days and temperatures falling rapidly below freezing in the fall (Natural-Resources-Canada, 1995), opportunities to include SICs are more restricted than suggested in Figure 2.1. In scenario 1 for example, a SIC would need to be planted after the first killing frost (often in October), and then grow and establish through the cold fall to survive the winter under the snow. This is possible with winter rye (*Secale cereale* L.) and is done to keep the ground covered and reduce erosion, but the fertility and soil improvement benefits are minimal. In order to leave the SICs in the ground long enough for them to provide a beneficial fertility effect, one either has to dedicate an entire season to the growth of the green manure or grow the SICs alongside the main crop at least for a limited period of time such as in scenarios 2, 3, 6, 9 and 10. These scenarios involve the use of intercrops as living or dead mulches in a main vegetable crop.

2.1.3. Intercropping, Living Mulches and Dead Mulches

A good way to include SICs on vegetable farms is the use of intercropping, a form of cultivation where two or more species are grown in close proximity to promote their agronomic interaction (Theunissen, 1994). Varghese (2000) suggests that intercropping can lead to economies of space, and could increase the productivity of vegetables. Research has demonstrated that certain associations have great potential in field crops and in vegetables (Newman, 1986; Szumigalski and Van Acker, 2005).

Living mulches are a type of intercropping where one would add a new type of plant in a vegetable growing system – not with the intent of harvesting it, but rather with the intent of deriving some other benefits (weed control, insect control, fertility) from this associated crop. Living mulch systems can thus be seen as a special "temporal niche" in which both the main vegetable crop, and an associated crop (often a SIC) grow actively together, and interact agronomically.

The nature of that interaction determines how beneficial, and how viable a given main crop/ living mulch association will be. Brainard and Bellinder (2004) report that living mulches often suppress neither crop nor weeds or else they suppress both. The competition that can happen between the main and associated crop, and the ways to control it, has been a main focus of living mulch research (Kloen and Altieri, 1990; Nicholson and Wein, 1983; Vanek et al., 2005). Many no-till researchers (Abdul-Baki et al., 1997; Bottenberg et al., 1997; Mitchell and Teel, 1977), simply kill the associated crop, this option is called a dead mulch.

In scenarios 4 and 6 in Figure 2.1, Sarrantonio (1992) proposes the use of dead mulches. A dead mulch is the remains of a crop that has senesced naturally, died naturally from exposure to cold temperature or that was chemically killed. The remains are left undisturbed on the surface of the ground, and the main crop is no-till planted or transplanted into the dead organic matter mulch. Benefits from the use of dead mulches include: increased moisture for the crop, reduction of temperature and moisture fluctuations, production of allelochemicals by certain mulch species, nitrogen supply in the case of legumes (such as hairy vetch), and potential for weed control that lasts from 30 to 75 days after the crop is killed (depending on density of cover; Masiunas, 1998).

For organic producers the use of dead mulches is restricted to plants that will either senesce naturally or that do not overwinter. Fall cover crops such as oats (*Avena sativa* L.) or field peas (*Pisum sativum* L.) are commonly used by organic growers in

Québec (Weil, 2005), but with rapid spring decomposition of the mulch cover, the long term soil improvement and fertility potential of standard dead mulch systems may be low.

A possibility that has been explored very minimally in the scientific literature (Kleinhenz et al., 1997; Wiens et al., 2006) is to use the clippings from living mulches and spread them over a crop row. It is very similar to the dead mulch described above, and it has the potential to be renewed with each subsequent mowing of the living mulch. The literature that follows shows the extent of the potential benefits that could be expected from controlled and well designed use of living and dead mulches in modern organic agricultural production.

2.1.4. Living and dead mulches and their various potential benefits

Organic growers know that cover crops and living mulches can help to reduce pest populations as well as improve the quality of the soil (Bottenberg et al., 1997). Reduction of insect damage in the main crop is a broadly studied impact of living mulches (Andow et al., 1986; Costello, 1994; Hooks and Johnson, 2001; Hooks and Johnson, 2002; Hooks and Johnson, 2003; Kloen and Altieri, 1990; Theunissen, 1994; Theunissen, 1997). Andow et al. (1986) suggest that early season chemical treatment for flea beetles in cabbage (*Brassica oleracea* var *capitata*) could be eliminated when living mulches are used. Costello (1994) has shown that aphid infestation was consistently less in broccoli grown in legume living mulch systems compared with clean cultivation.

Weed competition can also be reduced though the appropriate use of living mulches. Adequate choice of living mulch species, seeding date, and method of repression are all factors that need to be fine-tuned for optimisation of results (Brainard and Bellinder, 2004; Infante and Morse, 1996; Masiunas, 1998; Tessier and Leroux, 1994).

Addition of organic matter to the soil and a reduction in soil compaction by the use of living/dead mulches has been reported by Abdul-Baki (1997). Akobundu (1980) noted improved water infiltration rates, improved soil structure, and increased microbial activity. Byers et al. (1994) report intercropping bell peppers (*Capsicum annuum*) and pumpkins (*Cucurbita maxima*) with fescue (*Festuca arundinaecea*) has reduced soil and water loss, and reduced the concentration of herbicides in runoff waters. Biological nitrogen fixation by the associated crop (when it is a legume) is another benefit of these systems (Costello, 1994). An increase in biological N fixation in an agroecosystem is linked to reduced dependence of farmers on the fertilizer industry, on imported manures, and on other inputs. Reduced use of fertilizers, especially N fertilizers, is linked to reduced greenhouse gas emissions from agriculture. Overall, Theunissen (1997) estimates that there is a potential to develop intercropping on a commercial base for agricultural sustainability (Theunissen, 1997).

There are also potential drawbacks and negative impacts of living mulches that one must be aware of. The possibilities to deplete soil moisture, to lower spring soil temperatures, and to disrupt field operations are risks to be considered. The choice of species will determine if there are potential risks of allelopathy, of creating a habitat for harmful organisms, or even to create nitrate pollution from improper management of legumes (Sarrantonio, 1992).

To avoid these problems, the living mulch system must be carefully designed. The next section will explore the different agronomic aspects necessary to consider.

2.2. Agronomic aspects of living and dead mulches

2.2.1. Reducing competitive ability of living mulches

The more aggressive a living mulch, the greater its biomass production, and the greater potential benefits in terms of reduction of insect or weed pressure. However a downside to this aggressive growing behaviour is that the living mulch is also more likely to compete with the crop for light and water, and to reduce yields. Nicholson and Wien (1983) state that some compromise must be made between improving soil tilth, and limiting competition between crop and living mulch. The choice of crop, living mulch and management system are critical. Hooks and Johnson (2003) working with intercropping for cruciferous crops noted several management strategies such as proper fertilization and watering, using vigorous main crop, using a less competitive background crop, increasing the distance between main crop and intercrop, adjusting planting times of both crops, planting background crop in narrow strips, and suppressing background vegetation (mowing, herbicides) helped to optimize yields.

2.2.2. Effect of living and dead mulches on crop yields

2.2.2.1. Nitrogen contribution from the legumes

2.2.2.1.1. Experiments in grain corn

Most of the literature dealing with the use of living and dead mulches has dealt with grain corn. Eblehar et al (1984) used legume grown as winter cover crops (crop planted in the fall and killed in the spring – dead mulch) for 5 consecutive years in corn. Hairy vetch dead mulch supplied between 90 and 100 kg N/ha to grain corn per year and significantly improved yield. Mitchell and Teel (1977) tested hairy vetch and crimson clover as dead mulches in grain corn. They found that the N supply from an oat-vetch cover represented 87% of the total-N absorbed by the fertilized corn. Yields from hairy vetch and crimson were similar to those obtained when fertilizing with 112 kg N/ha. They also observed that about a third of the N contained in the mulch was released and used by the corn in the first season and that most of the N released (90%) came from the top growth. When used as winter cover crops (dead mulches), the legume was sufficient to supply between 90 and 112 kg N/ha to the corn crop. The finding that 90% of that N came from top growth suggest that if living mulch is grown throughout the year and mown regularly, the clippings applied as a mulch to the main crop could potentially supply a significant amount of nitrogen, depending on above-ground biomass production.

2.2.2.1.2. Experiments in vegetable production

Skarphol et al. (1987) tested the effect of 3 legume dead mulches [hairy vetch, crimson clover (*Trifolium incarnatum*) and Austrian winter pea (*Pisum sativum* spp arvense)] on the growth and yield of snap bean (*Phaseolus lunatus*) in a conventional and no-till system. They found that when dead mulch was used, fertilization with 90 kg N/ha was not necessary to achieve maximum yields. Abdul-Baki (1997) used dead soybean and millet (Panicum miliaceum) cover mulches in broccoli. They observed that the soybean dead mulch provided part of the N required by the broccoli crop, although it did not fully meet the crop's requirements.

In Taiwan, (Kleinhenz et al., 1997) grew several vegetables [including Chinese cabbage (*Brassica chinensis*)] year round using a permanent living mulch system for 4 years. Tropical legumes (*Macroptilium atropurpureum, Desmodium incanum, Alysicarpus vaginalis, Centrosema angustifolium*) were grown on the edges of permanent beds. Legumes were clipped throughout the season as needed, collected and either spread as a mulch or rototilled into the soil. Use of the legume clippings as a mulch increased cabbage yields in the long run, but not the first year. The management of this system had a number of weaknesses including the use of raised beds (design consideration and work load) , the proportion of crop plant to legume (not enough legumes to provide the required N), and the impracticality of rototilling clippings which made this system too expensive and labour intensive to implement.

Costello (1994) grew broccoli in clean cultivation and in 3 different leguminous living mulch treatments [white clover (*Trifolium repens)*, strawberry clover (*Trifolium fragiferum*) and birdsfoot trefoil (*Lotus corniculata*) + red clover (*Trifolium pratense*)]. A 10 cm band was rototilled into the mulch, and broccoli was planted in the strip. When no N fertilizer was applied, plots with living mulch yielded better than those in clean cultivation, suggesting N contribution from the legume. It is not clear if the N came from a transfer from the intercrop or from residual N in the tilled strips.

