

Developing management strategies to support sustainable production of lucerne in long-rotation cropping systems

by

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Declaration

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Abstract

Lucerne (*Medicago sativa* L.) forms part of long-rotation cropping systems integrated with livestock in the southern Cape of South Africa. The lucerne phase is five to seven years long, followed by five to seven years of cash crops. Lucerne swards allow for the integration of livestock into cropping systems, improved resource utilisation, is a high-quality animal forage, biologically fix N, break disease cycles and may improve subsequent cash crops yields. However, some farmers consider excluding lucerne from crop production cycles. The main reasons for the exclusion of lucerne swards are low summer and winter herbage production due to moisture stress and lucerne's natural winter dormancy. Low herbage production during these periods create considerable fodder flow deficits and make the management of fodder flow programmes challenging for farmers. The oversowing of dryland lucerne swards with annual winter growing forage crops, to create lucerne-based multiple species pastures, was investigated to determine if fluctuations in fodder flow programmes could be reduced through increased winter herbage production. Field experiments were conducted at Tygerhoek Research Farm (Riversonderend) during the 2018 and 2019 growing seasons. Both single species treatments and mixes were oversown into an existing lucerne base. Single species treatments included black oats, forage barley, stouling rye, Westerwolds ryegrass, forage radish and canola. Mixes consisted of various combinations of hybrid ryegrass, Italian ryegrass, forage barley, black oats, various annual *Medicago* and clover species, vetch and forage radish. The effect of oversown species and mixes on herbage production, pasture and soil quality was monitored for the duration of this study. Drought conditions after oversowing restricted the performance of the oversown species and mixes and had a knock-on effect that persisted for the duration of this trial. No treatment had a higher herbage yield to that of the control at any stage in the growing season ($p > 0.05$). Small grains and mixes that contain small grains did however show the most potential to improve herbage production, especially in late winter. Due to poor performance of oversown treatments, herbage samples mainly consisted of the lucerne base and ryegrass, both as an oversown species and weed. The relatively similar species composition from different treatments yielded no clear and or obvious treatment that improved pasture quality, however, grazing management ensured that all treatments were of a high quality at the time of sampling. Similar returns of organic matter, both quantitatively and qualitatively resulted in soil quality that was similar between all treatments. Different results may be obtained if oversown species establish well and this study should be replicated in years of normal rainfall distribution to fully comprehend how changes in pasture composition will affect herbage production and pasture quality. Soil physical, chemical and biological parameters should also be monitored over an extended period of time as changes in soil quality may take several years in Mediterranean climates.

Opsomming

In die suid Kaap van Suid-Afrika word lusern (*Medicago sativa* L.) in langrotasie-wisselboustelsels ingesluit en laat vir boere toe om hul boerdery te diversifiseer met 'n veekomponent. Die lusern-fase is vyf tot sewe jaar lank en word opgevolg deur vyf tot sewe jaar van kontantgewasse. Lusern hou 'n verskeidenheid voordele in vir boere. Buitendien dat dit toelaat vir 'n veekomponent, laat lusern boere ook toe om hul hulpbronne meer effektief te gebruik. Dit verseker 'n hoë gehalte weiding vir diere, stikstoffiksering, verhoed die oordrag van siektes en kan lei tot hoër opbrengste van die daaropvolgende kontant gewasse. Ten spyte van die voordele, oorweeg boere steeds om lusern uit hul wisselboustelsels weg te laat. Dit is hoofsaaklik as gevolg van lusern se lae produksie in somer en winter. In die somer word produksie beperk deur droë toestande. In die winter sal koel en koue toestande produksie beperk weens lusern se natuurlike winterdormansie. Groot produksie word fluktuasies maak dit moeilik vir boere om voervloeiprogramme effektief te bestuur. Die oorsaa van bestaande droëland lusernstande met eenjarige wintergewasse, om lusern-gebaseerde mengsels te vorm, is ondersoek om te bepaal of voervloei fluktuasies in die wintermaande verminder kan word. Veldproewe is uitgevoer op Tygerhoek proefplaas tydens die 2018 en 2019 groei seisoene. Enkelspesies as ook mengsels is in die bestaande lusern stand ingesaai. Enkelspesies het in gesluit swart hawer, voergars, stoelrog, Westerwolds raaigras, voerradys en canola. Mengsels het bestaan uit verskeie kombinasies van hibriede raaigras, Italiaanse raaigras, voergars, swart hawer, verskeie *Medicago* en klawerspesies, wieke en voerradys. Die invloed van die lusern-gebaseerde mengsels op produksie, weidingskwaliteit en grondkwaliteit is gemeet tydens hierdie studie. Buitengewone droë toestande na die oorsaai van die eenjarige wintergewasse het 'n deurlopende effek op die proef gehad. Geen behandeling het 'n hoër produksie as die kontrole gehad tydens enige tydperk in die groeiseisoen nie ($p > 0.05$). Kleingrane en mengsels wat kleingrane bevat het, het egter die meeste potensiaal gewys, veral in laat winter. Die swak vestiging van die oorgesaaide gewasse het daartoe gelei dat die spesiesamstelling soortgelyk was as gevolg van die groot lusern en raaigraskomponente. Die soortgelyke spesiesamestellings het verhoed dat daar onderskei kon word tussen behandelinge op 'n weidingskwaliteit vlak. Alle behandelings was egter van 'n hoë gehalte weidingskwaliteit toe monsters geneem is. Soortgelyke insette van organiese materiaal, beide in hoeveelheid en kwaliteit, het daartoe gelei dat daar geen verskille vir grondkwaliteit tussen enige behandelings was nie. Indien eenjarige gewasse goed vestig mag resulte verskil van die wat in hierdie studie verkry is. Hierdie studie moet herhaal word in jare waar reënval meer normaal is om die ten einde verstaan hoe produksie, weidingskwaliteit en grondkwaliteit beïnvloed kan word. Grondfisiese, -chemiese en -biologiese parameters moet ook oor 'n langer tydperk gemeet word aangesien grondkwaliteit stadig verander in 'n Mediterreense klimaat.

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List of Abbreviations

A	Area
ADF	Acid-detergent fibre
AM	Arbuscular mycorrhizal
ANOVA	Analysis of variance
B	Boron
BW	Bulk weight
C	Carbon
CA	Conservation agriculture
Ca	Calcium
Cl	Chloride
C-m	Clover-medic
CP	Crude protein
CSUP	Whole-community substrate utilisation profile
Cu	Copper
DM	Dry matter
E	Substrate evenness index
H'	Shannon-Weaver substrate diversity index
ha	Hectare
K	Potassium
L.	Linnaeus
ME	Metaboliseable energy
Mg	Magnesium
Mn	Manganese
N	Nitrogen
Na	Sodium
NDF	Neutral-detergent fibre
NFE	Nitrogen free extract
P	Phosphorus
PCA	Principle Component Analysis
pp	Pages
R-c	Ryegrass-clover

Rpm	Revolutions per minute
S	Sulphur
spp.	species
TDN	Total digestible nutrients
US\$	United States Dollar
Var.	Variety
viz.	Namely
Ww	Westerwolds
Zn	Zinc

Chapter 1

Introduction

1.1 Background

The southern Cape of South Africa is characterised by a Mediterranean-type climate and stretches from Botrivier in the West to George in the East. Annual rainfall varies from approximately 400 mm in the West to 700 mm in the East (van Heerden, 1976). Warm dry summers and cold wet winters are conducive to the cultivation of wheat (*Triticum aestivum*), barley (*Hordeum vulgare*), canola (*Brassica napus*) and lupins (*Lupinus* spp.) in crop rotation systems (Smith, 2014).

Conservation agriculture (CA), or at least some of the principles of CA, have been adopted by most farmers in the southern Cape. The principles of CA include 1) reduced tillage, where not more than 25% of the soil surface is disturbed, 2) the retention of crop residue and 3) making use of crop rotations (Verhulst et al., 2010). Farmers aim to build up soil organic matter through these principles (Smith, 2014). The incorporation of a pasture phase and livestock may further promote the build-up of soil organic matter (Chan et al., 2001). Pasture phases used in the region include annual medics (*Medicago* spp.) or perennial lucerne (*Medicago sativa* L.) (van Heerden and Tainton, 1987).

In the southern Cape region, lucerne is typically used as a leguminous pasture crop, as it is more productive than annual medic pastures in this region (van Heerden and Tainton, 1987). The perennial nature of lucerne allows it to be included in long-rotation cropping systems. A typical lucerne phase lasts five to seven years, followed by five to seven years of cash crops. Integrating a lucerne phase in long-rotation cropping systems complements CA practices as it does not only add to crop diversity, but the perennial nature supports low levels of soil disturbance and there is more soil cover compared to conventional farming practices. It may, however, be argued that lucerne is cultivated as a monoculture due to the typical lucerne phase being between five to seven years long. Crop rotation is an effective way to break disease cycles (Lamprecht et al., 2011). Crop rotation additionally assists farmers with weed management as grass weeds can be sprayed with a selective herbicide in legume or brassica fields and vice versa (MacLaren et al., 2019). Crop rotation will also result in improved crop yields when compared to monocultures. Lucerne serves as a tool to fix atmospheric nitrogen and will produce a high-quality forage for livestock. The lucerne phase will typically be followed by a winter grain. The effect of crop rotation along with nitrogen fixation by the lucerne may lead to an increased grain yield and quality (Bouton, 2012).

South Africa is a net importer of mutton with imports valued at 19 388 000 US\$ and exports at only 3 321 000 US\$ (FAO, 2019). This potentially leaves scope for the expansion of the sheep industry to supply local demand. The integration of livestock into cropping systems has various advantageous for farmers. Sheep integrated into long-rotation cropping cycles provides diversity and reduces financial vulnerability of farmers to crop failure (Crookes et al., 2017). The sale of livestock and animal products like meat and wool allow farmers to generate an income during times when crops perform poorly as well as reducing the risk of cash flow problems (Herrero et al., 2016). In extreme cases where crops fail, animals may graze these fields, referred to as sacrificial grazing, and ensure at least some income from the particular field. The integration of livestock may also reduce fertiliser requirements. Peyraud et al. (2014) reported that a herd of 200 ewes can provide enough nitrogen (N), phosphorus (P) and potassium (K) for a field of approximately 15 ha. The herd will excrete roughly 710 kg of N, 770 kg of P and 105 kg of K. While this may not substitute the need for fertiliser for the entire farm, it may reduce the amount that the producer needs to apply.

Crop rotations integrated with livestock also allow for better overall resource utilisation on farms. Areas that are not suited for crop production may be utilised by livestock and improve overall farm productivity and profitability (Bell and Moore, 2012).

1.2 Problem statement

Herbage production patterns from lucerne swards in the southern Cape often present challenges for farmers. Summer and early autumn production may be limited due to moisture stress and winter production will be low due to lucerne's natural winter dormancy. During spring and autumn, when temperatures are warmer and enough soil moisture is available, lucerne production is high. This considerable fluctuation in fodder availability to livestock is challenging for farmers. Lucerne also has a poor reseeding ability due to autotoxicity. Thus, if a sward is damaged or plant population decreases with age, the sward may not be financially viable. This has led to some farmers exploring the possibility of excluding lucerne from long-rotation cropping systems.

Oversowing lucerne swards with winter annual forage crops to create a lucerne-based multiple species pasture may present an opportunity to ensure a better distribution of fodder availability throughout the year. Complementary seasonal growth patterns of certain winter annual forage crops may help farmers to overcome fodder shortages during winter months (van der Colf et al., 2015). In this case, oversown species will start contributing to total herbage production from early winter and will likely maximally contribute in spring (Purser, 1981). Winter annual forage crops will die off in late spring or early summer and lucerne will likely be the only contributor to herbage production. Farmers must

however consider risks associated with multiple species pastures. Oversown annual species will grow vigorously in spring and can directly compete with the lucerne base. This may reduce longevity of lucerne or result in a reduction of the lucerne component's productivity during summer.

Lucerne is a high-quality feed and another risk is that overall pasture quality may decrease as farmers strive for increased herbage production. During winter months when winter annuals are still in a young physiological stage, they should still be of a high quality to livestock. As the winter annuals mature and maximally contribute in spring their quality will be lower due to an increase in fibre content. The change in quality is more severe in grasses compared to legumes. Farmers should keep this in mind when considering when to utilise multiple species pastures. Grass and brassica species may increase total herbage production, while lucerne and other legumes will ensure a high-quality forage.

Multiple species pastures are also more resistant to weed invasion (Deak et al., 2007; Papadopoulos et al., 2012; Sanderson et al., 2004, 2005; Tracy and Sanderson, 2004). If a pure lucerne pasture is weed prone, oversowing with winter annuals can be considered. If weeds are replaced with annual winter forage crops, quality can be better managed and higher quality compared to a weed invested lucerne sward could be ensured.

There may also be additional benefits as a lucerne-based multiple species pasture can ensure more complete resource utilisation. The complementary root patterns of lucerne and winter annual forage crops may ensure that resources in both the shallow and deep root spectrum are utilised. The lucerne base will also promote nutrient cycling. Nutrients extracted from deep within the soil will be returned to the surface as deep-rooted species leave organic matter on the soil surface to decompose (Sanderson et al., 2004). Lucerne-based multiple species pastures will be more resilient than monocultures and are more drought tolerant (Papadopoulos et al., 2012). This could ensure higher production in drought prone semi-arid areas (Deak et al., 2007; Sanderson et al., 2004).

Soil quality is likely to improve in lucerne based multiple species pastures. The return of additional residue, diverse root systems and quality of organic matter from winter annual forage crops will have multiple effects on soil. This may include the formation of stable aggregates (Verhulst et al., 2010), promote soil microbial activity (Habig et al., 2015) and promote the build-up of soil organic matter. Multiple species pastures should result in reduced soil nutrient losses (Sanderson et al., 2004) due to the more complete use of nutrients leaving less excess available for nutrient leaching. Multiple species pastures will also return a balanced mix of residue, with both a high and low C:N ratio to the soil. This will ensure that the breakdown of organic matter by soil microbes is not too rapid, leaving residue to protect soil from erosion, conserve soil moisture and provide habitat for soil microbes. At the same

time some residue will be broken down to release nutrients and build organic matter (United States Department of Agriculture, 2011). Synergistic relationships between plants and soil microbes may form and ecosystem functions may increase. There may however also be unforeseen consequences like diseases that can be carried over from the pasture phase to the cash crop phase.

1.3 Objective and aims

The aim of this study is to make the lucerne phase more sustainable and a viable option for farmers in the southern Cape of South Africa through oversowing annual winter crops into existing lucerne swards. Sustainability will be defined through the following three objectives:

- 1) The pasture must have the same or higher yield when compared to a pure lucerne sward.
- 2) Pasture quality must not be compromised by the inclusion of winter annual crops.
- 3) The soil quality must improve or be of the same level after the lucerne phase.

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Chapter 2

Literature review: Lucerne-based multiple species pastures in long-rotation cropping systems in the southern Cape of South Africa

2.1 Background

Lucerne (*Medicago sativa* L.) is one of the most widely cultivated perennial legumes worldwide (Bouton, 2012a; Gallego et al., 2011). Lucerne is popular due to high nutritional quality as an animal feed, high biomass production, nitrogen (N) fixation potential and its adaptability to a large spectrum of environments (Bouton, 2012a). The lucerne phase in long-rotation cropping systems may also allow for improved soil quality through improved soil structure (Durand, 1993), allow for the build-up of soil organic carbon content and reduce the risk of soil erosion. Lucerne may be grown under irrigation or rainfed conditions.

In the Mediterranean climate of the southern Cape of South Africa, annual grain or oilseed crops are cultivated as part of a crop rotation system with lucerne incorporated as a forage crop in long-rotation cropping systems. In these systems, lucerne will typically be grazed by livestock, but may also be cut for hay, ensiled or be incorporated into a silage mixture. Lucerne also provides farmers with the benefit of having a more diversified farming system as livestock may generate income in years when crops perform poorly and generate income through the sale of animals or animal products to buffer cash flow fluctuations.

2.2 Lucerne under rainfed conditions

Lucerne can be cultivated under rainfed conditions in semi-arid regions with an annual rainfall of 300 mm to 450 mm if swards are well established and management maintains an adequate plant density (Bowman et al., 2004). An adequate plant density may vary in different regions depending on rainfall and function of the lucerne sward. A plant density that is suitable for N fixation may not be suitable for grazing under typical stocking rates for the southern Cape of South Africa. In semi-arid regions, a lower plant population may be advantageous as competition between individual plants will be lower allowing for better plant and pasture performance (Durand, 1993). Lucerne is a drought tolerant plant (Bouton, 2012a; Malinowski et al., 2007) with the potential to utilise water in deeper soil layers than annual grasses (Raza et al., 2013). It has a tap root system that can grow as deep as 7 m below the soil surface, but roots are concentrated in the upper 15 cm of soil (Humphries and Auricht, 2001). Deep rooted species like lucerne are advantageous in dry summer months as they can obtain water that is

not available to shallow rooted crops, resulting in active leaf growth (Hopkins and Holz, 2006). Lucerne may also utilise water during the summer months when there are no annual crops planted, allowing farmers to utilise rainfall throughout the year (Hayes et al., 2010).

Lucerne longevity and production may be influenced by both managerial and environmental factors. Environmental factors include moisture stress, day length and temperature, while managerial factors will include stocking rate and grazing system. Lucerne will respond to relatively low levels of rainfall, but water stress can decrease production by up to 30% or more in extreme cases (Bouton, 2012a; Jun et al., 2014). Shorter day lengths and reduced temperatures are linked to decreased herbage production. Lucerne will grow optimally at a daytime temperature of around 25°C and a slightly lower temperature at night (Durand, 1993). Appropriate stocking rates in a rotational grazing system will ensure the best chance for a sward to remain productive with a plant population above the critical threshold (Teixeira et al., 2007).

2.2.1 Grazing management

Good grazing management is required to ensure that farmers get the best performance from lucerne swards. Rotational grazing results in higher yields than continuous grazing (Avondo et al., 2013) as plants have an opportunity to replenish taproot reserves (Burnett et al., 2018). Although maximum build-up of root reserves will take place at full flowering, this is not ideal in terms of grazing quality. A compromise that allows for a quality herbage and the build-up of reserves can be reached at around 10% flowering (Durand, 1993). Swards that are grazed too frequently, especially in summer, will result in the reduced assimilation of N and carbon that is used for shoot growth or may be stored in the plant roots (Teixeira et al., 2007). This will in turn have a negative impact on the regrowth cycles in winter and spring resulting in reduced herbage production (Durand, 1993). In severe cases, plants reserves may be exhausted and result in plant mortality. The ideal grazing regime will allow for the rapid defoliation of lucerne with livestock being removed before regrowth occurs. After the decapitation of apices, it will take roughly seven days for regrowth to commence (Cosgrove and White, 1990).

In the southern Cape region of South Africa, lucerne can be cut or grazed approximately every six weeks regardless of dormancy class (Oberholzer et al., 1993, Oberholzer and van Heerden, 1997). Depending on climatic conditions lucerne swards may be rested for slightly shorter periods during optimal growing conditions or longer periods during times of water stress. Pastures will rarely consist of a pure lucerne sward due to the infiltration of ryegrass (*Lolium spp.*) and other weeds (Durand, 1993). A grazing frequency that is too high will result in a decrease in the lucerne component of a pasture (Deak et al., 2007) and result in the infiltration of unwanted weeds. To avoid selective grazing,

a high stocking rate can be applied for a short duration of time in a technique known as mob grazing. A longer grazing period, of up to 12 days, is also viable at lower stocking rates. (Cosgrove and White, 1990). Lucerne herbage production will be reduced if grazed for too long, as palatable species are often selectively grazed leading to damage to apices. Determining optimum grazing duration may be challenging due to varying stages of plant maturity (Cosgrove and White, 1990). Individual lucerne plants in the same sward may be in different physiological stages due to variation in soil.

Due to low winter productivity of lucerne, the oversowing of annual winter crops into lucerne swards may be a viable option to ensure less fluctuation of forage availability. Oversown crops in different growth stages will have different nutritional compositions. This may allow livestock to select for their nutritional requirements as opposed to monocultures (Hopkins and Holz, 2006). Grazing management must remain in line with that of lucerne to ensure that the pasture remains viable for animal production for the desired time frame of five to seven years (Badenhorst, 2011). Mixed species pastures can lead to improved animal nutrition and reduce risk of nutritional linked diseases due to sudden shifts in forage quality (Humphries, 2012).

Livestock will have an impact of the composition of a pasture due to selective grazing, defoliation patterns, trampling and uneven distribution of excreta (Sanderson et al., 2005). Soil compaction has been raised as a concern when incorporating livestock into conservation agriculture systems. Soil compaction from livestock is not as severe as that from vehicles and heavy machinery, with livestock generally only impacting the top 10 cm of soil (Bell et al., 2014). Livestock returns nutrients to the pasture through urine and faeces. This is opposed to cutting and removal for hay or silage, where nutrients are removed from the pasture (Truter et al., 2015). Due to the significant impact that livestock may have on pastures, they must be incorporated into studies to ensure accurate and applicable results for farmers (Sanderson et al., 2004).

2.3 Lucerne physiology

2.3.1 Nitrogen fixation

Legumes can be incorporated into pastures or planted in crop rotations to fix N. Nitrogen fixation by legumes can be used by farmers to subsidise a portion of the N requirement of grasses and reduce the input of N fertiliser (Bouton, 2012b). Lucerne is a typical legume species used to subsidise some of the N requirement of mixed species pastures (Bouton, 2012b; Carlsson and Huss-Danell, 2003; Raza et al., 2013). This may lead to reduced input cost of N fertiliser for farmers. There are various factors that may influence the amount of N fixated by lucerne in pastures. Factors include herbage production,

persistence, age of the lucerne sward, grazing management, pests and diseases, water stress, soil N and competition from other species that use N like grasses (Ledgard and Steele, 1992). The accumulation of N in the soil due to N fixation triggers a negative loop resulting in reduced N fixation. This is due to it being more efficient for the plant to utilise N in the soil than to fixate N (Ledgard and Steele 1992). The dynamic relationship between pasture species and the effect of grazing will influence how the species composition of a pasture changes over time. Increased N fixation will lead to conditions more favourable for grasses. If grasses increase and deplete soil N levels, conditions may become more favourable for legumes.

Research on the amount of N that will be fixed by lucerne or multiple species pastures with both legumes and grasses have found varying results. Pastures consisting of a grass-legume mixture with a legume fraction of 50% to 70% have been found to be capable of transferring 60 kg N ha⁻¹ from the atmosphere to soil (Finn et al., 2013). Bell et al. (2014) reported that lucerne may fixate 50 to 120 kg N ha⁻¹year⁻¹. Under optimal conditions N fixation may be as high as 230 kg ha⁻¹year⁻¹ (Humphries and Auricht, 2001). Although there is large variation, roughly 20 kg of N is fixed per tonne of DM produced when comparing findings from across the world (Peoples et al., 2001). Unkovich et al. (2010) concluded that lucerne N fixation is 18.7 kg N per tonne DM produced. There may be an underestimation due to N fixed in nodules or in roots that are not accounted for. In lucerne-grass mixes a rough estimate may be made by using the formula (Carlsson and Huss-Danell, 2003):

$$\text{N fixed (in a lucerne – grass mixture)} = (0.021 \times \text{DM}) + 17 \quad (1)$$

Where DM was measured in kg ha⁻¹ year⁻¹.

Due to various factors, N fixation values referred to may be used as a guideline but can ultimately not replace direct measurements (Unkovich et al., 2010). If the N required and application of N through fertiliser is reduced in conjunction with a mixed species pasture that utilises the N, leaching may be reduced (Blanco-Canqui et al., 2015).

If there is no damage from overgrazing or through pests and disease then the most important factors relating to N fixation will be plant density, soil temperature and soil water (Bowman et al., 2002). As lucerne swards get older there is a tendency that less N will be fixed, but age alone will not be the determining factor (Bowman et al., 2002). As swards age the plant population of lucerne will decrease. If the plant population drops to below a threshold value of eight plants per square meter, N fixation will almost half (Bowman et al., 2002). A sward with a plant population of eight plants per square meter may however already be too low to support typical stocking densities in the southern Cape of South Africa. Nitrogen fixation may cease if soil temperature averages below 10°C at a depth of 10 cm

(Bowman et al., 2004). Water stress will limit root-nodule formation and thus limit N fixation (Humphries and Auricht, 2001). Bowman et al. (2004) reported that if lucerne plants were at wilting point for 10 days it can be assumed that nodules have been shed and even if lucerne plants recover from moisture stress, N fixation will not commence immediately. It will take roughly 20 days for the re-formation of nodules and N fixation to initiate (Bowman et al., 2004).

2.3.2 Dormancy groups

Lucerne cultivars may be classified as dormant to non-dormant relating to regrowth potential in the autumn months. Dormancy classes range from 1-11 with 1 being dormant, having the least potential for autumn regrowth and 11 non-dormant with the highest potential for autumn regrowth (Malinowski et al., 2007). Lucerne with a low dormancy tend to be more sensitive to damage from grazing and careful management is required to ensure sward persistence. This is due to the growth points of winter dormant cultivars being situated close to the soil surface and less likely to be damaged by grazing livestock compared to non-dormant cultivars where the growth points are higher off the soil surface and may be damaged by grazing livestock. Cultivars with a low dormancy class (more dormant) are traditionally associated with colder climates and cultivars with a high dormancy class (less dormant) are associated with warmer climates, however this may not be true for all cultivars and dormancy ratings (Malinowski et al., 2007). Plants with a high winter dormancy may not be ideal in climates with long growing seasons. They may however be ideal for multiple species pastures as it will reduce competition between lucerne and annual grasses (Humphries and Auricht, 2001). While dormancy will influence production, it is not the most important factor regarding productivity in a rainfed system. Environmental stresses, pests and diseases will have a more profound effect on production (Malinowski et al., 2007).

