

Evaluation of annual companion crops for the establishment of perennial
forages in Québec

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August 2021

A thesis submitted to McGill University in partial fulfillment of the
requirements of the degree of Master of Science

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Abstract

One of the main agricultural sectors in Canada is the dairy industry for which forages are used as a base feed. When perennial forages such as alfalfa (*Medicago sativa* L.) are solo seeded, their yield is often minimal, and they are not highly competitive against weeds. Seeding annual companion plant species along perennials is an approach to increase yields and control weeds during the seeding year. Our main objective was to evaluate the potential of different annual species used as companion crops for the establishment of perennial forages in a context of climate change. More precisely, six annual companion crops (i.e., Berseem clover [*Trifolium alexandrinum* L.], forage pea [*Pisum sativum* L.], annual ryegrass [*Lolium multiflorum* Lamarck], oat [*Avena sativa* L.], Japanese millet [*Echinochloa esculenta* (A. Braun) H. Scholz], and sudangrass [*Sorghum* × *drummondii* (Nees ex. Steud.) Millsp. & Chase]) were seeded with a mixture of alfalfa and timothy (*Phleum pratense* L.) at three different seeding dates and three contrasting sites in Québec. They were compared based on forage yields, botanical composition (i.e., alfalfa, timothy, companion crop and weeds proportions), nutritional value, weed control potential, and establishment of the perennial mixture. Across environments, oat, Japanese millet and sudangrass increased total forage yields in the seeding year (82 % increase on average) and helped control weeds (58 % decrease on average) for the first two seeding dates in the spring compared to alfalfa and timothy established without a companion crop. However, the stem density of perennials and the nutritional value of the mixture were reduced with sudangrass, followed by Japanese millet and oat in the seeding year compared to the control. Total forage yields were also reduced (26 % decrease on average) in the post-seeding year with these three annual companion crop treatments compared to the control.

For the August seeding, the use of oat resulted in the highest total forage yields, followed by Japanese millet compared to the control. However, similar results to the other seedings were observed (i.e., reduced perennial forages establishment and nutritive value). Thus, the use of sudangrass and Japanese millet as companion crops for the establishment of perennials would be appropriate in order to increase forage yields in the seeding year. Oat also provided high forage yields in the seeding year without affecting the other variables evaluated to the same extent. For example, aNDF concentrations were greater by 32 % with oat and 37 % with Japanese millet and sudangrass on average compared to the control. Peas offered intermediate forage yields and high-quality forages in the seeding year without having residual effects on total forage yields in the post-seeding year.

Résumé

L'industrie laitière est l'un des secteurs les plus importants au Canada, pour laquelle les plantes fourragères sont majoritairement utilisées. Lors du semis de plantes fourragères pérennes, comme la luzerne (*Medicago sativa* L.), les rendements sont minimaux et les plantes ne sont pas compétitives contre les mauvaises herbes. L'utilisation d'espèces annuelles comme plantes-abri avec des plantes pérennes est une approche qui peut permettre d'augmenter les rendements et réduire les mauvaises herbes lors de l'année de semis. Le but de cette expérience était d'évaluer le potentiel de différentes plantes annuelles comme plante-abri lors de l'établissement de plantes fourragères pérennes dans un contexte de changements climatiques. Précisément, six plantes-abri annuelles (i.e., trèfle d'Alexandrie [*Trifolium alexandrinum* L.], pois fourragers [*Pisum sativum* L.], ray-grass annuel [*Lolium multiflorum* Lamarck], avoine [*Avena sativa* L.], millet japonais [*Echinochloa esculenta* (A. Braun) H. Scholz], et herbe du Soudan [*Sorghum × drummondii* (Nees ex. Steud.) Millsp. & Chase]) ont été semées avec un mélange binaire de luzerne et fléole des prés (*Phleum pratense* L.) à trois dates différentes et trois sites avec différentes conditions pédoclimatiques au Québec. Les traitements ont été comparés par rapport aux rendements, composition botanique (i.e., proportions de luzerne, fléole des prés, plante-abri et mauvaises herbes), valeur nutritive, contrôle de mauvaises herbes et établissement du mélange binaire. À tous les environnements, l'utilisation de l'avoine, du millet japonais et de l'herbe du Soudan comme plantes-abri, semés en début de saison, ont augmenté les rendements fourragers totaux dans l'année de semis (de 82 % en moyenne) et permis de réduire les mauvaises herbes (de 57 % en moyenne), comparés au témoin du mélange de luzerne et fléole des prés semé sans plante-abri. Cependant, la valeur nutritive

des fourrages ainsi que le nombre de tiges de plantes pérennes ont été réduits dans l'année de semis avec l'utilisation de l'herbe du Soudan suivi du millet japonais et de l'avoine comme plantes-abri comparés au témoin semé sans plantes-abri. Les rendements fourragers totaux lors de l'année post-semis ont également été réduits (de 26 % en moyenne) avec l'utilisation de ces trois espèces de plantes-abri dans l'année de semis comparativement au témoin. En août, l'avoine suivi du millet japonais ont augmenté les rendements, mais ont eu les mêmes effets négatifs qu'aux dates de semis précédentes. Donc, l'utilisation de l'herbe du Soudan et du millet japonais comme plantes-abri pour l'établissement de plantes pérennes serait appropriée afin d'augmenter les rendements dans l'année de semis. L'utilisation de l'avoine comme plante-abri permet également d'augmenter les rendements tout en affectant moins les autres variables évaluées. Par exemple, l'utilisation de l'avoine comme plante-abri dans l'année de semis a augmenté les concentrations de aNDF de 32 % vs de 37 % en moyenne avec l'herbe du Soudan et le millet japonais comparés au témoin. Les pois utilisés en plante-abri ont permis d'obtenir des rendements fourragers totaux intermédiaires et des fourrages de haute qualité lors de l'année de semis sans avoir d'effets résiduels sur les rendements fourragers totaux dans l'année post-semis.

Acknowledgements

I would like to express my sincere gratitude to my supervisor Dr. Philippe Seguin, for giving me the opportunity to work on this project as well as his advice and guidance for all the field work, data analyses, writing and editing of my thesis, poster and papers, and his overall support throughout the past years.

I would also like to thank Dr. Caroline Halde, from Université Laval, and Céline Goerlette, from the Centre de Développement Bioalimentaire du Québec (CDBQ), for supervising the experimental sites at Saint-Augustin-de-Desmaures and La Pocatière, respectively. I am grateful for their comments and suggestions for articles, poster and presentations related to this project. I would like to thank Dr. Gaëtan F. Tremblay, from Agriculture and Agri-Food Canada (AAFC), for the laboratory analyses of the samples as well as his insight and suggestions on the data analysis and the different articles, poster and presentation related to this project. I am also grateful for the contribution of Huguette Martel and Ayitre Akpakouma from Ministère de l'Agriculture, des Pêcheries et de l'Alimentation du Québec (MAPAQ) for their insight on the initial experimental design as well as their comments on the different documents associated with this project. Thank you to Dr. Valério Hoyos-Villegas for serving on my research advisory committee.

I am greatly thankful for the different research assistants and technicians at the three sites for making my project one of their priorities. Particularly, I thank Geneviève Bégin (AAFC) for all the chemical analyses related to this project, Héroïse Henry (Université Laval) for taking care of the Saint-Augustin-de-Desmaures experimental site, Élisabeth Dubé (CDBQ) for taking care of the La Pocatière site, and Parghat Gopal, Marc Samoisette

and Caroline Matteau (McGill University) for helping me at the Sainte-Anne-de-Bellevue site. I would also like to thank all the graduate and summer students who helped out for the harvests and collection of data; particularly, my lab mates, John Battu, Sayanthika Thevakumaran and Xavier Godin. Finally, I want to thank my boyfriend, and all my friends and family that have shown me continuous support throughout the years.

I was a recipient of scholarships from the Natural Sciences and Engineering Research Council of Canada (NSERC), the Fonds de Recherche du Québec-Nature et Technologie (FRQNT), and McGill University. Ce projet a été réalisé en vertu du sous-volet 2.2 du programme Prime-Vert 2018-2023 et il a bénéficié d'une aide financière du Ministère de l'Agriculture, des Pêcheries et de l'Alimentation du Québec (MAPAQ).

Contribution of authors

This thesis is written in the form of a traditional thesis according to the “Guidelines Concerning Thesis Preparation” of McGill University and is ultimately based on a collaborative work between McGill University, Université Laval, CDBQ, AAFC, and MAPAQ. The protocol and experimental design presented in Chapter 3 (sections 3.1 and 3.2) were developed primarily by P. Seguin in collaboration with C. Georlette, C. Halde, G. F. Tremblay, H. Martel, and A. Akpakouma. The field work of the methods presented in sections 3.1 and 3.2 were performed and supervised by P. Seguin and S. St-Pierre-Lepage at Sainte-Anne-de-Bellevue, by C. Halde and her team at Saint-Augustin-de-Desmaures, and by C. Georlette and her team at La Pocatière. The chemical analyses for the five environments (section 3.3) were completed under the supervision of G. F. Tremblay. The statistical analyses in section 3.4 are the original work of S. St-Pierre-Lepage, with suggestions from P. Seguin. Data interpretation in Chapters 4 and 5 was done by S. St-Pierre-Lepage, with contributions from P. Seguin.

List of abbreviations

ADF	Acid detergent fiber
aNDF	Neutral detergent fiber assayed with a heat-stable α -amylase and sodium sulfite
CP	Crude Protein
DM	Dry matter
GDD5	Growing degree-days calculated using a 5°C basis
NDF	Neutral detergent fiber
RPD	Ratio of prediction to deviation
SD	Standard deviation of validation set
SEP(C)	Standard error of prediction corrected for bias
TN	Total nitrogen
VNRIS	Visible and near infrared reflectance spectroscopy

Chapter 1. Introduction

1.1 General introduction

In Québec, one of the main crops produced are perennial forages which are normally grown in mixtures. In the province, the most common mixture consists of alfalfa (*Medicago sativa* L.) and timothy (*Phleum pratense* L.), due to the fact that it is one of the mixtures with the greatest yield potential and nutritive value (Bélanger et al., 2014; Pomerleau-Lacasse et al., 2019). There are different strategies normally used to establish perennial forage species as they produce less during the seeding year compared to subsequent ones, especially in warmer and dryer conditions, and as they are less competitive compared to annual weed species, which can lead to weed encroachment. Weeds not only may reduce the establishment and yield of forage species but also have the potential to decrease the nutritional value of forage harvested depending on the species (Hoy et al., 2002; Sheaffer et al., 2014). The use of herbicides during the establishment of perennial forages is an option available to reduce weeds, however, this approach may be limited by herbicides selectiveness when a mixture of grass and legumes is established. Another strategy available to reduce weeds during the seeding year and to increase forage yields is to establish perennial forages with annual companion crops; most producers currently using oat (*Avena sativa* L.). While there is a growing interest for other annual species used as companion crops for the establishment of perennial species, such as warm-season grasses, cool-season grasses and legumes, there has been to date no systematic evaluation of the various options available across the Province of Quebec seeded at different times during the growing season (ranging from spring to late summer).

1.2 Objectives and research hypothesis

1.2.1 Overall objective

The main goal of this study is to evaluate six different annual companion crop species used for the establishment of perennial forage species seeded at three different times during the growing season.

1.2.2. Specific objectives

Our specific objectives are to compare the effects of the different treatments (annual companion crops and seeding dates) at contrasting sites on:

- a) Forage yield
- b) Botanical composition (i.e., alfalfa, timothy, companion crop and weeds proportions)
- c) Nutritional value
- d) Establishment and winter survival of alfalfa and timothy.

1.2.3 Hypothesis

We hypothesize that the use of warm-season grasses, such as sudangrass [*Sorghum × drummondii* (Nees ex. Steud.) Millsp. & Chase] and Japanese millet [*Echinochloa esculenta* (A. Braun) H. Scholz], as companion crops for the establishment of perennial forages will result in the highest forage yields in the seeding year, without negatively affecting the establishment and winter survival of perennial species when compared to the use of oat, berseem clover (*Trifolium alexandrinum* L.), ryegrass (*Lolium multiflorum*

Lamarck), forage pea (*Pisum sativum* L.), and a control of the alfalfa-timothy mixture seeded without a companion crop.

Chapter 2. Literature review

2.1 General background

2.1.1 Importance of forages

In Canada, especially in Québec and Ontario, the dairy industry is one of the main agricultural sectors. In 2019, this industry contributed to 25% of the total revenues for the agricultural sector in the province of Québec (MAPAQ, 2020). Forage crops are destined primarily to feed the ruminants at the base of this industry. Thus, it is not surprising that most of the cultivated land in Québec is grown with forage crops, with over 655 000 hectares used to grow forages (Institut de la Statistique du Québec, 2020). Although two thirds of the harvested forages are used as base feed for dairy production, the rest is also destined as feed to the beef-calf production, horse industry, and sheep production (MAPAQ, 2018).

2.1.2 Perennial forages in mixtures

Perennial forage crops have typically been grown either as solo-seeded stands or in mixtures. In Québec, perennial forages are usually grown in mixtures. The most widely grown forage legume in Québec is alfalfa, also known as the queen of forages. It is normally mixed with perennial grasses, the most popular one being timothy. This binary mixture results in high quantity and adequate quality forages for ruminants (Bélanger et al., 2014). Growing forages in mixtures has advantages over solo-seeded stands. For

instance, grasses in a mixture with alfalfa can help reduce erosion and weed encroachment. They also allow for more persistence of the mixtures over time since alfalfa is more easily winterkilled compared to timothy (Lackman, 2001). If the forage crops are ensiled, the process is quicker with a mixture compared to solo-seeded legume forages since the latter have a higher buffering capacity than forage grasses and, thus, require more time to reduce the pH at acceptable silage levels (i.e., $\text{pH} \leq 4$; Moore et al, 2020). If solo-seeded alfalfa was to be used in pasture, it could cause bloat to ruminants, adding grasses reduces the risks of bloating (Lackman, 2001).

2.2 Alfalfa

2.2.1 General description and benefits

Alfalfa is a perennial legume with a deep tap root. This tap root confers resistance to drought (CRAAQ, 2005). Growing alfalfa has multiple other benefits. For instance, it can help improve some physical soil characteristics such as the structural stability of the soil and its porosity (Rasse et al., 2000). If it is included in a rotation with grasses such as corn, it can reduce the incidence of diseases and parasites, such as pests, bacteria and virus, since it is from a different family (*Fabaceae*). This legume can also fix nitrogen through its nodules as it forms a symbiotic relationship with a rhizobium, *Sinorhizobium meliloti*. When it is well-managed, it can persist and be productive for at least three years in Québec and Ontario (CRAAQ, 2005; OMAFRA, 2009). Its primary use remains hay and silage making since it is a high-yielding forage crop. It is less often used in pastures since it can cause bloat in ruminants (Undersander et al., 2011). It is not an issue when given in silage since its proteins involved in the bloat process are degraded during ensiling (Majak et al.,

2001). It is, however, possible to select varieties that are more suited for grazing (Undersander et al., 2011).

2.2.2 General management

Although alfalfa is widely grown across Québec, it is not well suited to all types of soils and conditions. It does best in well drained and fertile soils. It also thrives when the soil pH is between 6.6 and 7. Thus, choosing a field with the proper conditions is a key to successfully growing alfalfa. Another factor for its success is its fertilization. It does require nitrogen for its establishment but to a lower extent compared to other species since it can fix this nutrient once its nodules are formed. However, phosphorous and potassium are important elements for its establishment. Phosphorous promotes root growth while potassium plays a role in alfalfa yield and persistence (Undersander et al., 2011). Furthermore, if boron levels are too low in the soil, it can limit its development and establishment. Thus, fertilizing in function of the soil nutrient levels can improve alfalfa establishment (CRAAQ, 2005; OMAFRA, 2009).

Alfalfa can either be solo-seeded or seeded in a mixture with grasses. The latter is favored in Québec since mixtures are more resilient to winters and have an improved persistence. In a mixture, it is locally recommended to seed alfalfa at a rate of 9 kg ha⁻¹ (CRAAQ, 2005). In general, alfalfa is chosen over other forage legumes due to its high yielding capacity and high quality (i.e., source of fiber and nutrients, such as calcium, that meet the requirements of dairy cows). Forage nutritive value is often determined based on certain key variables including neutral detergent fiber (NDF), acid detergent fiber (ADF) and crude protein (CP) concentrations. High fiber concentrations are inversely correlated

with forage quality (i.e., the higher the fiber concentrations, the lower the nutritive value). They are assessed using NDF, which includes three fiber components: hemicellulose, cellulose and lignin, and is negatively correlated with intake by ruminant animals (CRAAQ, 2005). Fiber concentrations can also be evaluated with ADF, which includes two fiber components: cellulose and lignin, and is negatively correlated with forage digestibility (CRAAQ, 2005). Crude protein estimates the total amount of protein available in the feed based on the quantity of nitrogen (N) present ($CP = N \times 6.25$), and is positively correlated with forage quality (CRAAQ, 2005). Ideally, alfalfa has a CP concentration of 20 %, ADF concentration of 30 % and NDF concentration of 40 % when it is harvested for the feeding of dairy cows. However, the CP, the energy and the nutrients' levels in the plants depend on their stage of maturity at harvest. To optimize forage yields and quality, it is suggested to harvest alfalfa at the early flower stage. At the early bud stage, the nutritive value of alfalfa is higher, but forage yields are reduced. Harvesting at an earlier stage of maturity can also reduce the persistence of the stand (OMAFRA, 2009). So, it is generally recommended to cut at the early flower stage of alfalfa for the first harvest, corresponding to the best compromise between yield, nutritive value, and persistence (OMAFRA, 2009; Pomerleau-Lacasse et al., 2019). Subsequent harvests can be taken at the same stage of maturity, which often corresponds to a period of approximately 30 to 40 days between each harvest (CRAAQ, 2005; OMAFRA, 2009). In Québec, this leads to 3 to 4 cuts per year on average (CRAAQ, 2005).

2.3 Timothy

Timothy is a bunch-type perennial grass that does not abundantly tiller. It is a widely grown grass across Québec since it is well suited to prevailing climatic conditions. For instance,

it survives cold winters well and is adapted to a cool and humid climate since it occurs naturally in environments between the 35° and 55° N latitude. It performs well in heavy humid soils with variable drainage and when the soil pH is between 6.0 and 6.5, but it can also tolerate slightly more acidic soils (CRAAQ, 2005). It can grow as tall as 80 to 100 cm high and is well suited for hay making, silage making and grazing, especially if the pasture is managed in a rotation. Timothy hay is primarily directed towards the horse industry (OMAFRA, 2009). It has a high palatability and quality early in the season, but as it matures, it loses nutritive value rapidly. The ideal stage of maturity at harvest is thus the boot stage, when the inflorescence of the plant emerges, or prior for this species. Since it has a limited tillering capacity, it does not compromise the establishment of legumes such as alfalfa and birdsfoot trefoil (*Lotus corniculatus* L.) when it is used in mixtures. It is locally recommended to seed this species at a rate of 7 kg ha⁻¹ when seeded in a mixture with alfalfa (CRAAQ, 2005; OMAFRA, 2009).