2.2.2.2. Effect of Living Mulches on Yields of Crucifers

Andow et al. (1986) looked at the effect of white clover intercrops on cabbage yields. They established the living mulch early in the spring, tilled 30 cm strips and transplanted cabbage into them. They did not repress or mow the mulch after they planted the cabbage. The more vigorous type of clover shaded the cabbage and reduced yield substantially. They found that the greater was the living mulch's dry weight, the smaller were the cabbage yields. Short stature living mulches such as dwarf white clover resulted in less above ground interaction between cabbage and mulch.

Hooks and Johnson (2001; 2002) studied the impacts of undersown (when a mulch is seeded after the main crop has been transplanted) yellow sweet clover (*Melilotus officinalis*) on broccoli. The sweet clover was not repressed, and it surrounded and choked the broccoli. Broccoli height was greater in intercrop, indicating competition for light. Intercropped broccoli biomass was lower and heads were 48% smaller compared to a monocrop.

Bottenberg et al. (1997) grew cabbage in a red clover living mulch. Red clover was spring seeded, and cabbage was transplanted in June. They found that because of competition for water, red clover living mulch plots had the lowest yields. Eberlein et al (1992) obtained similar results with corn grown in strips tilled into an alfalfa living mulch. Yields were lower than in bare ground when no irrigation water was applied. However, there was no difference in yields in an irrigated system.

Strategies exist to restrain living mulch growth and to eliminate competition for water and light between crop and mulch. If one or several of these strategies are used in an integrated, well-designed system, can researchers achieve with living mulches yields similar or better than those of monocrops?

Using ***overseeded*** (equivalent to undersown) legumes (red clover, hairy vetch and 'Dutch' white clover) in broccoli, Infante and Morse (1996) showed no difference in yields compared with a monoculture. The legumes were overseeded 2 days before broccoli transplantation and irrigation was used, but no further methods were used to control living mulch growth. Costello (1994) obtained increased yields in a living mulch because the design used included irrigation of the broccoli, mowing of the red clover living mulch, and broccoli was also planted in 10 cm wide tilled strips (83% soil coverage). Kleinhenz et al (1997) also increased yields of Chinese cabbage using living mulches by applying legume clippings to the cropped row. Our hypothesis is that a living mulch system using irrigation, mowing of the living mulch, tillage of a strip for the main crop and application of clippings to the crop row will allow for increased crucifer yields.

2.2.3. Legumes Versus Other Species of Living Mulches

Several kinds of crops can be used as living mulches. Their plant family, their growing habits, composition, and competitiveness will all influence the way they behave as living mulches, and what impact they will have on the main crop. The choice of the appropriate species will depend on local growing conditions and on the specific objectives pursued by the researcher or farmer. Masiuna (1998) in his review identifies red clover as one of the most promising crops to use as living mulches in brassicas. Red clover has a more erect growth habit than other types of clovers, and doesn't propagate vegetatively. It is thus less likely to run into the crop row and compete with the crop.

Kloen and Altieri (1990) designed an experiment where they intercropped mustard and broccoli. Mustard sown less than 7 days after the broccoli was transplanted reduced the yield of the latter. Use of a mustard reduced the cabbage aphid population but increased the predation by syrphid larvae, a beneficial insect. They report mustard served as a trap crop for broccoli pests probably due to its higher glucosinolate concentrations. *Brassica* species can therefore be used as intercrops with broccoli, but mainly to act as a trap crop, and reduce insect pest pressures on broccoli.

Cereal crops are also commonly used as mulches, more often as winter covers. In a 3 year experiment with sorghum, Hargrove (1986) tested 4 legume winter cover crops, and rye as dead mulches. He found that the production of dry weight biomass by rye was similar to that of subterranean clover, hairy vetch and common vetch, but was significantly lower than that of crimson clover. The N content of rye biomass was 3 to 4 folds less than that of the legumes (72 kg N/ha).

Cereals and grasses have also been used as spring seeded living mulches. Hughes and Sweet (1979) interplanted cabbage and beets with spring seeded rye, oats and ryegrass. Yields of cabbage and beet were significantly reduced by the mulches. They however found that these species provided excellent repression of broadleaf weeds.

Creamer et al. (1997) found that rye, barley, crimson clover and hairy vetch were easily killed by mowing. They found tall fescue, perennial ryegrass, and orchard grass did not establish well because they were out competed by other species present in the mixtures. They also found that red clover, white and yellow blossom sweet clover, and annual and perennial ryegrass could not be killed by mechanical means. Nicholson and Wien (1983) screened 30 grasses and 52 legumes in monocultures. From their trial they selected species with low height, high early density, and relatively high amounts of dry matter production to use as living mulch. In a trial with cabbage they found that competition between the intercrop and cabbage for water was critical. Tap rooted legumes took up more water and reduced cabbage yields compared with shallow rooted species.

In summary, it appears from our literature review that legumes have less potential to control weeds than some grass or cereal species, but they have a better potential to improve yields through N fixation. Proper growth control measures must be applied, and adequate species selection can help reducing competition with the main crop. Adequate watering seems to be a crucial element for the success of a brassica-legume intercrops. Most of the research has, however, been done in annual cropping systems, and one could wonder in what ways a perennial living mulch system would differ from the more commonly studied annual ones.

2.2.4. Perennial Versus Annual Living Mulches

Loy et al. (1987) planted squash and peppers in tilled strips in established mixed grass stands or in mixed grass/legume stands. When mulch was adequately repressed, and water was sufficient, they found that yields of squash and peppers grown in living mulches did not differ significantly from the clean cultivation treatments. They concluded that productivity per hectare in strip tillage perennial living mulch systems was close to that of clean cultivation and that the technique could allow vegetable production on land more prone to erosion.

To be able to improve vegetable yields with a legume living mulch, one must pay considerable attention to the reduction of the competitive ability of the living mulch, and must optimise the growth conditions of the main vegetable crop. Factors to consider are irrigation of the crop, mowing of the living mulch, and width of the tilled strip in which the main crop is established (Costello, 1994; Kleinhenz et al., 1997; Tessier and Leroux, 1994). In a system using perennial living mulches, these elements must be controlled even more carefully because of the greater competitive ability of the established perennial living mulch. Nicholson and Wien (1983) substantiate this statement when they state they would expect results to be different in years following establishment, where living mulches will be more competitive at the beginning of the growth period and require greater amounts of water.

2.3. Figure for Chapter 2

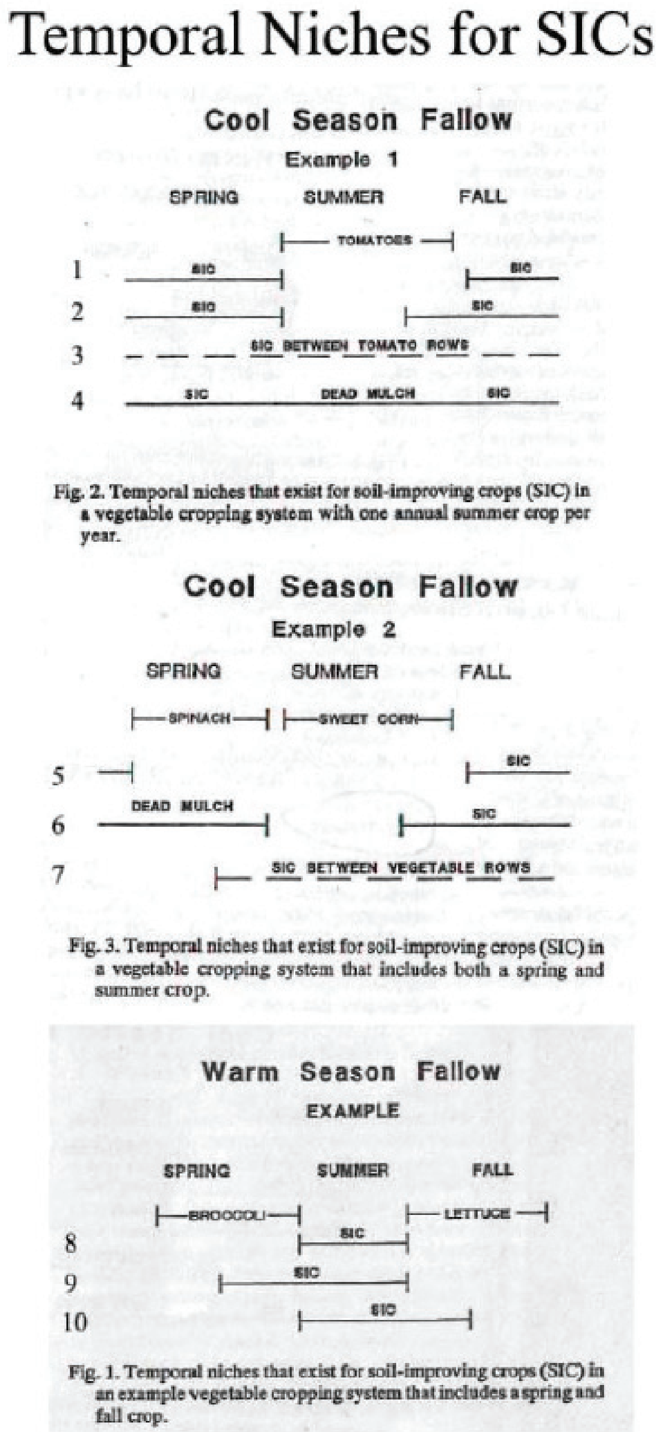

Figure 2.1: Temporal and spatial opportunities for SICs (Sarrantonio, 1992)
Different temporal options are numbered and are discussed in chapter 2

PREFACE TO CHAPTER 3

Chapter 2 reviewed the literature and demonstrated there were serious reasons to believe that perennial legume living mulches had the potential to become a plant-based alternative for the fertilization of organic vegetable crops. It was also highlighted that attention must be paid to reduce the competitive ability of the living mulches in order to avoid yield reductions. Techniques borrowed from other researchers to achieve better control of the living mulch include: proper irrigation of the main crop, rototilling of a strip in which vegetable is planted, and mowing of the living mulch. These techniques have been incorporated in this project's experimental design.

Chapter 3 presents the results of this project's main experiment. The experiment was conducted at the Horticulture Research Center, McGill University, Ste-Anne-de-Bellevue, and replicated over 2 years. Green manures and living mulches of alfalfa and red clover were studied for their impacts on broccoli yields, hollow stem, date of maturity, N uptake, and soil N availability. This manuscript is intended for publication in the International Journal of Vegetable Science with Dr. Philippe Seguin and Dr. Katrine Stewart as co-authors.