2.3.3 Longevity

Longevity or persistence of lucerne swards is dependent on the survival of individual plants within the sward (Teixeira et al., 2007). Persistence is important to farmers as a more persistent pasture will be more economically viable. More persistent swards distribute the high input cost and result in a lower average cost per year making the total cost of the rotation cheaper (Bouton, 2012a). In the southern Cape an acceptable duration of the lucerne phase will typically be five to seven years. Persistence is negatively affected by severe moisture stress, overgrazing and pests and diseases. Despite of its drought tolerance, very high temperatures and severe moisture stress may lead to low production and in extreme cases some plants may perish. Careful management is essential during these times as

not to damage lucerne swards. Lucerne does not respond well to continuous grazing and rotational grazing management is advised in all conditions. Poor establishment will result in swards that do not produce adequate herbage, reduced animal performance and reduced benefits associated with lucerne pastures. After establishment, the plant population will be at its highest level and will start to decrease over time. Good initial establishment of a pasture is essential to ensure longevity (Bouton, 2012a). Lucerne has a weak reseeding ability due to its natural autotoxicity. It requires good management practices that avoid overgrazing and damaging of the sward to ensure persistence (Bouton, 2012a; Humphries, 2012). If a sward is damaged and plants perish, it will not recover to the previous level of performance.

Lucerne herbage yield consists of various yield components that include plant population, individual shoot mass and shoots per plant (Volenec et al., 1987). As plants population declines over the swards lifetime these individual yield components may compensate for the reduced plant population (Teixeira et al., 2007). Smit and Terblanch (1994) stated that 80 to a 100 plants per m² is required for optimal production in swards younger than two years and that that plant density may be lower, roughly 60 to 70 plants, in older swards. Other authors have like Teixeira et al. (2007) have suggested a lower value of around 43 plants per m² for sufficient herbage production. Moot et al. (2012) suggested that the plant population threshold may be lower at around 30 to 45 plants per m² for sufficient herbage production. It is worth noting that plant population in lucerne swards may be underestimated as crowns of individual plants that grow near one another may fuse and look like a single plant. (Durand, 1993). If the main purpose of the lucerne sward is N fixation and not herbage production, a plant population as low as eight plant per m² may be acceptable (Bowman et al., 2002). Other biotic factors like pests, in particular nematodes, diseases and weeds can negatively impact longevity of lucerne swards and farmers must manage these risks according to best practice (Bouton, 2012b, 2012a). The main reason for plant mortality in lucerne swards are pests, diseases, high temperatures and moisture stress during summer months. Poor management practices in the previous growing season reduce resilience of swards and increase plant mortality.

2.3.4 Autotoxicity

Lucerne has a natural autotoxicity that prohibits the establishments of new seedlings in established lucerne swards. This mechanism may have served as a favourable survival tool in dry regions where lucerne originates from with limited resources like water (Chon et al., 2006). However, it provides challenges for modern farmers with diminishing plant populations in older lucerne swards. Autotoxicity is a specialised form of allelopathy where older plants inhibit seedlings of the same

species from establishing. The autotoxic chemicals of lucerne are found in fresh herbage, water-soluble and more concentrated lucerne shoots compared to roots (Chon et al., 2006). The chemicals have various effects that include delayed germination, inhibited lucerne root growth and reduced or lack of hairs on roots (Chon et al., 2006). The chemicals are water soluble and will typically be transferred from leaves, where the concentration is the highest, to the soil by rainfall washing or via old leaves that fall to the soil surface (Chon et al., 2006). The main effect on seedlings is that the autotoxic chemicals inhibit the development of a taproot (Chon et al., 2006). Numerous autotoxic chemicals for lucerne has been reported, but not have been proven to be the definitive cause of lucerne's autotoxicity. It may also be possible that autotoxicity is caused by the interaction or combination of several of these chemicals (Chon et al., 2006).

2.4 Multiple species pastures

The use of multiple species pastures is a strategy that may be adopted for sustainable intensification of pastures (Finn et al., 2013; Tracy and Sanderson, 2004). Multiple species pastures also known as mixed-species pastures, mixed herb leys or multi-species pastures contain a combination of species that may include legumes, grasses and/or herbs (Daly et al., 1996). Multiple species pastures tend to be dynamic with the botanical composition changing over time (Deak et al., 2007). Multiple species pastures can be divided into either a binary pasture, consisting of two species or diverse pastures consisting of three species or more.

Multifunctionality refers to a pasture that provides multiple services. These include weed suppression, environmental stability, reduced nutrient leaching and reduced fluctuation in herbage yields. This may lead to more effective use of available resources, improved herbage yields, reduced cost of weed suppression, better soil quality, increased soil organic carbon and a higher pasture stability against environmental stresses. While the number of species in a pasture does not determine the pasture's multifunctionality, multiple species pastures tend to have a greater multifunctionality than monocultures (Finney and Kaye, 2017). Multifunctionality is determined by the species' ability to utilise different niches and provide functional diversity. Some monocultures may provide the same level of multifunctionality than multiple species pastures (Finney and Kaye, 2017). Factors like soil improvement, seasonal distribution of yield, weed suppression, high quality herbage and increased biodiversity that can justify multiple species pastures even if total herbage production is lower (Sanderson et al., 2004).

It has been argued that for multiple species pastures to be considered, positive effects must be evident within a short period of time and performance must be comparable to the best current monocultures

(Finn et al., 2013). The flaws in this approach is that the best performing crop may not be planted in a specific year (Finn et al., 2013). In crop rotation systems as practised in the southern Cape it will be more beneficial to look at the most productive crop rotation cycle or the specific phase of the cycle that one wishes to improve. The value added from a systems approach may also not be measurable after a single year. Conservation agriculture and the incorporation of a multiple species pasture phase will over time allow for the build-up of soil organic carbon, weed suppression, breaking of disease cycles, increased yields and more stable yields during droughts. These positive effects will however only be apparent after a longer period and when looking at a systems approach as opposed to performance over a single growing season.

Management of lucerne-based multiple species pastures may be challenging. A fine balance must be maintained between lucerne not dominating the oversown species or the oversown species dominating lucerne compromising persistence (Humphries, 2012). Establishment of different species may also be challenging. Establishing a lucerne base and oversowing grasses and other crops into the sward in subsequent years has however been successful (Humphries, 2012). It will be important to apply management strategies that allow seedlings to germinate and emerge in the sward. This can either be done through grazing of lucerne prior to oversowing or oversowing crops with a high seedling vigour. Through grazing management, farmers may manipulate the species composition in a pasture. If grazing frequency is increased, the lucerne fraction tends to decline and if the grazing frequency is reduced, allowing for a longer recovery time, lucerne tends to increase (Humphries, 2012). A heavy stocking rate for short durations of time may also be favourable to lucerne as it will minimalise selection by animals. Selection may be a problem due to lucerne's high palatability. Improved animal performance may be achieved by oversowing annual crops into lucerne. Diverse pastures will also bring new challenges. Cost of establishment may increase, management may be more complex and planting seeds of different sizes may prove to be challenging (Smith et al., 2014). It may also be difficult to find the right combination of species for a specific area due to environmental factors and species interactions.

2.4.1 Species interaction

Competition for resources between lucerne and different oversown annual winter crops may influence production, weed suppression and establishment of the oversown species in the pasture. The growing season of different crops must be considered. If two species compete for limited resources during critical growth stages then the pasture performance will be compromised (Humphries, 2012). In the southern Cape, lucerne herbage production will start to increase in spring

when there is adequate soil moisture and temperatures start to rise. Oversown annual species may also grow vigorously during this time. These species include ryegrass, vetch (*Vicia* spp.) and clovers (*Trifolium* spp.). There may thus be direct competition between lucerne and these oversown crops for limited resources. This can lead to the lucerne component being reduced, but overall production being higher. When temperatures rise and rain becomes less frequent in late spring and early summer, there will be less water available and annual species will die off. The reduced lucerne component and reduced soil moisture may then have a negative impact on lucerne production in summer, when water is already a limited resource. The termination of annual winter crops may be considered to ensure minimal negative impact on summer production of lucerne. A careful balance must be maintained between competition for resources and creating an environment with no competition where weeds may capitalise. Both may have a negative impact on summer production as the typical weeds found in the area, like *Conyza sumatrensis*, will not positively contribute to herbage production or herbage quality. The same chemicals that lead to autotoxicity in lucerne may also have an allelopathic effect on some of the seedlings of oversown species. There may be allelopathic effects on both broadleaf and grass species. The effect on broadleaf species may however be more severe (Chon et al., 2006). The allelopathic effect is not strong enough to exhibit weed control (Chon et al., 2006), but may still result in reduced performance of oversown species.

2.4.2 Herbage production

Herbage yield in pastures depend on various interactions between cultivated crops and soil quality, soil composition, weather conditions, plant species interactions and grazing management (Deak et al., 2007; Hopkins and Holz, 2006). There are various theories and mechanisms used to explain improved performance of pastures with multiple species. Botanical composition changes as pastures mature and it is likely that the mechanism responsible for production in a specific grassland will also change over time (Sanderson et al., 2004). Diverse pastures have been linked to increased production by some authors (Finney and Kaye, 2017; Sanderson et al., 2004) while others have reported mixed results when comparing monocultures and multiple species pastures. Finn et al. (2013) reported that multiple species pastures outperformed the best performing monoculture in 60% of observed sites over multiple years and that 97% of multiple species pasture sites had outperformed the average monoculture. Daly et al. (1996) reported that multiple species pastures outperformed monocultures and binary pastures, but this was not the case for all sites. Facilitation or species compatibility, where the presence of one species promotes the growth and survival of another species, may result in improved performance (Sanderson et al., 2005, 2004). Tall plants may create microclimates underneath their canopy with lower soil temperature promoting growth of a species that might have

struggled otherwise (Sanderson et al., 2004). This can however also go the other way where one species is suppressed by the presence of another. Low growing species can be suppressed by taller grasses and legumes (Deak et al., 2007). The presence of one species of grass may inhibit or promote the growth of another non-grass (Papadopoulos et al., 2012). It is important to understand the inter-species relationship and species compatibility to ensure that input costs are kept at a minimum and money is not wasted on pasture mixtures that are destined to fail. Variation in results should not be surprising as multiple species pastures are complex. Soil quality, climatic condition, grazing or cutting management and species interaction will all have an effect on herbage production. Under optimal conditions for a specific monoculture, the monoculture is likely to outperform diverse pastures (Sanderson et al., 2004).

In the southern Cape, lucerne is planted as a monoculture in the pasture phase of crop rotations. Farmers sometimes find it challenging to cultivate lucerne as it has low herbage during two periods of the year. The first period is during mid-summer due to limited water under rainfed conditions. The second period is during winter when poor regrowth is observed due to low temperatures and lucerne's natural dormancy. Overseeding species into lucerne with different seasonal growth patterns is a possible way to increase annual and seasonal herbage production (Humphries, 2012, Badenhorst, 2011). The inclusion of winter growing grasses may increase the quantity of herbage while the legumes will ensure a high-quality feed. The additional herbage may also serve other functions similar to cover crops like a possible decrease in weeds as well as soil and wind erosion (Blanco-Canqui et al., 2015). Higher herbage yields can be attributed to the mixture of species being better adapted to grow throughout the growing season effectively extending the growing season when comparing diverse pastures to monocultures (Deak et al., 2007).

Productivity can peak with a relatively low number of species present (Hopkins and Holz, 2006). Species composition, species interaction, functional group utilisation and climatic conditions are the most important factors for herbage production (Deak et al., 2009, 2007; Finney and Kaye, 2017; Papadopoulos et al., 2012). Climatic conditions will still influence herbage yield, but the effect may not be as severe. Herbage yields may also increase due to an abundance of fast-growing weeds (Sanderson et al., 2004). The positive effect on production will in this case be short lived.

2.4.3 Weeds

Multiple species pastures can be planted to manage weed populations (Finney and Kaye, 2017). They have been known to decrease the abundance of weeds through competition for limited resources (Deak et al., 2007; Papadopoulos et al., 2012; Sanderson et al., 2005, 2004; Tracy and Sanderson,

2004). Niche differentiation may improve pasture performance as plants utilise different aspects of the available resources resulting in very little waste (Sanderson et al., 2004). Each species will dominate a specific part of the habitat, but no species can take over resulting in a wide range of species in the pasture (Deak et al., 2007). This complementary use of the available resources ensure that weeds cannot invade in the highly competitive environment (Tracy and Sanderson, 2004).

The cultivation of diverse pastures may result in condition that favour invasive weeds (Tracy and Sanderson, 2004). When a pasture is established, it is critical that establishment of the sown species will be rapid to ensure that weeds do not take advantage of good growing conditions and establish themselves in the pasture (Finn et al., 2013). A high producing vigorous species can be included in the pasture to limit the risk of weeds. (Sanderson et al., 2005, 2004). The inclusion of a species with vigorous growth reduces the likelihood that weeds will establish as the dominant species and outcompete sown pasture species for resources (Tracy and Sanderson, 2004). Species evenness is important as a pasture where species are evenly distributed is linked to higher multifunctionality and weed resistance (Finney and Kaye, 2017). This is as a result of resources being used more evenly resulting in a more competitive environment throughout the pasture, not allowing weeds to establish (Tracy and Sanderson, 2004). The duration of weed resistance will depend on the mixture of species planted. Finn et al. (2013) found that a pasture consisting a various legumes and grass species exhibited weeds suppression for at least three years. This was achieved despite some variation of pasture composition over the study period. Ultimately it is difficult to predict exactly what will happen due to the complexity of multiple species pastures as species composition will change over time. The outcome will depend on environmental response as well as the different species planted and how they interact (Blanco-Canqui et al., 2015). It is, however, widely agreed that multiple species pastures may be an effective way to suppress weeds if the correct combination of species for a specific area can be identified.

2.4.4 Effect of environmental stresses

Diverse pastures have been linked to better stability against environmental stresses through better resources utilisation by different functional groups (Sanderson et al., 2004; Tracy and Sanderson, 2004). The “insurance effect” may serve as a mechanism to ensure a stable pasture as even under extreme conditions or stress. If one crop does not perform due to suboptimal conditions, another may compensate and reduces the risk of crop failure (Sanderson et al., 2005). Drought tolerance of diverse pastures compared to cultivated monocultures of grassland is higher (Papadopoulos et al., 2012) and might be of importance to farmers in areas that are predicted to become more drought prone due to

climate change. In dry years, diverse pastures may produce more herbage than monocultures, while herbage production may be the same in years of normal rainfall distribution (Deak et al., 2007; Sanderson et al., 2004). Deep-rooted species may allow shallow rooted species to perform better during dry spells through a process known as hydraulic lift where water is brought closer to the surface from deep within the soil (Sanderson et al., 2004). This may leave more water near the surface and available to shallow rooted species. Nutrients extracted from deep within the soil is also returned to the surface as deep-rooted species leave organic matter on the soil surface to decompose in a process known as nutrient cycling (Sanderson et al., 2004).

Species composition of pastures may change over time and lead to the pasture responding differently to the environment (Deak et al., 2009). Selective grazing may further contribute to species composition change in pastures and make the estimation of grazing value as well as the reaction to climatic conditions of the pasture increasingly difficult (Deak et al., 2009). Finn et al. (2013) found that even though species composition may vary, a pasture that is dominated by one species making up to 70% of the pasture is comparable to a pasture that is relatively even. This was at sites in both Europe and Canada and results might prove to be difficult to replicate under drier South African conditions. If such a wide range of species composition does deliver all the benefits of multiple species pasture, it would make management for the producer much easier as species composition changing over time would not be a major concern.

2.4.5 Nutrient retention

Multiple species pastures will result in reduced soil nutrient losses (Sanderson et al., 2004). This may be because of the more complete use of nutrients leaving less excess available for nutrient leaching. Evenness of plant species distribution will be essential to ensure that nutrients throughout the pastures is utilised and to avoid uneven uptake of nutrients at different areas of the same pasture (Sanderson et al., 2004). Nitrogen fixation and weed suppression might also lead to management practices that use reduced levels of fertiliser and herbicides and reduce the risk of leaching.

2.4.6 Biodiversity

Monocultures have been linked with a reduction in biodiversity not only in the number of species in pastures, but also in the genetic variation in pastures as well as fauna and flora in surrounding landscapes (Hopkins and Holz, 2006). Adapting farm management strategies to only maintain high biodiversity may, however, not be financially viable (Hopkins and Holz, 2006). The loss of biodiversity may lead to the loss of multifunctionality of a pasture (Storkey et al., 2015). It is thus crucial for the

modern producer to strike a balance between what is financially viable and a healthy ecosystem. Some species may also positively contribute to one function and negatively to another further complicating the selection process (Storkey et al., 2015). For a diverse pasture to be viable, production must be comparable to that of the best performing pasture in the current crop-rotation. If production is lower, a diverse pasture may still be viable if there is improved grazing quality or better soil quality. Increased performance of plant species is related to the effective use of functional groups (Hopkins and Holz, 2006). In diverse pastures a variety of species is not as important as the utilisation of various functional groups. Multifunctionality will not only be determined by the utilisation of the various functional groups, but also by competitive dynamics of the different species in the pasture (Storkey et al., 2015). Selection of species that can coexist or promote each other are thus important (MaLaren et al., 2019)

2.4.7 Establishment

In lucerne-based multiple species pastures, the lucerne base will often be established a year prior to the oversowing of winter annual crops. These annual winter crops may be a single species or mixes of different grasses, legumes and brassicas. The oversowing of species mixes into pastures may be challenging due to seeds being different sizes. Seeding rate is also important and the incorrect balance between species may result in one species dominating the other to such an extent that very poor establishment takes place and the multifunctionality of the multiple species pastures never being utilised. Wortman et al. (2012) suggested that the seeding rates of mixtures should be proportional to that of the monoculture seeding rate. It is however possible that there is another optimal seeding rate. A fully additive seeding approach is impractical and not cost effective (Wortman et al., 2012). Thus, if a proportional seeding rates result in a pasture where one species dominates the other, one may consider to either reduce the seeding rate of the dominant species or increased the seeding rate for the dominated species. Oversown annual winter crops may have poor establishment in well-established lucerne pastures. Once oversown annual crops have emerged they may also struggle to compete against the established lucerne base if there are limited resources. Lucerne will have well developed root systems and if there is moisture stress it will outcompete young seedlings for available resources. This could in turn lead to oversown crops performing poorly due to poor initial establishment.

Production potential of oversown annual grasses may be manipulated through oversowing date (Botha et al., 2015, Badenhorst, 2011). Early oversowing dates may promote improved winter production, while oversowing from April to early May can allow for high spring production (Badenhorst, 2011). In the western regions of the southern Cape water is limited during late summer

and early autumn and farmers that plant too early will have a high risk of crop failure. Badenhorst (2011) conducted trials where lucerne was oversown with ryegrass in March and produced good winter production. The study included supplementary irrigation after oversowing and was able to compensate for low rainfall in March. For most farmers in the western regions of the southern Cape this may not be an option and it is possible that dry conditions during late summer and early autumn may be a limiting factor to boost winter production.

When long-term rainfall data is considered (Figure 2.1) for Tygerhoek Research Farm, it becomes apparent that the average rainfall in March, just over 30 mm, is similar to that of summer months. In summer months, moisture stress is a limiting factor and oversowing of annual winter crops will almost certainly result in crop failure. April has a potential for oversowing and establishing of pastures, but farmers may still run the risk of crop failure if rains are late. Oversowing in May is likely to lead to good establishment of winter annuals, but this may lead to a compromise in herbage production in winter months. Farmers must assess the situation and weigh up the risk before ultimately deciding when to oversow winter annual crops into the lucerne base. It is likely that a compromise in herbage production will outweigh the risk of crop failure.

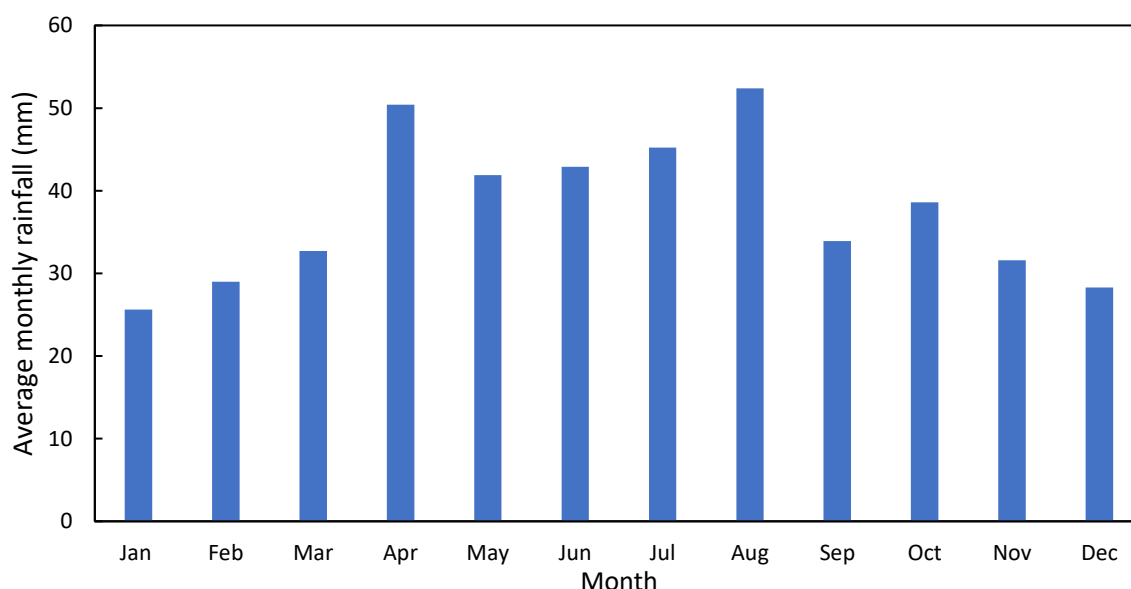


Figure 2.1 The long-term average rainfall at Tygerhoek Research Farm from 1965 to 2018

2.5 Lucerne quality and animal production

Crop-livestock integrated farming systems diversify farming activity and reduce risk for farmers (Bell et al., 2014). Long-term crop rotations integrated with pastures ensures income from livestock in years when crops underperform or market conditions are favourable. Livestock may also increase total farm production through utilisation of areas that are not suitable for crop production for various reasons.

This includes mountainous terrain, areas that may become waterlogged or too wet in winter for optimal crop production and soils that have generally performed poorly in the past.

2.5.1 Fodder flow

In long-rotation cropping systems a lucerne phase will often be included and allows for the incorporation of livestock. Lucerne is a highly productive legume, but when incorporated in grazed long-rotation cropping systems in the southern Cape, winter production remains low due to lucerne's winter dormancy. Production may also be limited in summer months due to moisture stress. In Mediterranean pastures a common trend will be that herbage production will gradually increase from the rains in late autumn to reach a maximum in late spring (Purser, 1981). Dryland lucerne will follow a similar trend as can be seen in Figure 2.2. It is worth noting that if there is sufficient rainfall during summer months herbage production may dramatically increase (Durand, 1993). It is typical for rainfed lucerne pastures to have an over-supply of forage in spring and forage shortages in winter, late summer and autumn (Bell et al., 2018).