One major downside to timothy is that its root system is shallow: 90% of the roots grow in the first 20 cm of the soil. Thus, it has a poor drought tolerance and does not do well in warmer and dryer conditions. This also means that, after the first harvest at the beginning of the summer, its regrowth is often limited due to dryer and warmer days in mid-summer. Its persistence can also be compromised depending on the harvest schedule: a more intensive cutting management (3 to 4 cuts per summer) can affect stand survival. When timothy is used, the nitrogen needs are higher than with solo-seeded legumes and thus must be managed accordingly to ensure successful establishment (CRAAQ, 2005).

2.4 Establishment challenges and strategies

In general, perennial forage species establish slowly during the seeding year. During that year, forage yields typically remain relatively low compared to subsequent post-seeding years. One problem that arises from the slow establishment of perennial forage seedlings is weed encroachment. When weeds compete with the perennial forages, it can lead to a decrease in the population and proportion of perennials harvested in the seeding year, as well as in post-seeding years (Spandl et al., 1999; Undersander et al., 2011). However, the total yield in the seeding year could remain similar to the yield in post-seeding years due to a greater proportion of weeds harvested. Furthermore, depending on the weed species, quantity and stage of maturity, the forage quality of the harvested material can be reduced, and drying time can be increased, as some weeds may take longer to dry to a target moisture content compared to some forage species (Spandl et al., 1999; Renz, 2015). Thus, controlling weeds during the seeding year is an important factor in the successful establishment of perennial forage species.

2.4.1 Herbicides

There are many strategies that can be used in order to control weeds in the seeding year of perennial forage species. Typically, alfalfa has been solo-seeded or seeded in mixtures with perennial forage grasses. In both cases, one of the strategies is to use herbicides to control weeds. To control perennial weeds, it is suggested to apply an appropriate herbicide the fall prior to the establishment of the perennials. If the herbicide is used in the spring when the perennial forages are seeded, it can delay seeding. In order to control annual weeds, there are many herbicide options when alfalfa is seeded, especially if solo-seeded. It is also

possible to incorporate an herbicide before planting alfalfa. For instance, when herbicides, such as Eptam or Treflan containing the active ingredients EPTC and trifluralin, respectively, are incorporated at a depth of 5 to 8 cm, they can help to control annual grasses and some annual broad-leaf weeds. If Eptam is not incorporated properly, it can, however, cause injuries to alfalfa. These herbicides cannot be used if alfalfa is seeded in a mixture with grasses since they would adversely affect the perennial forage grasses as well. It is also possible to apply certain herbicides to solo-seeded alfalfa after its emergence. Some examples include Butyrac (2,4-DB), Prowl H20 (pendimthalin) and glyphosate (in the case of Roundup Ready alfalfa). These herbicides can act on some annual and perennial broadleaf weeds as well as grass weeds and offer typically a good control against them. An herbicide containing the active ingredient bromoxynil can be used when alfalfa is not seeded alone, for example in mixtures with perennial forage grasses, to control broadleaf weeds after the emergence of the seedlings. It should not be applied when alfalfa has less than four trifoliate leaves. A period of 30 days after application needs to be respected before harvesting (Undersander et al., 2011). Weed control during the establishment of alfalfa-grass mixtures can thus be difficult to achieve.

2.4.2 Companion crops

Another strategy used to reduce weed encroachment during the establishment of perennial forage species is to use annual companion crops (Canevari et al., 2000; Undersander et al., 2011). In Eastern United States, it is estimated that around 60% of alfalfa is seeded with a companion crop (Spandl et al., 1999). Typically, in the Midwest, small-grain cereals such as oat (*Avena sativa* L.) have been used as companion crops for alfalfa. Companion crops present many advantages: reduction in weed competition for the establishing perennial

stand, reduction in soil erosion, and increase in total forage yield during the seeding year which would normally remain low (Hoy et al., 2002). However, there are also disadvantages to seeding perennial forage species with an annual companion crop. The annual species compete against weeds, but they also compete with alfalfa and perennial forage grasses for light, moisture and nutrients. This can result in a reduced contribution of the perennials to forage harvested in the seeding year as well as reduced plant density, and thus persistence in the long term (Hoy et al., 2002; Undersander et al., 2011). It is suggested to reduce the seeding rates of the annual companion crops or to harvest the mixture based on the maturity stage of the annual companion crop, earlier than the ones suggested for solo-seeded annual forage crops, in order to minimize their effects on the establishing perennial species (Spandl et al., 1999; Undersander et al., 2011).

2.4.3 Comparison of different establishment strategies

A study conducted by Hoy et al. (2002) compared different establishment strategies: multiple herbicides management strategies, oat companion crop harvested either for forage or grain, and a winter-killed mulch residue of fall seeded oat. In the seeding year, the greatest forage yields were obtained with the oat companion crop harvested for silage or for grain and with alfalfa seeded into the fall-seeded oat mulch, while some of the herbicides had negative effects on forage yields compared to the control of solo-seeded alfalfa without herbicide and even caused some injury to alfalfa. The best weed control was observed when alfalfa was seeded with oat or when alfalfa was solo seeded along with the use of an herbicide containing the active ingredient sethxydium. However, the use of an oat companion crop had adverse effects on the quality of the forage harvested: reduced quality at the second harvest, caused by the advanced stage of maturity of oat in the second

harvest compared to the other harvests that year. In the post-seeding year, there were no differences between treatments used in the seeding year on forage yield. Thus, companion crops did not affect forage yield in the post-seeding year more than the herbicide treatments or the mulch treatment and appeared to be a good option to establish alfalfa. Another study conducted by Spandl et al. (1999) compared the establishment of alfalfa seeded alone and in mixtures with different perennial forage grasses such as timothy with and without an oat companion crop. Similar to Hoy et al. (2002), in the seeding year, they observed that the treatments with the companion crop had significantly lower proportions of weeds in the forage harvested than when no companion crop was used. However, there were also lower proportions of perennial grasses in the forage harvested. The perennial forage grasses also contributed to a reduction of weeds compared to the solo-seeded alfalfa control treatment when they were seeded with alfalfa and without a companion crop but to a lesser extent than the oat companion crop. In the second post-seeding year, it was observed that the treatments established with a companion crop had higher proportions of weeds. This could be due to a reduced plant density of perennial forage grasses in the seeding year, due to the companion crop competition, and also due to the fact that grasses, in post-seeding years, were not able to compete as well with weeds compared to the ones established without the companion crop. Thus, in the long term, it is possible that the reduced plant density caused by the companion crop leads to higher proportions of weeds in the post-seeding years, even though it offers more weed control during the establishment.

2.5. Companion crop species description

2.5.1 Oat

There are different options when it comes time to choosing an annual companion crop. They can be grouped into a few categories: small-grain cereals, cool-season grasses, legumes, and warm-season grasses. Small-grain cereals such as oat have traditionally been used as companion crops. Oat is a small-grain spring cereal usually preferred because of its wide adaptation: it is best suited to loam and clay soils, but it can also do well in sandy soils compared to other types of cereals like wheat. It does well when the soil pH is between 5.8 and 7.0 (CRAAQ, 2005). Oat has a high nutritive value and palatability in the vegetative stage, but forage yields are lower than at later phenological stages. However, as it matures, it quickly loses quality, especially past the milk dough stage. Thus, if its use is intended for the nutrition of dairy cows, it is best to harvest at the boot stage (i.e., best compromise between forage nutritive value and quantity). This earlier removal can also help minimize the competitive effect of oat if used as a companion crop for the establishment of perennial species (CRAAQ, 2005; Undersander et al., 2011).

In many studies, it was observed that oat, when used as a companion crop, is competitive with weeds and thus provides good weed control. It also increases overall forage yields in the seeding year, but the proportion of perennials such as alfalfa are reduced (Peters, 1961; Becker et al., 1998; Sheaffer et al., 2014). Its high competitiveness has been shown to have an impact on the establishment of perennial forages: lower plant densities have been observed with treatments containing oat used as companion crop in comparison to alfalfa seeded without a companion crop, especially when conditions are

unfavorable for establishment such as in a drought year (Peters, 1961; Spandl et al., 1999; Canevari et al., 2000).

However, it does not mean that forage yields in the post-seeding year are affected. For instance, Sheaffer et al. (2014) observed no significant residual effect of oat in post-seeding years in one environment and even observed higher alfalfa yields for the oat companion crop treatment compared to an unweeded solo-seeded alfalfa control in post-seeding years for another environment. Oat did not have a significant effect on post-seeding years forage yields in the study of Hoy et al. (2002) reported earlier. Peters (1961) also reported that, when precipitations were adequate during the seeding year, the regrowth of alfalfa once oat was removed was equal to a non-weedy solo-seeded alfalfa control and forage yields were not affected later on. Others have reported that in order for oats to not significantly affect alfalfa establishment and its yields in the post-seeding years, and still provide weed control, oat's seeding rate should be reduced compared to solo-seeded oat (Lanini et al., 1991, Undersander et al., 2011). It is suggested to reduce oat's seeding rates by 30% (60 to 90 kg ha⁻¹ suggested) in order to minimize competition with establishing perennial forages (NSDU, 2018).

2.5.2 Annual ryegrass

Another option that has been long considered as a companion crop is annual ryegrass, also called Italian ryegrass (*Lolium multiflorum* Lamarck). Italian ryegrass is a biennial grass which usually does not survive under Québec winter conditions; thus, it often acts as an annual. Two types of cultivars exist: diploids and tetraploids. Usually, tetraploids do better as companion crops with legumes since they have thinner leaves (CRAAQ, 2005;

Scheinder and Undersander, 2008). It is well suited to soils with a pH greater than 6.0, but it can tolerate more acidic soils. It requires high amounts of nitrogen for optimal forage yields and quality (CRAAQ, 2005).

Studies conducted on the use of annual ryegrass as an annual companion crop have concluded that annual ryegrass provided high quality forage and high forage yields during the seeding year (Sulc et al., 1993; Wiersma et al., 1999; Coulman et al., 2019). In the study conducted by Wiersma et al. (1999), ryegrass produced more biomass during the seeding year than an oat companion crop. The opposite was observed by Sulc et al. (1993): it was reported that ryegrass generally produced less biomass during the seeding year than oat, but it still increased forage yields when compared to solo-seeded alfalfa. It was also more constant throughout the season while oat did not contribute much biomass after the first cut. For both studies, annual ryegrass had inconsistent effects on alfalfa yields in the post-seeding year. It was observed that when the conditions were favorable to a rapid ryegrass growth in the seeding year or with a high ryegrass seeding rate, alfalfa yields were reduced in the post-seeding year. This was not true when ryegrass grew moderately during the seeding year (Sulc et al., 1993; Wiersma et al., 1999). It was also shown by Coulman et al. (2019) that ryegrass reduced the perennial forage crops' populations, but their yields were not affected in the post-seeding years. Thus, a low seeding rate of annual ryegrass (i.e., 6 kg ha⁻¹) is suggested when used as a companion crop for the establishment of perennial species (Sulc et al., 1993; Wiersma et al., 1999; Schneider and Undersander, 2008).

2.5.3 Berseem clover

Berseem clover (*Trifolium alexandrinum* L.), also called Egyptian clover, is an annual legume with a combination of a deep tap root along with branched roots. It does best in well drained soils with a pH above 5.8 (Bélanger, 2019). Shrestha et al. (1998) evaluated solo-seeded berseem clover used as an emergency source of forage. They observed that it could be cut twice in the summer and provide high forage yields. It also had similar CP concentrations than solo-seeded alfalfa, but with higher fiber concentrations.

The use of berseem clover as a companion crop for the establishment of perennial species has also been suggested. It can improve forage yields during establishment by 10 to 40 % compared to solo-seeded alfalfa (Canevari et al., 2000). Nelson et al. (1965) reported that berseem clover performed best in cool environments and, if they were seeded early enough, they could yield similar to forage oat. Berseem clover also produces high quality forage with 18 % CP and low ADF concentration (Nelson et al., 1965; Bélanger, 2019). In the study of Nelson et al. (1965), berseem clover, however, often did not offer much weed control when used as a companion crop. The best weed control was obtained when it was seeded at a rate of 22 kg ha⁻¹. When established at this rate, it did not negatively affect alfalfa establishment nor forage yields in the post-seeding year. Contrary to the findings of Nelson et al. (1965), a trial conducted by Bélanger (2019) in Québec suggested that berseem clover did best when there were high precipitations and temperatures. They also suggested that when berseem clover is seeded at rates greater than 5 kg ha⁻¹, it reduces alfalfa establishment. Canevari et al. (2000) also recommended a seeding rate below 10 kg ha⁻¹. The seedlings can die if they are exposed at temperatures under 2°C, so, it is best to wait later in the spring before seeding berseem clover (Bélanger, 2019). In the case of

Nelson et al. (1965), lower seeding rates were not recommended since it did not help control weeds and the latter reduced alfalfa establishment.

2.5.4 Field pea

Peas (*Pisum sativum* L.) are also another species that can be used as a source of forage and as a companion crop for the establishment of perennial forage species. Peas are a legume that forms a symbiotic relationship with a rhizobium, *Rhizobium leguminosarum*, thus, it does not require much nitrogen fertilization (Owens and Carr, 2004). Boron can be toxic to peas, so this nutrient needs to be managed carefully (CRAAQ, 2013). They grow best in cool and moist environments, and preferably in drained soils with a pH between 4.2 and 8.7, ideally at 7.0 (Fleury, 2017). It has been demonstrated that field peas can be grown and harvested as forages to feed dairy cows. It contains high amounts of protein and starch, providing both energy and protein. When ensiled at the pod filling stage, it can thus make a suitable feed for dairy cows and could even replace other types of silage such as alfalfa silage for example (Mustafa and Seguin, 2004; Borreani et al., 2007). However, the pea protein is highly and rapidly degradable in the rumen (almost 80 %). This is not desirable since a high ratio of rapidly degradable protein can lead to losses of microbial protein and nitrogen when there is not sufficient energy available to the microbes in the rumen (Mustafa and Seguin, 2004).

Typically, when peas are used as annual companion crops for the establishment of perennial species, they are grown along with small-grain cereals. Peas are generally used to increase the quality of the forage mixtures through an increase in CP and a decrease in NDF concentrations (Owens and Carr, 2004; OMAFRA, 2009). It can also be used as a

companion crop by itself. Sheaffer et al. (2014) evaluated different companion crops for the establishment of alfalfa. Field peas increased forage yields compared to a solo-seeded alfalfa control but was the lowest producing companion crop when compared to others such as oat and ryegrass. It offered weed control in some of the environments evaluated and did not reduce the alfalfa yields in the post-seeding year; so, it led to a successful alfalfa establishment. Cupina et al. (2010) evaluated the establishment of red clover (*Trifolium pratense* L.), another perennial legume, with field peas. It was shown that field peas increased forage yields in the seeding year, especially at the first cut, and did not alter the red clover yields in the post-seeding year compared to a solo-seeded red clover control.

2.5.5 Japanese millet

In recent years, there has been increased interest in the use of warm-season annual grasses. This includes plants from the millet family. In Québec, one of the most suited millet species is Japanese millet (*Echinochloa esculenta* (A. Braun) H. Scholz]; CRAAQ, 2005). This grass does best in drained heavy soils, but also tolerates sandy soils, with a pH between 5.8 and 7.0. It is best adapted to regions with at least 2300 corn heat units (CHUs) and it should be seeded in a warm soil ($>12^{\circ}\text{C}$; OMAFRA, 2009). It requires high amounts of nitrogen in order to grow successfully. Japanese millet can be grazed when it is 25 to 30 cm high. It can also be harvested as hay or silage when it reaches 50 to 75 cm in height. When Japanese millet is harvested prior to the boot stage, it has good palatability and quality for ruminants (CRAAQ, 2005). When seeded on its own for hay, as an emergency source of forage for example, Lang (2001) reported dry matter (DM) forage yields of 12.4 Mg ha^{-1} while Peterson et al. (2008) reported DM forage yields up to 9.9 Mg ha^{-1} depending on the seeding date and the environment. It can produce hay with 16.6 % CP when harvested

multiple times during the growing season at the vegetative stage (Lang, 2001). Peterson et al. (2008) reported a range of 12 to 23 % CP, 46 to 54 % NDF, and 45 to 59 % total digestible nutrients (TDN). If Japanese millet is used as a companion crop, it is suggested to lower its seeding rate compared to solo-seeding situations (CRAAQ, 2005). However, little is currently known regarding the use of Japanese millet as a companion crop for the establishment of perennial forages.

2.5.6 Sudangrass

Sudangrass [*Sorghum × drummondii* (Nees ex. Steud.) Millsp. & Chase] is part of the sorghum family. It is a C₄ plant that can grow up to 1.8 m high. It has a high tolerance for dry conditions but does poorly in cooler situations. With climate change, there is a growing interest for sudangrass since it tolerates dry temperatures and low precipitations. It is mainly used as a forage source, either in pasture, or as a source of silage or hay, and is often used as an emergency forage source (Armah-Agyeman et al., 2002). However, sudangrass needs to be harvested with caution. When the plant is exposed to stress such as a frost or even a drought, it can cause an increase in the release of hydrocyanic acid, also called prussic acid. Such increase in prussic acid can be dangerous to animals. Prussic acid dissipates in two weeks and stressed sudangrass is safe to use if not consumed during this period. Hence it would not be recommended to feed sudangrass as a green chop (OMAFRA, 2012).

A study conducted in Southern Québec reported that sudangrass increased forage yields when used as a companion crop for alfalfa during the seeding year compared to a solo-seeded alfalfa control (Matteau et al., 2020). In some environments, it even resulted

in greater forage yields than the use of an oat companion crop. It had consistent forage yields throughout the season while oat was mostly only present at the first harvest and not at subsequent ones. It did not affect the alfalfa establishment in post-seeding years, however, it offered less weed control than oat, but still reduced the amounts of weeds in some environments compared to the solo-seeded alfalfa control. On the contrary, a study conducted in Nebraska reported that, while a sorghum-sudangrass hybrid did in fact improve forage yields in the seeding year and did offer some weed control, alfalfa yields in the post-seeding year were significantly reduced. They concluded that warm-season grasses may not be suited as a companion crop for the establishment of perennial forage legumes (La Vallie, 2020). Zhang et al. (2017) evaluated four different warm-season annual grasses including sudangrass and Japanese millet as solo-seeded emergency forage sources. Overall, sudangrass produced more biomass and better-quality forage than Japanese millet, especially if its seeding was delayed later in the summer. Japanese millet performed best when seeded earlier. Similarly, Peterson et al. (2008) observed that sudangrass provided more forage biomass and forage quality as an emergency forage source, especially when seeded in early June while Japanese millet performed best in early May plantings.