3. USE OF PERENNIAL LEGUMINOUS LIVING MULCHES AND GREEN MANURES FOR THE FERTILIZATION OF ORGANIC BROCCOLI.

3.1. Abstract

Perennial leguminous living mulches have the potential to provide several soil benefits sought for by organic farmers; but competition between the main crop and a living mulch has to be controlled to avoid reduced crop yields, and to maximize N benefits. Field experiments were conducted for two consecutive summers (2006-2007) in Sainte-Anne-de-Bellevue, Québec, Canada to evaluate the potential of red clover (*Trifolium pratense* L.) and alfalfa (*Medicago sativa* L.) as perennial living mulches or green manures for the fertilization of a nutrient demanding vegetable crop such as broccoli (*Brassica oleracea* var *italica* Plenk); and to test the effectiveness of mulch clipping management to increase nutrient transfers. Yield, broccoli N uptake, incidence of hollow stem, and available soil N were increased by both red clover and alfalfa green manures. Soil available N was increased by the alfalfa living mulch at both soil samplings, and by red clover living mulch at the first sampling. Competition from the living mulch however led to equal or reduced yields and broccoli N uptake for all living mulch treatments compared to the unfertilized control. Application of mulch clippings directly on the crop row did not increase nitrogen transfers. Alfalfa and red clover green manures have the potential to provide N in excess of recommended compost fertilization rates, while living mulches have a potential to supply N, but should be mown more frequently to reduce competition with the crop.

3.2. Introduction

In recent years, organic agriculture has been one of the most rapidly expanding sectors in the agri-food business (Cameron, 2004). This trend, observed at the consumer level, is confirmed by the 10.6 % increase in the number of Canadian organic producers between 2003 and 2004 (Macey, 2005). Many of these producers are changing their marketing techniques as well as their production methods to improve the ecological and economic viability of their farm operations. The Community Supported Agriculture (CSA) model has emerged as an alternative way to market ecological agricultural production. In this model, customers signup and commit to buy, for the duration of the growing season, a weekly share of mixed produce from the farm (Hendersen and Van En, 2007). In the province of Québec, the number of farms in the CSA farmer network has increased by 20% in 2008 (Équiterre, 2008).

Farmers producing for a CSA program have to grow a wide variety of nutrient-demanding vegetable crops. These farmers often specialize in vegetable production, and generally do not have the capacity to produce the manure required to meet crop nutrient needs (Weil, personal communication). Organic vegetable farmers often import fresh manure to compost on site. With recent environmental regulation changes in Quebec, many farmers are prevented from composting on-farm due to proximity to water courses or wells. Many of them will then turn to pelleted chicken manure sources. In all these cases, organic farms rely on the wastes of industrial and conventional agriculture to meet their fertility requirements, these sources may not be accepted by certification bodies in the future.

The organic philosophy puts high value on the minimization of off-farm inputs (Jordan, 2004) and also seeks to achieve natural plant nutrition (Lampkin, 1990). In an effort to reconcile current organic production practices with the theory and philosophy of the movement, there is a need to develop biologically-based alternatives for the fertilization of organic vegetables.

The use of cover crops and legumes for biological fixation of nitrogen is one of these alternative options; their soil benefits are of great interest to organic farmers (Vanek et al., 2005). Several researchers have shown and quantified the N benefits from the use of legume green manures (Nair et al., 1979; Triplett, 1962; Triplett et al., 1979). Guldan et al. (1997) established that alfalfa (*Medicago sativa* L.) and hairy vetch (*Vicia villosa* Roth) had fertilizer replacement values ranging between 78 and 140 kg N/ha when used as green manures. However, because of the short season, for successful use of green manures in Québec, one often has to fallow a field for an entire season. This has limited their use. Another possibility is the use of intercropping, a form of cultivation where two or more species are grown in close proximity to promote their agronomic interaction (Theunissen, 1994). Varghese (2000) suggests that intercropping can lead to economies of space, and could increase the productivity of vegetables. He however refers to growing two or several economical crops together. One can also intercrop a plant species which is not intended for sale with a vegetable crop. The use of a living mulch is a cropping design where soil improving crops such as legumes are intercropped with a main vegetable crop (Sarrantonio, 1992). To date, in the scientific literature, the competition between main and associated crop, and the ways to control it, have been the main focus of living mulch research (Kloen and Altieri, 1990; Nicholson and Wein, 1983; Vanek et al., 2005). Costello (1994) observed that broccoli (*Brassica oleracea* var *italica* Plenk) grown in a red clover (*Trifolium pratense* L.) living mulch had higher yields than an unfertilized control grown without a living mulch, suggesting N contribution from the legume.

Broccoli is a popular vegetable among CSA farmers in Québec. It is produced from transplants and has high nitrogen requirements, making it a good model crop for research with leguminous living mulches. This project was designed to study the potential of alfalfa or red clover as living mulches and green manures, and to compare them with the use of compost to fertilize organic broccoli.

3.3. Materials and Methods

3.3.1. Location, Soils, Crop History and Legume Establishment

The experimental site was located at the Horticulture Research Centre of the Macdonald Campus, McGill University, Sainte-Anne-de-Bellevue, Québec, Canada (lat. 45º 26'N long. 73º 56'W). The soil was a Gleyed Eluviated Eutric Brunisol. Fields (A and B) were not certified organic, but fertilizers and pesticides were last used in 2002. In 2003 and 2004, rye (*Secale cereale* L.) was grown in both fields as a cover crop, and no crop was grown in 2005 before the establishment of the legumes in field B. In June 2005 (Field A) and 2006 (Field B), alfalfa cv. 'Arrow' and red clover cv. 'Dolina' stands were established in plots targeted for living mulch and green manure treatments at a rate of at least 13 kg/ha with a tractor mounted *Brillion* seeder (Brillion, WI). To control weeds, the legumes were mowed several times, and bare ground plots were worked with a S-tine Triple-K cultivator (Kongskilde, Soro, Denmark).

3.3.2. Broccoli Variety, Greenhouse Practices, and Transplanting

Broccoli cv. 'Gypsy' was seeded on 11 May in 2006 and on 14 May in 2007 in 98 plug trays (35 cm^3/cell) in a greenhouse. Greenhouse conditions were 16h daylight period (supplemental lighting 800 $\mu mol \cdot m^{-2} \cdot s^{-1}$), relative humidity 65%, day/night temperatures 25/22°C, and shade cloths would cover the section if temperature rose above 28°C. The potting soil was a mixture of 500 L peat moss (Conrad Fafard Inc, Agawam, MA, USA), 500 L perlite (Perlite Canada Inc, St-Laurent, QC, Canada), 35 L "Humix" peat and shrimp compost (0.5-0.4-0.1) (Aquatrerie Inc, St-Fabien, QC, Canada), 60 L organic top soil (Canadian Garden, Saint-Laurent, QC, Canada) and 0,5 L "Mike Vegetable Grow" organic OMRI certified granulated fertilizer (8-4-5) (Premier Tech Biotechnology, Rivière-du-Loup, QC, Canada). One seed was placed per cell at a 0.75 cm depth. Flats were watered daily based on need, and fertigated three times starting 4 weeks after seeding with Neptune (2-4-0.5) fish emulsion (Engrais Acadie, Fortierville, QC, Canada) at a rate of 200 mL per cell ((in $mg \cdot L^{-1}$) 20 N, 40 P_2O_5, 10 K_2O). After 6 weeks, broccoli was transplanted to the field by hand.

3.3.3. Experimental Design and Treatments

The experiment consisted of 9 treatments arranged in a randomized complete block design with 4 replicates, including 3 treatments using alfalfa, 3 treatments using red clover and 3 treatments on bare ground. Treatments on bare ground were an unfertilized control (0N), a plot fertilized with compost (35.71 N, 38.92 P, 24.04 K, 6.76 Ca, 7.79 Mg mg/g) at the recommended dosage of 135 kg N/ha (COMP-135) and a plot fertilized at half the recommended dose, 68 kg N/ha (COMP-68). Compost was incorporated with a rototiller (Kubota, Osaka, Japan) one week prior to transplanting. For all other treatments, no additional compost or fertilizers were applied as soil tests indicated high levels of P and K (523.3 P, 300.1 K, 5474.7 Ca, 742.3 Mg in kg/ha) . For green manure (GM) plots, alfalfa or red clover was incorporated with a rototiller one and three weeks prior to transplanting in 2006 and 2007, respectively. For living mulch treatments, the mulch, either alfalfa or red clover, was mown one and three weeks prior to transplanting in 2006 and 2007, respectively and 30 cm strips were rototilled into the mulch. The strips were spaced 90 cm apart (center-to-center), and the broccoli was transplanted into these rototilled strips. The living mulch grew in the 60 cm inter-row, and was mown twice during the season when it reached a height of 35-50 cm. In the LM treatments, the clippings were left in the inter-row (RC-LM and ALF-LM). In the CLIP treatments, the clippings were raked and applied over the crop row (RC-CLIP and ALF-CLIP). Plots were 4.8 meters long by 4.2 meters wide and consisted of 4 rows spaced 0.45 m between plants and 0.9 m between rows for a density of 24,666 plants/hectare. Measurements were made on the soil, mulch or the 18 broccoli plants located in the middle of the plot.

3.3.4. Weeding, Irrigation and Agronomic Practices.

Weeding followed recommendations for commercial small scale organic vegetable production in Québec. Weeds were removed with a wheel hoe (Glaser, Switzerland) or with stirrup hoes (Glaser, Switzerland) 2, 4 and 7 weeks after transplanting (WAT). Bare ground and GM plots had all weeds removed. In living mulch plots (LM and CLIP plots), weeds were removed only in the broccoli row, not in the mulched inter-row. In 2006, a drip irrigation system (Toro, El Cajon, CA) was used; it was replaced in 2007 with a Naan mini-sprinkler irrigation system (Dubois Agrinovation, St-Rémi, Qc, Canada). Irrigation was used if natural precipitation levels were less than 20 mm per week. When necessary, irrigation was applied every second day for a period of four hours. To protect the crop from the swede midge (*Contarinia nasturtii* Keiffer) and the cabbage root fly (*Delia radicum*), the field was covered with floating row cover Agryl P-10 (10 $g{\cdot}m^{-2}$) (Dubois Agrinovation, St-Rémi, QC) immediately after transplanting. Except when measurements were taken or weeds removed, the field remained covered until bud initiation, 5 or 6 WAT. *Bacillus turingiensis* var *kurstaki* was used as a spray to control the imported cabbage worm (*Pieris rapae*).