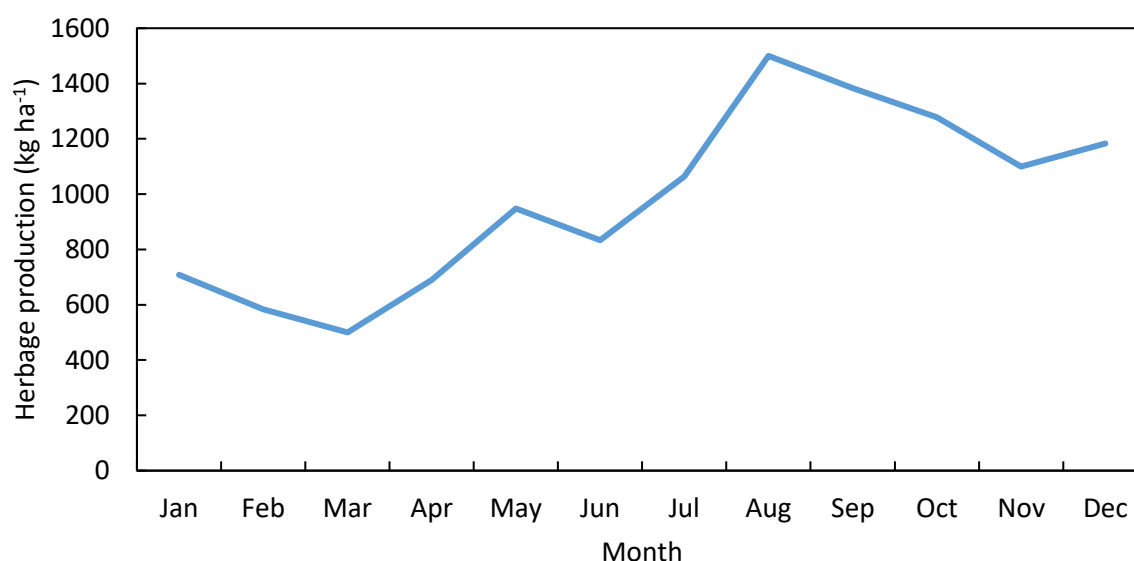


Figure 2.2 Average herbage production of lucerne in rainfed conditions at Tygerhoek Research Farm from May 1990 to March 1993. Adapted from Durand (1993)

Oversowing lucerne swards to create lucerne based multiple species pastures may present an opportunity to ensure more evenly spread forage for animal production. The lucerne production rate throughout the year can be seen in Figure 2.3. In winter when production rate is still relatively low due to cold temperatures, the complementary seasonal growth patterns of different annual winter crops may be a possible way to plug gaps in winter fodder flow programmes of farmers in this area (van der Colf et al., 2015). Lucerne will be the main herbage contributor from summer to autumn

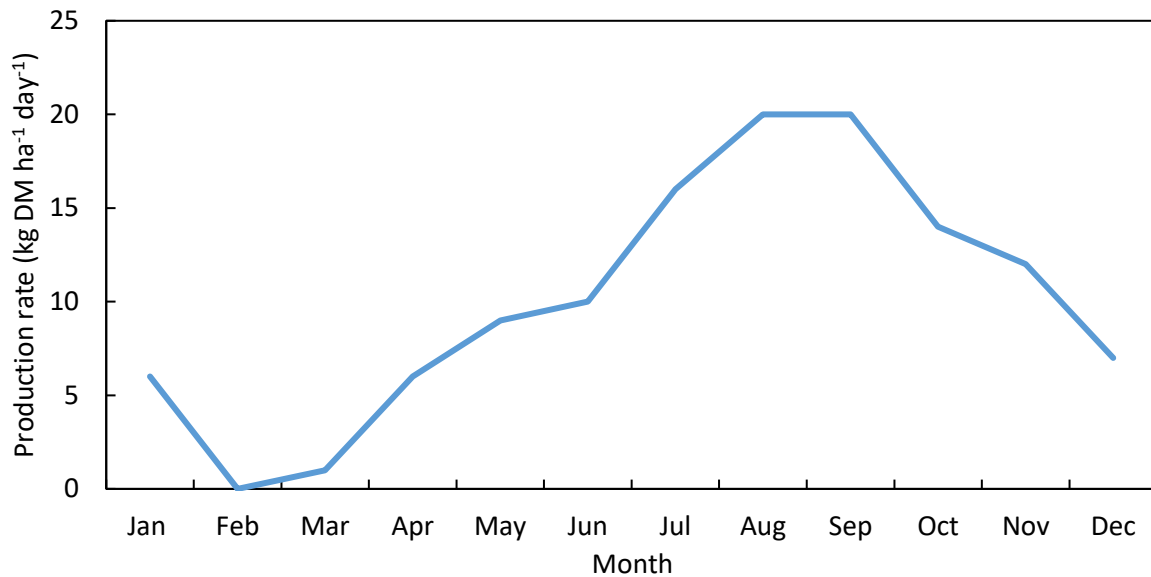


Figure 2.3 The production rate of a lucerne pasture at Tygerhoek Research Farm. Adapted from van Heerden (1976)

when oversown winter growing species are absent. Oversown species will start contributing to total herbage production from early winter and this contribution will increase to reach a maximum in spring (Purser, 1981). Grass and brassica species may increase total herbage production, while lucerne will ensure a high-quality forage. Careful consideration must be given to species interaction and how oversown winter crops may affect the lucerne fraction as well as how summer production may be influenced. When oversown species perish in summer, lucerne will once again be the main contributor to total herbage production. It is possible that the oversown species may reduce the lucerne fraction in the pastures and when oversown species die off production will be low due to reduced lucerne persistence. The production rate may also be restricted due to limited soil moisture.

2.5.2 Pasture quality

The management of pasture quality presents scope for small stock producers to improve financial viability (Lambert et al., 2000). Pasture quality varies throughout the year due to plant maturity, herbage composition, grazing management and seasonal variance. When evaluating a pasture, herbage production must not be the only criteria as pastures with a high herbage yield may still have livestock that underperform. Inadequate voluntary intake of forage is potentially one of the biggest limitations to animal production in South Africa and may be related to seasonal changes in pasture composition, forage availability and quality (Brand, 1996). The nutritive value of a pasture can be

defined as the animal response per unit intake (Purser, 1981). It is a function of herbage digestibility for either maintenance or production (Purser, 1981).

In lucerne based multiple species pastures, the lucerne base and numerous oversown species will all contribute differently in terms of forage quality and herbage production during different times of the year. In summer months, it is likely that the pasture will only consist of the lucerne base and possibly summer weeds. In autumn, after oversowing, winter and spring there will be contributions from the lucerne base and oversown grasses, legumes and brassicas. Grasses and brassicas will likely contribute to increased herbage production while legumes are more closely linked to a high forage quality.

Quality of grazing crops varies between grasses, legumes and brassicas. As plants mature their chemical composition will change and quality declines. This is attributed to an increase in fibre content which results in a decrease in both crude protein and digestibility. These changes will vary between different species. The changes in the chemical composition of legumes will not be as severe as in grasses and forage quality will remain relatively high. Fibre can be measured as acid detergent fibre (ADF) or neutral detergent fibre (NDF). Acid detergent fibre consists of cellulose and lignin and NDF consists of cellulose, hemicellulose and lignin. Fibre is not rapidly digested in the rumen and may be used to increase retention time of a feed. Too much fibre in the diet may limit animal intake and take up too much space in the rumen (Waghorn and Clark, 2004). Small stock has an upper limit to how much it can chew per day and will not be able to physically breakdown large fibrous material fast enough. This may limit animal production. Too little fibre is also negative as it may lead to nutritional diseases like acidosis. Fibre remains a key component to long-term rumen health. Scholtz et al. (2009) found a large variation in the fibre content of South African lucerne. The ADF varied from 21.3% to 47.3% with an average of 33.2%. Neutral detergent fibre had an even wider range varying from 28.9% to 65.9% with an average of 44.1%. These large variations may be due to a range of different factors that include physiological stage, grazing management or weed contamination of pastures.

The voluntary feed intake of legumes is higher compared to that of grasses (Brand, 1996). Legumes have a lower proportion of fibre compared to that of grasses, due to fewer cell wall constituents and this results in more rapid particle breakdown after maceration (Brand, 1996). There exists a narrow relationship between digestibility, voluntary intake and pasture quality. A decrease in digestibility will lead to reduced voluntary intake by livestock (Durand, 1993; Waghorn and Clark, 2004). Cell wall components are a good indicator of digestibility which is a good indicator to feed quality (Durand, 1993). This is due to cell wall components being negatively correlated to rumination rate, the breakdown of feed particles and through flow of feed (Durand, 1993). Of the cell wall components, lignin is the most limiting and may serve as a good indicator of feed quality (Durand, 1993).

In pastures there is a tendency that protein will decrease as plants mature. This is true for both legumes and grasses, but the effect is not as severe in legumes. Legumes also have a higher protein content compared to that of grasses. The low solubility of legume proteins compared to that of grasses may also be beneficial (Brand, 1996). The more rapid microbial degradation of grass proteins may lead to insufficient amino acid supply which in turn may result in reduced animal performance (Brand, 1996). Scholtz et al. (2009) determined the nutritive value of South African lucerne and found that the crude protein (CP) value ranges from 13.9% to 27.8%, with an average value of 20.7%. There is a tendency for more dormant cultivars to have a higher CP content than less dormant cultivars (Avcı et al., 2018).

Lucerne swards in an early growth stage tend to be of a higher quality than more mature swards (Testa et al., 2011). Leaves are of a higher grazing quality than stems due to protein being concentrated in leaves as well as leaves having a higher digestibility (Pecetti et al., 2001; Pembleton et al., 2010). This makes a higher leaf to stem ratio more desirable. As lucerne swards age, there is a reduced leaf to stem ratio reducing digestibility and quality of the pasture (Rogers et al., 2014). As lucerne plants mature, stems elongate and make up a larger percentage of total herbage. Leaves that are lower down on the stems may also die and fall off resulting in overall lower grazing quality (Cruywagen et al., 2011). The utilisation of young lucerne swards before they are well established may lead to reduced longevity and sward persistence. More mature swards will also have higher herbage yields than younger swards. Farmers must thus find a compromise between herbage yield, persistence and pasture quality. In the southern Cape, grazing of lucerne swards may commence roughly every six weeks without compromising sward persistence (Oberholzer et al., 1993). This is flexible and might be shorter during times of optimum growth and longer during times of moisture stress.

In lucerne based multiple species pastures, the species composition will change along with plant maturity and will impact nutritional value of the pasture (van der Colf et al., 2015). In summer months the main contributor to available herbage will be the lucerne base (Durand, 1993). Water stress may result in reduced herbage in lucerne swards, but the DM digestibility and crude protein will not be affected (Rogers et al., 2014). Moderately water stressed lucerne may also be in a younger physiological stage, resulting in a higher crude protein, due to a high leaf to stem ratio (Snaydon, 1972). If leaves have died and fallen off, leaving mostly fibrous stems, the grazing quality will be reduced. Waghorn and Clark (2004) reported that grazing quality may be good in spring when there is a high leaf to stem ratio, but that in dry summer months leaves may be converted to stems and seed formation can occur resulting in reduced herbage quality. As lucerne based multiple species pastures get older, there may also be a decrease in pasture quality. This is due to a reduced lucerne fraction

that may allow more weeds to infiltrate the pasture and reduce overall forage quality (Coruh and Tan, 2008). The oversowing of high-quality annual crops into maturing lucerne swards may be a viable option to improve pasture quality and provide livestock with a larger variety of species to graze.

Careful management is required to ensure that a suitable stocking rate is applied to pastures. A stocking rate that is too high results in competition among livestock that can lead to inadequate forage intake, reduced quality of intake and result in sub-optimal animal performance. In lucerne based pastures, rotational grazing should be applied to ensure long-term sustainability of pastures (Oberholzer et al., 1993, Oberholzer and van Heerden, 1997). If pastures are overstocked, excessive defoliation and selection by animals may result in a reduction of productive and palatable species and reduce pasture quality, resulting in reduced sustainability (Brand, 1996). A stocking rate that is too low will not be financially viable to the farmer. Livestock will also be able to selectively graze for palatable crops, decreasing their frequency and over time pasture quality will decrease as less favourable crops increase (Waghorn and Clark, 2004).

During various stages of the year, different aspects of a pasture may limit livestock performance. The lucerne base will provide forage throughout the year, although the quantity will vary depending predominantly on moisture stress in rainfed systems (van Heerden, 1976). Oversown species will start to contribute to the pasture as soon as seedlings germinate after autumn rains and contribute maximally during late spring. During winter months, there will be green herbage available in a young physiological stage and of a high grazing quality (Purser, 1981). The quantity of available herbage may however be limiting and restrict livestock performance. During spring, pastures will generally consist of a high-quality non-limiting herbage if appropriate stocking rates are applied (Purser, 1981)). In late spring and early summer, the feeding value of oversown species may rapidly decline as plants start to wilt. Direct measurements are not available, but during wilting, digestibility may be reduced by as much as 0.5% per day (Purser, 1981) Lucerne may still provide some form of high-quality herbage. This is likely to result in a forage that is quantitatively non-limiting, but a reduced quality of herbage may limit livestock production (Purser, 1981). During autumn, lucerne may once again provide green herbage of a good quality if there is enough moisture. There will however be no contribution from annual crops from the previous growing season and it is likely that both quantity and possibly quality of available herbage will limit livestock production (Purser, 1981)

2.5.3 Animal performance

Rumen microbes are required for the breakdown of consumed forages to volatile fatty acids which serve as the main energy source for ruminants (Brand, 1996). Nutritional requirements of livestock

vary depending on age, sex and reproductive stage. Crude protein levels must exceed 10% for animal maintenance, but a value closer to 19% is suitable for high producing livestock such as dairy cows (Waghorn and Clark, 2004). If there is too much protein in the diet, it may lead to energy losses and sub-optimal performance of animals due to the high cost of protein degradation. Lucerne is well suited as a forage for high producing animals due to its high protein content.

Due to the fluctuations in both available herbage and herbage quality, it can be expected that animal performance will also change along with the seasons. Sheep can be expected to gain around 80 to 120 grams in body weight per day during winter (Purser, 1981). This increases to 200 to 300 grams per day during spring. With summer comes reduced quality of herbage and weight gains may be reduced to 40 grams per day in early summer (Purser, 1981). In late summer and early autumn, when herbage quality and quantity is restricted, animals will use body reserves and this will result in small, but continued weight loss (Purser, 1981). Wool production will follow a similar trend. Production will rise from 8 to 14 grams per day to roughly 16 to 20 grams per day with the onset of winter and increased availability of herbage and improved herbage quality (Purser, 1981). In late spring and summer when quality of herbage deteriorates, wool production will decline to 8 grams per day per sheep.

Pastures will generally not allow livestock to perform to their full genetic potential. Livestock performance may be limited by energy intake as fibre may slow digestion and result in lower voluntary intake (Waghorn and Clark, 2004). Pure lucerne swards may have sufficient crude protein and digestible organic matter during most of the year, but levels may be insufficient during times when ewes are under severe stress, such as lactation (Brand, 1996) Supplementation may then be used to enhance livestock performance (Brand, 1996). For supplementary feeding to be economically viable, supplementation must not replace natural grazing of animal (Purser, 1981). Farmers may benefit the most from supplementation that result in increased animal intake and promote the efficient utilisation of digested nutrients (Purser, 1981). During times when forage and forage quality may be limiting, like autumn, focus should be on animal maintenance rather than supplementation for increased production (Purser, 1981).

2.6 Effects on soil quality

In recent decades there has been an adoption of one or more of the principles of conservation agriculture (CA) in the southern Cape. The principles of CA include 1) reduced tillage or no-till systems, where not more than 25% of the soil surface is disturbed, 2) the retention of crop residue, the amount of residue retained varies between cropping systems and 3) making use of crop rotations that may include cover crops (Palm et al., 2014; Verhulst et al., 2010). Other management practices that may

also be incorporated to improve soil quality include controlled trafficking to reduce soil compaction and yield loss as well as the incorporation of livestock into farming systems. Lucerne plays a critical role in long-rotation cropping systems in the southern Cape. The perennial nature of lucerne ensures that there will be no fallow period and no-tillage for the duration of the lucerne phase. The lucerne phase will also promote the incorporation of livestock in the system and reduce traffic by heavy machinery. This may influence soil physical, chemical and biological properties over time. The lucerne phase may however also be seen as a short-term monoculture within a long-term crop rotation system.

2.6.1 Physical soil properties

Physical characteristics of soil include soil structure and aggregation, porosity, hydraulic conductivity, water holding capacity, soil water balance and the level of soil erosion. Physical degradation of soils may be caused by removing soil cover, burning of organic materials, tillage, overgrazing and compaction by machine traffic (Palm et al., 2007). Physical soil properties may have a significant effect on lucerne yield even if chemical and biological properties are identical (Miretti et al., 2010). The inclusion of lucerne in long-rotation cropping systems inevitably leads to the inclusion of livestock in farming practices and the impact that livestock will have of soil properties must be considered.

2.6.1.1 Soil Structure and Aggregation

Soil structure and aggregation is the relationship between shape, size as arrangement of solids and voids in the soil, the continuity of these pores and their capacity to transmit both organic and inorganic substances that may include fluids and gasses (Verhulst et al., 2010). The stability of soil structure is the ability of its aggregates to remain stable under various stresses (Verhulst et al., 2010). Aggregate stability of soil is closely linked to the soil organic carbon content. Reduced input of organic matter resulting in reduced soil organic carbon will lead to reduced aggregate stability of soils (Blair and Crocker, 2000). In lucerne-based multiple species pastures, continuous soil cover with the addition of residues from annual winter crops will promote improved soil structure and aggregation due to various factors. The variety of different root systems, quality and quantity of organic matter returned to the soil from lucerne-based multiple species pastures will positively contribute to the formation of stable soil aggregates (Verhulst et al., 2010). Lucerne based multiple species pastures also protect the soil against the impact of rain and wind (Bell et al., 2011). The absence of tillage in the pasture phase will improve soil structure with larger soil aggregates due to tillage physically breaking down soil structure and the interruption of the formation of stable aggregates (Verhulst et al., 2010). Soil

degradation invariably starts with the removal of vegetation as it exposes the soil surface to the impact of rain drops, wind erosion and larger fluctuation of soil temperatures (Palm et al., 2007). Stocking densities that are too high, too high a grazing frequency and continuous grazing can all result in the removal of excessive amounts of herbage that may leave soil bare. This will result in the breakdown of soil structure and aggregation, which in turn will lead to reduced potential water infiltration (Palm et al., 2007). Grazing management is thus an important factor as poor grazing management will lead to reduced soil quality.

2.6.1.2 Soil Porosity

Soil pores can vary in shape and size and has an impact on soil water infiltration, drainage, gas exchange and root penetration (Verhulst et al., 2010). In lucerne swards the lack of tillage, reduced traffic by heavy machinery, animal component and continuous cover may all impact soil porosity. Pastures, with the exclusion of ryegrass, will improve macroporosity of soil (Chan et al., 2001). Porosity may be negatively influenced by tillage, overgrazing and traffic by machinery (Palm et al., 2007). The impact of no-till practices has had varying results according to literature. Studies have linked the introduction of no-till techniques to increased bulk density and reduced porosity, but generally this impact is restricted to the plough layer (Palm et al., 2014). An increase in bulk density and reduced porosity in pastures does not negatively impact water infiltration if sufficient soil cover is maintained (Palm et al., 2014). This may be due to an increase of biological activity and increased organic matter associated with pasture systems as roots penetrate the plough layer and biological activity increases (Chan et al., 2001). The deeper layers of soil will possibly have a lower bulk density due to reduced compaction from heavy machinery.

There is a concern that treading by livestock will increase soil strength and bulk density leading to reduced macroporosity and water infiltration (Bell et al., 2011). Stocking density, soil moisture, degree of soil cover and livestock species are all variables that will influence the extent of soil compaction. Appropriate stocking densities are essential as overgrazing may result in increased erosion and reduced soil cover. Cattle are more likely to result in soil compaction due to a larger hoof pressure per unit surface area when compared to small stock like sheep that are typically found in the southern Cape. The negative implications are however short lived and yield penalties may vary between no effect and less than 10%. This may be attributed to compaction being relatively shallow (10 cm to 15 cm), a low degree of compaction and compaction being rectified by the natural drying and wetting of soils (Bell et al., 2011). Soils that are wet or have a clay texture will be more susceptible to compaction and careful management is required to avoid soil damage that may reduce root penetration (Avondo

et al., 2013). Verhulst et al. (2010) concluded that the impact on soil porosity would vary depending on soil quality, traffic and input of soil organic matter.

2.6.1.3 Soil water balance

Soil water balance refers to water infiltration, runoff, evaporation and plant available water (Verhulst et al., 2010). Perennial pastures prevent the breakdown of aggregates, crust formation, provide permanent soil cover and requires less tillage. Deep rooted plants, like lucerne, allow water to infiltrate through old root channels. Water infiltration is likely to increase under pasture systems due to an increase in labile carbon that results in reduced surface sealing (Blair and Crocker, 2000). In lucerne based multiple species pastures the likely increase of residue from oversown winter crops may result in less exposure to heat and drying out of the soil surface (Palm et al., 2014; Verhulst et al., 2010). The threshold residue level is not known (Jun et al., 2014; Palm et al., 2014). The likely increase of residue will increase the time over which water may be absorbed leading to an increase water infiltration, reduce runoff and increase water available to plants (Palm et al., 2007; Verhulst et al., 2010). This provides farmers with a buffer against droughts and may result in more stable yields during years of poor rainfall. The omission of conventional tillage from pastures may further reduce water loss through evaporation due to less exposure to wind and heat.

2.6.1.4 Soil erosion

Soil erosion impacts various aspects of the soil including soil depth, soil nutrients, biota, organic matter and reduces soil productivity (Palm et al., 2007). Perennial lucerne pastures may result in reduced erosion when compared to annual pastures as there will not be a fallow period where soil is left bare and exposed to the elements. The benefits are both above and below the soil surface. On the surface, plant cover protects the soil from the direct impact of rain and wind ensuring that topsoil does not blow or wash away (Palm et al., 2014). Beneath the soil surface roots bind the soil, promoting soil structure and adding organic matter. The likely increase in organic matter returned to the soil also promotes the formation of stable aggregates. The lack of tillage on perennial pastures ensures that aggregates are not broken down, leading to a reduced risk of wind and water erosion (Palm et al., 2014).

2.6.2 Chemical soil properties

Soil chemical and physical properties are closely linked and changes in physical characteristics will often impact soil chemical properties. Soil chemical properties included soil organic carbon content and nutrient availability.

2.6.2.1 Soil organic matter

Soil organic matter can be described as the balance between the addition of organic matter and the decomposition thereof (Palm et al., 2007). Soil organic matter is an important component in a healthy soil as it supports biotic life and provides protection from both wind and water erosion (Avondo et al., 2013). Soil organic carbon is a component of soil organic matter and roughly 45 – 60% of soil organic matter consist of soil organic carbon (Lal, 2016). The replacement of natural vegetation with cropping systems and conventional tillage practices has resulted in a reduction of soil organic carbon and has negatively impacted soil quality (Blair and Crocker, 2000). Total soil carbon can be derived into three main functional pools. These are free particulate organic matter, occluded particulate organic matter and a heavy mineral bound fraction (Smith, 2014). For this study, the focus is free particulate organic matter, also known as labile carbon or active carbon. Active carbon is the carbon fraction that is available to microorganisms to be broken down and used as energy. It may also be used as an early indicator of soil organic carbon as this fraction will respond the rapidly to changes in soil management (Smith, 2014). Increased levels of active carbon in the short term will be translated to increased levels of soil organic matter in the long term due to increased inputs of above or below ground biomass (Haynes, 2005).

Soil organic carbon contributes directly to soil fertility through the release of inorganic nutrients and trace elements as it decomposes and indirectly increases cation exchange capacity, water holding capacity and improves soil structure (Smith, 2014). The same basic principles that will allow for the increase of soil organic carbon in conservation agriculture may be applicable in lucerne based multiple species pastures. Crop rotation, residue retention and good grazing management allow for the build-up of soil organic carbon while tillage, overgrazing and burning of residue will deplete soil organic carbon levels. Organic matter must be returned to the soil otherwise soil organic carbon will decrease over time. Soil organic carbon is crucial to ensure optimum ecosystem functionality (Lal, 2016) and is an indicator of soil quality. A minimum soil organic carbon content of 1 – 1.5% is needed to reduce the risk of soil degradation (Lal, 2015). A Soil organic carbon level of roughly 1.5 – 2% will ensure good soil structure and aggregation, good water holding capacity, nutrient retention, reduced erosion and increased biological activity (Lal, 2016; Peyraud et al., 2014). Soil organic carbon accumulates over

time and an increase in soil organic carbon levels will not be apparent immediately (Finney and Kaye, 2017). In semi-arid areas with rainfall of less than 500 mm per annum, the effects of increased soil organic carbon will take longer to be observed when compared to higher rainfall areas (Blanco-Canqui et al., 2015). This can be linked to climatic conditions, low yields, fallow periods and overgrazing (Smith, 2014).

Long-rotation cropping systems may increase soil carbon levels through increased input carbon due to higher biomass production attributed to breaking disease cycles and diversified root biomass input in both pattern and depth (Palm et al., 2014). The incorporation of perennial pasture phases will allow for the higher build-up of soil organic carbon when compared to both annual pastures and cereal monocultures (Chan et al., 2001). Legume based perennial pastures may further result in increased levels of soil organic carbon due to the frequent return of high-quality residue to the soil without fallow periods as fallow periods are known to reduce soil organic carbon (Blair and Crocker, 2000).

The C:N ratio of crop residue returned to the soil will have a significant impact on decomposition and nutrient cycling (United States Department of Agriculture, 2011). Soil microorganisms require a diet with a C:N ratio of 24:1 for maintenance and energy. A low C:N ratio will be broken down faster than a high C:N ratio. If residue with a lower C:N ratio than 24:1 are returned to the soil, microbes would rapidly break it down. Carbon is the soil microbes' source of energy and when carbon is depleted, there will be excess N in the soil that will be available to plants. If organic material with a higher C:N ratio than 24:1 is returned to the soil, soil microbes must find additional N to breakdown residue. Microbes will take up N in the soil and this may lead to N deficiencies for plants (United States Department of Agriculture, 2011). Typical grains that are cultivated in the southern Cape like wheat and oats will have C:N ratios of 80:1 and 70:1 respectively. Lucerne has a C:N range from 13:1 to 25:1 depending on the physiological stage. Annual legume hay will have a C:N ratio of around 17:1 and vetch may have a ratio as low as 11:1. A balance must be found between residue that is returned to soil to ensure that breakdown is not too rapid as residue is required to protect soil from erosion, conserve soil moisture and provide habitat for soil microbes (United States Department of Agriculture, 2011). At the same time these residues must be broken down to release nutrients and build organic matter (United States Department of Agriculture, 2011). This may be overcome by returning both material with a high C:N ratio and a low C:N ratio to the soil.