2.6 Project rationale

In recent years, forage producers in Québec have increasingly faced situations where they lacked forages due to droughts or winterkill associated with climate change. One way to address a forage shortage is to seed an “emergency” annual forage species. However, this approach only resolves a forage shortage problem on the short-term. Therefore, seeding emergency annual forage species as companion crops along with perennial forage species

could resolve the problem of forage shortage in the short term while also establishing new fields successfully to address the situation on the longer term. Other situations could require the seeding of annual forages as companion crops. For example, if a producer has limited amounts of forage stored from the previous year and is facing a possible shortage or if there was winterkill, he/she could decide to seed early in spring. Another option in these situations could be to wait longer and seed in June to use warm-season grasses for example. A producer could also decide to establish a new field in June after taking a harvest from what was remaining of a partially winterkilled field. Lastly, there could be a difficult summer where limited biomass was produced. Thus, one might want to seed annual species late in the summer in order to have sufficient feed for their animals during the winter (Figure 2.1).

Peterson et al. (2008) evaluated in the American Midwest multiple emergency forages crops at three different seeding dates. The performance of each crop varied depending on the seeding date. For example, sudangrass did better when seeded in early June while oat-pea mixtures and Japanese millet had higher forage yields when planted in early May. The proposed study aims at providing data to Québec producers on what and when different annual forage species can be seeded depending on their specific situation.

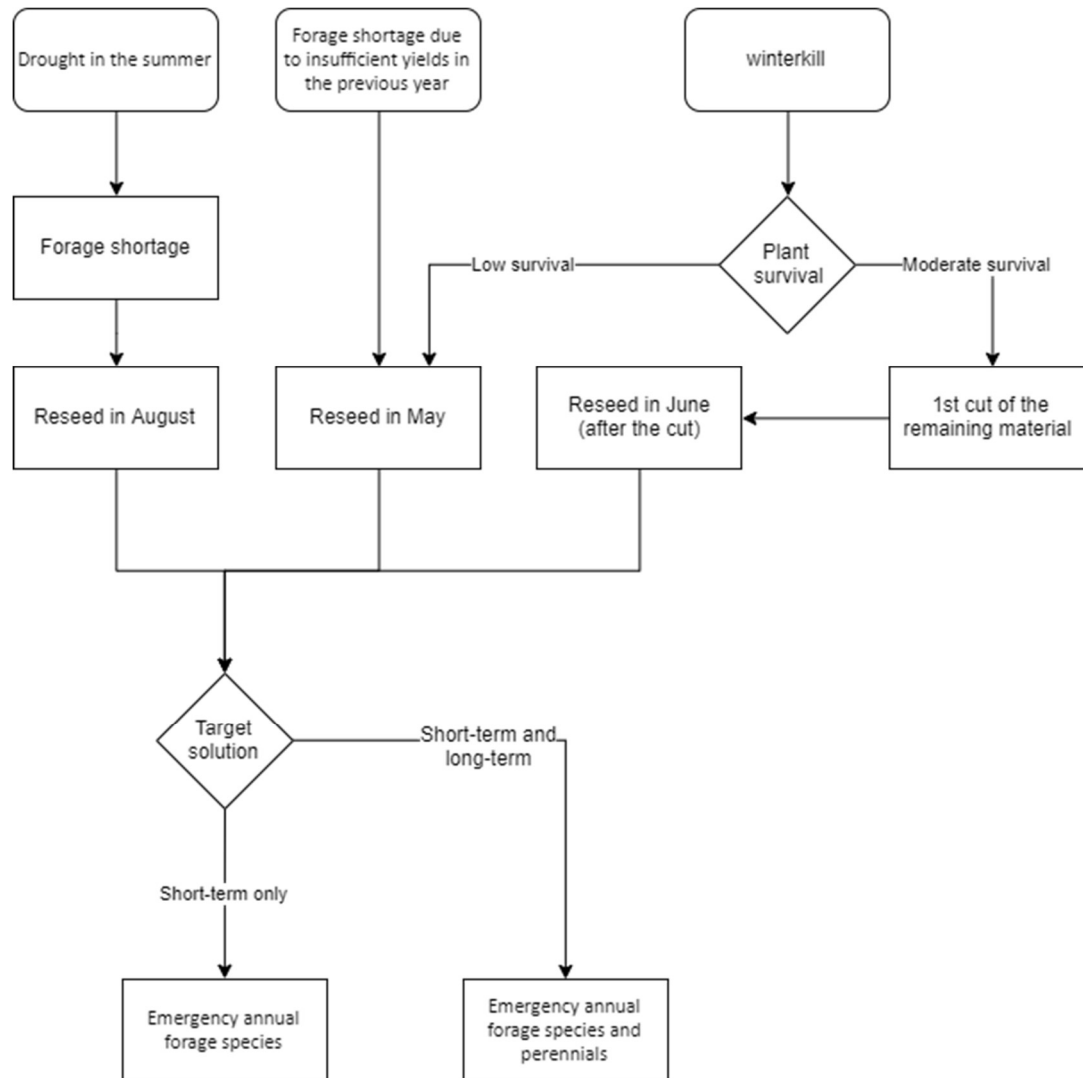


Figure 2.1. Possible forage crops seeding strategies based on different scenarios.

Chapter 3. Methodology

3.1 Sites and treatments description

An experiment was conducted in a total of five environments at three contrasting sites in Québec: Sainte-Anne-de-Bellevue (2136 cumulated growing degree days on a 5°C basis [GDD5]), seeded in 2019 on a Macdonald clay loam (45° 25' 34.0" N lat., 73° 55' 40.0" W long.) and 2020 on a Chateauguay clay loam (45° 25' 38.1" N lat., 73° 55' 47.2" W long.), Saint-Augustin-de-Desmaures (1821 cumulated GDD5), seeded in 2019 (46° 43' 50.0" N lat., 71° 31' 6.4" W long.) and 2020 (46° 43' 48.9" N lat., 71° 31' 8.1" W long.) both on a Saint-Antoine gravelly sandy loam, and La Pocatière (1506 cumulated GDD5), seeded in 2019 on a Kamouraska heavy clay soil (47° 21' 5.5" N lat., 70° 1' 51.5" W long.). Plots in each of these five environments were seeded at three different times during the cropping season, dates varying on the site: at Sainte-Anne-de-Bellevue and Saint-Augustin-de-Desmaures, plots were seeded in mid-May, mid-June and early-August, while they were seeded in early-June, early-July and early-August in La Pocatière. Plots were seeded at each of these times with an alfalfa-timothy mixture along with one of six annual companion crop species and were monitored for two consecutive years (i.e., the seeding and first post-seeding years). A binary mixture of alfalfa (cv. Acapella) and timothy (cv. Express) was seeded at rates of 9 and 7 kg ha⁻¹, respectively (CRAAQ, 2005) along with one of six companion crops: berseem clover (common), forage pea (cv. 4010), annual ryegrass (cv. Melquatro), oat (cv. CDC Haymaker), Japanese millet (common), and sudangrass (cv. Concerto BMR) seeded at rates of 5, 60, 6, 60, 20, and 20 kg ha⁻¹ on a pure live seed basis, respectively (Wiersma et al., 1999; Undersander et al., 2011; Anderson and Volesky., 2014; NSDU, 2018; Bélanger, 2019; Synagri, 2020; Matteau et al., 2020). A control which

consisted in the alfalfa-timothy mixture seeded without a companion crop was also included. Treatments were assigned to a randomized complete block design with split-plot restriction and three replicates (i.e., 3 seeding dates $\times$ 7 companion crop species treatments $\times$ 3 replications). Seeding dates treatments were assigned to main plots and companion crop treatments to sub-plots. Subplots varied in size depending on the site, but they all measured at least 1.3×5 m.

Seeding was done at a targeted depth of 10 mm and rows spaced at 18 cm using a Fabro seven-row seeder (Swift Current, SK, Canada) at Sainte-Anne-de-Bellevue, a Carter five-row seeder (Brookston, IN) at Saint-Augustin-de-Desmaures, and a Kincaid seven-row seeder (Haven, KS) at La Pocatière. Alfalfa and berseem clover seeds were respectively inoculated prior seeding with Nitragin Gold-alfalfa and Nitragin Gold-clover peat-based rhizobial inoculants (Bayer AG, Leverkusen, Germany). Depending on the respective sites and soil analyses, phosphorous, potassium and boron were applied according to local recommendations (CRAAQ, 2013) before seeding and in the spring of the post-seeding year. All sub-plots also received 50 kg N ha^{-1} before seeding (CRAAQ, 2013). Following the first cut in the seeding year, 50 kg N ha^{-1} was applied to the companion crop treatments which consisted of grass species (i.e., sudangrass, ryegrass, oat, and Japanese millet). In Sainte-Anne-de-Bellevue, Matador 120EC (Syngenta Crop Protection Canada, Guelph, ON, Canada), with the active ingredient lambda-cyhalothrin (120 g L^{-1}), was applied twice each year to control severe outbreaks of potato leafhopper (*Empoasca fabae*). Using a field sprayer, the insecticide was applied at the recommended rate (i.e., 83 mL ha^{-1}) on 17 July 2019, 15 August 2019, 2 July 2020, and 31 July 2020. No

herbicide was applied during experimentation in all environments in order to evaluate weed control provided by companion crops.

3.2 Data collection

Depending on the site, the seeding date, and companion crop treatment, plots were harvested one to three times in the seeding year and two to four times in the post-seeding year (Tables S2 to S5). During the seeding year, the first harvest was based on the optimal stage of the companion crops, according to local recommendations or previous research results. Sudangrass was thus harvested in the vegetative stage, once it reached a height of 1.2 m (SARE, 2007), ryegrass, in the vegetative stage, when it reached 25 cm (approximatively 7 weeks after seeding; Wiersma et al., 1999), forage pea when it reached the early pod fill stage (Mustafa and Seguin, 2004; Borreani et al., 2007), oat when it reached the boot stage (CRAAQ, 2005), berseem clover when it reached the full bloom stage (Bélanger, 2019), and Japanese millet, in the vegetative stage, when it reached a height of 50 to 75 cm (CRAAQ, 2005). The alfalfa-timothy control seeded without a companion crop was harvested when alfalfa reached the early flower stage (CRAAQ, 2005). The subsequent harvests in the seeding year and harvests in the post-seeding year were made when alfalfa reached the early flower stage when possible. When there was too much competition offered by the companion crop (i.e., with Sudangrass and Japanese millet) and, therefore alfalfa growth was stunted, harvest was done based on the companion crop stage of development and the number of GDD5 between each cut (at least 500 GDD5).

Harvest was done using an experimental flail mower to cut a surface area of at least 4×0.6 m at a height of 7 cm above ground, with the weight of the harvested material

recorded to determine forage yields. A 500-g sample of fresh material was sampled and dried at 55 °C in a forced-air oven until constant weight to determine forage yields on a dry matter basis. The dried samples from the seeding year were then grinded through a 1 mm-sieve using a Willey mill (Standard model 4, Arthur H. Thomas Co., Philadelphia, PA) and were preserved for laboratory analyses. Using cutters, a 50 × 50 cm area of a distinct permanent quadrat was sampled 7 cm above ground in every plot following each harvest. For all botanical samples, the species were separated by hand and then dried in a forced-air oven at 55 °C to constant weight in order to record weight on a dry matter basis and determine the proportion of each species (i.e., alfalfa, timothy, companion crop, and weeds) in each plot. Immediately prior harvest the phenological stages of the seeded plant species were also recorded (i.e., alfalfa, timothy, and companion crops). At the end of the season, in the fall, the number of alfalfa and timothy stems were counted in the permanent quadrats to assess their establishment. This was done again in the following spring to evaluate the winter survival of the perennial forage species.

3.3 Nutritional analyses

All grounded samples from seeding years were scanned by visible and near infrared reflectance spectroscopy (VNIRS) using a NIRsystem DS 2500 monochromator (Foss, Silver spring, MD) to select samples to be incorporated in calibration and validation sets. Nutritive values for all samples were then predicted by using forage reflectance spectrum and correlating them against nutritive values determined by wet chemistry. Laboratory analyses were performed on the calibration ($n = 75$) and validation ($n = 19$) sets of samples selected by the WinISI IV (version 4.0) software (Infrasoft International, LLC, Silver Spring, MD) from each seeding year to determine the following nutritive factors: neutral

detergent fiber assayed with a heat-stable α -amylase (aNDF), acid detergent fiber (ADF) and total nitrogen (TN) to estimate the crude protein (CP). The aNDF concentration was determined following the method of Mertens (2002) and using heat stable α -amylase and sodium sulfite. The ADF concentration was determined following the method of the Association of Official Analytical Chemists (AOAC, 1990). For both aNDF and ADF, the ANKOM filter bag technique was used (ANKOM Technology, Macedon, NY). For CP, it was estimated from TN using the equation $CP = TN \times 6.25$. Total N was determined according to the procedures used in Simili da Silva et al. (2014). First, the samples were mineralized in a mixture of sulfuric and selenious acids, then, N concentration was determined with an autoanalyzer (QuikChem 8000 Lachat Zellweger Analytics Inc., Lachat Instruments, Milwaukee, WI) using the method 13–107–06–2-E (Lachat Instruments, 2011). Upon correlating the calibration and validation sets to the VNIRIS prediction equation, the VNIRS predictions were considered excellent when the ration of prediction to deviation [$RPD = SD/SEP(C)$], where SD corresponds to the standard deviation of the reference data used in the validation set and [$SEP(C)$], to the standard error of prediction corrected for bias, was greater than 4 and successful when RPD was between 3 and 4 (Nie et al., 2009). All three attributes were deemed excellent or successful since their RPD were all higher than 5 or 3. From the nutritive values determined for each harvest, final data presented were annual weighted averages based on the individual contribution of each harvest to total annual forage yields.

3.4 Statistical analyses

Climatic conditions varied considerably among sites and years, in addition, the number of harvests during the seeding year varied depending on the site, thus data were analysed

separately for each environment. A two-way analysis of variance (ANOVA) was done using PROC GLM of the SAS software (SAS Institute, 2014). Replicates for each site were considered a random effect, seeding dates and companion crops effects were considered fixed effects. Since the seeding date $\times$ companion crop interactions were often significant, data was further analysed separately for each seeding date. Using the least square means difference (LSD) and Scheffé's adjustment for multiple comparisons, differences between means were determined and considered significant at the $P \leq 0.05$ level; only significant differences and effects are discussed herein.

Chapter 4. Results

4.1 Forage yields during the seeding year

Seeding date and companion crop treatment effects were significant at each site ($P < 0.001$), as well as a seeding date $\times$ companion crop interaction ($P \leq 0.05$) illustrating that companion crop performance differed depending on the seeding date (Tables 4.1 to 4.3). Overall, at Sainte-Anne-de-Bellevue and Saint-Augustin-de-Desmaures, annual total forage yields were similar for the first two seeding dates (i.e., mid-May and mid-June), which were significantly higher than the forage yields observed for the August seeding. At La Pocatière, the annual total forage yield for the first seeding date (i.e., early-June) was significantly higher than for the second seeding (i.e., late-June), which was also significantly higher than for the third seeding (i.e., early-August).

4.1.1 Sainte-Anne-de-Bellevue

During the 2019 seeding year, for the first two seeding dates (i.e., mid-May and mid-June), the use of sudangrass produced total forage yields which were 70 % higher than the control

seeded without a companion crop (7.10 vs. 4.18 Mg ha⁻¹ yr⁻¹ on average, respectively), while the use of oat and Japanese millet produced 34 % more than the control (5.60 Mg ha⁻¹ yr⁻¹ on average; Table 4.1). These companion crop treatments also reduced the total weed biomass and alfalfa yields by 67 % compared to the control on average. Timothy yields were minimal and no differences among treatments were observed. The use of peas produced similar total forage and alfalfa yields to the control while reducing weed biomass. For the third seeding date (i.e., August), all treatments produced higher yields than the control, except ryegrass, with the oat and Japanese millet treatments producing the highest total forage yields (average of 1.62 Mg ha⁻¹ yr⁻¹).

During the 2020 seeding year, similar trends were observed. For the first seeding date (i.e., mid-May), the use of sudangrass increased total forage yields by 93 %, followed by Japanese millet and oat treatments which increased forage yields by 46 %, on average, when compared to the control seeded without a companion crop (7.30, 5.87 and 5.18 vs 3.78 Mg ha⁻¹ yr⁻¹, respectively). However, half of the yields obtained with the oat treatment consisted of weeds. Alfalfa and timothy annual forage yields were reduced (67 and 61 %, respectively) with the use of sudangrass, oat and Japanese millet, while weed biomass was reduced with sudangrass and Japanese millet by 68 % on average compared to the control. For the second seeding date (i.e., mid-June), the use of sudangrass increased total forage yields the most, by 225 %, followed by Japanese millet which increased total forage yields by 136 % compared to the control (8.89 and 6.47 vs 2.74 Mg ha⁻¹ yr⁻¹, respectively). The use of these companion crops also reduced timothy annual yields and weed biomass by an average of 84 and 75 %, respectively. Alfalfa annual yields were minimal (i.e., < 340 kg ha⁻¹ yr⁻¹), and no differences were observed among treatments. Contrary to the 2019

seeding year, oat did not increase total forage yields for the second seeding date. This could be due to the severe drought which followed seeding (Figure S1) as oat is not particularly well adapted to drought conditions. For the first two seeding dates, the use of peas resulted in similar total forage yields, alfalfa and timothy yields than the control and higher weed biomass. For the third seeding date in 2020 (i.e., early August), similarly to 2019, the use of oat and Japanese millet resulted in the highest total forage yields compared to all other treatments (average of 4.58 Mg ha⁻¹ yr⁻¹). Peas and sudangrass also increased total forage yields (average of 2.59 Mg ha⁻¹ yr⁻¹) when compared to the control, berseem clover and ryegrass (0.76, 1.26 and 1.14 Mg ha⁻¹ yr⁻¹, respectively). The use of oat and Japanese millet also reduced timothy yields by 99 % and weed biomass by 95 % on average, when compared to the control. No differences in alfalfa yields were observed among treatments since they were minimal (i.e., < 160 kg ha⁻¹ yr⁻¹). For both seeding years, and all seeding dates, the use of berseem clover and annual ryegrass resulted in total forage yields which were comparable to the control in the majority of cases.

4.1.2 Saint-Augustin-de-Desmaures

During the 2019 seeding year, and for the first two seeding dates (i.e., late-May and mid-June), the use of oat increased total forage yields the most, by 60 % (6.48 Mg ha⁻¹ yr⁻¹), followed by Japanese millet and sudangrass, which increased total forage yields by 38 % on average compared to the control seeded without a companion crop (5.60 vs 4.06 Mg ha⁻¹ yr⁻¹, respectively; Table 4.2). The use of peas increased total forage yields by 27 % (4.86 Mg ha⁻¹ yr⁻¹) while berseem clover and annual ryegrass had higher yields (average of 20 %; 4.58 Mg ha⁻¹ yr⁻¹) compared to the control. No differences between treatments in alfalfa yields, timothy yields and weed biomass were observed for the first seeding date.