3.3.5. Broccoli Yield Measurement

Broccoli yields were taken from 18 plants in the two middle rows of each plot. Plants were harvested when heads had reached the mature marketable stage, grade 3 to 5 using a maturity index ranging from 1 to 7 modified from Sorensen and Grevsen (1994) (Table 3.1). Broccoli was cut to a standard 25 cm length and fresh weight and head diameter measured. Stems were inspected for hollow stem, a common physiological disorder in the brassicas that causes transversal splitting in the pith of the stem (Shattuck and Shelp, 1987). For each plot, two broccoli plants were shredded, dried at 50ºC for 72h in a forced-air dryer, and subsequently analysed for total N content with the True-spec N autoanalyser (Leco Corporation, Michigan, USA).

3.3.6. Living Mulch Biomass Productivity and N Content

Living mulch biomass samples were collected three times during the season. The first sampling was made on 12 June 2006 and 28 May 2007, before plots had been prepared and broccoli planted. Samples were collected with a forage harvester (Swift Machine and Welding inc., Swift Current, SK, Canada), and a 500 g sub-sample of biomass was kept for analysis. Sub-samples were dried in a forced-air dryer at 50ºC for 72h, weighed, ground, and analysed for total N. The remainder of the sample was returned to the GM, LM or CLIP plot it originated from. The next two samplings (in the LM and CLIP plots only) were taken during growth of the broccoli crop prior to mowing. Three random quadrats (50 x 50 cm) were used per plot. The biomass was hand harvested 6 cm from the ground using hand-scissors. Samples were dried, weighed and ÿÿtal ÿÿcontent was determined and used to com tÿÿ fstimates of organic N supplied by the mown biomass.

3.3.7. Soil Nitrogen Measurement

Extractable ammonium and nitrates in the soil were measured according to the procedures described by Maynard and Karla (1993). Soil was sampled at a depth of 0-20 cm within the row twice during the growing season: one week after transplanting and 3 weeks later. Soil samples were refrigerated at 4°C until extracted with a 2 M KCl solution. Soil was mixed in a 1:10 ratio with the KCl extractant for 30 minutes, and then filtered through Whatman 42 filter paper. The filtrate was analysed by colorimetry with a multi-channel Lachat (Milwaukee, WI, USA).

3.3.8. Plant Nitrogen and SPAD measurements

Plant N uptake was estimated through 3 measurements three, five, and seven WAT using a Minolta SPAD-502 chlorophyll meter (Soil Plant Analysis Development, Minolta, Japan). SPAD-meter readings are significantly correlated with applied N rates and plant N uptake in corn (Costa et al., 2001), and in vegetables (Sexton and Carroll, 2002; Swiader and Moore, 2002). SPAD readings were taken from several points on the fifth newest leaf of five randomly selected broccoli plants in each plot and were averaged. In 2006, readings were taken at ¼, ½, and ¾ of the leaf length on either side of the central vein, 1 cm from the edge of the leaf. In 2007, one measurement point was added at the tip of the leaf.

3.3.9. Statistical Analyses

Data was subjected to analysis of variance (ANOVA) using the general linear model (GLM) procedure. Differences between treatments were determined using a priori contrast analyses (SAS_Institute, 2002) and using the Student-Neuman-Keuls procedure. When variances were homogeneous, data from both years was pooled for analysis (Gomez and Gomez, 1984).

3.4. Results and Discussion

3.4.1. Broccoli earliness and yields, and occurrence of hollow stem

Broccoli from the GM treatments matured fastest (average of 65 days), followed by compost treatments (66 days), and living mulch and unfertilized control treatments (68 days) (Table 3.2). The difference in maturity between treatments could also be seen in the grading of the heads. Soil of the green manure plots might have had more available nitrogen than those of the other treatments. Increased nitrogen availability has previously been reported to accelerate broccoli development (Gutezeit, 1996; Vagen et al., 2004).

In both 2006 and 2007, head weights from both GM treatments were significantly heavier than the broccoli fertilized with compost. Head diameter of broccoli grown following ALF-GM was significantly greater than following RC-GM (23.3 and 22.8 cm respectively), while the latter was not significantly different from broccoli fertilized with COMP-135 (22.7 cm). Use of either GM or compost produced larger and heavier heads than any of the living mulch treatments, contrary to the findings of Costello (1994). In 2 treatments (RC-CLIP, ALF-LM) yields were significantly lower than in the unfertilized control. Bottenberg et al. (1997) obtained similar results with cabbage yields being reduced by a red clover living mulch. They identified moisture competition as the dominant factor in yield reduction. This suggests that even though great care has been taken in the present experiment to minimize all forms of competition between the main crop and living mulch (i.e., light, water and nutrients) some competition occurred. Vanek et al. (2005) noted that it was critical to reduce biomass production by the living mulch in the first 30 days after seeding pumpkins to reduce competition between the mulch and crop. Perhaps by reducing the mowing threshold to 25-35 cm rather than 35-50 cm, particularly for the first of the three mowings, we might have reduced competition sufficiently and hence increased yields in the LM and CLIP plots.

There was no significant difference in yields from the application of living mulch clippings (CLIP) over the crop row compared with the standard LM. These results are similar to those obtained by Wiens et al. (2006) who obtained positive yield increases in wheat only when they applied alfalfa clippings from an area twice that of the wheat plot. In the current experiment, where the LM covered 66% of the field surface, the amount of clippings applied to the crop row might not have been sufficient to significantly increase the yield of a high nitrogen demanding crop such as broccoli.

More heads in green manure and compost treatments exhibited hollow stem than the control. Bélec et al (2001) and Tremblay (1989) have shown that hollow stem is greater when high levels of N are available, as well as when broccoli plants have a fast growth rate (Hipp, 1974). It appears, based on our results, that using LM would slow down the rate of broccoli development, and reduce the incidence of hollow stem compared to full dosage of compost and green manures.

3.4.2. Nitrogen uptake of broccoli plants

SPAD readings taken 3 WAT indicated that GM treatments had the greatest chlorophyll levels and by extension, the greatest amounts of N in plant tissues. This was followed by the compost treatments, then by the LM and CLIP treatments which had significantly lower levels of chlorophyll and plant N (Table 3.3). SPAD readings taken two weeks later (5 WAT) suggest a general increase in plant N uptake for all treatments, but only GM treatments had significantly greater N uptake than the control. The third SPAD reading, taken at 7 WAT (in 2007) and 9 WAT (in 2006), did not reveal any significant differences between treatments. SPAD readings were thus most significant when performed at 3 or 5 WAT. SPAD readings suggest that at 3 WAT red clover LM, CLIP, and GM broccoli plants had taken up greater amounts of N than alfalfa LM, CLIP and GM respectively. This trend was reversed later in the season, as shown in SPAD II, and in the broccoli total N analysis. This might be explained by the possible slower mineralization of alfalfa compared to red clover. It has been demonstrated that alfalfa stems contain greater concentrations of lignin than red clover stems (Buxton and Russel, 1988), which slows microbial digestibility. N uptake by the broccoli was similar for the CLIP and LM treatments. In fact, none of our LM and CLIP treatments increased plant N uptake compared with the control. SPAD readings suggests that broccoli grown after a GM had a greater N uptake, which might explains their faster development and greater yields.

Broccoli grown after alfalfa and clover green manures had a significantly higher total N content (4.12 and 4.02 % respectively) compared with all other treatments (3.09 to 3.54 % N). Whether this higher N content of the GM broccoli translates into a higher nitrate content has not been determined in the present experiment. Zebarth et al (1995) have shown that high nitrogen fertilization levels can increase nitrate concentrations in broccoli tissues. High levels of nitrates in foods may be linked to health risks for consumers (Codex-Alimentarius-Commission, 1998).

3.4.3. Soil available nitrogen

All treatments increased the amount of soil available nitrogen compared with the unfertilized control. This was observed on 27 June at the beginning of the experiment (6.55 kg $N \cdot ha^{-1}$ for the control vs an average of 10.06 kg $N \cdot ha^{-1}$ for all other treatments; Table 3.4), and later in the season on 16 July (6.85 kg $N \cdot ha^{-1}$ vs 11.55 kg $N \cdot ha^{-1}$). At both dates, more N was mineralized in the form of NO_3 or NH_4 in ALF-GM and RC-GM plots (average of 16.86 kg $N \cdot ha^{-1}$) compared with all of the other treatments (average of 8,49 kg $N \cdot ha^{-1}$). At the start of the experiment, the addition of clippings (CLIP treatments) to the crop row initially immobilized nitrogen compared with the LM treatments without clippings. Kleinhenz et al. (1997) also observed that incorporation of legume biomass into the soil initially resulted in N immobilisation. Later, on 16 July, this effect was no longer observed, resulting in a lack of differences between the LM and the CLIP treatments. Furthermore, comparison of soil available nitrogen in ALF LM and CLIP treatments with that of RC LM and CLIP treatments suggested a greater N contribution from the ALF. Guldan et al. (1997) report that alflafa has a higher fertilizer replacement value than red clover. It is not clear however, why three weeks after transplanting, SPAD readings indicated greater plant N status in RC treatments compared to ALF treatments. It is possible that because most of the available N was in the form of NO_3 early in the season, it could have leached down the soil profile after rain, and been replenished more slowly during the first weeks of broccoli growth for the ALF treatments due to biomass characteristics.

This points to the potential risk of nitrate leaching, especially in the GM treatments, where very high levels of nitrates were measured in the soil. Careful use of GM is necessary to avoid ground water pollution. Research has shown it is essential to time tillage events to synchronize N mineralization with crop uptake (Carter et al., 1991). In that sense, one of the potential advantages of perennial living mulches is that after mowing, the N released from the decomposing biomass that is not taken up by the primary crop could be recovered at deeper levels of the soil by the well established root system of the secondary crop (the living mulch). This recovered N would be incorporated in the legume biomass, and be made available again for the main crop at a subsequent mowing.