Oversowing of annual winter crops into lucerne based perennial pasture may have similar advantages to that of cover crops. The root biomass production of lucerne may potentially be low as new roots are not formed and broken down annually (Smith, 2014). Increased biomass and root production from winter annuals will lead to increased levels of soil organic matter and crop residues

(Blanco-Canqui et al., 2015). The retention of crop residue is a crucial component to maintain or increase soil organic carbon content (Palm et al., 2014). Good grazing management is required to ensure enough residues is left on pastures. Overgrazing will negatively impact both pasture crops and soil quality. Reduced soil cover due to overgrazing and trampling will expose soil and increase risk for wind and water erosion. The return of nutrients to the soil through faeces and urine will result in a positive effect on soil organic carbon. If there is sufficient moisture available, faeces will be reincorporated back into the soil (Avondo et al., 2013).

The degradation of soil organic carbon levels will have multiple negative effects on soil quality and include aggregate breakdown, increased risk of soil erosion, low quality soils, poor yields and reduced profit (Blair and Crocker, 2000). It is possible to determine that certain activities are likely to increase soil organic carbon content, but the complex interactions of residues with the soil, the types of residues and the biological activity of the soil will make it difficult to determine the exact effect that any given practice may have on soil properties (Palm et al., 2014).

2.6.2.2 Nutrient availability

Sufficient nutrients are required to ensure that plants grow optimally and production potential of a pasture is reached. Nutrient depletion or the loss of soil fertility is one of the most common forms of soil chemical degradation (Palm et al., 2007). Reduced productivity as a result of nutrient depletion results in reduced biomass production which in turn leads to reduced input of soil organic matter (Palm et al., 2007). Grazing of a pasture may be advantageous when compared to cutting and haymaking as grazing allows for the return of most nutrients through faeces and urine. If nutrients are continuously removed without being replaced, it will result in nutrient depletion. This in turn has a negative impact on various other aspects such as soil biota. The opposite is also true if too much fertiliser is applied as this may result in nutrient leaching that may pollute water resources, nutrient imbalances or result in toxicity for plants. Nutrient depletion is more likely in developing countries and eutrophication more likely in developed countries (Palm et al., 2007). It is thus important to establish what is removed from soil or what is needed to maintain an appropriate nutrient balance and only supplement according to plant specific requirements.

2.6.3 Biological soil properties

Soil biota refers to soil microbes, nematodes, insects, earthworms and other living organisms in the soil. Each of these components of soil biota play a different role and contribute to a healthy soil. Soil health refers the state of the soil as a living entity and how it may affect plant health (Lal, 2016). Soil

microbes mainly consist of fungi and bacteria (Palm et al., 2014). Soil microbial biomass is an indicator of soil quality and it plays an important role in decomposition, nutrient cycling rates, contributes to soil aggregation and serves as an indicator to the soil water holding capacity (Palm et al., 2014, 2007; Verhulst et al., 2010). Soil microbes also play a major role in the breakdown of contaminants and may influence soil quality (Habig et al., 2015). Soil microbial biomass is dynamic and responds rapidly to changes in management (Verhulst et al., 2010). Larger organisms like earthworms also contribute positively to soil by maintaining both soil structure and nutrient cycling and performing other tasks such as creating soil aggregates and macropores (Palm et al., 2014).

Soil biota requires organic matter to thrive and biological degradation of soil is closely linked with reduced inputs of organic matter (Palm et al., 2007). Soil organic matter may be made up of several components that include animal and plant residues, microbial biomass, active or labile carbon and stable soil organic matter (Lal, 2016). Soil organic matter serves as soil biota's source of energy and carbon and is crucial for the biological functions of soil (Palm et al., 2007). Soil microbes decompose soil organic matter into nutrients that may then be taken up by plants (Habig et al., 2015). Farming techniques that promote the build-up of soil organic matter is thus likely to have a positive impact on soil biological factors and soil biota. The implementation of conservation agriculture increases biodiversity of soil biota (Palm et al., 2014) and the further incorporation of a pasture phase will result in an increase in soil fauna (Peyraud et al., 2014).

In lucerne based multiple species pastures there will be numerous benefits concerning soil microbes. Lucerne will ensure that there is always soil cover and that there are living roots in the soil throughout the year. The addition of winter annual crops oversown into a lucerne pasture ensures that there will also be root diversity. This root diversity in soil will promote soil microbe activity and can lead to synergistic relationships between microbes and plants (Habig et al., 2015). Continuous cover from perennial pastures and animal waste will promote microbial diversity in soils (Peyraud et al., 2014). This may further be enhanced by the addition of annual winter crops in lucerne based pastures. Increased build up of soil organic matter will lead to increased soil microbial activity (Habig et al., 2015). The quality and quantity of residue will however influence the degree of this increase in microbial activity (Habig et al., 2015). Healthy diverse microbial communities provide better resistance towards pests and diseases and improved ecosystem functions (Lal, 2016; Palm et al., 2014). Decreased soil disturbance due to no or reduced tillage in permanent pastures will promote soil biota as conventional tillage may physically kill organisms that would naturally till the soil like earthworms (Habig et al., 2015). Tillage results in the breakdown of soil structure, reducing porosity and infiltration of soils which leads to reduced soil biota activity (Palm et al., 2007). Other conventional farming techniques such as

burning of residues and the use of pesticides will further reduce life in soils. Burning of residues negatively impact soil biota as it destroys soil organic matter and soil organic carbon. A soil with high microbial diversity that is not tilled may be an effective biological way to control pests (Habig et al., 2015).

2.7 Summary

The literature provides evidence that multiple species pastures have the potential to improve long-rotation cropping systems in the southern Cape. Increased production, weed suppression, environmental stability, better nutrient retention, improved soil quality, N fixation, increased forage quality and reduced fluctuation in feed availability are possible outcomes of oversowing lucerne with annual winter crops. This prediction is based on agronomic principles, but research on lucerne based multiple species pastures in South African conditions remains rare. Accurate predictions on pasture performance cannot be made and more research is required to understand the complex and dynamic interactions between mixed species in pastures, soil and livestock.

2.8 References

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Chapter 3

The potential of lucerne-based multiple species pastures to increase herbage production in winter months and reduce fodder flow fluctuations in the southern Cape of South Africa

3.1 Introduction

The Mediterranean climate of the southern Cape is conducive to the cultivation of cool-season crops, including wheat (*Triticum aestivum*), barley (*Hordeum vulgare*), canola (*Brassica napus*) and lupins (*Lupinus spp.*) in crop rotation systems (Smith, 2014). These crop rotation systems have also allowed for the incorporation of lucerne (*Medicago sativa* L.) in long-rotation cropping systems, since about 25 to 35% of rainfall occurs during summer. Lucerne allows for the integration of livestock into the farming system. A typical lucerne phase is five to seven years and is followed by five to seven years of cash crop production. Lucerne has numerous advantages for farmers that include N fixation (Bouton, 2012a; Carlsson and Huss-Danell, 2003; Raza et al., 2013), high-quality animal feed (Badenhorst, 2011), utilisation of summer rainfall (van Heerden and Tainton, 1987), ensures that there are always living roots in the soil and may provide more soil cover than conventional farming practices. Sporadic summer rainfall has however resulted in summer herbage yields often being poor. In winter months, herbage production may also be limited due to lucerne's inherent winter dormancy. Warmer temperatures and sufficient rainfall results in lucerne production reaching a maximum in spring (van Heerden, 1976), while autumn production may be either high or low depending on the rainfall of any given year. Considerable fluctuations of available fodder make management and planning of fodder flow programmes challenging for farmers and some have investigated excluding lucerne from their crop rotation systems.

The oversowing of lucerne swards to create a lucerne-based multiple species pasture may present farmers with an opportunity to increase total herbage production and ensure improved distribution of available fodder for animals. The complimentary growth patterns of lucerne, winter grasses, small grains, brassicas, clovers and other legumes may help farmers overcome forage shortages in winter months (van der Colf et al., 2015). If additional herbage is produced, lucerne-based multiple species pastures may also suppress weeds (Deak et al., 2007; Sanderson et al., 2004, 2005).

Oversown winter annual species may serve a dual purpose as residue of oversown species may be utilised as additional fodder for animals and as a cover crop that covers the bare soil between individual lucerne plants. The additional soil cover and forage for animals may also benefit farmers in

the hot summer months. Additional cover may help to conserve moisture if there is a rainfall event and enable the lucerne base to utilise soil moisture over a longer period of time. Animals may graze the stubble if lucerne plants are small or wilted. It is however also possible that oversown annual species will have a negative impact on the lucerne base. Oversown annual species may deplete available soil moisture in spring and subsequently reduce lucerne production in summer and autumn. If oversown annual species compete vigorously during their growing season, it can also lead to a reduced lucerne stand density. This results in a situation where the oversown annual species replaces the lucerne base as opposed to increasing herbage production through complementing it (van der Colf et al., 2015). The reduced stand density or plant population of the lucerne base will not recover due to lucerne's poor reseeding ability as a result of its autotoxicity (Chon et al., 2006). This may then result in lower herbage production by the lucerne base in seasons when oversown annual species do not contribute toward herbage production.

The aim of this chapter was to establish whether lucerne-based multiple species pastures produced more herbage than dryland lucerne swards in the southern Cape. Two objectives were used to determine if lucerne-based multiple species pasture are more sustainable than lucerne monocultures:

- 1) The first objective was to ensure the same or higher yield when compared to a pure dryland lucerne sward and that oversown species contribute to total herbage production. If herbage production was not increased, it may be compensated for if there was an increase in soil or fodder quality to ensure economic viability.
- 2) The second objective was to establish whether there was a similar or improved herbage production pattern with less fluctuation compared to pure dryland lucerne swards.

3.2 Materials and methods

3.2.1 Site description

This study was conducted from April 2018 to August 2019 on Tygerhoek Research Farm (34°09'34" S, 19°54'53 E; elevation 168 m) near Riviersonderend in the southern Cape, South Africa. Tygerhoek has a Mediterranean climate with a mean annual rainfall of 453 mm (Table 3.1). Average temperatures (Table 3.2) in summer range from 14.4 – 29.6°C and in winter from 5.2 – 18.6°C.

Table 3.1 Monthly rainfall (mm) for the duration of the trial and long-term average rainfall (mm) for the trial site

Rainfall (mm)	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
2018	22.9	14.7	33.3	18.1	21.8	37.6	52.6	42.4	72.4	14.8	37.9	28.6
2019	17.1	33.6	298.7	16.4	15.9	49.2	54.6	8.2	-	-	-	-
Long-term	25.6	29.0	32.7	50.4	41.9	42.9	45.2	52.4	33.9	38.6	31.6	28.3

- Indicates that data falls outside of the study period

Table 3.2 Long-term average monthly minimum and maximum temperatures (°C) at the site of this study

Temperature (°C)	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Maximum	29.1	29.6	28.7	24.8	21.6	18.6	17.9	18.4	21.4	24.4	26.4	27.9
Minimum	15.9	16.4	15.1	12.3	9.4	6.1	5.2	5.7	7.6	10.2	12.4	14.4

For the purpose of this study, summer refers to December to February, autumn is from March to May, winter is from June to August and spring from September to November. The trial site consisted of shallow shale-derived soils with a loamy texture and contained a high concentration of coarse fragments (Smith, 2014). The soil form was Glenrosa (Orthic A – Lithocutanic B) (Soil Classification Working Group, 1991).

3.2.2 Experimental layout and treatments

The experiment was arranged as a split-plot design and replicated in four blocks. Whole plots covered an area of 2.5 x 48 m and sub-plots covered an area of 2.5 x 24 m. Whole plots (factor one) were planted with a cover crop, black oats (*Avena strigosa*), at a seeding rate of 40 kg ha⁻¹, or left fallow in 2016, the year prior to the establishment of the lucerne base. The cover crop was terminated at the end of the growing season of 2016. The lucerne cultivar used for this trial was L70 with a dormancy class of 7. The cultivar L70 was used as it is widely cultivated in the region and results of this trial are applicable to farmers in the area. Lucerne was planted in 2017 at a seeding rate of 12 kg ha⁻¹. Lucerne was grazed by sheep to approximately 50 mm above ground prior to oversowing.

Sub-plots (factor two) were oversown with 12 species or mixes into the two-year-old lucerne base in 2018. Oversown species (Table 3.3) were planted annually with a Kuhn NEO 13 no till disc planter by Rovic and Leers in early May in 2018 and late April in 2019. Planting dates were dependent on the availability of the planter and weather conditions. A thirteenth oversown combination was added to

Table 3.3 Species that were oversown into the lucerne base, treatment name, common name of oversown species, scientific name, cultivar and planting density

Treatment name	Oversown species	Scientific name	Cultivar	Plant density (kg ha ⁻¹)
Control	Control	<i>Medicago sativa</i> L.	-	-
Black oats	Black oats	<i>Avena strigosa</i> L.	Saia	40
Forage barley	Forage barley	<i>Hordeum vulgare</i>	Moby	60
Stooling rye	Stooling rye	<i>Secale cereale</i> L.	Barpower	25
Westerwolds ryegrass	Westerwolds ryegrass	<i>Lolium multiflorum</i> var. <i>westerwoldicum</i>	Maximas	20
Forage radish	Forage radish	<i>Raphanus sativus</i>	Tajuna	5
Canola	Canola	<i>Brassica napus</i>	Hyola 577CL	2
Clover-medic mix	Medic	<i>Medicago</i> spp.	Jester	2.5
	Medic	<i>Medicago</i> spp.	Paraggio	2.5
	Balansa Clover	<i>Trifolium michelianum</i>	Paradana	2.5
	Subterranean Clover	<i>Trifolium subterraneum</i>	Dalkeith	2.5
Ryegrass-clover mix	Hybrid ryegrass	<i>Lolium x boucheanum</i>	Shogun	20
	Red clover	<i>Trifolium pratense</i>	Barduro	6
<i>Trifolium</i> mix	Arrowleaf clover	<i>Trifolium vesiculsum</i>	Zulu II	2
	Berseem clover	<i>Trifolium alexandrinum</i>	Elite II	3
	Crimson clover	<i>Trifolium incarnatum</i> L.	Barduro	2
Diverse mix 1	Black oats	<i>Avena strigosa</i> L.	Saia	15
	Forage radish	<i>Raphanus sativus</i> L.	Tajuna	3
	Vetch	<i>Vicia villosa</i>	Haymaker	10
Diverse mix 2	Forage barley	<i>Hordeum vulgare</i>	Moby	20
	Italian ryegrass	<i>Lolium multiflorum</i> var. <i>italicum</i>	Tabu	4
	Persian clover	<i>Trifolium resupinatum</i>	Shaftal	4

factor two in 2019. A pure lucerne sward, similar to the control, was added, but was not sprayed for weeds. This was done to quantify what the effect of weeds if best practice weed management was not applied. Poor rainfall after establishment of oversown species (factor two) in 2019 led to a second planting in June 2019. After the first grazing event in late July, oversown species did not recover and all treatments consisted of mainly the lucerne base and a weed fraction. The decision was made not to continue for the remainder of 2019. Due to early termination of the trial, data was not included as it was only taken over a single herbage collection.

Supplementary irrigation was applied from June onward in attempt to mimic normal rainfall patterns, as it was very dry from planting onward. Approximately 30 mm was applied three times over the course of the winter months resulting in 90 mm total irrigation.

3.2.3 Grazing management

The pasture was managed according to guidelines for the lucerne base and ideal grazing would be at roughly 10% of flowering regardless of dormancy class (Oberholzer et al., 1993). Plots were mob grazed by sheep for effective defoliation and sheep were removed before regrowth commenced. The stocking rate varied slightly, but typically, a group of either 76 rams or 78 ewes were used to graze the trial. Grazing management was aligned with herbage sampling. Sheep grazed each replicate individually for one to two days, depending on how many animals were available and the amount of herbage produced. Sheep were removed from the pasture at night and reintroduced to continue grazing the following morning if needed. Once the pasture reached a height of approximately 50 mm, sheep were removed. Grazing frequency varied from 42 to 77 days. The variation was due to slow regrowth in times of moisture stress in late summer and early autumn.

3.2.4 Herbicide application

Herbicide application was done in line with best local practice. In early August of 2018 the lucerne control treatment was sprayed with Clethodim for grass weeds and with Flumetsulam for broadleaf weeds.

3.2.5 Herbage production

A composite aboveground biomass sample was collected from each plot by randomly placing six 0.25 m² quadrants per plot. During times when there was a pure lucerne sward (January to April), four sub-samples instead of six were cut to comprise a composite sample. The composite sample was mixed

thoroughly and a representative grab sample of roughly 300 g was then taken, dried at 60°C for 72 hours and the total herbage production calculated using equation 3.1.

$$\text{Herbage yield} = \frac{\text{BW} \times \text{DM}}{10 \times \text{A}} \quad (1)$$

where herbage yield was measure as kg ha⁻¹, BW was bulk weight in g, DM was dry matter content (%) and A was area in m².

3.2.6 Botanical composition

A representative grab sample of roughly 300 g was taken from the same composite sample used to determine herbage production and fractionated into four groups, i.e. lucerne, oversown, weed and detritus fractions. The lucerne fraction consisted of the pure lucerne base. The weed fraction included all plants that were harvested that was not lucerne or oversown into the sward. The oversown fraction was only the species that were oversown into that specific treatment. There could not be distinguished between oversown and volunteer ryegrass and in treatments where ryegrass was oversown, all ryegrass harvested was considered to be part of the oversown fraction. The detritus fraction included all dead material, but mainly consisted of old lucerne stems. Fractions were placed into separate paper bags and dried at 60°C for 72 hours to determine species contribution on a dry matter basis.

3.2.7 Statistical Analyses

The data was subjected to an analysis of variance (ANOVA) using General Linear Models Procedure (PROC GLM) of SAS software (Version 9.4: SAS Institute Inc., Cary USA). The fixed effects were specified as system, species and the interaction between system and species. The block effect were the random effects. Shapiro and Wilk test confirmed normality of the standardised residuals (Shapiro and Wilk, 1965). Fisher's least significant difference (LSD) was calculated at the 5% level to compare treatment means (Ott and Longnecker, 2010).

3.3 Results

There was no interaction ($p < 0.05$) between the main factors for oversown treatments or the lucerne fraction (Table 3.4), and therefore the main effect is discussed. There was however interaction for the weed ($p < 0.001$) and detritus ($p < 0.05$) fractions in July of year one of the study. This may indicate that the effect of the cover crop from two years prior to the study may have had an effect on the trial up to this point.

Table 3.4 Statistical significance of the effects of the system applied (S1), species (S2) and system-species (S1xS2) interaction for the course of this study. Statistical significance was set at ($p < 0.05$) and is highlighted in bold. ND indicates no data due to ANOVA that was not conducted as values were either zero (oversown, detritus and weed fraction) or 100% (lucerne fraction) as the pasture composition changed over time

		Production		Lucerne fraction		Oversown fraction		Detritus fraction		Weed fraction	
		F-statistic	P-value	F-statistic	P-value	F-statistic	P-value	F-statistic	P-value	F-statistic	P-value
Jul' 18	System (S1)	8.44	0.005	12.07	0.009	0.15	0.703	15.29	<0.001	18.17	<0.001
	Species (S2)	1.48	0.162	2.30	0.019	21.50	<0.001	0.95	0.497	12.61	<0.001
	S1 x S2	0.82	0.623	1.42	0.187	1.20	0.309	2.04	0.039	7.54	<0.001
Aug' 18	System (S1)	7.87	0.007	1.74	0.191	6.39	0.014	9.42	0.003	0.06	0.808
	Species (S2)	0.80	0.644	4.10	<0.001	55.94	<0.001	2.93	0.004	5.93	<0.001
	S1 x S2	0.71	0.729	0.69	0.744	1.32	0.238	0.74	0.701	0.56	0.853
Oct' 18	System (S1)	0.60	0.441	0.30	0.585	0.01	0.915	0.13	0.717	0.46	0.498
	Species (S2)	1.89	0.057	7.05	<0.001	45.48	<0.001	2.10	0.033	17.65	<0.001
	S1 x S2	0.82	0.619	0.64	0.789	1.00	0.455	0.36	0.968	0.84	0.603
Nov' 18	System (S1)	0.24	0.659	3.79	0.056	0.00	0.980	5.48	0.022	0.03	0.861
	Species (S2)	2.29	0.020	1.96	0.047	6.70	<0.001	2.58	0.009	2.15	0.029
	S1 x S2	1.93	0.052	1.38	0.202	1.34	0.226	1.18	0.321	1.10	0.379
Jan' 19	System (S1)	0.64	0.425	1.24	0.270	ND	ND	1.24	0.270	ND	ND
	Species (S2)	0.69	0.740	0.60	0.823	ND	ND	0.60	0.823	ND	ND
	S1 x S2	1.59	0.123	1.33	0.230	ND	ND	1.33	0.230	ND	ND
Mar' 19	System (S1)	7.21	0.009	ND	ND	ND	ND	ND	ND	ND	ND
	Species (S2)	1.63	0.110	ND	ND	ND	ND	ND	ND	ND	ND
	S1 x S2	1.50	0.152	ND	ND	ND	ND	ND	ND	ND	ND
Jul' 19	System (S1)	0.29	0.628	0.32	0.614	1.75	0.278	0.62	0.488	0.77	0.445
	Species (S2)	1.23	0.279	1.25	0.269	10.87	<0.001	2.84	0.003	5.68	<0.001
	S1 x S2	0.70	0.746	1.06	0.409	0.92	0.536	1.28	0.249	0.55	0.871

Throughout the growing season, May to October, similar herbage yields to the control ($p > 0.05$) were recorded (Figure 3.1). Mid-winter (July) herbage production ranged from 806 kg ha⁻¹ to 1199 kg ha⁻¹ with the control yielding 1078 kg ha⁻¹. All treatments were fractionated into four fractions viz. lucerne, oversown, weed and detritus fractions respectively. The control had a lucerne fraction of 76.4% (Table 3.5) and all treatments had a similar lucerne fraction to that of the control ($p > 0.05$), except for black oats and diverse mix 1 that had smaller lucerne fractions ($p < 0.05$). Due to the control not being oversown, the minimum oversown contribution for the duration of the trial was zero. The maximum oversown species contribution during mid-winter was 5.6% from the black oats treatment (Figure 3.2) and this was higher than all other treatments ($p < 0.05$), excluding Westerwolds ryegrass and diverse mix 2 ($p > 0.05$). Canola, stouling rye, forage radish, clover-medic mix, *Trifolium* mix and the control had the lowest oversown fractions ($p < 0.05$). Throughout this trial, volunteer ryegrass (*Lolium rigidum*) was the dominant weed species and there could not be distinguished between volunteer and oversown ryegrass. As a result, both were considered to be part of the oversown fraction where ryegrass was oversown. This resulted in treatments that contained ryegrass as an oversown species to have the lowest weed fraction ($p < 0.05$) throughout the course of the trial (Table 3.6). Mid-winter weed fractions ranged from 0.2% to 8.9%. All ryegrass-containing treatments had similar weed fraction to the lowest weed fraction ($p > 0.05$), but only Westerwolds ryegrass had a lower weed fraction to that of the control ($p < 0.05$). The control treatment had the highest detritus fraction of 21.3% and all other treatments had a similar detritus fraction ($p > 0.05$) (Table 3.7), except for canola that had a lower detritus fraction to that of the control ($p < 0.05$).

The late winter (August) herbage production range was similar to that of mid-winter (July) and ranged from 943 kg ha⁻¹ to 1228 kg ha⁻¹ (Figure 3.1). Oversown fractions contributed as much as 21.2% in diverse mix 2 and all treatments that contained ryegrass as an oversown species had a higher oversown fraction than all other treatments ($p < 0.05$). The control, *Trifolium* mix, forage radish, clover-medic mix and canola had the lowest or similar to the lowest oversown fractions during this period ($p < 0.05$). The control had the highest lucerne fraction ($p < 0.05$) (Table 3.5) with similar lucerne fractions for forage radish, canola and clover-medic mix ($p > 0.05$). The weed fraction ranged from 0.1% for Westerwolds ryegrass to 14.1% for forage barley (Table 3.6). The control and all treatments that contained ryegrass as an oversown species, recorded the lowest or similar to the lowest weed fraction ($p < 0.05$). All other treatments had the highest or similar to the highest weed fraction ($p < 0.05$). The control had the highest detritus fraction of 9.1% (Table 3.7). All other treatments had significantly lower detritus fractions ($p < 0.05$).