However, for the second seeding date, 40 % of the total forage yields obtained with the oat treatment consisted of weeds. Thus, the use of an oat companion crop did not reduce weeds at that seeding date, which were, in contrast, reduced with the use of sudangrass and Japanese millet by an average of 73 % compared to the control. The use of Japanese millet and sudangrass, however, also reduced alfalfa and timothy annual yields by 80 and 68 % on average compared to the control. The use of peas produced similar total forage, alfalfa and timothy yields, and lower weed biomass compared to the control. The use of annual ryegrass and berseem clover produced higher total forage yields, similar alfalfa and timothy yields and weed biomass. For the third seeding date (i.e., early August), no differences among treatments were observed for total forage and timothy yields. However, for the oat and Japanese millet treatments, on average, the companion crop contributed to an average of 76 % of the total forage yields compared to only 14 % on average for the other companion crop treatments. Alfalfa yields and weeds biomass were respectively reduced with the use of oat and Japanese millet, by 78 and 59 % on average, compared to the control while they were similar for the other companion crop treatments compared to the control.

During the 2020 seeding year, for the first seeding date (i.e., mid-May), no differences were observed among treatments for total forage, alfalfa, and timothy yields as well as for weed biomass. However, with oat, Japanese millet and sudangrass treatments the companion crop contributed to 45 % of the total forage yields, on average, compared to 4 % for the other companion crops species, the vast majority of the yield consisting of weeds. Such low contribution to total forage yields could be due to the heavy drought that followed seeding (Figure S1). For the second seeding date (i.e., mid-June), the use of Japanese millet increased total forage yields by 126 %, while sudangrass increased total

forage yields by 70 % compared to the control seeded without a companion crop (5.53 and 4.16 vs 2.44 Mg ha⁻¹ yr⁻¹, respectively). In the case of the oat treatment, 70 % of the total yields consisted of weeds. The drought conditions that prevailed possibly affected oat growth which was outcompeted by weeds (Figure S1). Sudangrass and Japanese millet treatments reduced weed biomass the most (average of 70 %) compared to the control. No differences among treatments for alfalfa and timothy yields were observed since they were in all cases minimal (i.e., < 350 and 80 kg ha⁻¹ yr⁻¹, respectively). The use of peas produced similar total forage yields and lower weed biomass compared to the control while the use of annual ryegrass and berseem clover produced similar total forage yields and weed biomass compared to the control. For the third seeding date (i.e., early August), no differences among treatments were observed. However, similarly to the 2019 seeding, the use of oat and Japanese millet tended to produce more total forage yields than the other treatments. The companion crops themselves contributed to 91 % of the total forage yields with oat, 81 % with Japanese millet, and 77 % with peas while the other treatments contributed to 30 % of the total forage yields on average.

4.1.3 La Pocatière

During the 2019 seeding year at La Pocatière, for the first two seeding dates (i.e., early-June and late-June), the use of sudangrass, oat and Japanese millet increased total forage yields by 93 % on average and resulted in the highest total forage yields when compared to the control seeded without a companion crop (average of 5.50 vs 2.85 Mg ha⁻¹ yr⁻¹; Table 4.3). For the first seeding date, compared to the control, oat reduced alfalfa yields the most (by 78 %), followed by sudangrass, Japanese millet and peas (average of 62 %). The use of peas, berseem clover and annual ryegrass produced similar total forage and alfalfa yields

compared to the control. There was no timothy present and no differences among treatments for weed biomass were observed. For the second seeding date, oat, Japanese millet and sudangrass reduced weed biomass by 91 % on average compared to the control. The use of peas produced intermediate yields (i.e., higher than the control and lower than the highest yielding companion crops) and reduced weed biomass, while berseem clover and annual ryegrass produced similar total forage yields and weed biomass compared to the control. No differences among treatments were observed for alfalfa and timothy yields, which remained minimal. For the third seeding date (i.e., early-August), only oat and peas treatments grew due to the weather conditions that preceded and followed seeding; there were no precipitations for two weeks prior to seeding and only a very strong precipitation a week after seeding as well as low temperatures ($< 20^{\circ}\text{C}$; Figure S1). Total forage yields were greater with oat compared to peas (3.14 vs $0.43 \text{ Mg ha}^{-1} \text{ yr}^{-1}$, respectively), most of the yield consisting of the companion crop itself. No other differences were observed among these two treatments.

4.2 Forage yields in the first post-seeding year

Seeding date, except at Saint-Augustin-de-Desmaures, and companion crop main effects were significant at each site ($P < 0.001$), with a seeding date $\times$ companion crop interaction being observed at Saint-Augustin-de-Desmaures and La Pocatière, illustrating that perennial forages performance in the post-seeding year differed depending on the seeding date and the companion crop used in the previous year ($P < 0.001$; Tables 4.5 and 4.6). There was, however, no seeding date $\times$ companion crop interaction at Sainte-Anne-de-Bellevue (Table 4.4). Despite a lack of interaction at this site, the results are presented according to each seeding date in order to preserve the uniformity of the data presented.

Overall, at Sainte-Anne-de-Bellevue, total forage yields for the first seeding date used in the previous year (i.e., mid-May) were higher than the one for the second seeding (i.e., mid-June) which was also higher than the one for the third seeding date (i.e., early-August). At La Pocatière, the total forage yields for the first seeding date (i.e., early-June) were higher than the second and third seeding dates (i.e., late-June and early-August), which did not regrow sufficiently to harvest in the post-seeding year.

4.2.1 Sainte-Anne-de-Bellevue

During the 2020 post-seeding year, for the plots seeded in 2019 at the first seeding date (i.e., mid-May), no differences among treatments for the total forage yields and weed biomass were observed. The use of sudangrass as a companion crop in the previous year, however, reduced alfalfa yields by 40 % compared to the control originally seeded without an annual companion crop (5.74 vs 9.49 Mg ha⁻¹ yr⁻¹), followed by oat, Japanese millet and peas, the use of which in the previous year reduced alfalfa post-seeding year yields by 24 % on average (average of 7.18 Mg ha⁻¹ yr⁻¹; Table 4.4). In contrast, timothy annual yields were increased with the use of sudangrass, oat and peas in the previous year by 67 % on average, followed by Japanese millet which increased timothy yields by 29 % compared to the control. This reflects that a reduction in alfalfa yield caused by the use of a companion crop in the previous year was compensated by greater timothy production, which was also observed in the stem density of both species in the spring (Table S1). Ryegrass was the only companion crop species that survived the winter and contributed to 7 % of the total forage yields. For the second original seeding date (i.e., mid-June), the use of a sudangrass companion crop in the previous year resulted in a 42 % reduction in total forage yields compared to the control, followed by oat and Japanese millet treatments which reduced

total forage yields by 27 % on average. Alfalfa yield and alfalfa stem density were also reduced with these three companion crop treatments (by 50 % with sudangrass, 36 % with oat and 28 % with Japanese millet on average; Tables 4.4 and S1). Weed biomass was highest with sudangrass compared to all other treatments (388 % higher than the control). Again, ryegrass survived the winter and contributed to 11 % of the total forage yields ($0.84 \text{ Mg ha}^{-1} \text{ yr}^{-1}$). No differences among treatments were observed for timothy yields. For the first two original seeding dates, the use of peas, berseem clover and ryegrass did not have residual effects on forage yields in the post-seeding year. For the third original seeding date (i.e., early-August), no differences were observed among treatments for total forage yields, timothy yields and weed biomass. However, the use of oat and Japanese millet at seeding reduced post-seeding year alfalfa yields by 56 % on average compared to the control (1.18 vs $2.68 \text{ Mg ha}^{-1} \text{ yr}^{-1}$). This was reflected in a reduction in alfalfa stem density (average of 78 % for oat and Japanese millet treatments; Table S1). Ryegrass survived the winter and contributed to 27 % of the total forage yields for the annual ryegrass treatment ($1.15 \text{ Mg ha}^{-1} \text{ yr}^{-1}$). The use of sudangrass, peas and ryegrass at seeding also reduced post-seeding year alfalfa yields compared to the control while the use of berseem clover had no residual effect.

4.2.2 Saint-Augustin-de-Desmaures

During the 2020 post-seeding year, for the first two original seeding dates (i.e., mid-May and mid-June), the use of sudangrass and Japanese millet in the preceding year reduced total post-seeding year yields by 22 % on average ($4.89 \text{ Mg ha}^{-1} \text{ yr}^{-1}$), followed by oat to a lesser extent since it reduced total forage yields by 13 % compared to the control originally seeded without a companion crop (5.49 vs $6.30 \text{ Mg ha}^{-1} \text{ yr}^{-1}$; Table 4.5). Ryegrass survived

and contributed minimally (0.63 % on average) to the total forage yields. No differences among treatments were observed for weed biomass. For the first seeding date, Japanese millet and sudangrass had residual effects on alfalfa yields in the post-seeding year, which were reduced by 26 % on average ($4.07 \text{ Mg ha}^{-1} \text{ yr}^{-1}$), while the use of oat reduced alfalfa yields by 14 % compared to the control (4.75 vs $5.53 \text{ Mg ha}^{-1} \text{ yr}^{-1}$). Timothy yields were higher with the use of Japanese millet and sudangrass in the previous year compared to the control (average of 76 %). A reduction in alfalfa yield was compensated to some degree by higher timothy yield. For the second seeding date, sudangrass and Japanese millet treatments respectively reduced alfalfa post-seeding year yields by 45 % on average ($3.17 \text{ Mg ha}^{-1} \text{ yr}^{-1}$), while oat treatment reduced alfalfa yields by 24 % compared to the control (4.35 vs $5.70 \text{ Mg ha}^{-1} \text{ yr}^{-1}$). No differences among treatments were observed for timothy yields. For the third initial seeding date (i.e., early-August), annual ryegrass survived and helped improve yields by 7 % compared to the other treatments. No other differences were observed among treatments for total forage, alfalfa and timothy yields, and weed biomass. However, total post-seeding forage yields tended to be lower with the use of oat and Japanese millet treatments in the preceding year, compared to the control (average of 25 %). For the third seeding date, the number of timothy stems was reduced by 57 % on average with oat, Japanese millet, and annual ryegrass treatments (Table S1). For all seeding dates, the use of peas, berseem clover and ryegrass at seeding did not have residual effects on the post-seeding year total forage yields, alfalfa and timothy yields compared to the control.

4.2.3 La Pocatière

During the 2020 post-seeding year, regrowth from plots originally seeded at the second (i.e., late-June) and third seeding date (i.e., early-August) was minimal, not being sufficient to harvest. For the third seeding date, only oat and peas treatments established successfully in 2019. This poor establishment in the August seeding was reflected in poor stem densities of alfalfa and timothy observed in the spring of the post-seeding year (i.e., 16 to 40 alfalfa stems per m² and 0 to 63 timothy stems per m²; Table S1). For the second seeding date, the number of stems of perennial forages was minimal in the fall of 2019 for all treatments and was even slightly lower in the spring of 2020 (i.e., 3 to 31 alfalfa stems per m² and 0 to 152 timothy stems per m² in the spring; Table S1 and Figure S8). Excessive competition from the companion crops in very dry conditions could be the cause of the poor establishment of perennial species which was reflected in poor post-seeding year forage growth. The presence of darksided cutworms (*Euxoa messoria*) was also observed in the spring, which could partly explain the lower number of stems observed in the spring 2020 compared to the fall 2019 (Figure S8). For the first original seeding date (i.e., early-June), total post-seeding year forage yields were reduced by 52 % (1.61 Mg ha⁻¹ yr⁻¹) with the use of sudangrass in the previous year, and by 28 % on average with the use of Japanese millet and oat compared to the control originally seeded without a companion crop (average of 2.42 vs 3.36 Mg ha⁻¹ yr⁻¹; Table 4.6). The use of peas, berseem clover and ryegrass at seeding did not have residual effects on total post-seeding year forage yields compared to the control. Missing data on the botanical composition during the second harvest rendered the calculation of annual yields for each component impossible. However, at the first harvest, no differences among treatments for alfalfa yields, timothy yields and weed

biomass were observed (Figure S6). Sudangrass significantly reduced total forage yields at the first harvest compared to all other treatments, followed by Japanese millet and oat.

4.3 Forage nutritive value in the seeding year

Seeding date and companion crop treatment effects were significant at each site ($P < 0.05$) and there was a seeding date $\times$ companion crop interaction observed at Saint-Augustin-de-Desmaures (2019 and 2020) and at La Pocatière for aNDF, ADF and CP concentrations, and at Sainte-Anne-de-Bellevue for aNDF and ADF (2020), and CP concentrations (2019 and 2020; $P < 0.01$). However, there were no significant seeding date effects at Sainte-Anne-de-Bellevue in 2020 and Saint-Augustin-de-Desmaures in 2019 (Tables 4.7 to 4.9). Overall, at Sainte-Anne-de-Bellevue, for the 2019 seeding, aNDF and ADF concentrations for the first seeding date (i.e., mid-May) were similar to the second seeding (i.e., mid-June) and were higher than the ones for the third seeding date (i.e., early-August) while a reverse trend was observed, in both 2019 and 2020, for CP concentration. At Saint-Augustin-de-Desmaures, ADF, and aNDF concentrations, in 2020, were similar for the first two seeding dates (i.e., mid-May and mid-June) and higher than the third seeding date (i.e., early-August) while the trend was opposite with CP concentration in both years. At La Pocatière, aNDF and ADF concentrations were higher with the second seeding date (i.e., late-June) compared to the first and third seeding dates (i.e., early June and August) which were similar, while the trend was the opposite with CP concentration.

4.3.1 Sainte-Anne-de-Bellevue

During the 2019 seeding year and for the first two seeding dates (i.e., mid-May and mid-June), the use of Japanese millet and sudangrass companion crop resulted in, on average,

higher average annual aNDF and ADF concentrations (33 and 16 %) compared to the control seeded without a companion crop. The use of these two companion crops for the establishment of the perennial species also resulted in a 35 % lower CP concentration, on average, compared to the control (Table 4.7). The use of oat resulted on average in higher aNDF and ADF concentrations, by 23 and 12 %, and lower CP concentration by 22 % compared to the control. The use of these three species thus produced a forage of lower nutritive value. For the third seeding date (i.e., August), the use of sudangrass, Japanese millet and oat produced a forage with, on average, a 64 % higher aNDF concentration, compared to the control. No differences were observed among treatments for ADF and CP concentrations.

During the 2020 seeding year and for the first seeding date (i.e., mid-May), the use of sudangrass resulted in aNDF and ADF concentrations that were 29 and 16 % higher, respectively, and CP concentration 21 % lower compared to the control. The use of oat and Japanese millet also resulted, on average, in 14 and 10 % higher aNDF and ADF concentrations and 9 % lower CP concentration compared to the control. For the second seeding date (i.e., mid-June), the use of sudangrass and Japanese millet on average resulted in 36 and 16 % higher aNDF and ADF concentrations compared to the control. The use of sudangrass also resulted in 38 % lower CP concentration, while the use of Japanese millet resulted in a 19 % reduction. Finally, for the third seeding date (i.e., early-August), sudangrass, Japanese millet and oat treatments produced a forage with higher aNDF and ADF concentrations, (53 and 37 % on average) and lower CP concentrations (45 % on average) compared to the control. For all seeding dates and years, except August 2020, the

nutritive values associated with the use of peas, berseem clover and annual ryegrass did not differ from the control.

4.3.2 Saint-Augustin-de-Desmaures

During the 2019 seeding year and for the first seeding date (i.e., mid-May), the use of oat and Japanese millet resulted, on average, in 40 and 17 % higher aNDF and ADF concentrations compared to the control seeded without a companion crop. The use of sudangrass resulted in 29 and 13 % higher aNDF and ADF concentrations, respectively, compared to the control (Table 4.8). The use of oat and Japanese millet also resulted in 21 % lower CP concentration on average compared to the control. For the second seeding date (i.e., mid-June), the use of sudangrass and Japanese millet resulted, on average, in 38 and 15 % higher aNDF and ADF concentrations compared to the control, while the use of oat resulted in 20 and 6 % higher aNDF and ADF concentrations. The use of sudangrass, Japanese millet and oat as companion crops also resulted in 16 % lower CP concentration on average compared to control. For the third seeding date (i.e., August), the use of oat and Japanese millet resulted, on average, in 49 and 20 % higher aNDF and ADF concentrations compared to the control. The use of oat and Japanese millet treatments also resulted in 21 and 11 % lower CP concentrations respectively compared to the control.

During the 2020 seeding year and for the first seeding date (i.e., mid-May), no differences among treatments were observed for the nutritional values due to the high proportion of weeds in each treatment. For the second seeding date (i.e., mid-June), the use of Japanese millet and sudangrass respectively resulted in 41 and 31 % higher aNDF concentrations, and 20 and 10 % lower CP concentrations compared to the control. The use

of Japanese millet and sudangrass also resulted in 20 % higher ADF concentration on average compared to the control. For the third seeding date (i.e., early-August), the use of oat and Japanese millet resulted, on average, in 83 and 49 % higher aNDF and ADF concentrations compared to the control. They also resulted in 32 and 23 % lower CP concentrations respectively when compared to the control. For all seeding dates and years, the nutritive values associated with the use of peas, berseem clover and annual ryegrass were similar to the control.

4.3.3 La Pocatière

During the 2019 seeding year and for the first seeding date (i.e., early-June), the use of sudangrass resulted in higher aNDF and ADF concentrations (75 and 32 %, respectively) and lower CP concentration (44 %) compared to the control seeded without a companion crop (Table 4.9). The use of oat and Japanese millet treatments resulted, on average, in 57 and 32 % higher aNDF and ADF concentrations, and 32 % lower CP concentration compared to the control. The use of peas, berseem clover and annual ryegrass resulted in comparable aNDF and ADF, and lower CP concentrations as the control in the majority of cases. For the second seeding date (i.e., late-June), the use of berseem clover resulted in the highest aNDF and ADF concentrations, compared to all other treatments, followed by the use of annual ryegrass which resulted in higher aNDF and ADF concentrations (average of 15 and 9 %, respectively) compared to the control. The use of berseem clover and annual ryegrass also resulted in 19 % lower CP concentrations on average compared to the control. The lower nutritive value of the berseem clover and annual ryegrass treatments could be due to the high proportion of weeds in these treatments. The use of peas resulted in the lowest aNDF and highest CP concentrations compared to all the other treatments. For the

third seeding date (i.e., early-August), the use of oat resulted in 32 and 7 % higher aNDF and ADF concentrations and 41 % lower CP concentration on average when compared to the use of peas as a companion crop.

4.4 Tables

Table 4.1. Seeding year forage dry matter yield at Sainte-Anne-de-Bellevue, QC, Canada of an alfalfa-timothy mixture seeded in two different years and fields at three different dates with six different companion crops.