3.4.4. Legume biomass characteristics

In 2006, from the first to the third mowing, there was a strong reduction in the quantity of legume biomass produced and in the N applied to the broccoli crop through the clippings (Table 3.5). In 2007, the amount of biomass produced declined between the first and third mowing, but to a much lesser extent than in 2006. The living mulch biomass N content at the second and third mowings was also lower 2007 compared with 2006. Differences in living mulch productivity and N content between years are probably linked to the difference in the irrigation system used in both years. In 2006 a drip irrigation system supplied water at the base of the broccoli crop, and no water was directly applied to the living mulch. Living mulch plants might have been stressed for water, and thus had a reduced growth rate and greater N content. Halim et al. (1989) found that crude protein increased in alfalfa stems as drought stress increased. In 2007, a sprinkler irrigation system watered both the broccoli crop and the living mulches equally. With less water limitation living mulches grew more vigorously, and had a lower nitrogen content in their tissues.

In 2007, alfalfa supplied more N than red clover at each mowing. In 2006, data for the second mowing is less conclusive due to patchiness of the living mulch strips. We were not able to constantly obtain a full 50 x 50 cm quadrat that was completely covered in vegetation. This patchiness introduced error in the final estimate calculations. At the final mowing in 2006, the productivity of red clover appeared to be greater than alfalfa. This is probably due to the drip irrigation, and to a differential response to water stress between red clover and alfalfa. Peterson et al. (1992) found exposure to drought reduced alfalfa yields by 33%, but by only 13% for red clover. This tendency was not observed in 2007 when a sprinkler irrigation system was used.

The vigorously growing living mulches produced large quantities of biomass, particularly in 2007. The significant reduction in yields for some LM and CLIP treatments compared with the control can be attributed to the competition for light. Competition for light probably prevented the LM and CLIP treatments from growing and absorbing the nitrogen delivered through the mowing of the living mulch biomass.

3.5. Conclusions

Perennial living mulches of ALF or RC have potential to provide N to their associated main crop through the mowing of their biomass. Application of biomass clippings to the crop row (CLIP) did not increase nitrogen transfer. Soil available nitrogen was significantly improved in our experiment for the LM treatments at the first soil sampling, and for the alfalfa treatments at the second sampling. In our experiment, yields and nitrogen uptake from the LM and CLIP broccoli were either reduced or equal to that of unfertilized control because of competition by the living mulch. Both the alfalfa and red clover green manures were able to supply sufficient N for the broccoli, and to increase soil N availability in excess of the recommended compost dose. In the case of broccoli, this increased N supply from GM translated into greater yields, more incidence of hollow stem, higher SPAD readings, greater plant tissue total N content, and faster maturity. Green manures of alfalfa or red clover constitute a biologically-based alternative for the fertilization of organic vegetables, but should be used with care to minimize risks of nitrate leaching, and to avoid increasing the concentration of nitrates in organic foods. The potential of living mulches to reduce nitrate leaching and nitrate contents of organic vegetables in comparison with green manures should be further investigated. More research is also needed to determine how many times, at what stage of growth, and at which height perennial leguminous mulches should be mown to avoid the crop-intercrop interaction that was observed in this experiment.

3.6. Figures and Tables

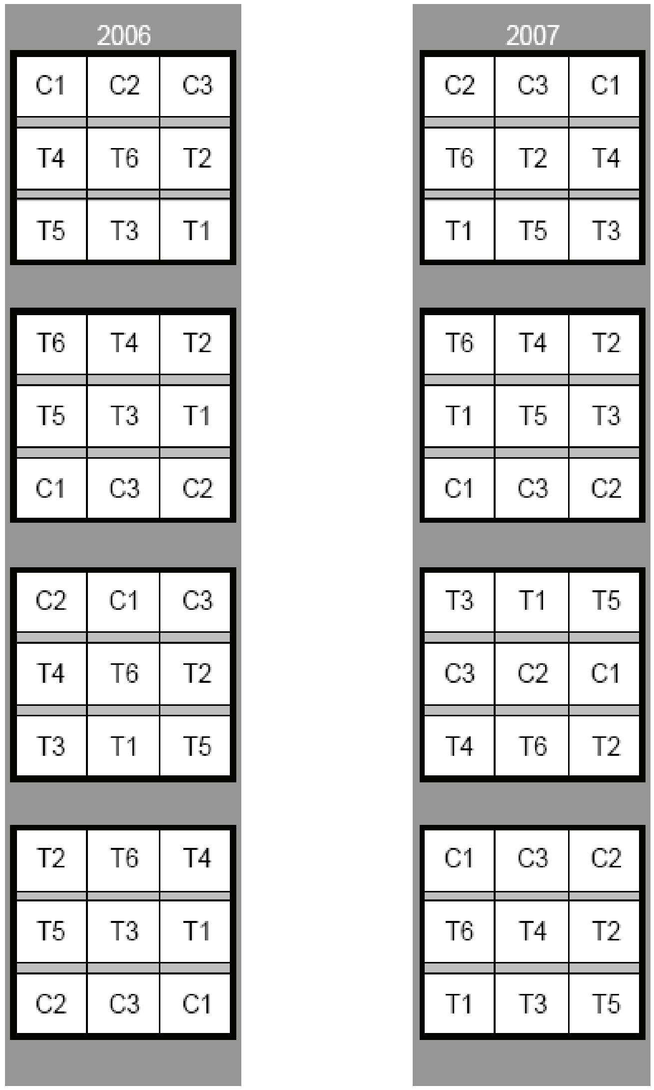

FIGURE 3.1. Experimental design and randomized location of plots for experiment comparing the effect of different living mulch, compost and green manure treatments on broccoli cv. 'Gypsy' grown in Southwestern Quebec in 2006 (left) and 2007 (right).
C1 - control, no compost ;
C2 - full compost dose, 135 kg N/ha ; C3 - Half dose compost, 68 kg N/ha ;
T1 - Alfalfa living mulch, clippings applied ; T2 - Red clover living mulch, clippings applied
T3 - Alfalfa living mulch, no clippings ; T4 - Red Clover living mulch, no clippings ;
T5 - Alfalfa green manure ; T6 - Red Clover green manure

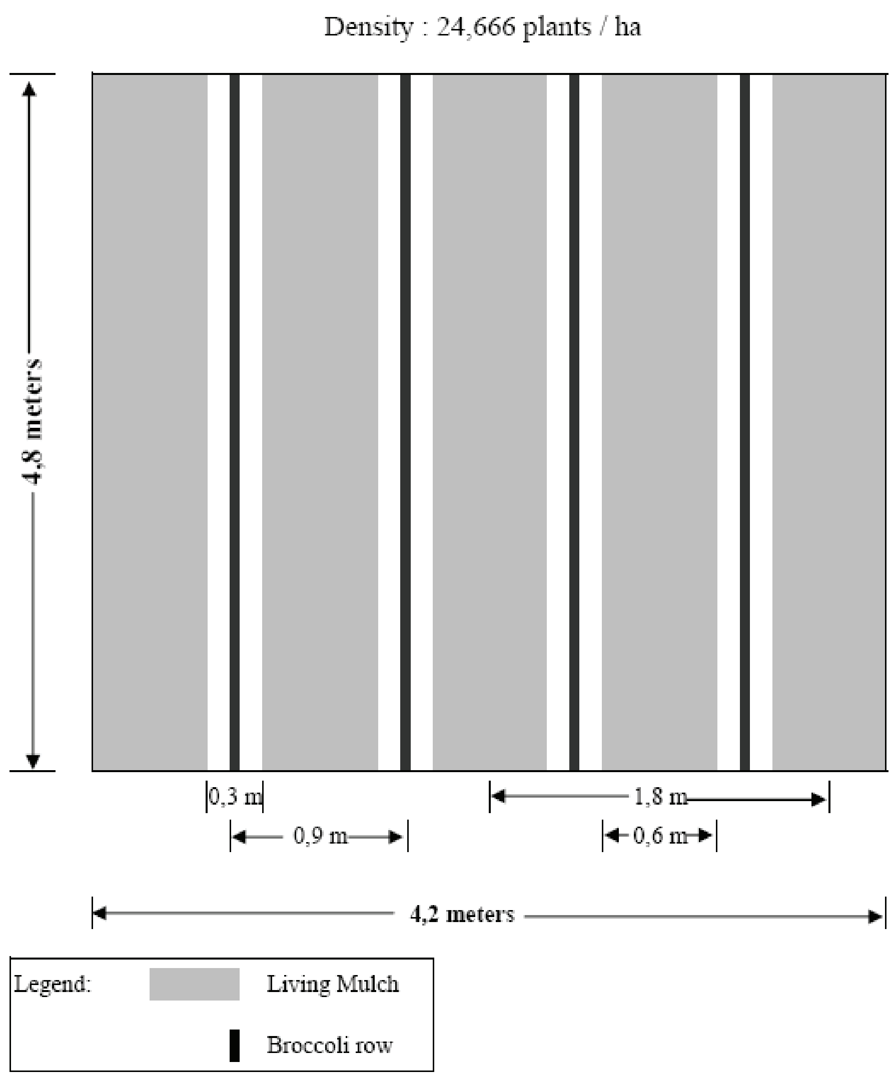

FIGURE 3.2. Plot layout for experiment studying effect of living mulches on organic broccoli cv. 'Gypsy' grown in southwestern Québec in 2006 and 2007

TABLE 3.1: Grading system for broccoli heads, modified from Sorensen and Grevsen (1994)

1	head small, clearly very immature
2	head not fully mature, florets small
3	head firm and marketable, optimum density and floret size
4	head firm and marketable, but either density of floret size not optimal
5	head firm, but discoloured, passed optimal marketable stage
6	head slightly loose
7	head loose, florets stretching

TABLE 3.2: Effect of composts, green manures and living mulches on yield parameters, number of plants with hollow stem, and days to maturity of organic broccoli cv. 'Gypsy' grown in southwestern Québec in 2006 and 2007 (data averaged for both years).