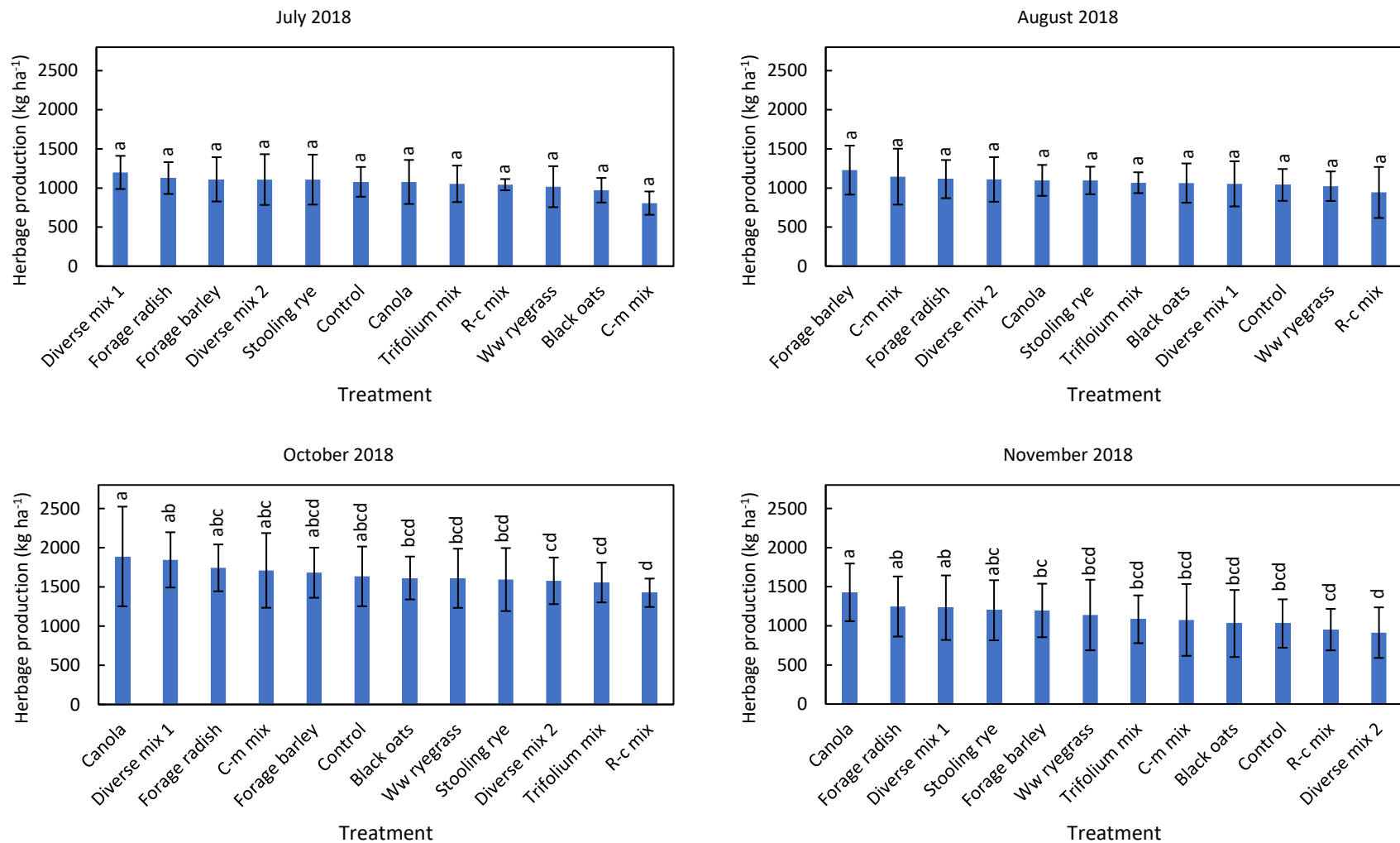


Figure 3.1 Herbage production from July 2018 to November 2018. Oversown treatments that are different to each other do not share the same letter. Significance levels were set at ($p < 0.05$), excluding October where significance was set at ($p < 0.1$). C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass

Table 3.5 Lucerne fraction (%) means for treatments for the course of this study. No common letter indicates significant differences at ($p < 0.05$) between treatments per cut

Treatment ^a	Jul'18	Aug'18	Oct'18	Nov'18	Jan'19	Mar'19	Apr'19	Jul'19
Canola	76.9 ^{ab}	81.2 ^{abc}	70.2 ^b	79.4 ^{abcd}	84.1 ^a	100.0 ^a	100.0 ^a	71.8 ^a
C-m mix	79.4 ^a	80.8 ^{abc}	58.7 ^{cd}	77.3 ^{bcd}	83.7 ^a	100.0 ^a	100.0 ^a	81.3 ^a
Control	76.4 ^{ab}	87.4 ^a	86.7 ^a	86.9 ^a	84.0 ^a	100.0 ^a	100.0 ^a	80.6 ^a
Forage barley	73.9 ^{abc}	69.9 ^e	55.0 ^d	77.2 ^{bcd}	83.7 ^a	100.0 ^a	100.0 ^a	68.0 ^a
Forage radish	78.9 ^a	84.5 ^{ab}	60.4 ^{cd}	81.4 ^{abc}	83.7 ^a	100.0 ^a	100.0 ^a	75.6 ^a
R-c mix	77.0 ^{ab}	76.9 ^{cde}	70.3 ^b	82.3 ^{abc}	87.0 ^a	100.0 ^a	100.0 ^a	73.5 ^a
<i>Trifolium</i> mix	77.0 ^{ab}	78.2 ^{bcd}	61.2 ^{bc}	77.6 ^{bcd}	84.1 ^a	100.0 ^a	100.0 ^a	73.0 ^a
Diverse mix 1	69.7 ^c	71.2 ^{de}	58.6 ^{cd}	75.7 ^{cd}	81.4 ^a	100.0 ^a	100.0 ^a	69.5 ^a
Diverse mix 2	71.8 ^{bc}	72.9 ^{de}	56.6 ^d	72.3 ^d	85.4 ^a	100.0 ^a	100.0 ^a	69.3 ^a
Black oats	70.5 ^c	75.5 ^{cde}	57.4 ^d	78.5 ^{bcd}	86.0 ^a	100.0 ^a	100.0 ^a	65.9 ^a
Stooling rye	73.7 ^{abc}	76.4 ^{cde}	66.9 ^b	85.0 ^{ab}	82.6 ^a	100.0 ^a	100.0 ^a	73.3 ^a
Ww ryegrass	74.5 ^{abc}	75.2 ^{cde}	60.7 ^{bcd}	80.9 ^{abc}	85.7 ^a	100.0 ^a	100.0 ^a	68.8 ^a

^a C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass

Means within columns with no common superscript differed significantly ($p < 0.05$)

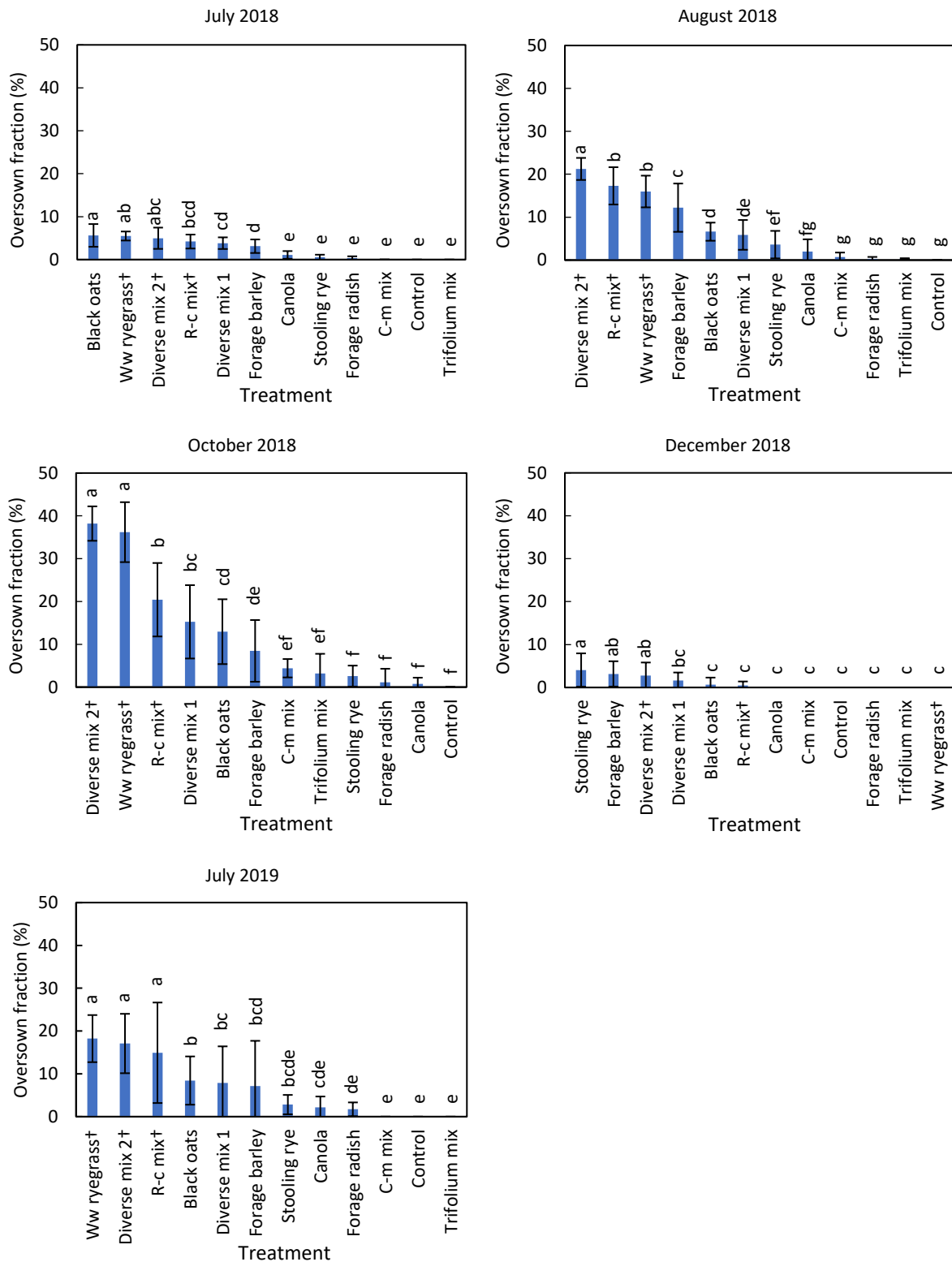


Figure 3.2 Oversown species fraction contribution for data collection cycles when oversown species were present. Treatments that do not share the same letter are different to each other ($p < 0.05$). C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass
 † Treatments contain ryegrass as an oversown species and volunteer ryegrass was considered as part of the oversown fraction

Table 3.6 Weed fraction (%) means of treatments for the course of this study. No common letter indicates significant differences at ($p < 0.05$) between treatments per cut

Treatment ^a	Jul'18	Aug'18	Oct'18	Nov'18	Jan'19	Mar'19	Apr'19	Jul'19
Canola	2.4 ^{def}	12.5 ^a	26.1 ^b	0.0 ^c	0.0 ^a	0.0 ^a	0.0 ^a	19.4 ^a
C-m mix	2.6 ^{cdef}	12.0 ^a	33.1 ^{ab}	1.0 ^a	0.0 ^a	0.0 ^a	0.0 ^a	19.1 ^a
Control	2.3 ^{def}	3.5 ^{bc}	8.6 ^c	0.7 ^{abc}	0.0 ^a	0.0 ^a	0.0 ^a	10.4 ^{bc}
Forage barley	4.1 ^{cd}	14.1 ^a	27.5 ^{ab}	0.2 ^{bc}	0.0 ^a	0.0 ^a	0.0 ^a	18.0 ^{ab}
Forage radish	2.4 ^{def}	10.1 ^{ab}	36.0 ^a	0.7 ^{abc}	0.0 ^a	0.0 ^a	0.0 ^a	15.6 ^{ab}
R-c mix [†]	0.7 ^{fg}	0.6 ^c	1.2 ^c	0.0 ^c	0.0 ^a	0.0 ^a	0.0 ^a	3.2 ^{cd}
<i>Trifolium</i> mix	4.4 ^c	14.0 ^a	29.5 ^{ab}	0.2 ^{bc}	0.0 ^a	0.0 ^a	0.0 ^a	14.7 ^{ab}
Diverse mix 1	7.0 ^b	11.0 ^a	24.2 ^b	0.0 ^c	0.0 ^a	0.0 ^a	0.0 ^a	16.5 ^{ab}
Diverse mix 2 [†]	1.9 ^{efg}	0.5 ^c	0.5 ^c	0.0 ^c	0.0 ^a	0.0 ^a	0.0 ^a	2.3 ^d
Black oats	3.0 ^{cde}	12.3 ^a	28.1 ^{ab}	0.0 ^c	0.0 ^a	0.0 ^a	0.0 ^a	17.9 ^{ab}
Stooling rye	8.9 ^a	13.9 ^a	28.4 ^{ab}	0.8 ^{ab}	0.0 ^a	0.0 ^a	0.0 ^a	11.8 ^{ab}
Ww ryegrass [†]	0.2 ^g	0.1 ^c	0.5 ^c	0.0 ^c	0.0 ^a	0.0 ^a	0.0 ^a	1.8 ^d

^a C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass

[†] Treatments contain ryegrass as an oversown species and volunteer ryegrass was not considered to be a weed

Means within columns with no common superscript differed significantly ($p < 0.05$)

Table 3.7 Detritus fraction (%) means of treatments for the course of this study. No common letter indicates significant differences at ($p < 0.05$) between treatments per cut

Treatment ^a	Jul'18	Aug'18	Oct'18	Nov'18	Jan'19	Mar'19	Apr'19	Jul'19
Canola	16.4 ^b	4.3 ^b	3.0 ^{abc}	13.5 ^{cde}	15.9 ^a	0.0 ^a	0.0 ^a	6.7 ^{bc}
C-m mix	18.1 ^{ab}	3.9 ^b	2.9 ^{abc}	21.7 ^{ab}	16.3 ^a	0.0 ^a	0.0 ^a	8.6 ^{bc}
Control	21.3 ^a	9.1 ^a	4.7 ^a	12.2 ^{de}	16.0 ^a	0.0 ^a	0.0 ^a	9.0 ^b
Forage barley	18.1 ^{ab}	4.9 ^b	2.2 ^{bc}	19.5 ^{abc}	16.3 ^a	0.0 ^a	0.0 ^a	6.9 ^{bc}
Forage radish	18.3 ^{ab}	5.2 ^b	2.5 ^{bc}	15.8 ^{bcde}	16.3 ^a	0.0 ^a	0.0 ^a	7.1 ^{bc}
R-c mix	18.1 ^{ab}	5.2 ^b	2.9 ^{abc}	17.3 ^{abcde}	13.1 ^a	0.0 ^a	0.0 ^a	8.4 ^{bc}
<i>Trifolium</i> mix	18.6 ^{ab}	5.5 ^b	3.8 ^{ab}	21.4 ^{ab}	15.9 ^a	0.0 ^a	0.0 ^a	9.0 ^{bc}
Diverse mix 1	18.1 ^{ab}	4.3 ^b	1.9 ^{bc}	22.7 ^{ab}	18.6 ^a	0.0 ^a	0.0 ^a	6.2 ^{bc}
Diverse mix 2	20.8 ^a	4.6 ^b	2.4 ^{bc}	23.9 ^a	14.6 ^a	0.0 ^a	0.0 ^a	11.3 ^b
Black oats	21.0 ^a	5.1 ^b	1.6 ^c	18.0 ^{abcd}	14.0 ^a	0.0 ^a	0.0 ^a	3.7 ^c
Stooling rye	17.8 ^{ab}	6.1 ^b	2.2 ^{bc}	10.1 ^e	17.4 ^a	0.0 ^a	0.0 ^a	8.6 ^{bc}
Ww ryegrass	18.1 ^{ab}	5.0 ^b	1.1 ^c	19.1 ^{abcd}	14.3 ^a	0.0 ^a	0.0 ^a	7.5 ^{bc}

^a C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass

Means within columns with no common superscript differed significantly ($p < 0.05$)

As temperatures increased in early to mid-spring (October), herbage production peaked. Canola had the highest herbage yield, producing 1889 kg ha⁻¹ (Figure 3.1). This was higher than black oats, Westerwolds ryegrass, stooling rye, diverse mix 2, *Trifolium* mix and ryegrass-clover mix ($p < 0.1$). All other treatments had similar to the highest herbage production ($p > 0.1$). The control had the highest lucerne fraction ($p < 0.05$) of 86.7%. The oversown fraction reached a maximum contribution of 38.2% from diverse mix 2 and treatments that contained ryegrass as an oversown species had higher oversown fractions than all other treatments ($p < 0.05$), excluding diverse mix 1 ($p > 0.05$) (Figure 3.2). Clover-medic mix, *Trifolium* mix, stooling rye, forage radish, canola and the control treatment had the lowest oversown fraction ($p < 0.05$). Ryegrass-containing treatments as well as the control had lower weed fractions than all other treatments. Forage radish had the highest weed fraction as shown in Table 3.6 ($p < 0.05$). Clover-medic mix, *Trifolium* mix, stooling rye, black oats and forage barley had similar weed fractions to forage radish ($p > 0.05$). The control treatment had the highest detritus fraction of 4.7% (Table 3.7). *Trifolium* mix, canola, ryegrass-clover mix and clover-medic mix had similar detritus fractions to the control ($p > 0.05$). *Trifolium* mix and the control were the only treatments that had a higher detritus fraction than the lowest detritus yielding treatments ($p < 0.05$). From late spring 2018 (November) to oversowing in autumn 2019 (April), there were four data collection events. During late spring canola produced more herbage than the control (Figure 3.1) ($p < 0.05$). Herbage production ranged from 912 kg ha⁻¹ to 1428 kg ha⁻¹ (Figure 3.1). Stooling rye had the highest oversown fraction ($p < 0.05$) with forage barley and diverse mix 2 yielding similar oversown fractions ($p > 0.05$). Canola, clover-medic mix, the control, forage radish, *Trifolium* mix and Westerwolds ryegrass all had an oversown fraction of zero and ryegrass-clover mix, black oats and diverse mix 1 all yielded similar oversown fractions ($p > 0.05$). No oversown species survived the grazing cycle in late spring and the oversown fraction was zero until oversowing in the subsequent growing season. The control had the highest lucerne fraction (Table 3.5) in late spring and stooling rye, ryegrass-clover mix, forage radish, Westerwolds ryegrass and canola had similar lucerne fractions ($p > 0.05$). Black oats, *Trifolium* mix, clover-medic mix, forage barley, diverse mix 1 and diverse mix 2 had lower lucerne fraction to that of the control ($p < 0.05$). From summer onward, there were no differences recorded for the lucerne fraction until oversowing commenced in the subsequent growing season ($p > 0.05$). The weed fraction followed a similar trend (Table 3.6) and there were no differences ($p > 0.05$) up to oversowing in autumn. The detritus fraction (Table 3.7) for stooling rye, canola, forage radish, and ryegrass-clover mix were similar to that of the control ($p > 0.05$), the detritus fractions for all other treatments were higher than that of the control ($p < 0.05$) during late-spring. From the summer data collection onward, no differences for the detritus fractions were measured ($P > 0.05$).

All treatments had similar herbage yields during summer (Figure 3.3) ($p>0.05$). Early autumn, mainly in March of 2019, had an herbage production range of 1475 kg ha⁻¹ for *Trifolium* mix to 1148 kg ha⁻¹ for diverse mix 2 (Figure 3.3). Westerwolds ryegrass and diverse mix 2 produced the lowest herbage ($p>0.05$) but no treatment was different to the control ($p>0.05$). In mid-autumn, in April 2019, pre-oversown measurements produced 606 kg ha⁻¹ for all treatments and no differences were recorded ($p>0.05$).

For year one of the trial, May 2018 to April 2019, totalled a maximum herbage production of 8157 kg ha⁻¹ by canola and a minimum herbage production of 6911 kg ha⁻¹ by ryegrass-clover mix (Figure 3.4). The control produced 7535 kg ha⁻¹. No treatment was significantly different to the control. When considering the average seasonal production of all treatments, spring had the highest seasonal production followed by winter, autumn and summer (Figure 3.5) ($p<0.05$).

Year two of the trial started with oversowing in April 2019. Winter production, from May 2019 to July 2019, in year two ranged from 771 kg ha⁻¹ for forage radish to 575 kg ha⁻¹ for *Trifolium* mix (Figure 3.3). The control yielded 768 kg ha⁻¹ and no differences were recorded ($p>0.05$). Lucerne fractions (Table 3.5) were similar between the control and other treatments ($p>0.05$). The ryegrass-containing treatments (Westerwolds ryegrass, diverse mix 2 and ryegrass-clover mix) had higher oversown fractions than all other treatments ($p<0.05$). (Figure 3.2). The control, *Trifolium* mix, clover-medic mix, forage radish, canola and stouling rye produced the lowest or similar to the lowest oversown fraction ($p<0.05$). The weed fraction ranged from 19.4% for canola to 1.8% for Westerwolds ryegrass (Table 3.6). Canola and clover-medic mix had a higher weed fraction than the control ($p<0.05$) while diverse mix 2 and Westerwolds ryegrass had significantly less weeds. The detritus fraction ranged from 11.3% for diverse mix 2 to 3.7% for black oats. Black oats had a lower detritus fraction than that of the control ($p<0.05$). All other treatments had a similar detritus fraction to that of the control.

Due to very low rainfall in year two the trial oversown species had perished by late August 2019. By the end of August 124.2 mm of rain was received during the growing season compared to the long-term average of 235.7 mm. This resulted in a lucerne base with very little oversown species and a decision was made not to make further measurements for year two of the trial as no differences between treatments were expected.

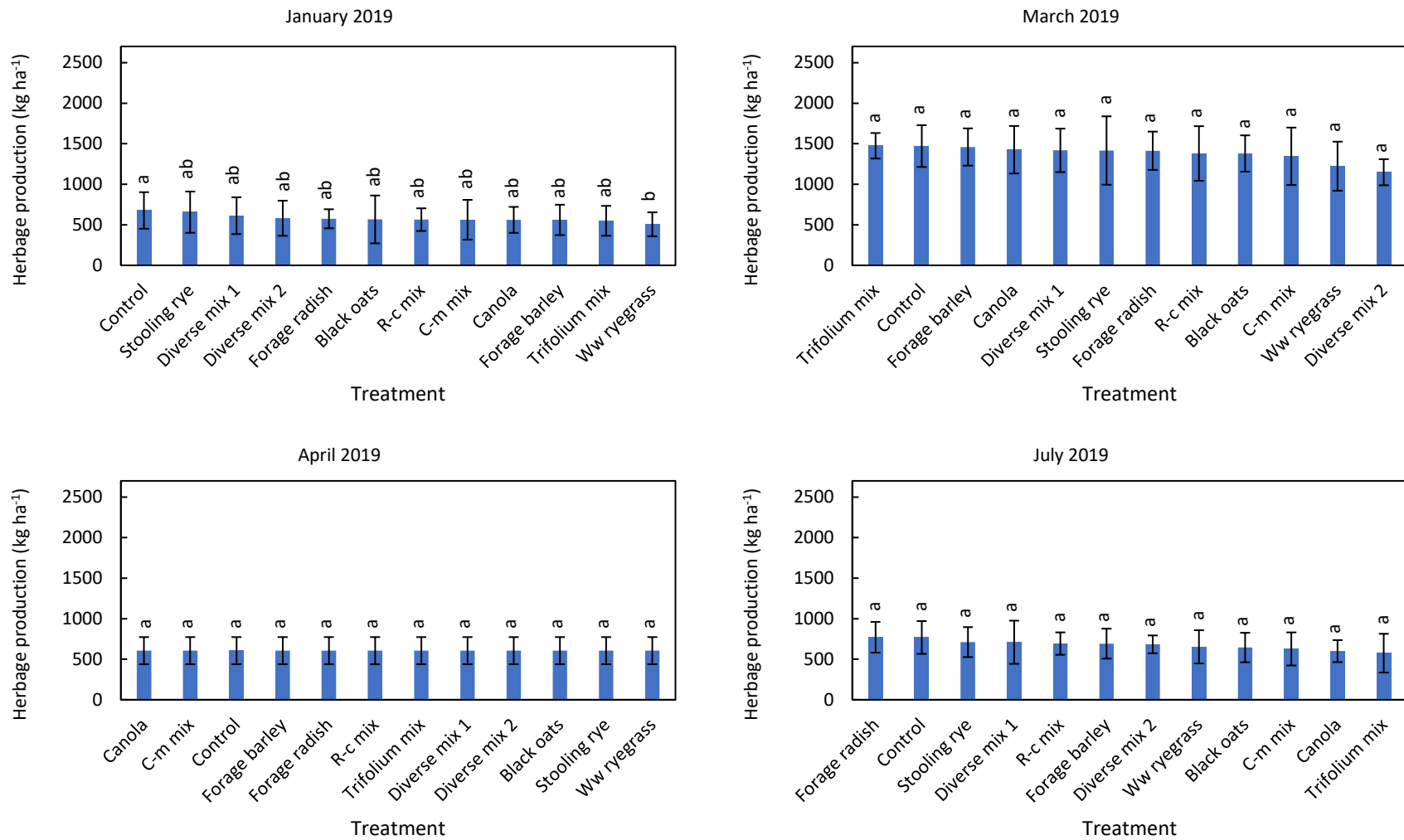


Figure 3.3 Bar graphs that depict herbage production from January 2019 to July 2019. Treatments that are different to each other do not share the same letter. Herbage production was statistically analysed between treatments within the same data collection and not between collections. Significance levels were set at (p<0.05). C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass

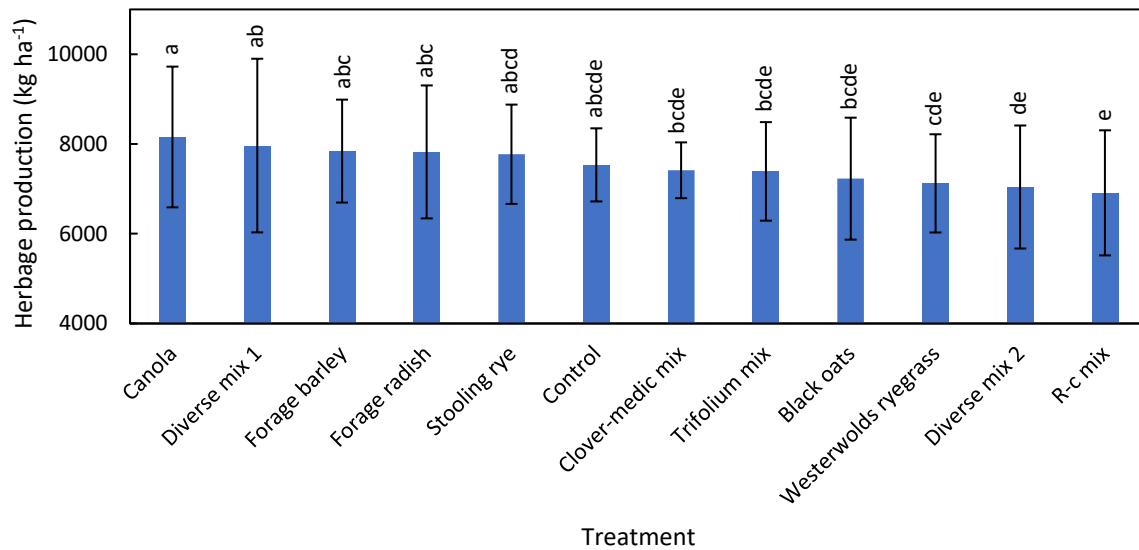


Figure 3.4 Total herbage production for year one of this trial. Treatments that share a letter are not different to each other. Significance indicated at ($p < 0.05$)

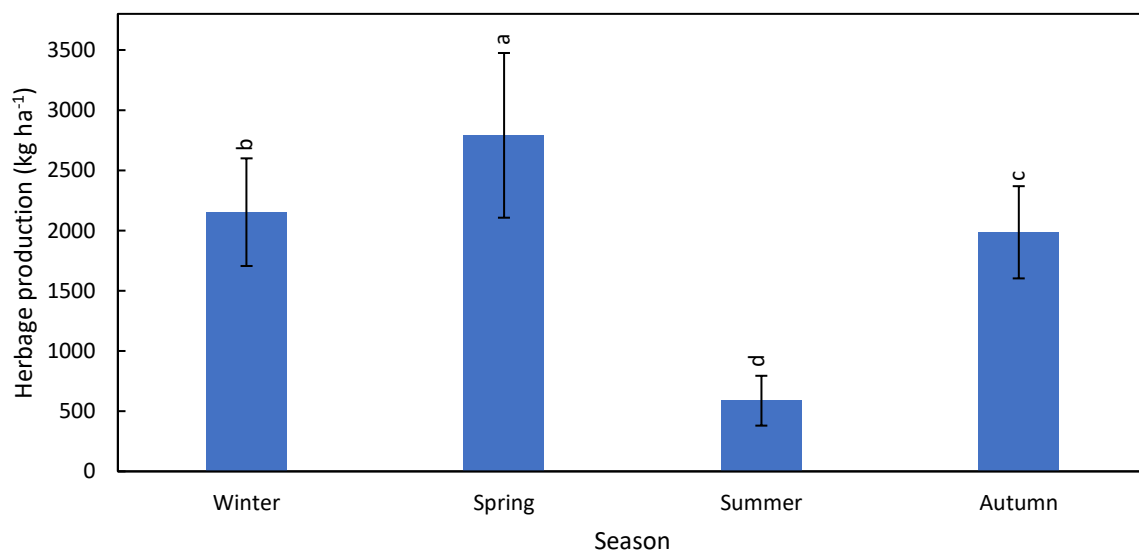


Figure 3.5 Combined average seasonal herbage production of all treatments. Treatments that share a letter are not different to each other. Significance indicated at ($p < 0.05$)

3.4 Discussion

3.4.1 The effect of cover crops sown two years prior to oversowing

In July of year one of this study, there was interaction ($p < 0.05$) between the system and oversown treatment for the weed and detritus fractions. The subsequent data collection in August 2018, showed no interaction ($p > 0.05$), which may indicate that the effect of the cover crops lasted up to this point. Cover crops are typically used in crop rotation cycles to help manage weeds (MacLaren et al., 2019).

The results from this study indicate that the effect of cover crops may possibly last until early in the growing season two years after initial establishment. The applied system did not have an effect on the plant population and it is thus not expected to have a long-lasting effect on weed suppression or the detritus fraction.

3.4.2 Mid-winter herbage production (July)

All treatments, including the control, produced similar herbage yields throughout the growing season ($p > 0.05$). Current deficits in fodder flow programmes were not abridged by oversowing of annual winter growing crops into the lucerne-base. Herbage production patterns were similar to that described by van Heerden (1976) for dryland lucerne swards in the southern Cape, despite oversowing. Similar herbage production in mid-winter is likely due to planting date as herbage production of lucerne-based multiple species pastures can be manipulated with different planting dates (Badenhorst, 2011; Botha et al., 2015). Oversowing from January to March will ensure that there is high herbage production during winter months in kikuyu-ryegrass pastures (Botha et al., 2015) and similar results could be obtained in lucerne-based multiple species pastures. Early oversowing of dryland lucerne-based multiple species pastures may however not be viable due to rainfall patterns and the high likelihood of crop failure. The onset of the rainy season in the region where this study was conducted, has been late from at least 2016 (MacLaren et al., 2019) and this means that oversowing in mid to late April would probably not have been possible in any of the past four growing seasons. This may be a factor to consider when assessing whether lucerne-based multiple species pastures are a viable option to reduce fodder flow deficits in especially early to mid-winter. Long-term average rainfall does however suggest that oversowing would be possible from mid-April onward in years of normal rainfall distribution. The small oversown fraction of zero to 5.6% in mid-winter was due to small individual plants and is a consequence of moisture stress after germination, competition from the lucerne base and late oversowing in May. Other factors such as competition (Humphries et al., 2004), allelopathy (Chon et al., 2006), lucerne cultivar (Humphries et al., 2004) and soil moisture may also have had an effect, but there could not be distinguished between them at such an early stage.

3.4.3 Late winter herbage production (August)

Herbage production similar to that of the control in late winter ($p > 0.05$) was likely caused due to a range of different factors. In the treatments oversown with ryegrass (Westerwolds ryegrass, ryegrass-clover mix and diverse mix 2), there could not be distinguished between oversown and volunteer

ryegrass, leading to all ryegrass being considered to be part of the oversown fraction. This resulted in the ryegrass-containing treatments having the largest oversown fraction ($p < 0.05$) as well as the lowest weed fraction ($p < 0.05$). Despite the higher oversown fractions ($p < 0.05$), herbage production was similar to that of the control ($p > 0.05$). Competition for resources such as sunlight, soil water and nutrients likely resulted in yield penalties for the oversown fractions (Egan and Ransom, 1996; Fukai and Trenbath, 1993; Harris et al., 2007; Humphries, 2012) and the lucerne base (Humphries et al., 2004). Total herbage production was thus similar to that of the control ($p > 0.05$). Allelopathy may also have reduced ryegrass performance. Lucerne's allelopathic effect can reduce the root length of ryegrass by up to 65% (Zubair et al., 2017) and reduced oversown species performance (Chon et al., 2006).

Treatments that consisted of small grains or mixes with small grains, excluding ryegrass (black oats, forage barley, stouling rye and diverse mix 1), had similar yield to that of the control ($p < 0.05$), but the factors influencing herbage production might have been different to that of ryegrass-containing treatments. Oversown fractions were lower than that of the ryegrass-containing treatments ($p < 0.05$). Oversewing in late April and early May should result in oversown species making a reasonable contribution toward herbage production in late winter (Botha et al., 2015) if oversown species established well. There can thus be concluded that oversown treatments of small grain and mixes with small grains were insufficient to make a noteworthy contribution toward total herbage production in late winter. The lucerne fraction for the above-mentioned treatments was lower than that of the control ($p < 0.05$) and weed fractions were also the highest or similar to the highest ($P < 0.05$). The complex nature of pastures means that a number of different factors could be linked to poor establishment of these treatments. After oversewing in early May of 2018, there was a three-week period without rainfall. Dry conditions after oversewing likely resulted or contributed to poor or reduced germination of oversown species (Sharma, 1973). Rainfall during the growing season of annual crops will have a strong influence on annual species performance (Harris et al., 2007). Lucerne has also been known to dry out the soil profile and this will restrict oversown species, especially in dry years (Bell et al., 2014; Harris et al., 2007). The competition for moisture may have limited herbage production of the oversown fractions and made the pasture susceptible to invasion by wild ryegrass. Competition from wild ryegrass may then have suppressed the small grains that did establish. Seeds may also have been exposed to predation through microorganisms in soil or macro-organism like helmeted Guinea fowl (*Numida meleagris*).

Treatments that contained brassicas, clovers and medics also had herbage production similar to that of the control in late winter ($p > 0.05$), despite having the lowest oversown fraction contributions

($p < 0.05$). The low oversown fraction made treatment susceptible to ryegrass invasion and herbage contributions mainly consisted of weed and lucerne fractions. Poor establishment of brassicas may be due to competition for limited resources, especially soil moisture. Lucerne may have a distinct advantage to compete against oversown treatments as it had a well-established root system compared to small roots from newly emerged seedlings. Allelopathy may also have suppressed oversown annual winter growing species. Allelopathy has been reported on grains (Mamolos and Kalburtji, 2001), grasses (Chon et al., 2006; Zubair et al., 2017) and brassicas (Chon et al., 2006). The allelopathic effect on brassicas tends to be more severe than on grasses (Chon et al., 2006). In field conditions, it is however almost impossible to distinguish between the effects of competition and allelopathy (Mamolos and Kalburtji, 2001). The oversown fraction of brassicas may also have been underestimated. Forage radish and canola will typically have a lower plant density than small grains or grasses. Herbage production may however be compensated through larger individual plants. Random cutting may miss these larger, but sparser plants and result in underestimation of brassica fractions. The low growing morphology of clovers and medics make them vulnerable to being suppressed by taller grasses and legumes (Deak et al., 2007). Higher sowing rates are likely to increase herbage production (McCormick et al., 2012) and may be considered for species that performed poorly like brassicas, clovers and medics. Cutting of herbage samples simulate a typical height at which sheep would graze (roughly 50 mm). The low growing morphology of clovers may also lead to underestimation as they may grow below the height at which samples were collected. Low temperatures during the growing season may also have suppressed clover growth.

3.4.4 Spring herbage production (October)

The combination of increased temperature and regular rainfall in September resulted in the herbage production peaks by all treatments. This was similar to production curves reported by Van Heerden (1990) for Tygerhoek Research Farm over a three year period. Oversown fractions ranged from zero to 38%, but as was the case in late winter, did not result in increased herbage production ($p > 0.05$). The ryegrass-containing treatments' higher oversown fractions ($p < 0.05$) replaced the lucerne base. Competition for limited resources was the likely cause of the oversown fraction replacing rather than complimenting the lucerne base that resulted in similar herbage yields ($p < 0.05$). Ryegrass, whether it was oversown or volunteer, showed exceptional ability to compete with lucerne. It is possible that ryegrass could follow a sigmoidal curve to reach a dynamic equilibrium even higher than the range of 20% to 38% recorded for this study. Van Heerden (1990) recorded legume fractions as low as roughly 35% and 50% in lucerne swards that were unsprayed and sprayed to control ryegrass invasion.

Of the small grain containing treatments, diverse mix 1 and black oats followed similar herbage production patterns as described by Purser (1981) for annual winter growers in Mediterranean pastures. The oversown fraction of forage barley peaked in late winter and this may be due to grazing management. A grazing regime that was tailored for lucerne in the Western Cape was followed (Oberholzer et al., 1993) and this may not have been optimal for all treatments (Dove et al., 2015). Some of the forage barley plants may have started to enter into the reproductive phase opposed to diverse mix 1 and black oats that was still mainly in the vegetative phase. This may explain why some forage barley plants (still vegetative) recovered after grazing (Bell et al., 2014) while those that had already reached to the reproductive stage did not (Radcliffe et al., 2012). The oversown fraction of stouling rye also declined from late winter to spring. It must be noted that the oversown fraction of stouling rye in late winter was only 3.6%. The low stouling rye fraction is an indicator of poor initial establishment and that it had an effect throughout the growing season. Competition for limited resources from the lucerne base, the grazing programme and weed invasion all likely suppressed stouling rye.

The possible reasons for poor brassica performance have already been discussed earlier in this chapter. Grazing management may however have been a contributing factor to poor brassica performance. Stem elongation and flowering had started when late winter samples were taken prior to grazing. The grazing of brassicas in the reproductive phase will not allow plants to fully recover and yield penalties are inevitable (Radcliffe et al., 2012). Clover and medic production ranges increased from zero to 0.7% in winter to 3.2% to 4.4% in spring. It is likely that both medics and clovers were suppressed by the lucerne base (Deak et al., 2007) as they have similar herbage production patterns (van Heerden, 1990, 1976). Further competition for limited resources by volunteer ryegrass may have been a contributing factor to clover and medics suppression.

3.4.5 Late spring to autumn herbage production (November to April)

From late spring 2018 to summer 2019 there were two data collection events. The canola treatment produced significantly more herbage than the control during late spring (November). The higher herbage production cannot be linked to any specific fraction as all fractions were similar to that of the control ($p > 0.05$). By mid-summer (January), the control yielded the highest herbage ($p > 0.05$).

In March of 2019, more than 260 mm of rain was received in a 48-hour period. This resulted in an almost immediate response from the lucerne base and within four weeks, the control treatment produced 1471 kg ha⁻¹. This was similar the highest production of 1475 kg DM ha⁻¹ by *Trifolium* mix ($p > 0.05$). Diverse mix 2 and Westerwolds ryegrass produced less herbage than the control ($p > 0.05$).

The lower herbage production ($p>0.05$) could be linked to competition for limited resources in the previous growing season. By the end of April, the trial site was dry and production dropped to under 610 kg ha^{-1} for all treatments and no significant differences were recorded.

3.4.6 Year two winter herbage production

In year two of the trial, there was only a single data collection event, due dry conditions. Initial oversowing in late April resulted in crop failure due to drought condition and oversown species required a second oversowing in June. Trends were similar to those in year one of the trial. Oversown species made limited contribution in winter due to small individual plants as a result of the planting date. The ryegrass-containing treatments made the highest oversown fraction contribution ($p<0.05$), followed by small grains and small grain mixes with clover, medics and brassicas making the lowest oversown fraction contribution ($p<0.05$). Herbage production was lower than in year one as a result of moisture stress. After grazing, oversowing species did not recover and no further measurements were taken.

3.4.7 Lucerne dormancy, persistence and grazing management

The lucerne cultivar used for this this trial was L70 with a dormancy class of 7. L70 was used as it is widely cultivated in the region and results of this trial are applicable to farmers in the area. A dormancy class of 7 is semi-winter active. A more dormant lucerne cultivar may be considered as more dormant lucerne cultivars will not limit oversown species to the same degree as winter active cultivars (Humphries et al., 2004). This trial was also conducted during years two and three of the lucerne sward's five to seven-year lifespan. As lucerne swards age, plant population will decline (Bouton, 2012b; Humphries, 2012) and make lucerne less competitive allowing for higher production from oversown species (Egan and Ransom, 1996; Harris et al., 2008). The allelopathic effect of the lucerne base should also be considered when oversowing lucerne swards. Brassicas may be more sensitive to allelopathic effects (Chon et al., 2006) and may not be the most suitable crop to be oversown into lucerne swards. Ryegrass has shown potential to make meaningful contributions toward total herbage production, however the benefit of oversown pastures will only be realised if the oversown component compliments the lucerne base rather than replaces it (van der Colf et al., 2015). It seemed that ryegrass replaced the lucerne base rather than compliment it and this may hamper pasture production in the following summer and autumn. Grazing management was tailored for lucerne in the southern Cape. It is possible that different grazing intervals may be more suited to different oversown species (Dove et al., 2015). For this trial, different grazing management regimes were not practically

viable but may present opportunity for future research. Grazing management that allows for a higher legume fraction, or a more persistent lucerne base, is likely to ensure better pasture without compromising lucerne longevity (Deak et al., 2009). For the purpose of this study, it was crucial not to damage the lucerne base.

In Australia, oversowing of lucerne swards with small grains have shown promising results (Egan and Ransom, 1996). Despite a 56% rainfall shortfall in typical planting months, April and May, the small grains and small grain mixes showed potential without compromising lucerne performance in the following summer and autumn. Small grains showed regrowth despite low potential for regrowth due to limited soil moisture (McCormick et al., 2012). Forage barley declined from late winter to August and this was likely due to an unfavourable grazing programme. Further research is required for diverse mix 1, black oats and forage barley to determine whether these treatments may reduce the fodder flow gaps farmers currently face. In older lucerne swards, when the lucerne base will be less competitive, treatments that struggled in the young lucerne sward may also perform better. If a sward is going to be terminated, ryegrass may also be considered as future herbage production is irrelevant.

Restrictions in planting date due to climatic conditions make it unlikely that lucerne-based multiple species pasture will be able to reduce the fodder flow gap throughout the whole of winter. A combination of resources may be a viable solution to bridge winter fodder flow gaps rather than a single resource such as lucerne-based multiple species pastures. There is potential to supplement forage needs of animals in late winter with lucerne-based multiple species pastures while potentially supplementing early winter forage needs with dual-purpose crops (Bell et al., 2018; Bell and Moore, 2012; Kelman and Dove, 2009; Moore, 2009; Radcliffe et al., 2012), however further research under South African conditions is required.

3.5 Conclusion

For a lucerne-based multiple species pasture to be considered, total production must at the very least be similar to that of the control with oversown species contributing to herbage production. The distribution of herbage production must also be of such a nature that there is reduced fluctuation of available fodder in fodder flow programmes for farmers. Total herbage production and fodder flow was similar to that of the control for year one of the trial, although some of the ryegrass-containing treatments tended to have a lower herbage production in the following summer and autumn ($p>0.05$). Due to the complex nature of pastures, lucerne cultivar, sward persistence, plant population, dormancy class, allelopathy, planting date, moisture stress, grazing management or a combination of these factors may all have influenced performance of oversown treatments. Further research on the

above-mentioned factors are needed over a period of time to fully understand the potential and possible value of lucerne-based multiple species pastures. At this stage small grains and mixes with small grains show the most potential. There is also potential to investigate how dual-purpose crops, lucerne-based multiple species pastures and pure lucerne swards may be incorporated to develop a diversified fodder flow programme that will ensure fodder throughout the year in mixed livestock-grain farms in the southern Cape of South Africa.

3.6 References

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Chapter 4

Quality of lucerne-based multiple species pastures for sheep production in the southern Cape of South Africa

4.1 Introduction

Crop-livestock integrated farming systems are typically found in the southern Cape of South Africa. The Mediterranean climate is conducive to the cultivation of cash crops (Smith, 2014) and livestock integration is seen as a form of diversification for farmers (Bell et al., 2014). Long-rotation cropping systems will typically consist of five to seven years of cash crops followed by a lucerne (*Medicago sativa* L.) phase of a similar length. Large deficits in fodder flow programmes due to herbage production patterns have however resulted in some farmers excluding lucerne from rotation systems. The oversowing of dryland lucerne swards with annual winter growing crops was investigated to determine if fodder flow deficits that farmers typically face can be reduced. To fully understand the value of lucerne-based multiple species pastures for grazing animals, herbage production as well as pasture quality should be considered. The management of pasture quality has the potential to improve the financial viability of small stock producers (Lambert et al., 2000). It is important for farmers to understand how oversowing of annual winter growing crops into a high-quality lucerne base (Bouton, 2012; Humphries, 2012) will affect overall pasture quality. Pasture quality is dynamic and will change throughout the course of the growing season due to species composition (Avci et al., 2018; Deak et al., 2007), grazing management (Deak et al., 2009) and physiological stage of individual plants (Avci et al., 2018; Humphries, 2012; Purser, 1981).

Insufficient voluntary nutrient intake is the most restricting factor regarding animal production in South Africa and may be directly linked to seasonal pasture quality and availability (Brand, 1996). In winter months in the southern Cape, the quantity of available forage rather than quality is likely to be the limiting factor regarding animal production (Purser, 1981) and therefore, a forage that is of a lower quality may still be viable if herbage production is increased. However, if herbage production remains similar in lucerne-based multiple species pastures, herbage quality must remain high to ensure that animal production is not compromised. Legumes, like the lucerne base, are typically linked to high quality forage while grasses and small grains may be linked to increased herbage production. If oversown species replace the lucerne base rather than compliment it, the potential of the pasture will not be realised (van der Colf et al., 2015). Increased species diversity can reduce crude protein (CP) and digestibility, while increasing neutral-detergent fibre (NDF) if lower quality species displace high quality species (Deak et al., 2007). Grazing management of lucerne-based multiple species pastures

may be challenging as a specific grazing cycle will favour either the lucerne base or the oversown fraction. The nutritive quality of one of the fractions may be optimal, but if the other fraction may be low and reduce overall pasture quality (Deak et al., 2009). If grazing management focusses on the legume content, pasture quality might be upheld (Deak et al., 2009).

Traditional pure lucerne swards will also pose a risk if it is grazed as is, as it is known to cause bloat (Azad et al., 2019). Lucerne-based multiple species pastures may also be a useful tool for farmers to reduce the risk of bloat in small stock (Sottie et al., 2014; Wang et al., 2012).

Herbage production of lucerne swards is low in winter months in the southern Cape of South Africa. Lucerne-based multiple species pastures have the potential to reduce the current fodder flow deficit, especially in late winter (Chapter 3). The chemical composition and digestibility of lucerne-based multiple species pastures in winter and spring was determined to establish if changes in species composition will reduce forage quality and possibly limit animal production.

4.2 Materials and methods

Site description, experimental layout and treatments, establishment methods, grazing management and herbicide application are discussed in Chapter 3.

4.2.1 Sample collection and analyses

Six sub-samples were randomly cut per plot from quadrants with a surface area of 0.25 m² (0.25 m x 1 m) to comprise a sample on 28 August 2018 for winter and on 10 October 2018 for spring. A representative grab sample of roughly 300 grams was taken from the main sample. Grab samples were dried at 60°C for 72 hours and milled with a 1 mm sieve. Chemical analysis of winter forage samples was conducted by the animal nutrition laboratory at the Western Cape Department of Agriculture. Proximal analysis was conducted to determine dry matter (DM), ash, CP, crude fat and nitrogen free extract (NFE) according to the Association of the Official Analytical Chemists (AOAC, 2016). Crude fibre was determined according to Goering and Van Soest (1970) and Acid-detergent fibre (ADF) and NDF were analysed according to Van Soest et al. (1991).

Data obtained from chemical analysis for the winter herbage samples were then used as a baseline for near-infrared reflectance spectroscopy (NIRS) calibration. Spring samples were scanned by the animal nutrition laboratory at the Western Cape Department of Agriculture for DM, ash, CP, crude fat, crude fibre, ADF, NDF and NFE. Total digestible nutrients (TDN) and metabolisable (ME) were then calculated by the following equations (Davie, 1988):

$$\text{TDN (\%)} = (0.8 \times \% \text{CP}) + (0.4 \times \% \text{crude fibre}) + (0.9 \times \% \text{NFE}) + (0.9 \times 2.25 \times \% \text{crude fat}) \quad (1)$$

$$\text{ME (MJ kg}^{-1}\text{)} = (\text{TDN} \times 14.95)/100 \quad (2)$$

4.2.2 Statistical analyses

The data was subjected to an analysis of variance (ANOVA) using General Linear Models Procedure (VEPAC) of Statistica (version 13.5.0.17)(TIBCO Software Inc., 2018). Fixed effects were species, season and species season interaction and random effects were blocks. Shapiro and Wilk test confirmed normality of the standardised residuals (Shapiro and Wilk, 1965). Fisher's least significant difference (LSD) was calculated at the 5% level to compare treatment means (Ott and Longnecker, 2010).

4.3 Results and Discussion

Chemical analyses were conducted for a variety of quality parameters and statistical significance of species, season and the interaction between species and season is depicted in Table 4.1. Energy, TDN, CP, NDF and ADF remain the most important quality parameters when determining lucerne grazing quality (Avci et al., 2018) and will be the main focus of this section.

Table 4.1 Statistical significance of the effects of species oversown (S1), season (S2) and species-season interaction for nutritive value parameters from August to October of year one of this study. Statistical significance was set at ($p < 0.05$) and is highlighted in bold

Parameter†	Species (S1)		Season (S2)		S1 x S2 Interaction	
	F-statistic	P-value	F-statistic	P-value	F-statistic	P-value
DM (%)	10.02	<0.001	8.98	0.058	8.35	<0.001
Ash (%)	2.25	0.035	49.90	0.006	1.38	0.231
Crude protein (%)	5.59	<0.001	1.35	0.329	2.17	0.043
Crude fibre (%)	1.78	0.100	4.52	0.124	2.37	0.027
Crude fat (%)	1.08	0.403	2.91	0.187	1.30	0.269
NDF (%)	3.05	0.006	0.72	0.457	3.72	0.002
ADF (%)	1.86	0.082	31.07	0.011	2.23	0.037
ME (MJ kg ⁻¹)	7.06	<0.001	11.32	0.436	5.92	<0.001
TDN (%)	7.08	<0.001	9.41	0.055	5.94	<0.001

† DM = dry matter, NDF = neutral detergent fibre, ADF = acid detergent fibre, ME = metabolisable energy, TDN = total digestible nutrients

4.3.1 Dry matter

Dry matter values ranged from 84.69 – 91.35% over both seasons (Table 4.2). The high DM values are due to sample preparation that including drying, milling and storing prior to analysis. Values are similar to those recorded by Scholtz et al. (2009).