	2019 seeding					2020 seeding				
	Total	CC†	Alfalfa	Timothy	Weeds	Total	CC	Alfalfa	Timothy	Weeds
CC at the establishment	-----Mg DM ha ⁻¹ yr ⁻¹ -----									
Seeding date 1	21 May 2019					19 May 2020				
Control (none)	4.62c‡	.	1.91a	0.26	2.45a	3.78d	.	0.61a	0.02bc	3.15bc
Berseem clover	3.79d	0.44c	1.63ab	0.23	1.49b	4.60cd	0.05e	0.71a	0.03ab	3.81ab
Peas	4.41cd	1.84b	1.22bc	0.20	1.15bc	4.76c	1.03d	0.62a	0.04a	3.07c
Annual ryegrass	4.82c	0.20c	2.18a	0.23	2.22a	4.72c	0.05e	0.60a	0.02bc	4.04a
Oat	5.91b	4.48a	0.83c	0.12	0.48d	5.18bc	2.17c	0.25b	0.01c	2.75c
Japanese millet	6.09b	4.39a	0.75c	0.09	0.85cd	5.87ab	3.76b	0.25b	0.01c	1.86d
Sudangrass	6.82a	4.74a	0.91c	0.16	1.01bcd	7.30a	5.83a	0.10b	0.01c	1.37d
SEM	0.23	0.25	0.20	0.04	0.21	0.29	0.29	0.01	0.01	0.23
Seeding date 2	13 June 2019					15 June 2020				
Control (none)	3.75cde	.	1.56a	0.01	2.19b	2.74c	.	0.25	0.01a	2.48a
Berseem clover	3.34e	0.22c	1.51a	0.01	1.60bc	2.34c	0.01c	0.28	0.01ab	2.04ab
Peas	3.44de	1.33c	1.00ab	0.02	1.09cd	2.96c	0.28c	0.34	0.01a	2.33a
Annual ryegrass	4.59bcd	0.11c	1.34a	0.01	3.13a	2.40c	0.04c	0.25	0.01a	2.09ab
Oat	4.92bc	3.91b	0.42bc	0.01	0.57d	2.68c	0.99c	0.29	0.00bc	1.39bc
Japanese millet	5.47b	4.54b	0.24c	0.00	0.69d	6.47b	5.89b	0.05	0.00c	0.53d
Sudangrass	7.37a	6.15a	0.27c	0.00	0.96cd	8.89a	8.08a	0.09	0.00bc	0.71cd
SEM	0.39	0.50	0.20	0.01	0.29	0.61	0.52	0.06	0.01	0.24
Seeding date 3	13 August 2019					31 July 2020				
Control (none)	0.05d	.	0.02	0.02	0.01	0.76c	.	0.16	0.18a	0.42ab
Berseem clover	0.44d	0.40d	0.02	0.00	0.01	1.26c	0.27c	0.12	0.14ab	0.72ab
Peas	0.93bc	0.89d	0.02	0.01	0.01	2.59b	1.95b	0.13	0.13ab	0.38bc
Annual ryegrass	0.08d	0.04bc	0.01	0.01	0.01	1.14c	0.54c	0.13	0.10abc	0.36bc
Oat	1.66a	1.64d	0.00	0.00	0.01	4.68a	4.65a	0.01	0.00c	0.02d
Japanese millet	1.58ab	1.57ab	0.00	0.00	0.00	4.49a	4.45a	0.01	0.00c	0.03d
Sudangrass	0.30cd	0.27cd	0.01	0.01	0.01	2.58b	2.41b	0.03	0.03bc	0.10cd
SEM	0.22	0.23	0.01	0.01	0.01	0.26	0.20	0.04	0.04	0.09
ANOVA (source of variation)	<i>P</i> -value									
SD	***	**	***	***	***	***	ns§	***	**	***
Companion	***	***	***	*	***	***	***	***	***	***
SD × Companion	***	***	***	*	***	***	***	*	*	***

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P > 0.05$).

Table 4.2. Seeding year forage dry matter yield at Saint-Augustin-de-Desmaures, QC, Canada of an alfalfa-timothy mixture seeded in two different years and fields at three different dates with six different companion crops.

	2019 seeding					2020 seeding				
	Total	CC†	Alfalfa	Timothy	Weeds	Total	CC	Alfalfa	Timothy	Weeds
CC at the establishment	-----Mg DM ha ⁻¹ yr ⁻¹ -----									
Seeding date 1	23 May 2019					15 May 2020				
Control (none)	3.82d	.	1.81	0.33	1.68	3.14	.	0.74	0.06	2.34
‡										
Berseem clover	4.57c	0.89c	1.98	0.26	1.43	2.90	0.04b	0.43	0.05	2.38
Peas	4.86c	2.54b	0.93	0.41	0.98	3.59	0.32b	0.38	0.06	2.82
Annual ryegrass	4.60c	0.27c	1.97	0.32	2.05	3.79	0.02b	0.46	0.03	3.28
Oat	6.61a	4.62a	0.87	0.29	0.83	4.18	1.78a	0.36	0.02	2.02
Japanese millet	6.11a	2.58b	1.77	0.18	1.59	4.02	1.68a	0.19	0.01	2.14
Sudangrass	5.83b	3.12b	1.46	0.26	0.98	3.90	2.01a	0.23	0.01	1.65
SEM	0.24	0.32	0.31	0.10	0.50	0.38	0.35	0.22	0.02	0.42
Seeding date 2	19 June 2019					15 June 2020				
Control (none)	4.29c	.	1.54a	0.12bc	2.64a	2.44de	.	0.20	0.00	2.23a
Berseem clover	4.83b	0.48c	1.31a	0.09cd	2.96a	2.80cde	0.19d	0.35	0.00	2.26a
Peas	4.49c	1.82b	1.14ab	0.13abc	1.40b	2.34e	0.39cd	0.30	0.00	1.65b
Annual ryegrass	5.14b	0.10c	1.63a	0.20a	3.20a	2.96cde	0.04d	0.33	0.00	2.59a
Oat	6.35a	2.48b	1.11ab	0.18ab	2.57a	3.14cde	0.76c	0.18	0.00	2.19a
Japanese millet	5.30b	4.26a	0.20c	0.04d	0.80b	5.53a	4.83a	0.17	0.00	0.53c
Sudangrass	5.16b	4.06a	0.42bc	0.04d	0.63b	4.16b	3.12b	0.14	0.08	0.83c
SEM	0.20	0.25	0.25	0.03	0.32	0.18	0.14	0.06	0.03	0.17
Seeding date 3	1 August 2019					8 August 2020				
Control (none)	1.96	.	0.55a	0.08	1.33a	0.18	.	0.12	0.02	0.04
Berseem clover	2.05	0.16b	0.47a	0.05	1.37a	0.49	0.15	0.23	0.04	0.07
Peas	2.18	0.60b	0.37ab	0.06	1.15ab	0.95	0.74	0.13	0.04	0.04
Annual ryegrass	2.19	0.21b	0.46a	0.05	1.48a	0.27	0.09	0.10	0.03	0.05
Oat	2.46	1.95a	0.10b	0.00	0.41c	1.67	1.52	0.08	0.02	0.05
Japanese millet	3.06	2.23a	0.14b	0.00	0.69bc	1.11	0.89	0.14	0.04	0.03
Sudangrass	2.38	0.44b	0.34ab	0.04	1.56a	0.44	0.11	0.23	0.04	0.05
SEM	0.39	0.32	0.09	0.02	0.19	0.06	0.06	0.04	0.01	0.02
ANOVA (source of variation)	<i>P</i> -value									
SD	**	**	*	**	*	***	**	ns§	ns	***
Companion	***	***	***	ns	***	***	***	ns	ns	***
SD × Companion	*	***	*	ns	***	***	***	ns	ns	**

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P > 0.05$).

Table 4.3. Seeding year forage dry matter yield at La Pocatière, QC, Canada of an alfalfa-timothy mixture seeded at three different dates with six different companion crops.

CC at the establishment	2019 seeding				
	Total	CC†	Alfalfa	Timothy	Weeds
	-----Mg DM ha ⁻¹ yr ⁻¹ -----				
Seeding date 1	10 June 2019				
Control (none)	4.14c‡	.	3.68a	0.00	0.46
Berseem clover	3.71cd	0.92cd	2.37ab	0.00	0.42
Peas	3.99c	2.27bc	1.51bc	0.00	0.22
Annual ryegrass	3.02d	0.01d	2.56ab	0.00	0.46
Oat	6.29a	5.33a	0.81c	0.00	0.15
Japanese millet	5.22b	3.56ab	1.45bc	0.00	0.22
Sudangrass	6.65a	4.47a	1.88bc	0.00	0.30
SEM	0.31	0.61	0.49	0.00	0.15
Seeding date 2	28 June 2019				
Control (none)	1.56c	.	0.36	0.09	1.11ab
Berseem clover	1.80c	0.00c	0.17	0.13	1.50a
Peas	3.35b	2.90b	0.03	0.00	0.42bc
Annual ryegrass	1.57c	0.16c	0.51	0.03	0.87abc
Oat	4.91a	4.74a	0.05	0.03	0.10c
Japanese millet	4.91a	4.83a	0.02	0.00	0.06c
Sudangrass	5.03a	4.86a	0.02	0.00	0.16bc
SEM	0.34	0.11	0.14	0.04	0.32
Seeding date 3	2 August 2019				
Control (none)	.	.	.	.	.
Berseem clover	.	.	.	.	.
Peas	0.43b	0.40b	0.01	0.02	0.00
Annual ryegrass	.	.	.	.	.
Oat	3.14a	2.96a	0.02	0.00	0.16
Japanese millet	.	.	.	.	.
Sudangrass	.	.	.	.	.
SEM	0.10	0.09	0.00	0.00	0.03
ANOVA (source of variation)	<i>P</i> -value				
SD	***	***	***	ns§	*
Companion	***	***	**	ns	*
SD × Companion	***	***	**	ns	*

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P > 0.05$).

Table 4.4. First post-seeding year forage dry matter yield at Sainte-Anne-de-Bellevue, QC, Canada of an alfalfa-timothy mixture originally seeded at three different dates with six different companion crops.

2019 seeding harvested in 2020					
	Total	CC†	Alfalfa	Timothy	Weeds
CC at the establishment	-----Mg DM ha ⁻¹ yr ⁻¹ -----				
Seeding date 1	21 May 2019				
Control (none)	10.94	.	9.49a‡	1.30cd	0.14
Berseem clover	8.87	0.00b	7.79ab	1.04d	0.05
Peas	9.25	0.00b	7.14bc	2.05ab	0.06
Annual ryegrass	9.48	0.69a¶	7.62abc	1.15cd	0.02
Oat	9.65	0.00b	7.19bc	2.37a	0.09
Japanese millet	8.96	0.00b	7.22bc	1.68bc	0.05
Sudangrass	7.93	0.00b	5.74c	2.11ab	0.09
SEM	0.58	0.07	0.64	0.20	0.03
Seeding date 2	13 June 2019				
Control (none)	8.97a	.	8.33a	0.59	0.05b
Berseem clover	7.34b	0.00b	6.14b	1.14	0.05b
Peas	7.73ab	0.00b	6.15b	1.49	0.09b
Annual ryegrass	7.71ab	0.84a	6.38b	0.45	0.04b
Oat	6.46bc	0.00b	5.35bc	1.01	0.11b
Japanese millet	6.65b	0.00b	6.00b	0.56	0.08b
Sudangrass	5.24c	0.00b	4.12c	0.89	0.22a
SEM	0.46	0.10	0.51	0.24	0.03
Seeding date 3	13 August 2019				
Control (none)	4.77	.	2.68a	1.96	0.12
Berseem clover	4.49	0.00b	2.35ab	1.93	0.21
Peas	3.95	0.00b	1.87abc	1.95	0.13
Annual ryegrass	4.32	1.15a	1.65bc	1.23	0.29
Oat	3.02	0.00b	1.05c	1.89	0.08
Japanese millet	2.83	0.00b	1.31c	1.32	0.20
Sudangrass	3.89	0.00b	1.59bc	2.12	0.19
SEM	0.43	0.17	0.33	0.32	0.07
ANOVA (source of variation)	<i>P</i> -value				
SD	***	ns§	***	**	ns
Companion	***	***	***	***	ns
SD × Companion	ns	ns	ns	ns	ns

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P > 0.05$).

Table 4.5. First post-seeding year forage dry matter yield at Saint-Augustin-de-Desmaures, QC, Canada of an alfalfa-timothy mixture originally seeded in two different years and fields at three different dates with six different companion crops.

2019 seeding harvested in 2020					
	Total	CC†	Alfalfa	Timothy	Weeds
CC at the establishment	-----Mg DM ha ⁻¹ yr ⁻¹ -----				
Seeding date 1	23 May 2019				
Control (none)	6.33ab‡	.	5.53a	0.75c	0.04
Berseem clover	6.14abc	0.00b	5.39ab	0.72c	0.02
Peas	6.25ab	0.00b	5.28ab	0.95bc	0.02
Annual ryegrass	6.40a	0.04a¶	5.49a	0.81bc	0.06
Oat	5.76bcd	0.00b	4.75bc	0.95bc	0.06
Japanese millet	5.28d	0.00b	3.99d	1.24ab	0.05
Sudangrass	5.57cd	0.00b	4.15cd	1.39a	0.02
SEM	0.20	0.01	0.23	0.14	0.01
Seeding date 2	19 June 2019				
Control (none)	6.27a	.	5.70ab	0.51	0.06
Berseem clover	6.48a	0.00b	5.82ab	0.64	0.02
Peas	6.67a	0.00b	5.92ab	0.69	0.05
Annual ryegrass	6.00a	0.03a	5.07bc	0.87	0.04
Oat	5.22b	0.00b	4.35c	0.77	0.11
Japanese millet	4.55bc	0.00b	3.23d	1.23	0.09
Sudangrass	4.16c	0.00b	3.10d	1.03	0.03
SEM	0.23	0.00	0.24	0.17	0.03
Seeding date 3	1 August 2019				
Control (none)	5.63	.	4.83	0.78	0.02
Berseem clover	5.60	0.00b	4.63	0.96	0.02
Peas	4.80	0.00b	3.64	1.14	0.02
Annual ryegrass	5.08	0.33a	4.16	0.49	0.09
Oat	4.45	0.00b	4.12	0.30	0.03
Japanese millet	3.94	0.00b	3.49	0.43	0.03
Sudangrass	5.08	0.00b	4.38	0.68	0.02
SEM	0.45	0.04	0.40	0.17	0.02
ANOVA (source of variation)	<i>P</i> -value				
SD	ns§	ns	ns	*	ns
Companion	***	***	***	*	ns
SD × Companion	*	***	***	**	ns

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P > 0.05$).

Table 4.6. First post-seeding year total forage dry matter yield at La Pocatière, QC, Canada of an alfalfa-timothy mixture originally seeded in early-June 2019 with six different companion crops.

2019 seeding harvested in 2020	
CC† at the establishment	-----Mg DM ha ⁻¹ yr ⁻¹ -----
Seeding date	10 June 2019
Control (none)	3.36a‡
Berseem clover	2.80abc
Peas	3.01abc
Annual ryegrass	3.17ab
Oat	2.48bc
Japanese millet	2.35cd
Sudangrass	1.61d
SEM	0.26

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

Table 4.7. Seeding year nutritive attributes values at Sainte-Anne-de-Bellevue, QC, Canada of an alfalfa-timothy mixture seeded in two different years and fields at three different dates with six different companion crops.

	2019 seeding			2020 seeding		
	aNDF¶	ADF	CP	aNDF	ADF	CP
CC† at the establishment	-----g DM kg ⁻¹ yr ⁻¹ -----					
Seeding date 1	21 May 2019			19 May 2020		
Control (none)	394b‡	287b	173ab	421cd	302c	150b
Berseem clover	413b	287b	162b	457bc	339ab	137bc
Peas	388b	275b	188a	410d	306c	169a
Annual ryegrass	407b	281b	168b	419cd	311c	151b
Oat	489a	325a	137c	491b	343ab	139bc
Japanese millet	511a	325a	129c	470b	322bc	134c
Sudangrass	506a	314a	109d	544a	351a	118d
SEM	10	6	5	13	9	5
Seeding date 2	13 June 2019			15 June 2020		
Control (none)	401c	270c	177a	388b	303b	159b
Berseem clover	415c	279bc	182a	384b	289b	183a
Peas	431c	290bc	173a	391b	308b	179a
Annual ryegrass	432c	277bc	165ab	366b	287b	178a
Oat	487b	300b	134bc	391b	305b	174ab
Japanese millet	544a	334a	127c	513a	350a	129c
Sudangrass	561a	328a	94d	545a	354a	99d
SEM	17	9	11	13	7	6
Seeding date 3	13 August 2019			31 July 2020		
Control (none)	255b	158	285	357c	266 ^e	238a
Berseem clover	271b	186	279	403b	299cd	187c
Peas	304b	225	261	392b	305c	213b
Annual ryegrass	289b	190	267	400b	285d	208bc
Oat	403a	244	216	549a	380a	124d
Japanese millet	450a	252	223	553a	360b	134d
Sudangrass	400a	234	256	532a	353b	134d
SEM	20	14	20	10	6	8
ANOVA (source of variation)	<i>P</i> -value					
SD	***	***	***	ns§	ns	*
Companion	***	***	***	***	***	***
SD × Companion	ns	ns	*	***	***	***

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P > 0.05$).

¶For all nutritive attributes, values presented consist in the annual weighted averages based on the individual harvest contribution to the total annual forage yields.

Table 4.8. Seeding year nutritive attributes values at Saint-Augustin-de-Desmaures, QC, Canada of an alfalfa-timothy mixture seeded in two different years and fields at three different dates with six different companion crops.

	2019 seeding			2020 seeding		
	aNDF¶	ADF	CP	aNDF	ADF	CP
CC† at the establishment	-----g DM kg ⁻¹ yr ⁻¹ -----					
Seeding date 1		23 May 2019			15 May 2020	
Control (none)	366c‡	290e	153ab	436	308	156
Berseem clover	396c	309d	145abc	402	287	168
Peas	397c	315cd	161a	430	305	160
Annual ryegrass	389c	307d	144abc	417	327	168
Oat	531a	343a	112d	460	315	160
Japanese millet	497ab	334ab	129cd	467	315	160
Sudangrass	471b	327bc	136bc	482	318	156
SEM	13	5	6	22	14	8
Seeding date 2		19 June 2019			15 June 2020	
Control (none)	374c	284c	149ab	404d	298e	159a
Berseem clover	366c	281c	158a	422d	308de	159a
Peas	360c	280c	164a	419d	312cd	160a
Annual ryegrass	364c	282c	154a	438cd	326cd	155ab
Oat	448b	301b	132bc	473c	332bc	147ab
Japanese millet	522a	331a	122c	571a	363a	127c
Sudangrass	509a	321a	120c	529b	350ab	142b
SEM	11	5	6	12	7	5
Seeding date 3		1 August 2019			8 August 2020	
Control (none)	331c	270c	209bc	255d	211e	286a
Berseem clover	347bc	273bc	219ab	301c	240d	258c
Peas	350bc	279bc	230a	351b	277c	262c
Annual ryegrass	341c	271bc	206bc	300c	223e	265bc
Oat	485a	322a	165e	458a	303b	194e
Japanese millet	500a	327a	186d	474a	327a	219d
Sudangrass	374b	284b	199cd	304c	247d	278ab
SEM	10	5	5	9	5	5
ANOVA (source of variation)				<i>P</i> -value		
SD	ns§	ns	**	*	**	***
Companion	***	***	***	***	***	***
SD × Companion	***	***	*	***	***	***

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P > 0.05$).