		DTM[z]	Yield			Hollow stem
		days	Head Grade	Head Weight g	Head diameter cm	%
Treatments						
1	Control - no fertilization (0N)	68.1a[y]	3.00c	604c	20.5d	35.8bc
2	Compost[x] - 135 kg N / ha (COMP-135)	66.3b	3.48ab	707b	22.7b	51.0ab
3	Compost - 68 kg N / ha (COMP-68)	66.7b	3.29bc	681b	22.1c	43.8abc
4	Alfalfa living mulch + clippings (ALF-CLIP)	68.3a	3.06c	597c	20.9d	28.3bcd
5	Clover living mulch + clippings (RC-CLIP)	68.4a	3.07c	526d	19.9e	7.9d
6	Alfalfa living mulch (ALF-LM)	68.5a	3.06c	531d	19.8e	24.7cd
7	Clover living mulch (RC-LM)	68.6a	3.18c	579c	20.6d	24.1cd
8	Alfalfa Green Manure (ALF-GM)	64.2d	3.58a	755a	23.3a	60.7a
9	Clover Green Manure (RC-GM)	65.0c	3.50ab	750a	22.8b	59.4a
Contrasts						
	1 vs 2,3,4,5,6,7,8,9	**** [w]	***	***	****	NS
	3 vs 4,5,6,7,8,9	NS	NS	****	****	NS
	2 vs 4,5,6,7,8,9	**	**	****	****	*
	1,2,3 vs 4,5,6,7,8,9	NS	NS	****	****	*
	4,6,8 vs 5,7,9 - RC vs ALF	NS	NS	NS	NS	NS
	4,5 vs 6,7 - LM vs CLIP	NS	NS	NS	NS	NS
	8,9 vs 4,5,6,7 - GM vs LM+CLIP	****	****	****	****	****
	4,5,6,7 vs 1,2,3 - LM vs COMP	****	***	****	****	****

z Days to maturity

y Values in a column followed by the same letter are not significantly different

x Compost: 35,71 N, 38,92 P, 24,04 K, 6,76 Ca, 7,79 Mg mg/g

w NS, *, **, ***, **** : Non-significant or significant at P≤ 0.05, 0.01, 0.001, 0.0001 respectively

TABLE 3.3: Effect of composts, green manures and living mulches on Minolta SPAD™ meter measurement of leaves and total nitrogen content of organic broccoli cv. 'Gypsy' grown in southwestern Québec in 2006 and 2007 (data averaged for both years)

	Minolta SPAD™ measurements			Total N content
	SPAD I [z]	SPAD II [y]	SPAD III [x]	%
Treatments				
1 Control - no fertilization (0N)	57.1bc[w]	64.1cd	66.7ab	3.00c
2 Compost[v] - 135 kg N / ha (COMP-135)	58.7b	65.0bcd	68.5ab	3.15c
3 Compost - 68 kg N / ha (COMP-68)	58.2b	65.6bc	65.8b	3.22c
4 Alfalfa living mulch + clippings (ALF-CLIP)	54.6d	63.6cd	67.1ab	3.31c
5 Clover living mulch + clippings (RC-CLIP)	55.8cd	63.9cd	67.1ab	3.24c
6 Alfalfa living mulch (ALF-LM)	55.0d	64.3cd	67.0ab	3.31c
7 Clover living mulch (RC-LM)	56.3cd	63.0d	67.3ab	3.22c
8 Alfalfa Green Manure (ALF-GM)	60.4a	68.0a	69.1a	3.92a
9 Clover Green Manure (RC-GM)	61.1a	66.5ab	67.8ab	3.63b
Contrasts				
1 vs 2,3,4,5,6,7,8,9	NS[u]	NS	NS	****
3 vs 4,5,6,7,8,9	NS	NS	NS	*
2 vs 4,5,6,7,8,9	**	NS	NS	**
1,2,3 vs 4,5,6,7,8,9	*	NS	NS	****
4,6,8 vs 5,7,9 - RC vs ALF	*	NS	NS	*
4,5 vs 6,7 - LM vs CLIP	NS	NS	NS	NS
8,9 vs 4,5,6,7 - GM vs LM+CLIP	****	****	NS	****
4,5,6,7 vs 1,2,3 - LM vs COMP	****	***	NS	*

[z] Minolta SPAD™ meter measurement performed on July 10th 2006 and July 11th 2007
[y] Performed on August 1st 2006 and July 24th 2007
[x] Performed on August 24th 2006 and August 8th 2007
[w] values in a column followed by the same letter are not significantly different
[v] Compost: 35,71 N, 38,92 P, 24,04 K, 6,76 Ca, 7,79 Mg mg/g
[u] NS, *, **, ***, **** : Non-significant or significant at P≤ 0.05, 0.01, 0.001, 0.0001 respectively

TABLE 3.4: Effect of composts, green manures and living mulches on soil available nitrogen (NO_3 and NH_4) measured on 27 June and 16 July 2007 for organic broccoli cv. 'Gypsy' grown in southwestern Québec in 2007

Treatments	June 27th kg N / ha	July 16th kg N / ha
1 Control - no fertilization (0N)	6.55e[z]	6.85c
2 Compost[y] - 135 kg N / ha (COMP-135)	7.02de	9.36c
3 Compost - 68 kg N / ha (COMP-68)	7.25de	8.40c
4 Alfalfa living mulch + clippings (ALF-CLIP)	7.69de	12.16b
5 Clover living mulch + clippings (RC-CLIP)	7.61de	7.11c
6 Alfalfa living mulch (ALF-LM)	9.93c	11.45b
7 Clover living mulch (RC-LM)	8.27d	9.20c
8 Alfalfa Green Manure (ALF-GM)	17.20a	17.41a
9 Clover Green Manure (RC-GM)	15.54b	17.27a
Contrasts		
1 vs 2,3,4,5,6,7,8,9	**** [w]	****
3 vs 4,5,6,7,8,9	****	****
2 vs 4,5,6,7,8,9	****	****
1,2,3 vs 4,5,6,7,8,9	****	****
4,6,8 vs 5,7,9 - RC vs ALF	***	****
4,5 vs 6,7 - LM vs CLIP	***	NS
8,9 vs 4,5,6,7 - GM vs LM+CLIP	****	****
4,5,6,7 vs 1,2,3 - LM vs COMP	****	***

[z] Values in a column followed by the same letter are not significantly different

[y] Compost: 35,71 N, 38,92 P, 24,04 K, 6,76 Ca, 7,79 Mg mg/g

[w] NS, *, **, ***, **** : Non-significant or significant at P≤ 0.05, 0.01, 0.001, 0.0001 respectively

TABLE 3.5: Effect of living mulch and green manure treatments on N applied to organic broccoli cv. 'Gypsy' grown in southwestern Québec in 2006 and 2007.

	1st Mowing[z]			2nd Mowing[y]			3rd Mowing[x]		
	DM[w]	% N	N applied	DM	% N	N applied	DM	% N	N applied
Year / Treatment	$t \cdot ha^{-1}$		$kg\ N \cdot ha^{-1}$	$t \cdot ha^{-1}$		$kg\ N \cdot ha^{-1}$	$t \cdot ha^{-1}$		$kg\ N \cdot ha^{-1}$
2006									
Alfalfa living mulch + clippings (ALF-CLIP)	4,00	3.5	140	0.751	5.29	39.7	0.380	5.41	20.5
Clover living mulch + clippings (RC-CLIP)	3.59	3.40	122	0.818	5.21	42.6	0.604	5.09	30.8
Alfalfa living mulch (ALF-LM)	4.24	3.33	141	0.898	4.95	44.4	0.402	5.25	21.1
Clover living mulch (RC-LM)	3.57	3.47	124	0.693	5.08	35.2	0.676	4.96	33.5
Alfalfa Green Manure (ALF-GM)	3.99	3.49	139	NA[v]	NA	NA	NA	NA	NA
Clover Green Manure (RC-GM)	3.55	3.27	116	NA	NA	NA	NA	NA	NA
2007									
Alfalfa living mulch + clippings (ALF-CLIP)	4.28	3.34	143	3.25	3.68	119.8	1.30	4.84	63.0
Clover living mulch + clippings (RC-CLIP)	4.15	3.31	137	2.69	3.63	97.8	1.36	4.05	54.9
Alfalfa living mulch (ALF-LM)	4.58	3.18	146	3.28	3.59	118.0	1.29	4.97	64.3
Clover living mulch (RC-LM)	4.09	3.18	130	2.56	3.76	96.3	1.25	4.26	53.3
Alfalfa Green Manure (ALF-GM)	4.25	3.26	138	NA	NA	NA	NA	NA	NA
Clover Green Manure (RC-GM)	4.34	3.37	146	NA	NA	NA	NA	NA	NA

[z] Performed on June 12th 2006 and May 28th 2007

[y] Performed on July 5th 2006 and July 3rd 2007

[x] Performed on July 20th 2006 and July 23rd 2007

[w] Dry matter produced by the leguminous secondary crop (alfalfa or red clover)

[v] Not applicable

PREFACE TO CHAPTER 4

In chapter 3 the yield and N impacts of the use of green manures and living mulches were described. It was concluded that green manures have a strong potential to increase yields, N uptake and N availability. The living mulch management used in this experiment however did not limit competition as much as was expected. It led to reduced yields and N uptake, even though soil N availability was increased. Fertility enhancement is a novel objective for living mulches, yet the design proposed here did not allow to achieve it; therefore modified management systems should be proposed.

However, living mulches are more frequently used for insect control than for fertility enhancement. In this experiment, the main method for insect control was the use of floating row cover. The use of a floating row cover had never been tested with living mulches for their combined effect on insect pests and the possible interactions between the two control methods. Chapter 4 presents the results of *P. rapae* counts that were conducted during the two years of our experiment in Ste-Anne-de-Bellevue. This chapter will be submitted for publication with Dr. Philippe Seguin and Dr. Katrine Stewart as co-authors.

4. EFFECT OF THE COMBINATION OF LIVING MULCHES AND ROW COVERS ON ABUNDANCE OF *PIERIS RAPAE* L. IN ORGANIC BROCCOLI GROWN IN SOUTHWESTERN QUÉBEC.