Table 4.2 Dry matter (%) and ash (%) values of treatments taken in August (winter) and October (spring)

Treatments†	Dry matter (%)		Ash (%)	
	Winter	Spring	Winter	Spring
Control	90.62 ± 1.94	89.79 ± 1.21	10.95 ± 1.04	9.22 ± 0.41
Canola	91.05 ± 1.30	88.99 ± 2.55	10.89 ± 0.33	9.48 ± 0.46
C-m mix	89.95 ± 1.93	89.40 ± 2.63	10.95 ± 0.65	9.72 ± 0.73
Forage barley	89.87 ± 1.76	86.79 ± 1.38	10.80 ± 0.65	9.25 ± 0.27
Forage radish	90.28 ± 1.82	85.31 ± 3.18	10.87 ± 0.39	9.05 ± 0.49
R-c mix	89.65 ± 1.66	87.65 ± 2.32	10.68 ± 0.52	9.37 ± 0.45
<i>Trifolium</i> mix	90.14 ± 2.00	84.69 ± 2.33	10.63 ± 0.37	9.38 ± 0.42
Diverse mix 1	90.10 ± 1.54	89.26 ± 2.28	11.66 ± 1.79	9.56 ± 0.37
Diverse mix 2	90.33 ± 2.07	91.26 ± 1.05	11.05 ± 0.76	9.72 ± 0.41
Black oats	90.20 ± 1.29	85.87 ± 1.51	10.84 ± 0.71	8.91 ± 0.11
Stooling rye	91.10 ± 1.89	86.33 ± 3.15	10.73 ± 0.55	9.11 ± 0.47
Ww ryegrass	91.35 ± 1.40	89.20 ± 1.01	10.75 ± 0.48	9.69 ± 0.32

† C-m = clover-medic mix, R-c = ryegrass clover mix, Ww ryegrass = Westerwolds ryegrass

4.3.2 Ash

Ash is an indicator of all the inorganic constituents of the forage. The ash range was 10.63 – 11.66% for winter and 8.91 – 9.72% for spring (Table 4.2). Values are within the range reported by Scholtz et al. (2009) for South African lucerne. There were no differences between treatments ($p > 0.05$). Ash values will typically decrease with plant maturity (Karayilanli and Ayhan, 2016) and may explain why spring has a lower ash compared to winter ($p < 0.05$).

4.3.3 Crude protein

Crude protein values ranged from 23.02 – 27.31% over both seasons with the control yielding a CP fraction of 26.60% in winter and 26.40% in spring (Table 4.3). The control treatment was equal to the highest CP fraction regardless of season and no treatment had a higher CP fraction to that of the control ($p > 0.05$) at any stage in this study. Winter CP for forage barley, diverse mix 1, diverse mix 2 black oats and Westorwolds ryegrass was lower than the control in winter ($p < 0.05$). All spring oversown treatments, excluding *Trifolium* mix, had a lower CP than the control in spring ($p < 0.05$). The control had the highest lucerne fraction ($p < 0.05$) throughout the study (chapter 3). In multiple species pastures, increased legume fractions generally resulted in increased CP (Deak et al., 2009; Stout et al., 1997) and the same trend was observed in this study.

Table 4.3 Crude protein values (DM-basis) of treatments taken in August (winter) and October (spring). Significance levels were set at ($p < 0.05$). Treatments that share a letter are not significantly different

Treatment†	Crude protein (%)	
	Winter	Spring
Control	26.60 ^{abde} ± 1.83	26.40 ^{abc} ± 0.94
Canola	27.31 ^{ad} ± 2.11	24.45 ^{ef} ± 2.00
C-m mix	25.95 ^{bcef} ± 1.90	24.04 ^{fgh} ± 2.30
Forage barley	25.00 ^{cgh} ± 3.01	24.19 ^{efg} ± 1.84
Forage radish	26.48 ^{abdef} ± 1.52	24.58 ^{ef} ± 2.03
R-c mix	25.44 ^{bcefg} ± 2.42	24.38 ^{egh} ± 1.80
Trifolium mix	25.78 ^{bcefg} ± 2.61	25.32 ^{abcde} ± 1.47
Diverse mix 1	25.27 ^{cgh} ± 2.65	24.00 ^{fgh} ± 2.12
Diverse mix 2	25.04 ^{cgh} ± 2.23	23.02 ^h ± 1.25
Black oats	25.20 ^{cgh} ± 2.33	24.45 ^{ef} ± 1.47
Stooling rye	25.52 ^{bcefg} ± 2.42	24.96 ^{def} ± 1.72
Ww ryegrass	25.22 ^{cgh} ± 2.71	23.25 ^{gh} ± 0.82

† C-m = clover-medic mix, R-c = ryegrass clover mix, Ww ryegrass = Westerwolds ryegrass

Crude protein was high in both seasons compared to studies conducted by Brand (1996), Avci et al. (2018) and Karayilanli and Ayhan (2016) that yielded crude protein ranges of 7.0 – 15.5%, 17.4 – 19.7% and 19.6 – 19.9% respectively. The low CP levels recorded by Brand (1996) was likely due to the low lucerne fraction, roughly 20%, due to ryegrass invasion and may give insight to how pasture quality may deteriorate if good grazing management of the lucerne base in lucerne-based multiple species pasture is not applied. Crude protein was within the range described by Scholtz et al. (2009), 13.9 – 27.8%, for South African lucerne. Although a wide range of species was oversown into the lucerne base, almost all treatments were dominated by lucerne and ryegrass (oversown and volunteer) (Chapter 3). The combination of a relatively high lucerne fraction combined with a grazing regime that kept lucerne in young physiological stage (Machado et al., 2007) likely led to relatively high CP values (Humphries, 2012) for this study. A contributing factor was that the lucerne base was still relatively young as CP has been known to decline in older lucerne swards (Coruh and Tan, 2008; Testa et al., 2011).

4.3.4 Crude fat

Crude fat was similar between all treatments ($p > 0.05$) with values ranging between 1.78 – 2.37% throughout both seasons (Table 4.4).

Table 4.4 Crude fibre (%) and crude fat (%) values (DM-basis) of treatments taken in August (winter) and October (spring) during year one of this experiment

Treatments†	Crude fibre (%)		Crude fat (%)	
	Winter	Spring	Winter	Spring
Control	17.86 ^{dfghij} ± 1.55	17.85 ^{ej} ± 0.68	2.22 ± 0.29	2.37 ± 0.15
Canola	17.43 ^{hij} ± 1.17	19.22 ^{abcdfg} ± 1.46	2.21 ± 0.11	1.88 ± 0.37
C-m mix	17.58 ^{ghij} ± 1.29	19.38 ^{abcdf} ± 1.12	2.35 ± 0.68	1.94 ± 0.39
Forage barley	17.79 ^{fghij} ± 2.50	19.22 ^{abcdfg} ± 1.02	2.18 ± 0.28	1.94 ± 0.27
Forage radish	16.96 ^j ± 1.73	18.78 ^{abcdefghi} ± 1.63	2.19 ± 0.30	2.11 ± 0.37
R-c mix	17.15 ^{ij} ± 1.09	18.60 ^{abcdefghij} ± 1.43	2.27 ± 0.27	2.04 ± 0.36
Trifolium mix	18.00 ^{cd fghij} ± 1.39	18.34 ^{befghij} ± 1.00	2.26 ± 0.28	2.05 ± 0.34
Diverse mix 1	17.24 ^{ij} ± 1.17	19.51 ^{acd} ± 1.19	2.26 ± 0.17	1.84 ± 0.32
Diverse mix 2	17.52 ^{hij} ± 1.74	19.65 ^{ac} ± 0.89	2.24 ± 0.21	1.78 ± 0.33
Black oats	17.59 ^{ghij} ± 1.72	18.75 ^{abcdefghi} ± 0.91	2.29 ± 0.44	2.18 ± 0.19
Stooling rye	19.26 ^{abcd fgh} ± 1.19	19.01 ^{abe} ± 1.58	2.21 ± 0.22	1.98 ± 0.41
Ww ryegrass	16.97 ^j ± 0.94	18.92 ^{abcdefgh} ± 0.43	2.25 ± 0.17	2.00 ± 0.20

† C-m = clover-medic mix, R-c = ryegrass clover mix, Ww ryegrass = Westerwolds ryegrass

4.3.5 Crude fibre, Acid-detergent fibre and Neutral detergent fibre

Fibre content of the forage was analysed for as crude fibre (Table 4.4), ADF and NDF (Table 4.5). The main focus will be on ADF and NDF. Crude fibre values ranged from 16.96 – 19.65% over seasons. In winter, all treatments had a similar crude fibre ($p>0.05$), except for stooling rye that had a higher crude fibre fraction ($p<0.05$). For spring all treatments had a similar crude fibre fraction ($p>0.05$), excluding the control that had a lower crude fibre fraction ($p<0.05$). The ADF range from winter to spring for this study was 18.33 – 24.42%. Although there was interaction between oversown species and season, all treatments had a lower ADF in spring compared to all ADF fractions recorded for winter ($p<0.05$). The general trend for ADF is that it will increase with sward age and increased plant maturity (Karayilanli and Ayhan, 2016). The control treatment in winter had the highest ADF fraction ($p<0.05$) and only stooling rye for winter was similar ($p>0.05$). No spring treatment was different to the control in spring ($p>0.05$). The higher ADF from the control in winter may be linked to reduced species diversity compared to other treatments, as Deak et al. (2009) reported that increased species diversity may result in reduced ADF.

Table 4.5 ADF (%) and NDF (%) values (DM-basis) of treatments taken in August (winter) and October (spring). Significance levels were set at ($p<0.05$). Treatments that share a letter are not significantly different

Treatments†	Acid-detergent fibre (%)		Neutral-detergent fibre (%)	
	Winter	Spring	Winter	Spring
Control	24.42 ^a ± 2.18	19.43 ^{de} ± 0.73	29.83 ^{abcdefg} ± 2.76	27.85 ^g ± 1.29
Canola	21.95 ^c ± 1.74	19.34 ^{de} ± 0.46	28.05 ^f ± 2.36	31.52 ^{abcf} ± 2.36
C-m mix	22.04 ^c ± 1.23	19.82 ^d ± 1.05	28.44 ^f ± 1.83	31.56 ^{abcf} ± 1.83
Forage barley	23.00 ^{bc} ± 1.69	18.88 ^{de} ± 0.36	30.54 ^{abcdeg} ± 3.20	30.99 ^{bcd} ± 3.20
Forage radish	22.41 ^c ± 2.94	18.68 ^{de} ± 0.66	28.89 ^{bcdefg} ± 2.23	28.55 ^g ± 4.27
R-c mix	22.87 ^{bc} ± 2.99	18.81 ^{de} ± 0.34	29.49 ^{bcdefg} ± 3.04	30.44 ^{bcdef} ± 3.72
Trifolium mix	22.00 ^c ± 1.90	18.33 ^e ± 0.28	27.86 ^f ± 2.19	29.39 ^{deg} ± 3.04
Diverse mix 1	22.72 ^c ± 2.13	19.36 ^{de} ± 0.55	29.41 ^{bcdefg} ± 2.65	32.38 ^{ab} ± 2.97
Diverse mix 2	22.10 ^c ± 2.87	19.77 ^d ± 0.41	29.42 ^{bcdefg} ± 2.26	33.42 ^a ± 2.86
Black oats	22.17 ^c ± 1.57	18.68 ^{de} ± 0.23	29.32 ^{bcdef} ± 2.83	28.86 ^{eg} ± 2.28
Stooling rye	24.21 ^{ab} ± 2.20	18.71 ^{de} ± 0.52	30.53 ^{abcdeg} ± 3.50	30.05 ^{def} ± 4.74
Ww ryegrass	22.21 ^c ± 1.52	19.07 ^{de} ± 0.26	28.01 ^f ± 1.59	30.97 ^{bcd} ± 0.98

† C-m = clover-medic mix, R-c = ryegrass clover mix, Ww ryegrass = Westerwolds ryegrass

The ADF range for both seasons were within the range or slightly lower than values recorded by Scholtz et al. (2009) for South African lucerne swards. Acid-detergent fibre was lower than values recorded by Brand (1996). Lower ADF values are likely due to a lower grass fraction in this study as volunteer ryegrass and weeds comprised, on average, 80% of the pasture in the study conducted by Brand (1996). Individual species contribution rather than species diversity is more important regarding nutritive value of pastures (Deak et al., 2007). Increased grass fractions are typically characterised by

increased fibre content (Deak et al., 2009). Acid-detergent fibre was relatively low in this study and is likely due to the lucerne base being in a young physiological stage (Avci et al., 2018; Karayilanli and Ayhan, 2016) at the time of sampling.

Diverse mix 2 for spring had the highest NDF fraction of 33.42% ($p < 0.05$). The control, forage barley and stouling rye for winter and canola, clover-medic mix and diverse mix 1 for spring had similar NDF fractions ($p > 0.05$). The control for spring had the lowest NDF fraction of 27.85% ($p < 0.05$), with black oats and forage radish for spring having similar NDF fractions as well as the control, forage barley, forage radish, ryegrass-clover mix diverse mix 1, diverse mix 2 and stouling rye ($p > 0.05$) for winter.

The NDF range was lower than that recorded by Brand (1996) and Avci et al. (2018) and similar to the lowest values recorded by Scholtz et al. (2009) and Karayilanli and Ayhan (2016). Lower NDF values compared to Brand (1996) and Avci et al. (2018) are likely due to a smaller grass fractions (Deak et al., 2009) in this trial. Neutral-detergent fibre has an inverse relationship with winter dormancy of lucerne (Karayilanli and Ayhan, 2016). More winter active cultivars are linked to increased levels of NDF. The NDF values recorded in this trial were lower than values recorded by Avci et al. (2018) for winter dormant lucerne and did thus not play a major role in NDF content. Low NDF values can thus be linked to the lucerne being the dominant contributor to forage as well as being in a young physiological stage at the time of sample collection.

4.3.6 Metabolisable energy

Westerwolds ryegrass in winter had the highest ME value of 9.6 MJ kg⁻¹ ($p < 0.05$) (Table 4.6). Canola and forage radish for winter as well as the control and diverse mix 2 for spring were similar to the highest ME ($p > 0.05$). *Trifolium* mix in spring had the lowest ME value of 8.73 MJ kg⁻¹ ($p < 0.05$) and stouling rye as well as forage radish for spring were similar ($p > 0.05$). Metabolisable energy was higher than ME recorded by Karayilanli and Ayhan (2016) in pure lucerne swards and may be due to lucerne in this study being in a younger physiological stage. As lucerne plants mature, ME will decrease (Karayilanli and Ayhan, 2016). This is likely due to a decreased leaf to stem ratio in older plants (Machado et al., 2007). Metabolisable energy values were lower than those recorded by Machado et al. (2007). This is likely due to grazing management by Machado et al. (2007) that used high density grazing with the specific management goal of keeping plants in a young physiological stage. High density grazing was used in this trial, but the focus was to ensure sward persistence rather than maintain a high forage quality.

Table 4.6 ME (MJ kg⁻¹) and TDN (%) values (DM-basis) of treatments taken in August (winter) and October (spring). Significance levels were set at (p<0.05). Treatments that share a letter are not significantly different

Treatments†	Metabolisable energy (MJ kg ⁻¹)		Total digestible nutrients (%)	
	Winter	Spring	Winter	Spring
Control	9.39 ^{bcd} ± 0.39	9.51 ^{ab} ± 0.10	62.61 ^{bcd} ± 2.59	63.62 ^{ab} ± 0.68
Canola	9.47 ^{abcd} ± 0.20	9.21 ^{ef} ± 0.19	63.18 ^{abcd} ± 1.31	61.61 ^{de} ± 1.29
C-m mix	9.35 ^{bcd} ± 0.34	9.24 ^{def} ± 0.26	62.36 ^{bcd} ± 2.25	61.80 ^{de} ± 1.73
Forage barley	9.33 ^{bcd} ± 0.32	8.96 ^{gh} ± 0.18	62.22 ^{bcd} ± 2.15	59.94 ^{fg} ± 1.20
Forage radish	9.41 ^{abcde} ± 0.29	8.84 ^{hi} ± 0.26	62.80 ^{abcd} ± 1.95	59.16 ^{gh} ± 1.77
R-c mix	9.37 ^{bcd} ± 0.28	9.12 ^{fg} ± 0.17	62.51 ^{bcd} ± 1.86	61.02 ^{ef} ± 1.14
Trifolium mix	9.37 ^{bcd} ± 0.23	8.73 ⁱ ± 0.26	62.52 ^{bcd} ± 1.51	58.39 ^h ± 1.72
Diverse mix 1	9.29 ^{bcd} ± 0.38	9.22 ^{ef} ± 0.24	61.99 ^{bcd} ± 2.50	61.64 ^{de} ± 1.57
Diverse mix 2	9.38 ^{bcd} ± 0.32	9.46 ^{abc} ± 0.10	62.60 ^{bcd} ± 2.16	63.27 ^{abc} ± 0.69
Black oats	9.40 ^{bcd} ± 0.28	8.95 ^{bcd} ± 0.14	62.68 ^{bcd} ± 1.86	59.90 ^{fg} ± 0.93
Stooling rye	9.39 ^{bcd} ± 0.28	8.93 ^{bcd} ± 0.21	62.63 ^{bcd} ± 1.87	59.72 ^{fg} ± 1.43
Ww ryegrass	9.60 ^a ± 0.22	9.27 ^{cdef} ± 0.16	64.06 ^a ± 1.45	60.02 ^{cde} ± 1.04

† C-m = clover-medic mix, R-c = ryegrass clover mix, Ww ryegrass = Westerwolds ryegrass

4.3.7 Total digestible nutrients

Total digestible nutrients ranged from 64.06% for Westerwolds ryegrass in winter to 58.39% for *Trifolium* mix in spring (Table 4.6). Canola and forage radish in winter and the control and diverse mix 2 in spring were not different to Westerwolds ryegrass in winter (p>0.05). Forage radish, and stooling rye in spring were not different to *Trifolium* mix in spring (p>0.05). Total digestible nutrients will decrease with plant maturity (Humphries, 2012; Karayilanli and Ayhan, 2016) and it is not surprising that TDN for this study was higher than that recorded by Karayilanli and Ayhan (2016) for lucerne in an older physiological stage. In Mediterranean pastures, digestibility of pastures tends to be high in winter and spring followed by a rapid decline in late spring as plants mature (Purser, 1981). Reduced digestibility linked to reduction in highly digestible parts and a reduced leaf to stem ratio.

4.4 Conclusion

Lucerne-based multiple species pastures' forage quality could be influenced by a number of factors that include physiological stage, oversown species, pasture composition, climatic factors, edaphic factors such as soil conditions, lucerne base age, diseases, insects, weeds and lucerne cultivar (Scholtz et al., 2009). The high lucerne fraction as well as oversown and volunteer ryegrass (Chapter 3) were the dominant species that influenced pasture quality between treatments in this trial. Crude protein (Avcı et al., 2018; Brand, 1996; Karayilanli and Ayhan, 2016) and TDN (Karayilanli and Ayhan, 2016) was high, while ADF (Brand, 1996; Scholtz et al., 2009) and NDF (Avcı et al., 2018; Brand, 1996; Karayilanli and Ayhan, 2016; Scholtz et al., 2009) was low. The young physiological state of the lucerne

base likely resulted in high protein and digestibility (Humphries, 2012) and low NDF and ADF (Deak et al., 2009). There was no treatment that clearly outperformed all others and all treatments in terms of pasture quality as described in this study.

The data presented in this chapter is also only a snapshot of the lucerne phase in long-rotation cropping systems and different results may be obtained in different years. Nutritive characteristics are likely to change as the lucerne base matures and lucerne plant population declines over different years. This may in turn lead to changes in pasture composition and thus possibly changes in pasture quality. Pasture quality is dynamic and likely to change within the same growing season as it progresses due to changes in pasture composition and plant physiological stage (Deak et al., 2007). Good grazing management that ensures that the pasture is in a young physiological stage (Machado et al., 2007), without comprising future herbage production is an important management tool to ensure pasture quality is upheld. Further research is required to monitor forage quality over the whole growing season. During winter months when feed shortages are typically experienced, quantity of feed rather than quality of feed is likely to be the most restricting factor regarding animal production (Purser, 1981). The value of forage quality is best understood when it can be related to livestock production (Sollenberger and Cherney, 1995) and livestock should be integrated into trials to establish how animal production will be influenced.

4.5 References

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Chapter 5

The influence of lucerne-based multiple species pastures on soil quality in the southern Cape of South Africa

5.1 Introduction

Dryland lucerne swards are typically found in long-rotation cropping systems in the southern Cape of South Africa (Smith, 2014; van Heerden and Tainton, 1987). The perennial nature of lucerne ensures that there are living roots in the soil throughout the year, permanent soil cover and the pasture phase allows for reduced soil disturbance over a long term. Lucerne will also fix atmospheric nitrogen (N). Reduced soil disturbance in pasture phases also allow for the build-up of soil aggregates as aggregates are not physically broken down (Wright et al., 2007) and aggregate formation is not interrupted (Verhulst et al., 2010). Perennial pastures have also been linked to increased soil organic C sequestration, compared to grain monocultures and annual pastures (Chan et al., 2001) as a result of the continuous return of organic matter to the soil. Increased soil organic C content can, in turn, lead to improved ecosystem functionality (Lal, 2016), improved water infiltration (Blair and Crocker, 2000) and accommodate increased soil microbial activity (Peyraud et al., 2014).

Dryland lucerne swards may however also provide challenges to farmers relating to soil quality. Soil physical degradation is closely linked to bare soil that is exposed to the elements (Palm et al., 2007). Low herbage production during certain times of the year (van Heerden, 1976) can lead to exposed soil, and result in low inputs of organic matter. This creates an environment not conducive to the build-up of soil C and aggregate formation. Lucerne also has a natural autotoxicity (Chon et al., 2006) and once an individual plant dies, it will not be replaced. This may lead to bare patches within the pasture, particularly in older swards, and leave the pasture vulnerable to weed invasion or other typical effects of bare soils such as reduced levels of soil organic C (Blair and Crocker, 2000). Reduced soil organic C content can lead to increased water runoff, reduced water infiltration, reduced microbial activity and reduced formation of soil aggregates. It may also be argued that the lucerne phase is a monoculture within a crop rotation system and that the benefits of crop rotation will be omitted for the duration of the lucerne phase.

The oversowing of dryland lucerne swards to form lucerne-based multiple species pastures may overcome some challenges farmers currently face. The return of diverse above and below ground biomass from both lucerne and annual crops, additional residue from winter growing annuals

(Badenhorst, 2011) and quality of the organic matter returned will all have an impact on soil physical, chemical and biological components.

Lucerne is a perennial legume and will not form as many new roots that can decompose such as those from the oversown annual species (Smith, 2014). The breakdown of roots from annual winter growing crops allows for the build-up of soil organic C due to increased soil organic matter inputs. Increased soil C promotes soil microbial activity (Habig et al., 2015), which can result in the secretion of bonding materials (Smith, 2014). The higher root biomass production from grasses combined with the different microbial populations from legumes (Haynes and Beare, 1997) contribute to the formation of stable soil aggregates. The diverse range of residues returned to the soil from lucerne-based multiple species pastures will include residue with both a high and low C:N ratio. The fraction with a low C:N ratio will be broken down more rapidly, returning nutrients to the soil and build soil organic matter, while the fraction with a higher C:N ratio will protect the soil from erosion, conserve soil moisture and provide habitat for soil microbes (United States Department of Agriculture, 2011).

The oversowing of annual winter growing crops may cover typical bare areas in lucerne swards that cannot be re-established due to lucerne's autotoxicity. The return organic matter to the soil on these previously bare areas can reduce the risk of both evaporation and soil erosion (Blanco-Canqui et al., 2015). Lucerne's autotoxicity may however have an allelopathic effect on some of the oversown species that results in poor establishment (Chon et al., 2006).

Due to the relatively slow rate at which soil physical and chemical properties like soil aggregate formation and the build-up of soil organic C takes place in semi-arid cropping systems (Blanco-Canqui et al., 2015; Smith, 2014), biological indicators can be used as tools to detect early changes in soil or as an indicator of soil health (Sekgota, 2018). Arbuscular mycorrhizal (AM) fungi is a mycorrhizal type that can form symbiotic relationships with crops commonly found in agriculture and has been linked to soil aggregate stability, C deposition and improved soil structure (Sekgota, 2018). Due to AM fungi's sensitivity to changes in the soil it can be used as indicators of soil degradation. When AM fungi is broken down, it releases a brown coloured glycoprotein known as glomalin and the concentration of glomalin can be linked to the activity of AM fungi (Sekgota, 2018). Glomalin serves as an adhesive and is linked to binding within microaggregates and macroaggregates (Wright et al., 2007).