¶For all nutritive attributes, values presented consist in the annual weighted averages based on the individual harvest contribution to the total annual forage yields.

Table 4.9. Seeding year nutritive attributes values at La Pocatière, QC, Canada of an alfalfa-timothy mixture seeded in 2019 at three different dates with six different companion crops.

	aNDF§	ADF	CP
CC† at the establishment	-----g DM kg ⁻¹ yr ⁻¹ -----		
Seeding date 1	10 June 2019		
Control (none)	310d‡	252d	230a
Berseem clover	340cd	266cd	183b
Peas	354c	273c	233a
Annual ryegrass	320d	255cd	230a
Oat	508a	331a	151bc
Japanese millet	469b	308b	161bc
Sudangrass	542a	333a	128c
SEM	11	6	14
Seeding date 2	28 June 2019		
Control (none)	529d	368b	142bc
Berseem clover	610a	400a	116cd
Peas	407e	310c	178a
Annual ryegrass	606ab	398a	114d
Oat	493d	307c	175a
Japanese millet	541cd	333c	165ab
Sudangrass	553abc	324c	146b
SEM	22	10	9
Seeding date 3	2 August 2019		
Control (none)	.	.	.
Berseem clover	.	.	.
Peas	359b	270	270a
Annual ryegrass	.	.	.
Oat	474a	290	160b
Japanese millet	.	.	.
Sudangrass	.	.	.
SEM	5	4	7
ANOVA (source of variation)	<i>P</i> -value		
SD	**	**	**
Companion	***	***	***
SD × Companion	***	***	***

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§For all nutritive attributes, values presented consist in the annual weighted averages based on the individual harvest contribution to the total annual forage yields.

Chapter 5. Discussion

Across environments, companion crop species maximizing total seeding year forage yields varied depending on the seeding date. Overall, the use of oat, Japanese millet and sudangrass as companion crops for the establishment of an alfalfa and timothy mixture tended to maximize seeding year total forage yields (3.90 to 7.30 Mg ha⁻¹ yr⁻¹ depending on the site and year; average of 47 % increase across environments and species compared to the control seeded without a companion crop) when seeded early in the season (Tables 4.1 to 4.3). For a seeding in late spring or early summer, forage yields were the greatest with the use of Japanese millet and sudangrass, (4.16 to 8.89 Mg ha⁻¹ yr⁻¹; average of 118 % increase), while with an early August seeding, yields were the greatest with the use of oat and Japanese millet (1.11 to 4.68 Mg ha⁻¹ yr⁻¹; average of 1043 % increase). For the early and late spring/early summer seedings, peas generally produced intermediate yields (2.34 to 4.83 Mg ha⁻¹ yr⁻¹; average of 17 % increase) as well as for the August seeding (0.93 to 2.59 Mg ha⁻¹ yr⁻¹; average of 643 % increase).

Oat, which is the species most commonly used in Quebec as a companion crop for the establishment of perennial forages, performed better with earlier spring and early August seedings, but did not perform as well when seeded in late spring or early summer. When oat was used as a companion crop for a seeding in mid-June or early-July in dry conditions, weeds contributed approximately to half of the forage yields obtained. As oat is a cool-season grass, these results are consistent with reduced growth and competitiveness against weeds in dryer and warmer conditions that are characteristic of late spring or early summer (Canevari et al. 2000; CRAAQ, 2005). Our results thus correspond to the locally

suggested seeding times which are to seed oat in spring or in August (CRAAQ, 2005; OMAFRA, 2021a).

Sudangrass performed well when used as a companion crop for the first two seeding dates, but particularly well when seeded in late spring or early summer, which is consistent with results from Matteau et al. (2020) obtained in Southern Quebec. Japanese millet performed well across all dates, but especially well in late spring and August in the two southernmost locations. This is in agreement with observations of Peterson et al. (2008) in the American Midwest, where Japanese millet had consistent yields across a wide range of seeding dates (i.e., from early-May to early-July). Japanese millet although did not grow when seeded in August in the northernmost location in the present study (i.e., La Pocatière). Thus, Japanese millet seems adapted to a wide range of conditions but appears not to tolerate lower temperatures ($< 20^{\circ}\text{C}$), which is consistent with typical needs of C_4 species that require high growing degree day accumulation and higher base temperatures (CRAAQ, 2005).

Peas produced intermediate total forage yields at some environments and seeding dates compared to the other companion crops, which is in agreement to results reported by Sheaffer et al. (2014). Overall, berseem clover and annual ryegrass performed poorly across sites and years contributing minimally (i.e., $< 1 \text{ Mg ha}^{-1} \text{ yr}^{-1}$) to seeding year total forage yields. In a study previously conducted in Québec on the use of berseem clover as a companion crop for the spring establishment of alfalfa, a contribution of $1 \text{ Mg ha}^{-1} \text{ yr}^{-1}$ to biomass from berseem clover itself was reported (Bélanger, 2019). Our results are consistent with the ones reported in that study for the first seeding date while berseem clover contribution to total forage yields were even lower in this experiment for later

seedings. Low forage yields associated to the berseem clover treatment were due to its poor growth in the climatic conditions over the two seeding years and poor competitiveness against weeds. Low seeding year total forage yields resulting from the use of annual ryegrass as a companion crop for the establishment of perennial forages was previously documented in certain environments by Sulc et al. (1993) and Matteau et al. (2020) and were even lower in this study ($<540 \text{ kg ha}^{-1} \text{ yr}^{-1}$). However, the use of annual ryegrass as a companion crop has also been reported in some cases to result in high total forage yield (Coulman et al., 2019) that could even be greater than the use of an oat companion crop (Wiersma et al., 1999), contrary to the results obtained in this study.

Sudangrass and Japanese millet were the only companion crops that contributed consistently to total forage yields at each of the two or three harvests of the seeding year, other companion crops contributing only to the first harvest, their regrowth being minimal or, in the case of annual ryegrass, the growth at each growth cycle being minimal (Figures S2 to S4). Consistent forage yield contributions over multiple harvests for sudangrass were previously reported by Matteau et al. (2020), while Japanese millet has been reported to be a multiple harvests crop (OMAFRA, 2021a). While the presence of the companion crop only during the first growth cycle allows for the perennial crop to recover upon the first harvest and could thus be an advantage (Undersander et al., 2011), not having a consistent botanical composition throughout the season could be undesirable as the nutritive value of the forage could vary across harvests which can be an issue in terms of ration formulation (Matteau et al., 2020).

When oat, Japanese millet and sudangrass were seeded in their respective ideal times as discussed above, they not only maximized total seeding year forage yields but also

minimized weeds growth and contribution to yield (average of 54 % reduction compared to the control seeded without a companion crop across environments and seeding dates), which has previously been reported for oat and sudangrass (Peters, 1961; Becker et al., 1998; Sheaffer et al., 2014; La Vallie et al., 2020). Peas intermediately controlled weeds (average of 21 % reduction), similarly to results reported by Sheaffer et al. (2014). Berseem clover and annual ryegrass treatments did not differ from the control seeded without a companion crop offering very minimal weed control, in part due to their limited growth and biomass production. This has also been previously observed by Nelson et al. (1965) and Matteau et al. (2020) for berseem clover and annual ryegrass, respectively.

Despite the positive impact on total forage yields and weed suppression, the use of oat, Japanese millet, and sudangrass as companion crops to establish perennial forages resulted in the production of a forage with an overall lower nutritive value when compared to the other treatments (average across environments and seeding dates of 35 and 18 % higher aNDF and ADF concentrations, and 20 % lower CP concentration; Tables 4.7 to 4.9). Peterson et al. (2008) also previously reported that the use of sudangrass and Japanese millet as solo-seeded crops could result in the production of forage of a lower nutritive value when compared to the use of other annual crop species. The use of an oat companion crop resulted in a forage with a nutritive value that was higher when compared to the use of sudangrass in the majority of cases, contrary to observations by Matteau et al. (2020). Sudangrass produced forage with higher nutritive value than oat in the August seeding, since sudangrass was removed from the field earlier than its predetermined phenological stage due to the frost in the fall (i.e., sudangrass was in the vegetative stage, but did not

reach its targeted height). The forage nutritive values observed following the use of oat in the present study are in accordance with previous ones such as Hoy et al. (2002).

The companion crop species that increased total forage yields and resulted in a forage with a high nutritive value similar to the control established without a companion crop, according to the attributes evaluated, was peas. Thus, forage peas appear to provide a compromise in terms of forage yields and nutritive value as previously reported by Cupina et al. (2010) and Sheaffer et al. (2014). However, rumen protein degradability with peas is high, and solely using peas as a companion crop may not be appropriate as it could cause issues in the rumen, potentially resulting in high N loss (Mustafa and Seguin, 2004). Moreover, peas tended to lodge in all experiments, thus harvestability could be another issue with using forage peas as a companion crop.

While the use of oat, Japanese millet and sudangrass as companion crops may have maximized total forage yields, it also reduced alfalfa and timothy yields (by 57 and 54 % on average across sites and seeding dates compared to the control seeded without a companion crop), as well as their stem densities in the seeding year in some environments and for some seeding dates. Ideally, alfalfa stem densities should be above 400 stems per m² (Undersander et al., 2011). In this case, alfalfa stem densities were below that level in some cases. Such negative effects resulting from the use of a companion crop was previously reported following the use of oat and sudangrass as companion crops (Becker et al., 1998; Spandl et al., 1999; Canevari et al., 2000; La Vallie et al. 2020; Matteau et al. 2020). This negative effect persisted in the post-seeding year: either total forage yields and/or alfalfa yields, and stem density being reduced in all environments and seeding dates, except the August seeding in one location (Tables 4.4 to 4.6 and S1). In fact, total post-

seeding forage yields were reduced, on average, across the three environments by 16, 22 and 29 % (average of 22 %) with the use of oat as a companion crop in the seeding year at the different seeding dates (from 1st, mid-may, to 3rd, August), by 22, 27 and 35 % (average of 28 %) with Japanese millet at the three seeding dates and by 31, 38 and 14 % (average of 27 %) with sudangrass at the respective seeding dates compared to the control originally seeded without a companion crop. Thus, although the averages for each may not differ statistically, the use of sudangrass appears riskier for the first two seeding dates, Japanese millet for late spring/early summer and late summer seedings, and oat for late summer seedings.

In the case where alfalfa yields were reduced, timothy yields were increased. Timothy appeared to compensate the loss of alfalfa and improve the persistence of the mixture, which is normally a benefit of seeding perennial grasses with legumes (Lackman, 2001). The alfalfa stem densities in the spring and total post-seeding year yields were lower with sudangrass, followed by Japanese millet and oat to a lower extent. This illustrates that the companion crops can have a negative impact on the establishment of the perennial forages and that this effect can persist in post-seeding years.

Contrary to Matteau et al. (2020) and similarly to La Vallie et al. (2020), sudangrass had most important negative effects on stem densities of alfalfa and total forage yields for the first two original seeding dates. The greater effect observed with sudangrass could be due to the dry conditions observed in the seeding year, which would have promoted the companion crop growth and increased competition with the perennial forages compared to the oat companion crop which has difficulties growing in these conditions (Figure S1; Canaveri et al., 2000).

The use of the other companion crops, berseem clover, annual ryegrass and forage peas, did not affect perennials in the post-seeding year as previously reported by Canevari et al. (2000) and Bélanger (2019) for a berseem clover companion crop, Wiersma et al. (1999) for an annual ryegrass companion crop, and Cupina et al. (2010) and Sheaffer et al. (2014) for a pea companion crop. The residual effects with the use of sudangrass, Japanese millet and, to a lesser extent, oat, (i.e., lower post-seeding year total forage yields, lower alfalfa stem density in some environments, and lower timothy stem density at one location and seeding date) were most important in the initial post-seeding year harvest and decreased with the subsequent harvests, except in the northernmost location (Figures S5 and S7). This could be due to the capacity of alfalfa to produce multiple stems from a single plant that can branch off the axillary buds, which would explain the increase in the number of stems in the fall at one location (data not shown; Casler and Undersander, 2019).

The negative impact on the establishment for perennials was more evident for the August seeding: all treatments reduced the establishment of the perennial forages compared to the control seeded without a companion crop. Thus, the use of companion crops does not appear to be a good option if seeding is to take place later in the season (i.e., in August), which is typically not recommended locally as companion crops compete for resources, such as moisture, and can thus reduce the perennial forages establishment (OMAFRA, 2021b). This being said, if producers had a lack of forage due to poor growth during the season, oat was shown to produce moderate to high yields when seeded in August at all locations and could thus potentially be solo seeded to provide a source of emergency forage. This could also be the case with Japanese millet in southernmost locations of Quebec. These results remain valid for the different seeding rates and harvest stages that

we used and could be different if the seeding rates were lowered for example or, if crops were harvested earlier.

When looking at the total forage yields over the two growing seasons (i.e., 2019 seedings harvested in 2019 and 2020), the effects of the companion crop treatments differed slightly depending on the site. However, in general, when weeds were excluded, the total forage yields of the two growing seasons were similar for all treatments at every site (data not shown). Thus, this reinforces that depending on the producer's situation, it could be adequate to use more risky companion crop species in the seeding year, such as sudangrass (i.e., increases yields greatly in seeding year, but reduced post-seeding total forage yields by 27 % on average and up to 52 % in one environment), and have potentially lower yields in the post-seeding year if a producer was in need of high yields in a particular year and situation.

Chapter 6. Conclusions

In conclusion, contrary to the initial hypothesis, oat produced similar yields to sudangrass and Japanese millet depending on the seeding date. It was confirmed that these two companion crop treatments maximized yields along with oat compared to all the other companion crop treatments. Contrary to the hypothesis, this experiment demonstrated that sudangrass, followed by Japanese millet and oat to lower extent, can have a negative effect on the establishment of the desired perennial species when they are seeded at their respective ideal period, (i.e, oat for spring or late summer seedings, Japanese millet throughout the growing season, and sudangrass for late spring and early summer seedings), which will be confirmed with the first post-seeding year of the 2020 seedings that have yet

to be evaluated. Thus, the choice of a companion crop will be specific for the needs of a producer and there is no single solution. The use of berseem clover and annual ryegrass did not help to substantially increase yields in the seeding year although they did not affect negatively the establishment of perennial forages. When there is a need for forage biomass quickly in the growing season and overall high yields for that season, oat, sudangrass or Japanese millet can be seeded early, in the spring, as companion crops for the establishment of an alfalfa-timothy mixture. A later seeding, from late spring to early summer, could be done with the use of sudangrass or Japanese millet as companion crops and maximize total seeding year forage yields. However, the use of sudangrass, followed by Japanese millet and oat as companion crops in these periods can have a negative impact on the establishment of perennials and residual effects in the first post-seeding year. This has to be considered if a producer aims at establishing forages for several years since the field may not persist as long in time, although the stem density observed was around the one suggested for a productive alfalfa field (≥ 400 stems per m^2). The use of peas in the seeding year in some environments produced similar or higher yields than the control while reducing weed biomass and did not have residual effects in the first post-seeding year. Thus, it could be considered as an alternative: producing intermediate yields while not negatively affecting the establishment of the perennial species. When forage yields are low during the growing season and there is insufficient forage stored, oat, at all locations, or Japanese millet, in southernmost locations, could potentially be solo seeded in August to provide an emergency forage source. Their use as companion crops, as well as any other companion crop, would not be recommended since they have important negative effects on the establishment of perennial forages when seeded in late summer. Sudangrass,

Japanese millet and oat, provided lower quality forages than other annual companion crops such as peas. In a situation where producers need high quality forage, peas appear to be the most appropriate, although it produces lower forage yields than other companion crops, while sudangrass and Japanese millet appear less appropriate in terms of quality. Oat provided moderate quality forage and could be used as a compromise between quality and high quantity forage source. Peas could potentially be added to the mixture with oat, to increase quality, as it is often suggested (OMAFRA, 2009). The oat-pea mixture could thus produce a high forage yield and quality source although it was not evaluated in this present study.

Chapter 7. References

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Chapter 8. Supplemental tables and figures

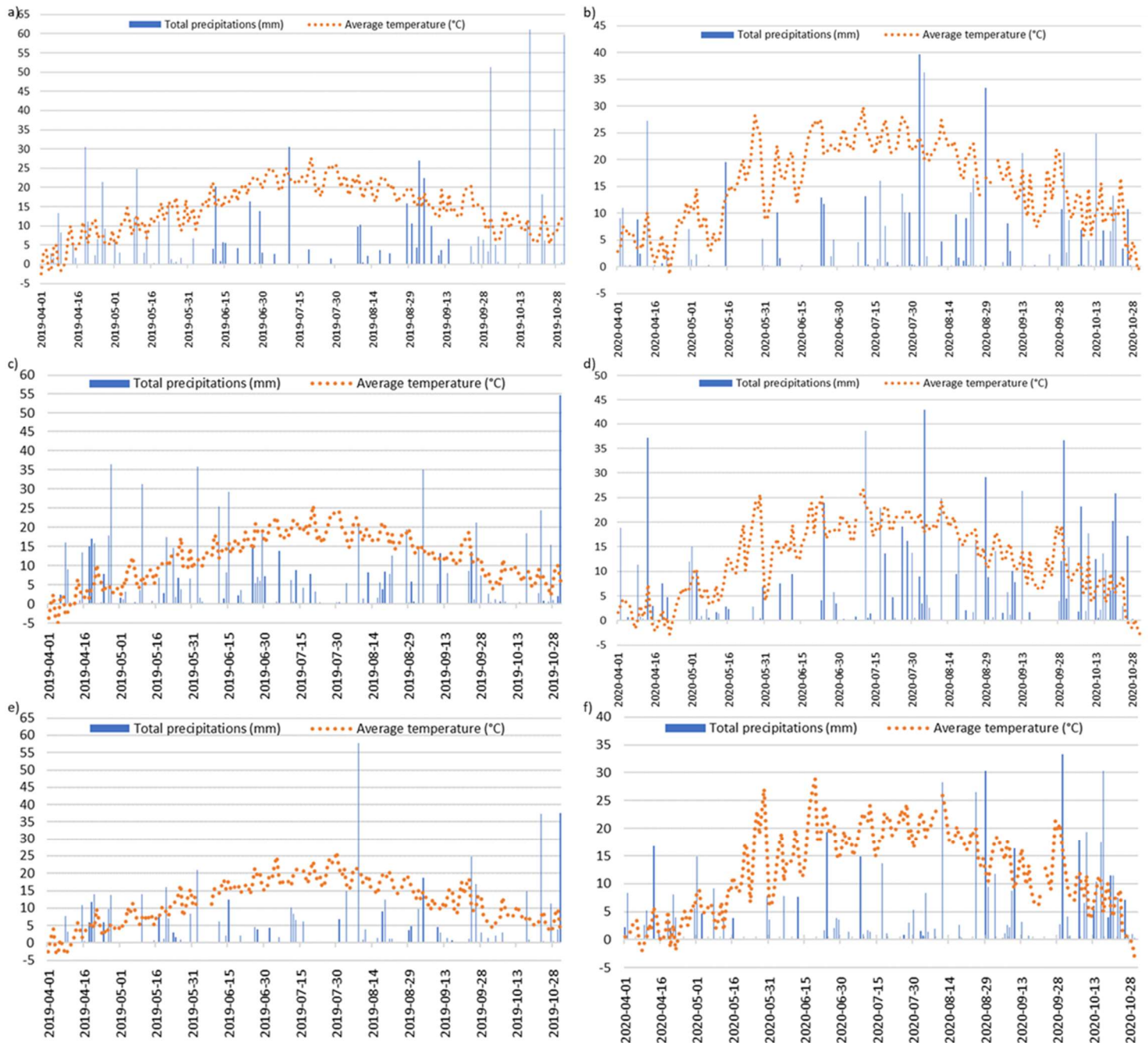


Figure S1. Average daily temperature and total precipitations during the growing season, from April 1st to October 31st at Sainte-Anne-de-Bellevue, QC, Canada, a) in 2019 and b) 2020, at Saint-Augustin-de-Desmaures, QC, Canada, c) in 2019 and d) 2020 and, at La Pocatière, QC, Canada, e) in 2019 and f) 2020. Adapted from Environment Canada (2020).