4.1. Abstract

Floating row covers and living mulches were tested in a combination for their potential to reduce imported cabbage worm (*P. rapae*) in organic broccoli in 2006 and 2007. It was shown that for two consecutive years, using a combination of row cover and living mulches (red clover and alfalfa) increased *P. rapae* larvae abundance in the broccoli compared to the use of the row cover with bare ground treatments. When row cover and living mulches were used together, the creation of a sheltered microenvironment appeared to be a more important factor in the choice of a site for oviposition by *P. rapae* than plant nutrient content and chlorophyll level of the plants. When floating row covers are removed frequently, which provides opportunity for *P. rapae* introduction under the row cover, the use of living mulches can increase the abundance and provide better conditions for the development of *P. rapae* larvae.

4.2. Introduction

Control of insect pests is one of the major challenges faced by organic farmers (Grubinger, 1992). Several methods and strategies have been developed by organic farmers to control insect pests including bio-pesticides and physical barriers. Spunbonded polyester and polyethylene floating row covers were originally used to modify environmental parameters such as light, soil and air temperature (Wells and Loy, 1985; 1993) but their usefulness to reduce insect pressure has also become apparent to researchers. Row covers were used to reduce aphid spp. and whitefly (*Bemisia tabacci*) and their associated viruses in cantaloupe (*Cucumis melo* L.; Perring et al., 1989) and zucchini (Cucurbita pepo L.; Walters, 2003; Webb and Linda, 1992). In Brassicaceae, they were successful in reducing cabbage maggots (*Delia radicum* L.) in cauliflower (*Brassica oleracea* var *botrytis*; Millar and Murray, 1988). Row covers have also been shown to reduce Lepidoptera in cabbage (*Brassica oleracea* var *capitata*; Evans et al., 1997) and in broccoli (*Brassica oleracea* var *italica* Plenk; Adams et al., 1990). Another strategy used by organic farmers is agroecosystem diversification. The use of living mulches has been studied as a way to increase agroecosystem diversity in the field, and to reduce insect damage in crops (Kloen and Altieri, 1990; Theunissen, 1994). Andow (1986) found that a rye (*secale cereale* L.) living mulch reduced flea beetle damage in cabbage. Costello (1994) used red clover (*Trifolium pratense* L.) living mulches to reduce aphid infestations in broccoli. Hooks and Johnson (2001; 2004) noted that lepidopterous pest infestations were reduced in broccoli by the use of clover living mulches. They hypothesized that this was due in part to a greater abundance of predatory spiders (Hooks and Johnson, 2002). Several mechanisms have been proposed to explain the lower pressure of herbivores in living mulches. The natural enemy hypothesis states that parasitoids and predators will occur in greater abundance and diversity in living mulches due to greater resource supply (Hooks and Johnson, 2001). The impact of habitat diversification on brassica growth parameters is another proposed explanation. It has been shown that during oviposition imported cabbage worm (*P. rapae*) females respond to leaf water content (Renwick and Radke, 1983), chlorophyll level (Hovanitz and Chang, 1964), and nutrient concentration (Myers, 1985). The objective of this study was to determine the effect of the combination of agrotextile floating row covers and living mulches on imported cabbage worm populations in organic broccoli.

4.3. Materials and Methods

4.3.1. Location and Experimental Design.

The experimental site was located at the Horticulture Research Centre, Macdonald Campus, McGill University, Ste-Anne-de-Bellevue, Québec, Canada (lat. 45º 26'N long. 73º 56'W). The soil was a Gleyed Eluviated Eutric Brunisol. The experiment consisted of 9 treatments arranged in a randomized complete block design. Five treatments were on bare ground: control (0N), turkey manure compost (35.71 N, 38.92 P, 24.04 K, 6,76 Ca, 7,79 Mg mg/g) at a dose of 135 kg N/ha (COMP-135), turkey manure compost at 68 kg N/ha (COMP-68), alfalfa green manure (ALF-GM) and red clover green manure (RC-GM). Four treatments were using living mulches: red clover living mulch (RC-LM), alfalfa living mulch (ALF-LM), and red clover living mulch + clippings (RC-CLIP) or alfalfa living mulch + clippings (ALF-CLIP). In June 2005 and 2006, alfalfa (*Medicago sativa* L.) cv. 'Arrow' and red clover cv. 'Dolina' stands were established. In June 2006 and 2007, broccoli cv. 'Gypsy' was transplanted in plots fertilized with either compost, green manures or living mulches.

In living mulch treatments, the legume stand established the previous year was mown with a flail mower (Kubota, Osaka, Japan) on 13 June 2006 and 12 June 2007. Thereafter, 30 cm wide strips were tilled using a modified rototiller (Kubota, Osaka, Japan). Broccoli was transplanted by hand in these strips that were 90 cm apart. The living mulch grew in the 60 cm inter-row, and was mown twice during the season using a brush cutter (Husqvarna, Stockolm, Sweden) when it reached a height of 35-50 cm. The first mowing took place on 9 July 2006 and 4 July 2007. In the LM treatments, the clippings were left in the inter-row. In the CLIP treatments, the clippings were raked and applied over the crop row. Plots were 4.8 meters long by 4.2 meters wide and consisted of 4 rows spaced 0.45 m between plants and 0.9 m between rows for a density of 24 666 plants/hectare.

4.3.2. Insect Control and Counts

The field was covered with a floating row cover (10 m × 90 m) Agryl P-10 (10 $g·m^{-2}$) (American Agrifabrics, Alpharetta, GA) immediately after transplanting on 21 June 2006 and 18 June 2007. The row cover was secured with sandbags. The field remained covered until bud initiation, but the row cover was frequently removed for short periods of time to allow for weeding and to perform measurements. On 6 and 8

July 2006 and 3 and 8 July 2007, the number of imported cabbage worm larvae (*Pieris rapae*) was determined on the 18 broccoli plants located in the middle of each plot. After counting, *Bacillus turingiensis* var *kurstaki* was sprayed (140 g/ha) on 9 July 2006 and 11 July 2007. Broccoli were also sprayed just before harvest on both years.

4.3.3. SPAD measurements

Plant chlorophyll and N-status was measured on 11 July 2006 and 9 July 2007 using a Minolta SPAD-502 chlorophyll meter (Soil Plant Analysis Development, Minolta, Japan). SPAD readings were taken from several points on the fifth newest leaf of five randomly selected broccoli plants in each plot and were averaged. In 2006, readings were taken at ¼, ½, and ¾ of the leaf length on either side of the central vein, 1 cm from the edge of the leaf. In 2007, one measurement point was added at the tip of the leaf.

4.3.4. Statistical Analyses

Data was subjected to analysis of variance (ANOVA) using the general linear model (GLM) procedure (SAS_Institute, 2002). Since variance was homogeneous, data from both years was pooled for the analysis (Gomez and Gomez, 1984). Means were compared, and effectiveness of treatments was determined using the Student-Neuman-Keuls procedure. Single degree of freedom contrasts were also used to conduct preplanned comparisons of specific treatment combinations.

4.4. Results and Discussion

4.4.1. Insect abundance

There were significantly more (i.e., 4 times as many) imported cabbage worm larvae in the living mulch plots compared with the other treatments (Table 1). These results are contrary to the findings of several researchers (Bottenberg et al., 1997; Hooks and Johnson, 2001; 2002; 2004) who reported that living mulches significantly reduced number of lepidopterous pests in broccoli or cabbage. Similarly, our results are contrary to the findings showing the effectiveness of row covers to exclude lepidopterous pests from broccoli and cabbage (Adams et al., 1990; Evans et al., 1997). The frequent removal of the row cover for field measurements and weeding operations however might have increased the chance of introducing *Pieris rapae* under the row cover, and once under the cover the *P. rapae* was probably able to lay eggs and multiply. It is not known if row covers are as effective in controlling *P. rapae* in organic systems as they are in conventional systems. In organic systems, weeding is done mechanically or manually (which necessitates the periodic removal of the row cover), contrary to conventional systems where weeds can be removed chemically while leaving the row cover in place. However, this would not account for the difference in the abundance of *P. rapae* between living mulches and bare ground treatments.

The simultaneous use of both row cover and living mulch together may explain the results we observed. For both years, the period when the imported cabbage worm was ovipositing and initial larval development occurred before the living mulch was mown. At this time, the living mulch was higher (35-50 cm) than the broccoli plants (10-15 cm). The row cover was floating over the living mulch, and it did not touch or disturb the broccoli in the living mulch plots. Broccoli plants also grew straighter and taller during the first month in the living mulch plots as the mulch supported the cover above the broccoli. Under windy conditions, unsupported agrotextile row covers can cause abrasions if the material rubs the leaf surface which can limit crop growth (Wells and Loy, 1985; 1993). The living mulch combined with the row cover most likely created a micro-environment where the *P. rapae* could more easily lay their eggs and where the early instar of the larvae could grow and feed undisturbed. Hooks and Johnson (2002) reported that although the number of early instar imported cabbageworm were greater in monoculture than in living mulches, the number of late instar was greater in the living mulch because of a greater survival of the early instar stages in the intercropped habitats

4.4.2. Chlorophyll level of broccoli

Broccoli growing in living mulches had significantly lower SPAD readings than broccoli growing in compost or green manure plots (Figure 1). Living mulch broccoli therefore had a lower chlorophyll level, and thus most likely a lower nitrogen content. Myers (1985) has shown that *P. rapae*'s ovipositional response was influenced by the macronutrient content of the host plant. He reports that *P. rapae* could recognize the physiological state of brassica plants and preferentially laid eggs on plants that had higher N and P content, a greener color, and higher transpiration rate. Hovanitz and Chang (1964) also showed that green was more attractive to *P. rapae* for oviposition compared with white, yellow, red, blue and various combinations. It is believed that a host plant with a lower nutrient content would be less attractive to the butterfly for egg laying. Our results however show that there were less larvae in broccoli from the green manure and compost treatments which had greater chlorophyll levels and nitrogen content. We suppose it is because the sheltered environment created by the combination of row cover and living mulch was more important than the physiological state of the plant for *P. rapae* oviposition site selection. Gossard and Jones (1977) have shown that unfavourable weather conditions inhibited *P. rapae* flight and prevented egg-laying.