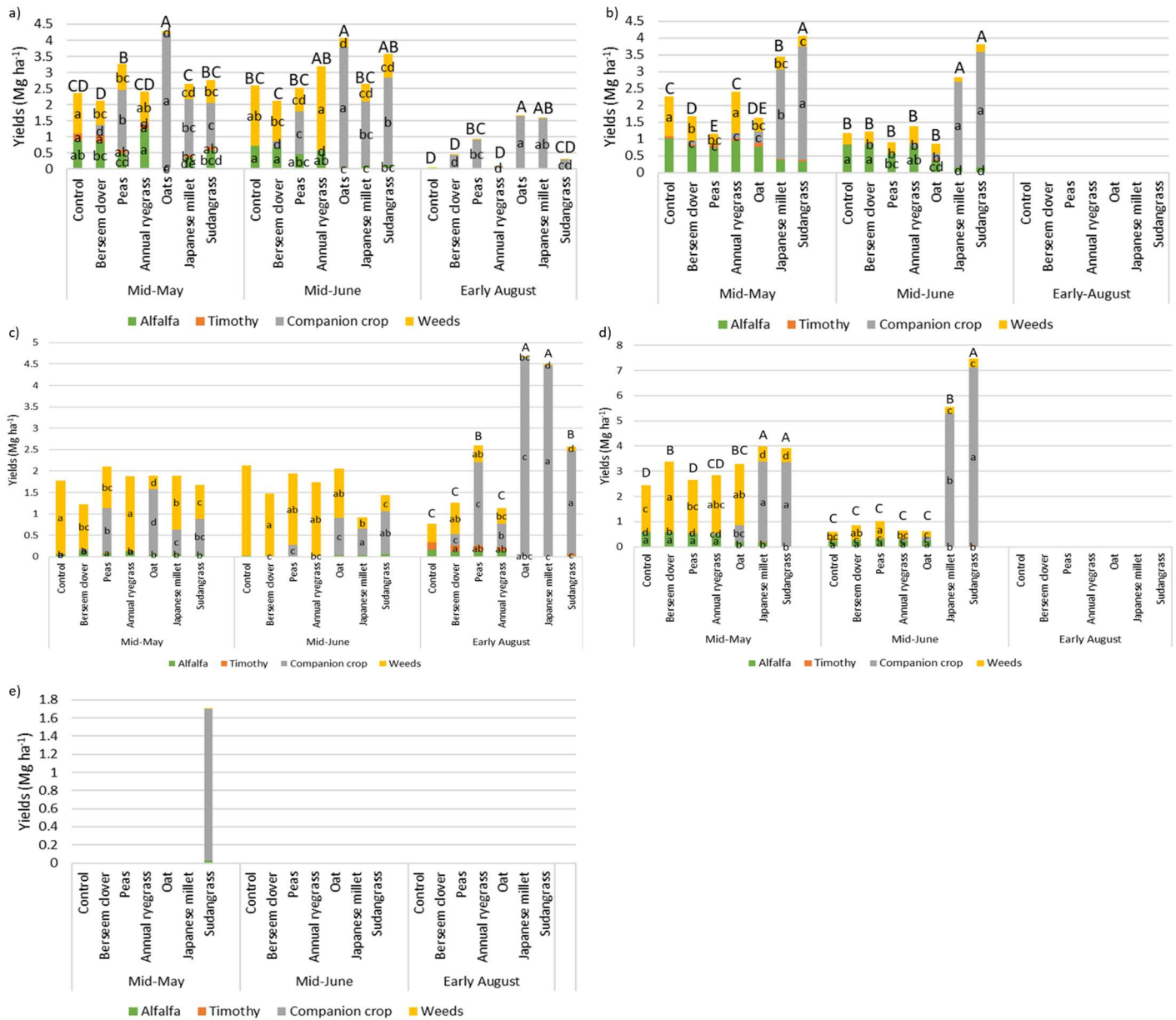


Figure S2. Seeding year forage dry matter yield at Sainte-Anne-de-Bellevue, QC, Canada of an alfalfa-timothy mixture seeded at three different dates with six different companion crops in 2019 a) harvest 1 and b) harvest 2, and in 2020 c) harvest 1, d) harvest 2 and e) harvest 3. For a given seeding date, total forage yield means for each companion crop followed by the same capital letter are not significantly different according to LSD (0.05). Within a forage component (i.e., alfalfa, timothy, companion crop or weeds) and for a given seeding date, means (corresponding to bars) with the same letter are not significantly different according to LSD (0.05).

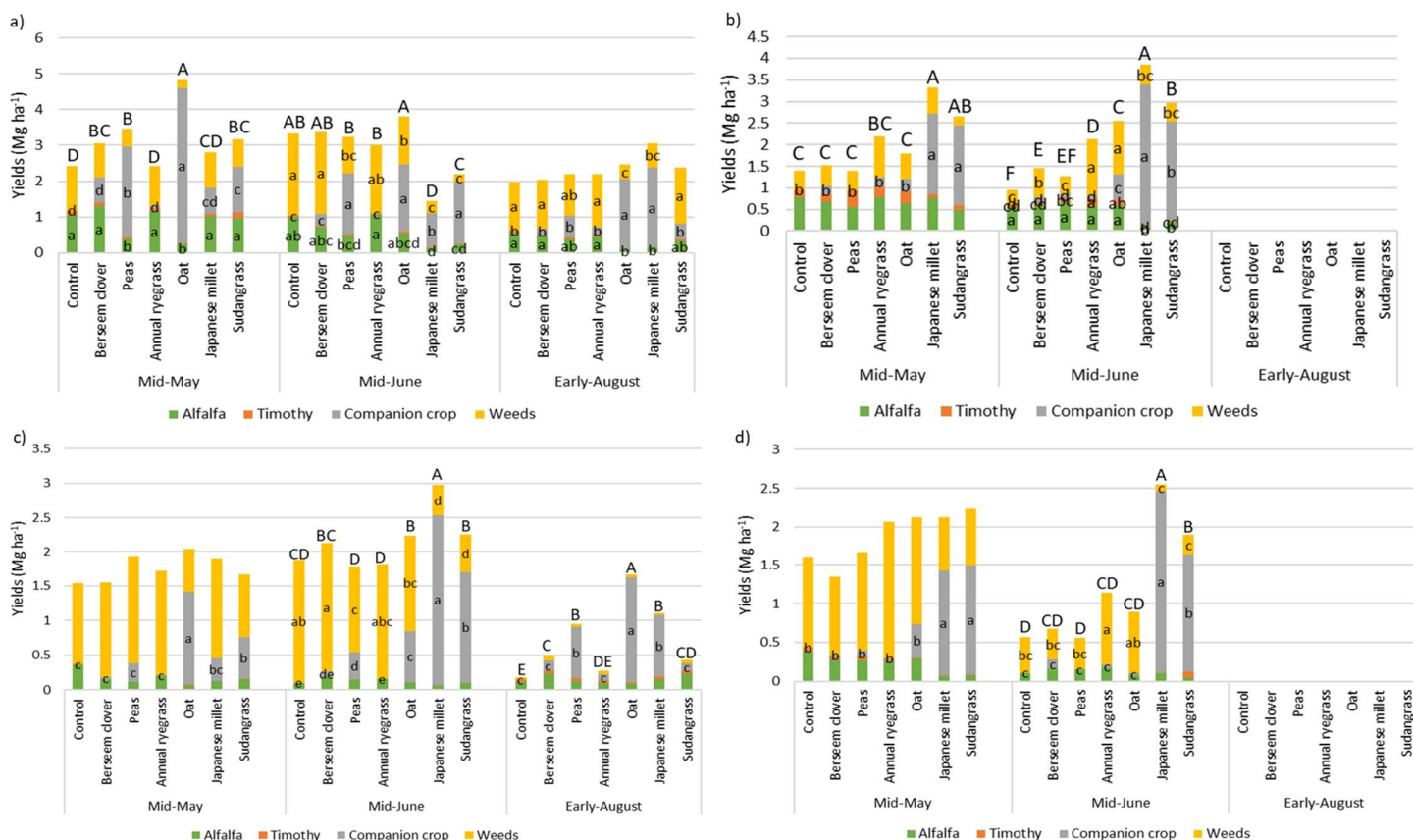


Figure S3. Seeding year forage dry matter yield at Saint-Augustin-de-Desmaures, QC, Canada of an alfalfa-timothy mixture seeded at three different dates with six different companion crops in 2019 a) harvest 1 and b) harvest 2, and in 2020 c) harvest 1 and d) harvest 2. For a given seeding date, total forage yield means for each companion crop followed by the same capital letter are not significantly different according to LSD (0.05). Within a forage component (i.e., alfalfa, timothy, companion crop or weeds) and for a given seeding date, means (corresponding to bars) with the same letter are not significantly different according to LSD (0.05).

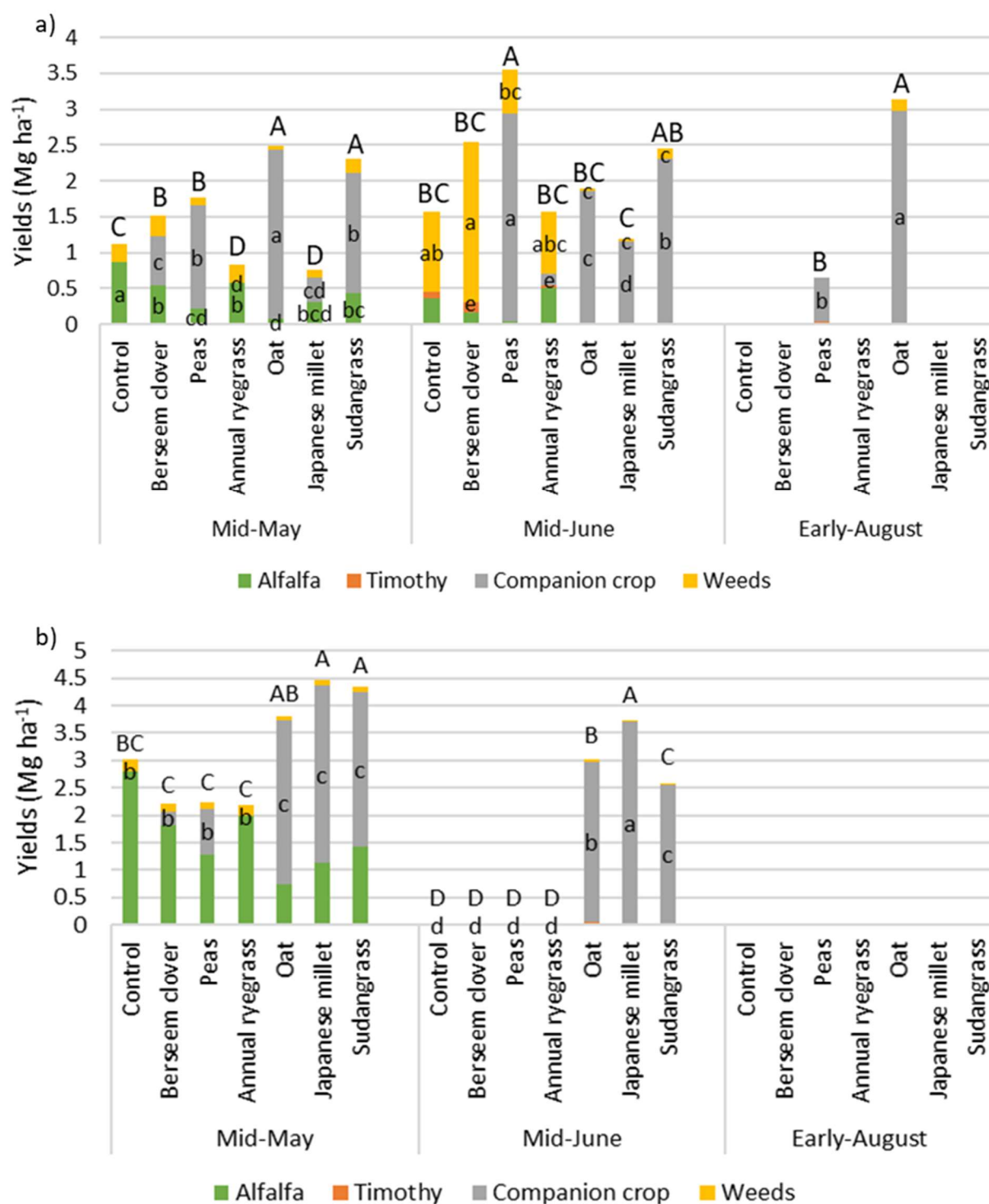


Figure S4. Seeding year forage dry matter yield at La Pocatière, QC, Canada of an alfalfa-timothy mixture seeded at three different dates with six different companion crops in 2019 a) harvest 1 and b) harvest 2. For a given seeding date, total forage yield means for each companion crop followed by the same capital letter are not significantly different according to LSD (0.05). Within a forage component (i.e., alfalfa, timothy, companion crop or weeds) and for a given seeding date, means (corresponding to bars) with the same letter are not significantly different according to LSD (0.05).

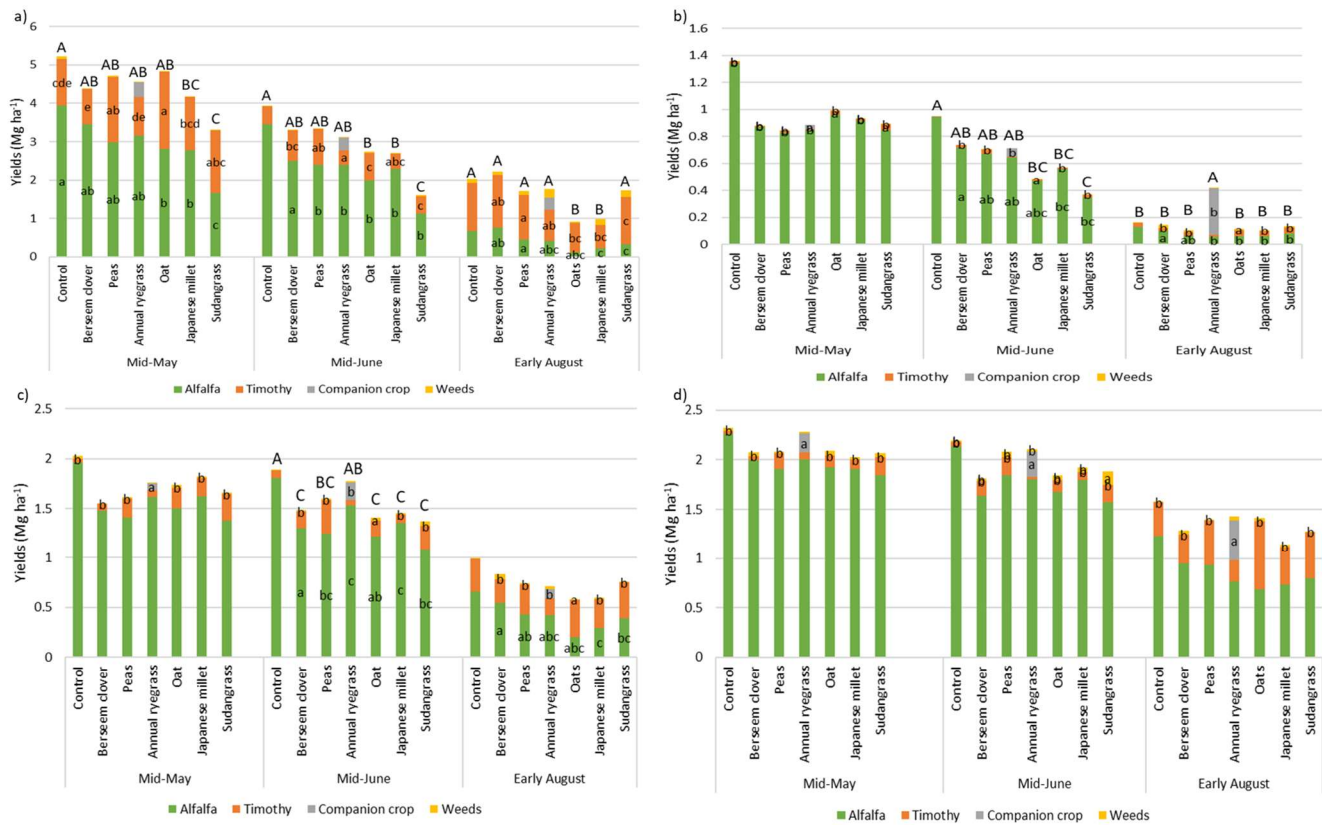


Figure S5. First post-seeding year forage dry matter yield at Sainte-Anne-de-Bellevue, QC, Canada of an alfalfa-timothy mixture seeded at three different dates with six different companion crops in 2019 a) harvest 1, b) harvest 2, c) harvest 3, and d) harvest 4.

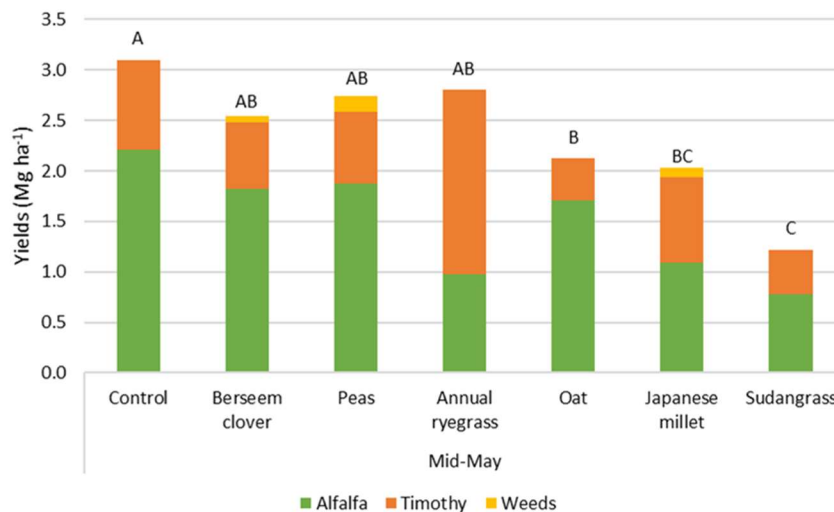


Figure S6. First post-seeding year forage dry matter yield at La Pocatière, QC, Canada of an alfalfa-timothy mixture seeded at three different dates with six different companion crops in 2019 at the harvest 1. For a given seeding date, total forage yield means for each companion crop followed by the same capital letter are not significantly different according to LSD (0.05). Within a forage component (i.e., alfalfa, timothy, companion crop or weeds) and for a given seeding date, means (corresponding to bars) with the same letter are not significantly different according to LSD (0.05).

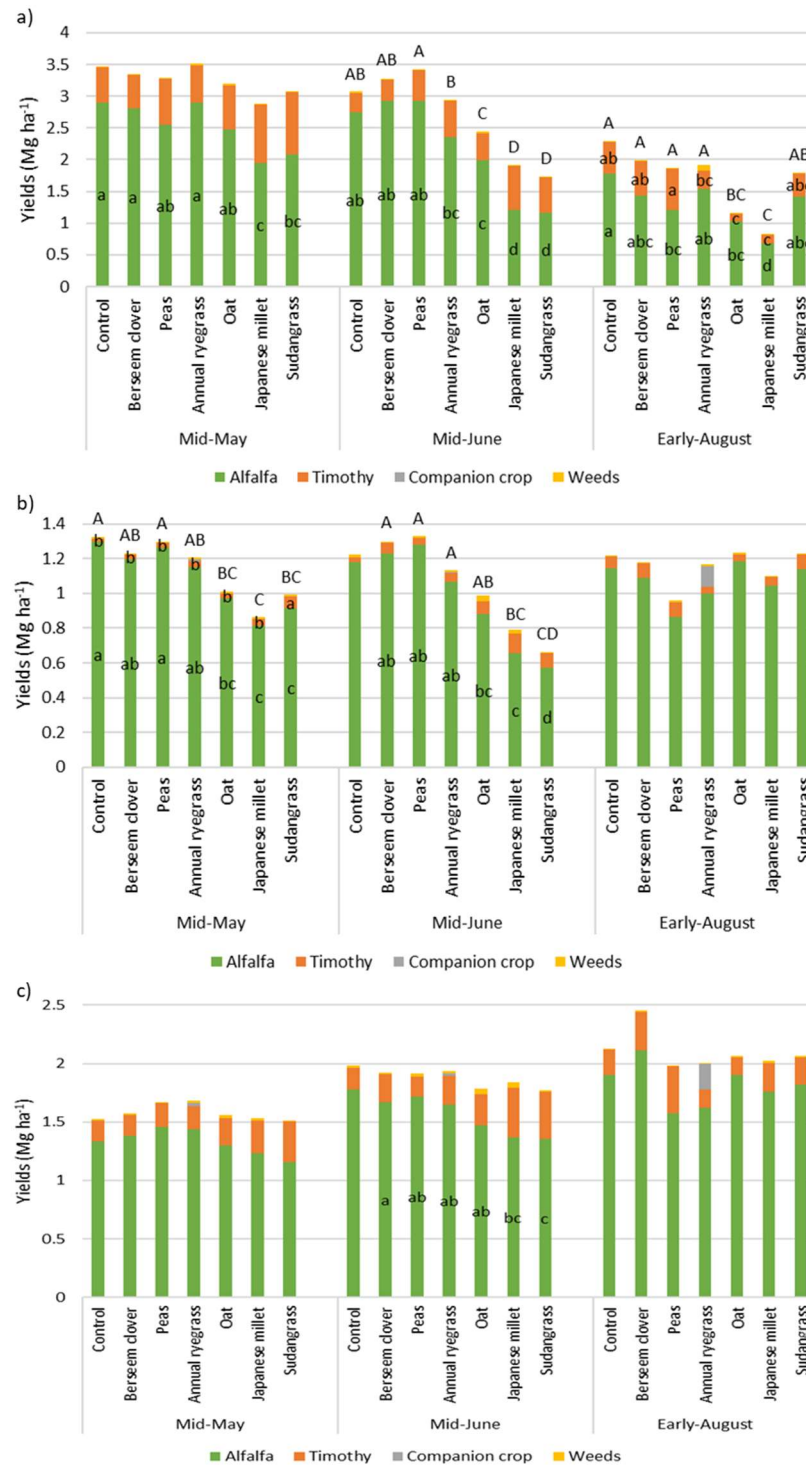


Figure S7. First post-seeding year forage dry matter yield at Saint-Augustin-de-Desmaures, QC, Canada of an alfalfa-timothy mixture seeded at three different dates with six different companion crops in 2019 a) harvest 1, b) harvest 2, and c) harvest 3. For a given seeding date, total forage yield means for each companion crop followed by the same capital letter are not significantly different according to LSD (0.05). Within a forage component (i.e., alfalfa, timothy, companion crop or weeds) and for a given seeding date, means (corresponding to bars) with the same letter are not significantly different according to LSD (0.05).

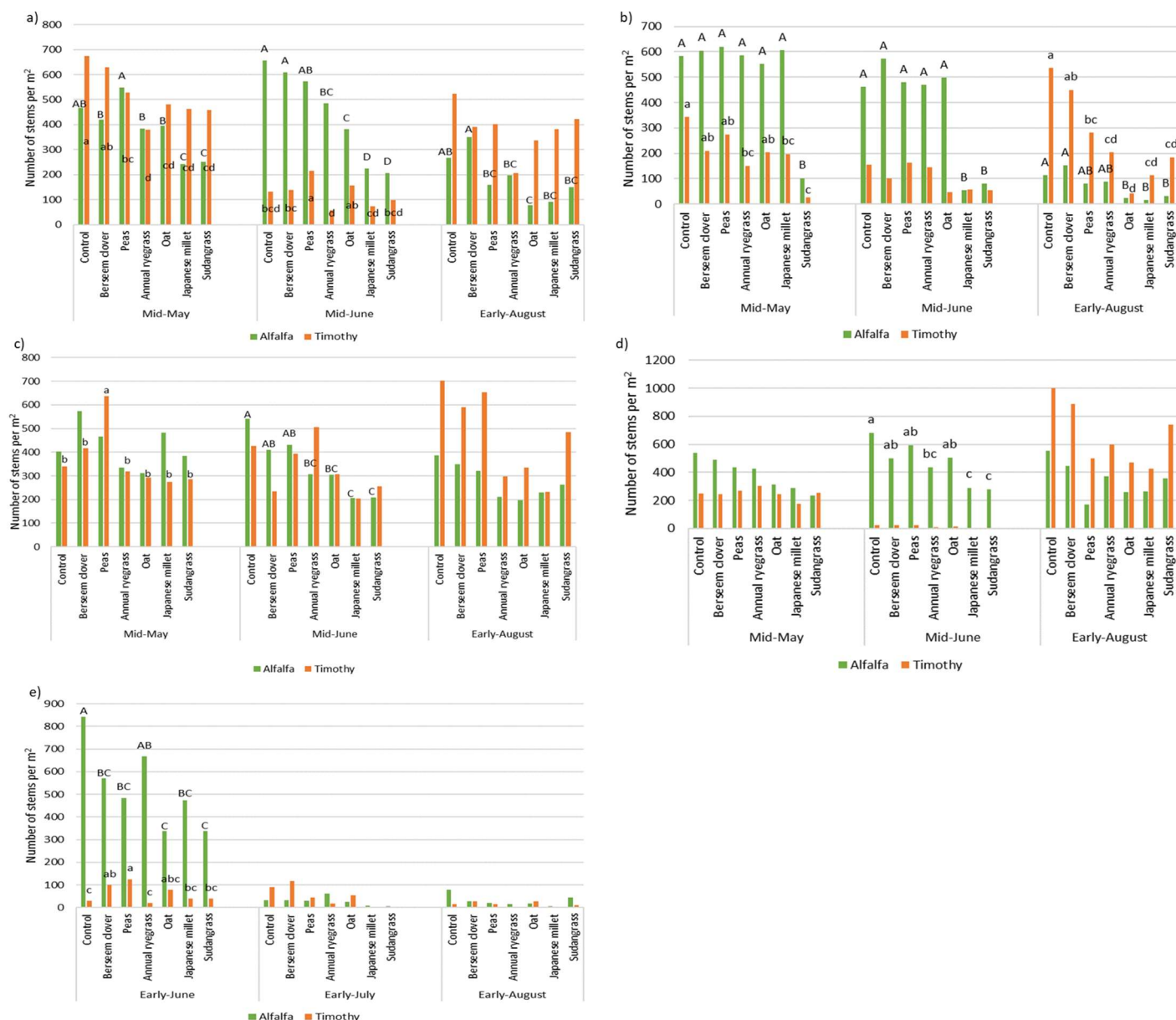


Figure S8. Alfalfa and timothy stem densities in the fall of the seeding year seeded at three different dates with six different companion crops at Sainte-Anne-de-Bellevue, QC, Canada a) in 2019 and b) in 2020, at Saint-Augustin-de-Desmaures, QC, Canada c) in 2019 and d) in 2020, and at La Pocatière, QC, Canada e) in 2019. For a given seeding date, total forage yield means for each companion crop followed by the same capital letter are not significantly different according to LSD (0.05). Within a forage component (i.e., alfalfa, timothy, companion crop or weeds) and for a given seeding date, means (corresponding to bars) with the same letter are not significantly different according to LSD (0.05).

Table S1. Stem densities of perennial species in the spring of the post-seeding year at three sites (Sainte-Anne-de-Bellevue, Saint-Augustin-de-Desmaures and La Pocatière, QC, Canada) of an alfalfa-timothy mixture seeded in different at three dates with six different companion crops.

	Sainte-Anne-de-Bellevue		Saint-Augustin-de-Desmaures		La Pocatière	
	2019 seeding		2019 seeding		2019 seeding	
	Alfalfa	Timothy	Alfalfa	Timothy	Alfalfa	Timothy
CC† at the establishment	-----Stems per m ² -----					
Seeding date 1	21 May 2019		23 May 2020		10 June 2019	
Control (none)	815ab‡	833b	868	740	817	60
Berseem clover	823a	894b	1021	651	633	144
Peas	636bc	1046a	872	1063	424	136
Annual ryegrass	693ab	550c	817	739	668	41
Oat	493cd	1050a	764	944	477	95
Japanese millet	649abc	843b	679	821	493	92
Sudangrass	406d	870b	663	856	385	60
SEM	62	50	106	98	101	37
Seeding date 2	13 June 2019		19 June 2020		28 June 2019	
Control (none)	1031a	393cd	999	599	3	152
Berseem clover	874ab	523bc	828	447	15	53
Peas	668bc	685a	768	595	4	39
Annual ryegrass	763abc	265d	720	687	28	19
Oat	669bc	599ab	656	591	31	71
Japanese millet	738c	318d	391	613	3	4
Sudangrass	532c	335d	512	652	13	0
SEM	93	52	119	76	8	32
Seeding date 3	13 August 2019		1 August 2020		8 August 2019	
Control (none)	445a	919	676	861a	44a	37
Berseem clover	431ab	764	529	708abc	13bc	63
Peas	280abc	769	469	996a	16bc	12
Annual ryegrass	267bcd	638	473	441bc	9c	0
Oat	89e	565	352	341c	1c	23
Japanese millet	105de	612	231	320c	7c	3
Sudangrass	198cde	860	459	829ab	40ab	23
SEM	57	119	85	132	9	25
ANOVA (source of variation)	<i>P</i> -value					
SD	**	ns§	*	*	**	ns
Companion	***	***	***	**	*	ns
SD × Companion	ns	*	ns	**	*	ns

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

†CC, companion crop. For the control treatment, the alfalfa-timothy mixture was established without a companion crop.

‡Within columns and for a given seeding date, means for each companion crop followed by the same letter are not significantly different according to LSD (0.05).

§ns, not significant ($P>0.05$).

Table S2. Harvest dates and growing degree-days between harvests of an alfalfa-timothy mixture seeded in two different seeding years and fields at three different dates with six different companion crops at Sainte-Anne-de-Bellevue, QC, Canada.

	Control† (none)	Berseem clover	Peas	Annual ryegrass	Oat	Japanese millet	Sudangrass
2019 seeding harvested in 2019							
Seeding date 1	21 May 2019						
Harvest 1	15 July	15 July	23 July	15 July	15 July	15 July	15 July
GDD5‡	708	708	843	708	708	708	708
Harvest 2	30 August	30 August	9 Sept.	30 August	9 Sept.	30 August	30 August
GDD5	720	720	690	720	1087	720	720
Seeding date 2	13 June 2019						
Harvest 1	2 August	30 July	6 August	2 August	2 August	23 July	23 July
GDD5	808	754	871	808	808	622	622
Harvest 2	17 Sept.	17 Sept.	17 Sept.	17 Sept.	17 Sept.	17 Sept.	17 Sept.
GDD5	590	640	527	590	590	768	768
Seeding date 3	13 August 2019						
Harvest 1	16 October	16 October	16 October	16 October	16 October	16 October	16 October
GDD5	635	635	635	635	635	635	635
2020 seeding harvested in 2020							
Seeding date 1	19 May 2020						
Harvest 1	16 July	9 July	13 July	16 July	9 July	16 July	9 July
GDD5	915	777	864	915	777	915	777
Harvest 2	26 August	26 August	26 August	26 August	26 August	26 August	14 August
GDD5	707	842	759	707	842	707	655
Harvest 3	-	-	-	-	-	-	15 October
GDD5	-	-	-	-	-	-	558
Seeding date 2	15 June 2020						
Harvest 1	7 August	7 August	7 August	7 August	7 August	23 July	23 July
GDD5	976	976	976	976	976	702	702
Harvest 2	15 Sept.	15 Sept.	15 Sept.	15 Sept.	15 Sept.	15 Sept.	15 Sept.
GDD5	486	486	486	486	486	1302	1302
Seeding date 3	31 July 2020						
Harvest 1	15 October	15 October	15 October	15 October	15 October	15 October	15 October
GDD5	809	809	809	809	809	809	809

†For the control treatment, the alfalfa-timothy mixture was established without any companion crop.

‡GDD5, cumulated growing degree-days calculated using a 5°C basis from seeding to harvest 1, and between harvests.

Table S3. Harvest dates and growing degree-days between harvests of an alfalfa-timothy mixture seeded in two different seeding years and fields at three different dates with six different companion crops at Saint-Augustin-de-Desmaures, QC, Canada.

	Control† (none)	Berseem clover	Peas	Annual ryegrass	Oat	Japanese millet	Sudangrass
2019 seeding harvested in 2019							
Seeding date 1				23 May 2019			
Harvest 1	22 July	22 July	22 July	22 July	22 July	22 July	22 July
GDD5‡	676	676	676	676	676	676	676
Harvest 2	16 October	16 October	16 October	16 October	16 October	13 Sept.	13 Sept.
GDD5	790	790	790	790	790	619	619
Seeding date 2				19 June 2019			
Harvest 1	15 August	15 August	15 August	15 August	15 August	29 July	29 July
GDD5	795	795	795	795	795	550	550
Harvest 2	16 October	16 October	16 October	16 October	16 October	13 Sept.	13 Sept.
GDD5	447	447	447	447	447	514	514
Seeding date 3				1 August 2019			
Harvest 1	10 October	10 October	10 October	10 October	10 October	10 October	10 October
GDD5	615	615	615	615	615	615	615
2020 seeding harvested in 2020							
Seeding date 1				15 May 2020			
Harvest 1	15 July	15 July	15 July	15 July	15 July	15 July	15 July
GDD5	727	727	727	727	727	727	727
Harvest 2	20 August	20 August	20 August	20 August	20 August	20 August	20 August
GDD5	534	534	534	534	534	534	534
Seeding date 2				15 June 2020			
Harvest 1	7 August	7 August	7 August	7 August	7 August	7 August	7 August
GDD5	728	728	728	728	728	728	728
Harvest 2	9 October	9 October	9 October	9 October	9 October	9 October	9 October
GDD5	554	554	554	554	554	554	554
Seeding date 3				8 August 2020			
Harvest 1	9 October	9 October	9 October	9 October	9 October	9 October	9 October
GDD5	540	540	540	540	540	540	540

†For the control treatment, the alfalfa-timothy mixture was established without any companion crop.

‡GDD5, cumulated growing degree-days calculated using a 5°C basis from seeding to harvest 1, and between harvests.

Table S4. Harvest dates and growing degree-days between harvests of an alfalfa-timothy mixture seeded in 2019 at three different dates with six different companion crops at La Pocatière, QC, Canada.

	Control† (none)	Berseem clover	Peas	Annual ryegrass	Oat	Japanese millet	Sudangrass
2019 seeding harvested in 2019							
Seeding date 1	10 June 2019						
Harvest 1	29 July	29 July	29 July	29 July	29 July	29 July	29 July
GDD5‡	638	638	638	638	638	638	638
Harvest 2	13 Sept.	13 Sept.	13 Sept.	13 Sept.	13 Sept.	13 Sept.	13 Sept.
GDD5	513	513	513	513	513	513	513
Seeding date 2	28 June 2019						
Harvest 1	16 October	16 October	13 Sept.	16 October	14 August	14 August	14 August
GDD5	1134	1134	960	1134	662	662	662
Harvest 2	-	-	-	-	4 October	4 October	4 October
GDD5	-	-	-	-	420	420	420
Seeding date 3	2 August 2019						
Harvest 1	16 October	16 October	16 October	16 October	16 October	16 October	16 October
GDD5	619	619	619	619	619	619	619

†For the control treatment, the alfalfa-timothy mixture was established without any companion crop.

‡GDD5, cumulated growing degree-days calculated using a 5°C basis from seeding to harvest 1, and between harvests.

Table S5. Harvest dates and growing degree-days between harvests in the post-seeding year of an alfalfa-timothy mixture seeded in 2019 at three different dates with six different companion crops and at three contrasting sites in Quebec, Canada.

	Sainte-Anne-de-Bellevue	Saint-Augustin-de-Desmaures	La Pocatière
2019 seedings harvested in 2020†			
Harvest 1	11 June	19 June	22 June
GDD5‡	442	535	412
Harvest 2	7 July	15 July	19 August
GDD5	416	364	827
Harvest 3	7 August	20 August	-
GDD5	567	534	-
Harvest 4	14 Sept.	-	-
GDD5	464	-	-

†In the post-seeding year, all seedings were harvested at the same dates. In La Pocatière, only the first seeding had sufficient growth for harvest.

‡GDD5, cumulated growing degree-days calculated using a 5°C basis from 1 April to harvest 1 in the post-seeding year, and between harvests.