4.5. Conclusions

In this experiment, we have shown that on average, over two consecutive years, *P. rapae* larvae were more abundant in red clover and alfalfa living mulches than they were in the control, green manure or compost treatments. When row cover and living mulches are used together, the creation of a sheltered microenvironment appears to override plant nutrient content and chlorophyll level in the choice of a site for oviposition by *P. rapae*, and provides better conditions for the development of *P. rapae* larvae. Other factors that are know to influence oviposition such as the size of the plants (Jones and Ives, 1979) and percent leaf moisture (Renwick and Radke, 1983) were not measured in this experiment and should be further investigated. The general effectiveness of row covers to exclude *P. rapae* in organic systems where weeding is done mechanically or manually should also be tested.

4.6. Figures and Tables

TABLE 4.1: Effect of composts, green manures and living mulches on average number of imported cabbage worm (*Pieris rapae*) larvae in organic broccoli cv. 'Gypsy' grown in southwestern Québec in 2006 and 2007 (data pooled).

Treatments			Insects per plot
1 control - no fertilization (0N)			2.38b[z]
2 compost[y] - 135 kg N / ha (COMP-135)			2.38b
3 compost - 68 kg N / ha (COMP-68)			1.38b
4 alfalfa living mulch + clippings (ALF-CLIP)			8.88a
5 clover living mulch + clippings (RC-CLIP)			9.63a
6 alfalfa living mulch (ALF-LM)			9.38a
7 clover living mulch (RC-LM)			8.50a
8 alfalfa Green Manure (ALF-GM)			2.50b
9 clover Green Manure (RC-GM)			2.63b
Contrasts			
4,6,8 vs 5,7,9	-	RC vs ALF	NS
4,5 vs 6,7	-	LM vs CLIP	NS
8,9 vs 4,5,6,7	-	GM vs LM+CLIP	****
4,5,6,7 vs 1,2,3	-	LM vs COMP	****

[z] Values in a column followed by the same letter are not significantly different

[y] Compost: 35,71 N, 38,92 P, 24,04 K, 6,76 Ca, 7,79 Mg mg/g

[x] NS, *, **, ***, **** : Non-significant or significant at P≤ 0.05, 0.01, 0.001, 0.0001 respectively

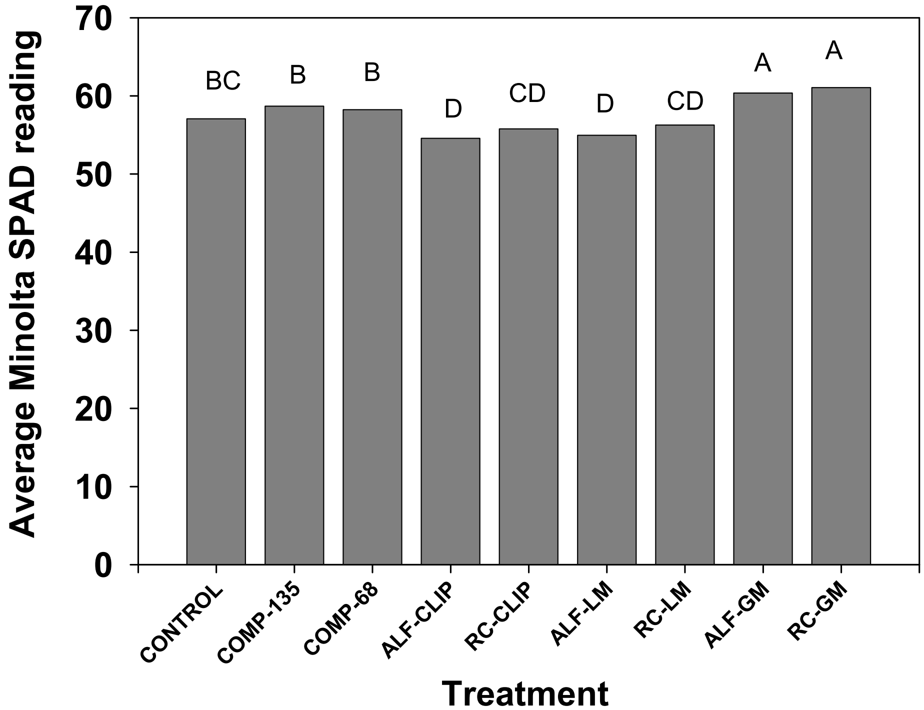

Figure 4.1: Effect of composts (COMP), and alfalfa (ALF) or red clover (RC) green manures (GM) and living mulches (LM) on Minolta SPAD meter measurement of organic broccoli cv. Gypsy grown in Soutwestern Québec in 2006 and 2007 (data pooled).

Minolta SPAD performed 10/07/2006 and 11/07/2007

COMP-138 and COMP-68 : turkey manure compost at 135 and 68 kg N/ha respectively

ALF-CLIP and RC-CLIP : alfalfa and red clover living mulches + clippings respectively

5. SUMMARY AND OVERALL CONCLUSIONS

Living mulch research has so far focused on the use of living mulches to reduce insect pest abundance, or on the ways to reduce competition between main crop and living mulch. Little work has been done to explore the potential to use leguminous living mulches to increase nitrogen supply in cropping systems, and to quantify their ability to meet crops requirements. This project was undertaken to study the use of perennial leguminous living mulches and green manures as an alternative, biologically-based, means of fertilizing organic vegetable crops.

Our first hypothesis was that the use of leguminous green manures or living mulches would result in broccoli yields, hollow stem incidence, plant nitrogen uptake and soil available nitrogen similar to those of plants fertilized with compost. This hypothesis was proven false. Our results demonstrate that alfalfa and red clover green manures increased broccoli yields and nitrogen availability. They provided greater broccoli head weight than controls (both unfertilized and fertilized with compost), and the alfalfa green manure also provided greater broccoli head diameter than controls. Both green manures led to a greater incidence of hollow stem disorder in broccoli than the unfertilized control, but were not significantly different from the compost fertilized broccoli. Maturation and harvest happened faster in the green manure broccoli compared to controls and to other treatments. At the beginning of the experiment Minolta SPAD readings were greater in green manure broccoli, indicating greater chlorophyll levels and N uptake. Total N content of the harvested heads was also greater for green manure broccoli. More N was available in the soil of the green manure plots compared with all other treatments and controls. Our estimates show that the biomass that was tilled into the soil in the green manure treatments contributed 139 kg N/ha for alfalfa and 131 kg N/ha for red clover. It is clear that green manures of alfalfa and red clover can supply N to a broccoli crop in excess of the recommended dosages, and that they can warrant satisfactory yields in organic cultivation systems.

Results were very different in the case of living mulches. Living mulches reduced broccoli yields, even though they increased N availability. Broccoli grown in living mulches had a head weight and head diameter equal or lower to that of the unfertilized control because of the important competition that occurred between the main crop and the living mulch, especially in 2007. Living mulches however reduced hollow stem, and delayed harvest compared with fertilized controls and green manures. Early in the season, SPAD readings were lower for living mulch broccoli, indicating reduced N uptake, probably because of competition by the living mulch. Total N content of harvested broccoli from living mulch plots was similar to that of all controls. Soil N availability varied among living mulch treatments, but indicated a tendency to increase N availability in the soil compared with controls, especially for the alfalfa living mulch treatments. Our estimates indicate that the partial rototilling and the mowing of the living mulch biomass over the season has returned 265 kg N/ha to the soil for alfalfa and 240 kg N/ha for red clover. Living mulches therefore appear to have the potential to supply N to a broccoli crop, but cropping practices have to be modified from the ones used in this experiment to limit competition between main crop and living mulch. More rapid and more frequent mowing in the early stages of growth of the broccoli could lead to better yields and results.

Results confirm our second hypothesis, which was that the nitrogen supplied from alfalfa green manures and living mulches would be greater than that of similarly managed red clover green manures and living mulches. Soil N measurements and our estimates of the N contribution from the biomass show that alfalfa has the potential to supply more N than red clover when used as a green manure or a living mulch.

Our third hypothesis, which suggested that applying living mulch clippings to the row would improve broccoli yields, plant nitrogen status and soil available nitrogen compared to a standard living mulch design was rejected. There were no differences in contrasts comparing LM and CLIP treatments, with the exception of one soil available N measurement, where results indicated greater N availability in the LM treatment compared with the CLIP.

Our last hypothesis was also rejected. It suggested that living mulches may reduce *P. rapae* pressure in the broccoli crop. We have shown that when living mulches are used in combination with row covers, the creation of a sheltered microenvironment leads to increased abundance and greater development of *Pieris rapae* in the broccoli.

6. RECOMMENDATIONS FOR FUTURE RESEARCH

This project has demonstrated the strong potential that perennial leguminous green manures have to supply N to organic vegetable crops; but as always in science, promising opportunities can turn out to be threatening or detrimental if misused. Our results show there was a much greater concentration of available N in the soil after an alfalfa or red clover green manure, which could lead to nitrate leaching and water resource pollution. Researchers should quantify the risks of nitrate leaching associated with the use of these techniques, and design cropping systems that minimize the threats to our natural ecosystems and drinking water resources. Another aspect that merits further investigation is the fact that broccoli heads had a higher N content following a green manure. It was not determined in this project whether the heads also had a higher nitrate content, but as nitrates in foods represent a risk to human health, research projects should be designed to test the nitrate content of broccoli and other vegetables produced with green manures, living mulches, and other organic sources of nutrients.

Our conclusion that living mulches can increase nitrogen availability in the soil also leads to a lot of new research possibilities. How could the system used in this project be modified and improved to minimize the competition between main and associated crop, and to maximize the nutrient benefits? The system we had designed incorporated irrigation, tilled strip and mowing in the hopes of minimizing competition, but was not successful, probably because the mowing was not aggressive enough. More research is needed to determine how many times, at what stage of growth, and at which height perennial leguminous mulches should be mown to avoid the crop-intercrop interaction that was observed in this experiment.

With regards to our findings about the interaction between the use of row covers and living mulches, and its impact on *P. rapae* abundance; we would suggest that more research be done to investigate the general effectiveness of row covers to exclude *P. rapae* in organic systems, where weeding is done mechanically or manually (implying periodic removal of the row cover). It is not clear whether *P. rapae* was introduced under the row cover in this experiment due to the frequent removal of the floating row cover for experimental purposes or if normal weeding practiced on organic farms would also lead to the introduction of the insect under the cover. In the case where row covers retain a certain efficacy under normal practices, then the use of some form of perennial living mulches to replace hoops and to reduce the crop damage due to the friction of the cover on the crop plants should be further investigated.

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