There exists a variety of molecular techniques to study microbial populations and microbial activity (Janvier et al., 2007). Functional diversity of soils may serve as an indication of soil microbial populations as they relate to the actual or potential activities of organisms that contribute to ecosystem dynamics. Nutrient cycling of C, N and P is a fundamental soil function and can be used to

assess the relative activity of soil microbial populations. Using this context, microbial community level physiological profiles and enzymatic assays may be used to determine soil microbial populations. Soil microbial enzymes serve crucial biochemical functions in the decomposition of organic matter and nutrient cycling (Janvier et al., 2007). Enzymatic activities or soil microbial activity in relation to the cycling of C and N and the release of inorganic P in soil have been used to evaluate the fertility of the soil to describe ecosystem functions. β -glucosidase activity is an indicator of soil quality due to the role it plays in catalysing the hydrolysis biodegradation of various β -glucosides in plant debris decomposing in the ecosystem. Phosphatases are believed to play important roles in P cycles as evidence has shown that these are correlated with P stress and plant growth. Phosphatase activity has also been correlated to the soil P state with inorganic P having an inverse effect on phosphatase production. Urease activity regulates the N supply to plants. Due to the influence of pH, temperature, organic matter content and soil moisture on microbial enzymatic activity, these are considered early indicators of ecosystems stress and indicators of soil degradation when compared to slow changing soil properties such as organic matter.

The aim of this chapter was to establish whether lucerne-based multiple species pastures can be used to improve soil quality when compared to dryland lucerne swards in the southern Cape. The following three objectives were used to determine if lucerne-based multiple species pasture resulted in better soil quality than lucerne monocultures:

- 1) to establish if the lucerne-based multiple species pasture affected soil aggregate stability as an indicator of dynamic soil physical quality
- 2) to establish whether lucerne-based multiple species pastures affected soil C dynamics
- 3) to determine if lucerne-based multiple species pastures affected soil microorganism activity and diversity

5.2 Material and methods

Site description, experimental layout and treatments, establishment methods, grazing management and herbicide application are discussed in Chapter 3.

5.2.1 Sampling and analyses for standard soil fertility tests

Soil cores (45 mm diameter) were taken in June 2018 and April 2019. Six sub-samples were taken from sub-plots and composited for a representative sample. Samples were taken at depths of 0 – 150 mm and 150 – 300 mm. Standard chemical analyses were conducted by the soil laboratory of the Western

Cape Department of Agriculture to establish baseline soil characteristics (Table 5.1). Analyses included pH (KCl) using a 1:2.5 soil to solution ratio and base cations (Ca, Mg, Na and K) with a potassium chloride and citric acid solution, respectively as set out by the Non-Affiliated Soil Analysis Work Committee (1990). Standard procedures were used to determine extractable P (citric acid), S (calcium phosphate), B (hot water), Cu, Mn and Zn (di-ammonium EDTA) (Non-Affiliated Soil Analysis Work Committee 1990). Electrical resistance was determined by the method set described by the United States Salinity Laboratory Staff (1954). Dry combustion was used to determine total C with a Eurovector Elemental Analyser (Eurovector Instruments & Software, Italy).

5.2.2 Sampling and analyses for soil physical, chemical and biological parameters

Soil samples were taken in October of 2018. Six sub-samples were taken per treatment at a depth of 0 – 150 mm to comprise each sample. Soil cores (45 mm) were collected with a hammer and pipe and stored in a cool dry place until the time of analysis. Due to dry conditions and a low oversown fraction in October 2019, no soil samples were taken for soil physical, chemical and biological parameters.

5.2.2.1 Aggregate stability

Aggregate stability was determined by using the wet sieving technique according to Kemper and Rosenau (1986). The aggregate stability for was determined at a depth of 0 – 150 mm for all treatments. The technique is based on the principle that unstable aggregates will break down easily when submerged in water. Analysis was performed using the Eijkelkamp E-365-08.13 wet sieving apparatus (Eijkelkamp Agrisearch Equipment, Netherlands). Four grams of soil of duplicated samples of each replicate in each treatment were analysed. The aggregates in the sieve were lowered and raised in distilled water for three minutes with these cans containing water unstable aggregates. The remaining aggregates were lowered and raised in NaOH at 2 g L⁻¹ for ten minutes using a rubber-tipped glass rod until all aggregates disintegrated. These cans contained the water stable aggregates. Both cans of stable and unstable aggregates were dried in the oven. The mass of the material in each can was determined.

5.2.2.2 Active carbon

Soil active C was determined by Western Cape Department of Agriculture Laboratories. Samples were collected at a depth of 0 – 150 mm and 2.50 g of soil was analysed for active C by the technique described by Weil et al. (2003). Soil samples were oxidised with 0.02 M KMnO₄ in 1 M CaCl₂ (pH 7.2) and colorimetric measurement of non-reduced Mn at 550 nm was recorded.

Table 5.1 Baseline chemical soil analysis of cover and no-cover sub-plots from year one and year two of the trial at depths of 0 – 150 mm and 150 – 300 mm. Soil nutrient status was sufficient for lucerne production

	0 – 150 mm				150 – 300 mm			
	2018		2019		2018		2019	
	Cover	No-Cover	Cover	No-Cover	Cover	No-Cover	Cover	No-Cover
pH (KCl)	5.8	5.4	6.0	6.0	6.1	5.8	6.0	6.0
Resistance (Ohm)	518	530	718	898	660	768	778	1088
Ca (mg kg ⁻¹)	1640	1200	1160	1240	940	760	920	980
Mg (mg kg ⁻¹)	276	192	192	180	336	228	240	300
Na (mg kg ⁻¹)	113.3	92.8	102.3	77.0	240.8	151.8	169.0	132.5
K (mg kg ⁻¹)	222.3	211.8	219.0	232.0	125.8	163.3	174.5	158.8
Cation exchange capacity (cmol kg ⁻¹)	11.8	9.1	8.5	8.7	8.9	6.8	7.7	8.3
P (mg kg ⁻¹)	77.8	74.8	63.3	72.3	21.8	35.8	42.3	30.5
Cu (mg kg ⁻¹)	1.0	1.1	ND	ND	0.8	1.0	ND	ND
Zn (mg kg ⁻¹)	1.8	2.3	ND	ND	0.8	1.1	ND	ND
Mn (mg kg ⁻¹)	112.1	151.5	ND	ND	75.5	112.6	ND	ND
B (mg kg ⁻¹)	0.7	0.6	0.5	0.5	0.7	0.7	0.6	0.6
S (mg kg ⁻¹)	ND	ND	9.4	9.2	ND	ND	11.2	9.1
C (%)	1.8	1.2	1.5	1.4	0.8	0.8	1.1	0.8

ND Indicates that measurements were not taken

5.2.2.3 Glomalin

Easily extractable glomalin was determined in the soil with a modified technique of Wright and Upadhyaya (1998, 1996) as described by Janos et al. (2008). Easily extractable glomalin in 1 g of soil was measured and placed in 15 mL centrifuge tubes. It was suspended in 20 mM sodium tri-citrate (pH 7.0) solution and then extracted in an autoclave at 121°C for 45 minutes. After autoclaving, the tubes were immediately centrifuged at 5000 rpm for 15 minutes. The supernatants were transferred into 2 mL Eppendorf tubes and stored at 4°C for quantification (Janos et al., 2008). The concentration of easily extractable glomalin (mg g^{-1}) was quantified according to the Bradford assay and bovine serum albumin (BSA) from Fermentas (# R1281) was used for the standards. BSA standard solutions with concentrations from 1.25 – 5.00 mg mL^{-1} were used. Duplicates of 5 μL of BSA standards, blank (water) as negative control and test samples were pipetted into 96 well microtiter plates. A volume of 250 μL of Bradford Coomassie brilliant blue dye (Bio-Rad) was added to each well and left at 25°C for 10 – 15 minutes to allow for colour development. Absorbance was measured using a spectrophotometer (SpectraMax Plus 384) at 595 nm. Absorbance was corrected for the blank and the values of the standards readings were calculated.

5.2.2.4 Soil microbial diversity

Whole-community substrate utilisation profiles (CSUP) were assessed when carbon sources were utilised. Soil samples were diluted with sterile distilled water and inoculated into Biolog EcoPlates™ (Biolog® Inc., Hayward, USA) that contained 31 carbon sources and a control, each with three replicates. The plates were incubated at 28°C. Respiration of carbon sources by microbial populations reduced the tetrazolium dye, causing a colour change which was measured twice daily over a period of 5 – 10 days at 590 nm to determine average well colour development. The functional diversity of soil microbial populations was determined by the amount and equitability of carbon substrates metabolised as indicators of richness and evenness. The Shannon-Weaver substrate diversity index (H') was used to quantify the functional diversity of soil microbial communities through the utilisation of different carbon sources on Biolog EcoPlates™. The substrate evenness index (E) was used to determine the variation between species in the soil microbial community (Magurran, 1988). The Evenness index (E) is a derivative of the Shannon-Weaver index and serves as an indication of how abundant species are within a soil microbial community. Evenness index values range between 0 and 1 and the closer the value is to 1, the more evenly distributed the different species in the microbial population.

5.2.2.6 Soil microbial activity

The ability of the present soil microbial population to obtain carbon, P (under low, and neutral to high pH conditions) and N, was assayed through measuring the activities of β -glucosidase, acid and alkaline phosphatase and urease activities in the soil. β -Glucosidase, acid phosphatase and alkaline phosphatase activities were calculated by determining the release of *p*-nitrophenyl after incubation of soil with *p*-nitrophenyl glucoside and *p*-nitrophenyl phosphate, respectively. Urease activity was determined where released ammonia was measured after the incubation of soil samples with a urea solution. Results were then calculated with reference to the standard calibration curve.

5.2.2.7 Statistical analyses

The data was subjected to an analysis of variance (ANOVA) using General Linear Models Procedure (VEPAC) of Statistika (version 13.5.0.17)(TIBCO Software Inc., 2018). Fixed effects were species, season and species season interaction and random effects were blocks. Shapiro and Wilk test confirmed normality of the standardised residuals (Shapiro and Wilk, 1965). Fisher's least significant difference (LSD) was calculated at the 5% level to compare treatment means (Ott and Longnecker, 2010).

5.3 Results and discussion

There was no interaction ($p > 0.05$) between the main factors, system (cover or no cover crop) and species oversown for the measured soil physical, chemical and biological parameters (Table 5.2). For this chapter of the study there is thus considered to be only twelve treatments i.e. canola, clover-medic mix, control, forage barley, forage radish, ryegrass-clover, *Trifolium* mix, diverse mix 1, diverse mix 2, black oats, stouling rye and Westerwolds ryegrass.

Table 5.2 Analysis of variance (ANOVA) results of the effects of the system applied (S1), i.e. soil cover or no cover, oversown species (S2) and system-species (S1xS2) interaction for soil parameters measured for the course of this study.

Parameter	System (S1)		Species (S2)		S1xS2 interaction	
	F-statistic	P-value	F-statistic	P-value	F-statistic	P-value
Aggregates (%)	0.25	0.652	0.83	0.606	1.15	0.341
Active C (mg kg ⁻¹)	2.38	0.220	0.58	0.837	1.27	0.261
Glomalin (mg g ⁻¹)	0.17	0.708	0.52	0.880	1.48	0.159
Shannon index (H')	0.04	0.853	0.63	0.793	1.02	0.442
Species evenness (E)	0.38	0.582	0.73	0.702	0.65	0.780
β-glucosidase (μg g ⁻¹ h ⁻¹)	0.22	0.672	1.29	0.251	0.84	0.603
Acid Phosphatase (μg g ⁻¹ h ⁻¹)	2.11	0.242	0.65	0.780	1.53	0.145
Alkaline Phosphatase (μg g ⁻¹ h ⁻¹)	0.01	0.939	0.98	0.476	0.54	0.869
Urease (μg g ⁻¹ h ⁻¹)	0.48	0.537	0.39	0.955	0.70	0.730

5.3.1 Aggregate stability

Aggregate stability ranged from 48.7% for forage radish to 60.1% for *Trifolium* mix (Table 5.3). The control had an aggregate stability of 54.5% and no treatment differed from the control ($p > 0.05$). Smith (2014) reported an aggregate stability of approximately 52% at a depth of 0 – 50 mm at Tygerhoek for a pure lucerne sward while Smit (2018) reported aggregate stabilities of 34% and 42% for mainly cereal and mainly leguminous mixtures, respectively at a depth of 0 – 150 mm. The aggregate stability recorded for all treatments of this trial may be higher than that recorded by Smit (2019) due to a number of reasons. Aggregate stability is sensitive to soil disturbance (Verhulst et al., 2010; Wright et al., 2007) and pasture phases will typically have less soil disturbance than cash crop or annual cover crop phases. The return of diverse biomass from legumes and grasses may also have promoted soil microbial activity in the current trial (Haynes and Beare, 1997) and the build-up of soil organic matter, which are both conducive for the formation of soil aggregates.

Soil aggregate stability was similar to those recorded by Smith (2014) in an eleven-year-old lucerne sward. This may indicate that the reduced soil disturbance will only promote aggregate formation to around 52 – 55% on Tygerhoek Research Farm where this study and that of Smith (2014) was conducted. Increased inputs of soil organic matter may be needed to promote further aggregate formation. It further seems that soil disturbance due to oversowing did not have an impact on soil aggregates. Oversowing either did not physically breakdown soil aggregates or the disc planter only disturbed the top few centimetres and this did not have an effect on samples that were taken down to 150 mm. The higher aggregate stability of *Trifolium* mix compared to forage radish ($p > 0.05$) may be due to increased inputs of belowground organic matter. Both treatments had similar oversown fraction contribution (Chapter 4), but the clover fraction may have been underestimated due to the

Table 5.3 Soil physical, chemical and biological measurements at a depth increment of 0 – 150 mm for year one of this study. Different letters indicate significant differences within columns ($p < 0.05$)

Treatment†	Physical soil properties	Chemical soil properties	Biological soil properties								
			Aggregates (%)	Active C (mg kg^{-1})	Glomalin (mg g^{-1})	Biodiversity		Enzyme activity ($\mu\text{g g}^{-1} \text{h}^{-1}$)			
						Shannon (H')	Evenness (E)	β -glucosidase	Alkaline Phosphatase	Acid Phosphatase	Urease
Canola	55.0 ^a	297.3 ^a	1.71 ^a	2.18 ^a	0.81 ^a	1748 ^a	1532 ^a	5962 ^a	111 ^a		
C-m mix	57.6 ^a	283.5 ^a	1.74 ^a	1.91 ^a	0.70 ^a	1548 ^a	1680 ^a	5672 ^a	89 ^a		
Control	54.5 ^a	292.3 ^a	1.71 ^a	1.99 ^a	0.70 ^a	1530 ^a	1442 ^a	4965 ^a	100 ^a		
Forage barley	55.4 ^a	280.8 ^a	1.88 ^a	2.10 ^a	0.74 ^a	1582 ^a	1301 ^a	5369 ^a	101 ^a		
Forage radish	48.7 ^a	316.2 ^a	1.73 ^a	1.89 ^a	0.68 ^a	1803 ^a	1572 ^a	5264 ^a	109 ^a		
R-c mix	53.1 ^a	300.9 ^a	1.42 ^a	2.01 ^a	0.76 ^a	1770 ^a	1492 ^a	6044 ^a	111 ^a		
<i>Trifolium</i> mix	60.1 ^a	320.6 ^a	1.54 ^a	2.06 ^a	0.72 ^a	1803 ^a	1715 ^a	5492 ^a	104 ^a		
Diverse mix 1	51.6 ^a	288.6 ^a	1.61 ^a	2.19 ^a	0.75 ^a	1750 ^a	1395 ^a	6187 ^a	106 ^a		
Diverse mix 2	56.5 ^a	303.1 ^a	1.51 ^a	2.03 ^a	0.70 ^a	1766 ^a	1492 ^a	5380 ^a	108 ^a		
Black oats	56.9 ^a	296.2 ^a	1.62 ^a	1.94 ^a	0.73 ^a	1641 ^a	1290 ^a	5904 ^a	97 ^a		
Stooling rye	56.4 ^a	299.9 ^a	1.62 ^a	2.20 ^a	0.79 ^a	1647 ^a	1306 ^a	5720 ^a	102 ^a		
Ww ryegrass	53.0 ^a	300.6 ^a	1.61 ^a	2.03 ^a	0.70 ^a	1420 ^a	1306 ^a	5076 ^a	98 ^a		

† C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass

morphology of clovers. The *Trifolium* mix may also have supported different microbial populations to that of forage radish and this could have contributed to the differences in soil aggregates (Haynes and Beare, 1997). The inputs of root biomass were not measured, but it is likely that annual oversown species would lead to higher below ground inputs of organic matter. The annual formation and breakdown of roots (Smith, 2014) along with the different microbial populations from a variety of different crops in lucerne-based multiple species pastures may promote aggregation over time (Smith, 2014; Verhulst et al., 2010).

5.3.2 Active Carbon

Active C content for this study was lower than values recorded by Smith (2014) for a 11-year-old dryland lucerne sward in the southern Cape. Active C for this study ranged from 280 mg kg⁻¹ to 320 mg kg⁻¹ (Table 5.3) with no differences between any treatments ($p > 0.05$). Smith (2014) recorded values ranging from 620 mg kg⁻¹ and 1700 mg kg⁻¹ at depths ranging from 0 – 200 mm. Differences in active C fraction may be due to seasonal variation (Haynes, 2005) as Smith (2014) took samples in winter while samples for this study were taken in late spring. Different inputs of above and below ground organic matter at sample collection may also have contributed to some of the variation (Smith, 2014). Active C is very sensitive to crop effects on soil organic matter and reflects short-term changes (Smith, 2014).

No differences between any treatments ($p > 0.05$) were likely due to similar herbage production for all treatments and the control ($p > 0.05$) (Chapter 4) and it is expected that treatments had similar inputs of soil organic matter. At this stage there were also two dominant species, lucerne and *Lolium* spp., leading to the quality of organic matter returned also being similar. Similar fractions of active C are thus explained by similar input of soil organic matter, both quantitatively and qualitatively.

5.3.3 Glomalin

Glomalin concentration ranged from 1.4 mg g⁻¹ to 1.9 mg g⁻¹ (Table 5.3) with no treatment being different to any other ($p > 0.05$). A glomalin concentration below 2.5 mg g⁻¹ is considered to be low (Sekgota, 2018). Glomalin concentration is linked to soil organic C (Wright et al., 2007) and the similar glomalin concentrations are not surprising due to similar quantitative soil organic matter inputs as well as similar active C content in the soil. A higher diversity of residue returned to the soil is however likely to lead to increased abundance of AM fungi (Daniell et al., 2001). The low glomalin concentrations indicate low AM activity in the soil.

Soil disturbance and monocultures (Daniell et al., 2001) will reduce the AM fungal populations, while low numbers of AM spores and absence of colonisation (Sekgota, 2018) may also lead to low AM abundance. It is unlikely that low AM abundance was caused by soil disturbance during oversowing for this study. Low AM abundance is likely due to a low number of AM spores and low colonisation. Inoculation of oversown species with an AM fungal inoculant may lead to increased AM fungal populations (Sekgota, 2018). Similar glomalin concentrations from different treatments were due to either the effect of the oversown fraction that was too small or not enough time was allowed for glomalin to increase (Sekgota, 2018).

5.3.4 Microbial diversity

A Principle Component Analysis (PCA) was attempted to demonstrate the effect of different treatments on the active bacterial diversity through carbon source utilisation (Table 5.4). There were no clear trends or groupings and a mere 7% of variation was declared by treatment factors. This means that there were other factors than those incurred by the treatments that determined microbial diversity. Results depicted the average carbon resource utilisation of the soil microbial populations present and indicate very few differences between treatments. This implies that there was little change in microbial population functioning when sampling took place.

Shannon-Weaver index values ranged from 1.9 to 2.2 for this study (Table 5.3) and there were no differences recorded between treatments ($p > 0.05$). Values typically range between 1.5 – 3.5 (Magurran, 1988) and values increase as both richness and evenness increase. The range of values fall within the normal range. Evenness values for this study ranged from 0.68 to 0.81 (Table 5.3) and there were no differences between any treatments ($p > 0.05$). The largest difference recorded ($p > 0.05$) was between canola (0.81) and forage radish (0.68). Although oversown fraction contributions were similar ($p > 0.05$), canola had a higher lucerne fraction ($p < 0.05$), while forage radish had a higher weed fraction ($p < 0.05$). The weed fraction consisted mainly of *Lolium* spp. (Chapter 4). This may indicate that if there is a larger ryegrass fraction in a lucerne sward, soil microbes may be more concentrated in certain areas of the soil.

Table 5.4 Carbon source utilisation of treatments at a sampling depth of 0 – 150 mm

Treatment†	Amines	Amino Acids	Carbohydrates	Carboxylic acids	Esters	Phosphorylated compounds	Polymers
Canola	0.96 ± 1.36	0.45 ± 0.25	1.02 ± 0.59	0.89 ± 0.41	2.96 ± 2.20	1.83 ± 2.39	1.16 ± 0.75
C-m mix	0.34 ± 0.37	1.10 ± 0.71	0.53 ± 0.52	1.56 ± 0.46	3.34 ± 3.09	0.30 ± 0.58	0.51 ± 0.46
Control	0.07 ± 0.14	0.89 ± 0.68	1.37 ± 0.98	0.91 ± 0.48	2.52 ± 1.84	0.33 ± 0.41	1.14 ± 0.92
Forage barley	0.17 ± 0.28	1.11 ± 0.84	0.75 ± 0.42	0.92 ± 0.54	3.14 ± 2.82	0.87 ± 1.68	1.40 ± 1.25
Forage radish	0.15 ± 0.20	0.54 ± 0.73	1.14 ± 0.85	1.45 ± 0.46	3.23 ± 2.96	0.55 ± 0.89	0.52 ± 0.47
R-c mix	0.01 ± 0.02	0.99 ± 0.85	0.87 ± 0.41	1.10 ± 0.53	3.19 ± 2.83	0.03 ± 0.05	1.46 ± 0.91
<i>Trifolium</i> mix	0.07 ± 0.14	0.52 ± 0.47	1.45 ± 0.69	0.71 ± 0.33	2.98 ± 2.14	1.04 ± 1.63	1.54 ± 2.13
Diverse mix 1	0.44 ± 0.53	1.34 ± 0.94	1.00 ± 0.37	0.84 ± 0.52	3.66 ± 1.75	0.91 ± 1.21	0.52 ± 0.45
Diverse mix 2	0.17 ± 0.28	0.59 ± 0.50	0.78 ± 0.47	1.04 ± 0.45	6.67 ± 5.06	0.82 ± 1.05	1.01 ± 1.01
Black oats	0.37 ± 0.53	0.77 ± 0.78	1.42 ± 0.36	0.92 ± 0.43	3.42 ± 1.81	0.51 ± 0.77	0.74 ± 0.70
Stooling rye	0.38 ± 0.69	0.50 ± 0.63	1.48 ± 0.51	0.97 ± 0.51	2.26 ± 1.98	0.66 ± 1.03	1.14 ± 0.73
Ww ryegrass	1.80 ± 3.98	0.72 ± 0.47	1.10 ± 0.55	1.01 ± 0.67	1.86 ± 1.33	0.96 ± 1.33	0.63 ± 0.40

† C-m mix = clover-medic mix, R-c mix = ryegrass-clover mix, Ww ryegrass = Westerwolds ryegrass

Increased pasture diversity leads to increased soil microbial diversity that will in turn lead to a higher functional diversity and the utilisation of different functional groups (Stephan et al., 2000). Pasture composition has a prominent effect on the functional composition of soil microbial communities due to different root exudates from different plant species (Garbeva et al., 2004; Stephan et al., 2000). The dominance of both lucerne and *Lolium* spp. at the time of sampling, likely overshadowed any possible effect that oversown treatments may have had. Similar pasture composition lead to similar plant community decomposition as well as quality and quantity of material available to soil microbes (Bissett et al., 2011). This may explain why no differences between treatments were recorded for microbial diversity.

5.3.6 Soil enzyme activities

Soil enzymes fulfil a crucial role in catalysing decomposition of soil organic matter and nutrient cycling in soils (Verhulst et al., 2010). The activities of four enzymes were assayed to evaluate ecosystem functions. β -glucosidase, alkaline and acid phosphatase and urease were analysed.

β -glucosidase ranged from 1420 $\mu\text{g g}^{-1}\text{h}^{-1}$ for Westerwolds ryegrass to 1803 $\mu\text{g g}^{-1}\text{h}^{-1}$ for forage radish (Table 5.3). The control treatment had β -glucosidase activity of 1530 $\mu\text{g g}^{-1}\text{h}^{-1}$ and no treatment was different to that of the control ($p>0.05$). The β -glucosidase activity for this study were lower than activity recorded by Swanepoel et al. (2014) on irrigated kikuyu-ryegrass pastures in the southern Cape.

Urease activity ranged from 89 $\mu\text{g g}^{-1}\text{h}^{-1}$ to 111 $\mu\text{g g}^{-1}\text{h}^{-1}$ (Table 5.3) and no differences were recorded ($p>0.05$). Swanepoel et al. (2014) recorded higher urease activity that ranged from 122 – 269 $\mu\text{g g}^{-1}\text{h}^{-1}$ at a depth increment of 0 – 200 mm in irrigated kikuyu ryegrass pastures in the southern Cape.

The activity of the assayed enzymes was used as an indicator of the potential of the soil microbial communities to convert substrates from their organic form to plant available macro- and micronutrients. The higher the enzymatic activity, the faster the turnover from organic substrates to plant available nutrients. While there were some differences between different treatments for β -glucosidase and alkaline phosphatase, no clear trends in microbial activities could be identified

5.4 Conclusion

The lack of clear trends or increased aggregate stability, active C and microbial activity after year one of this trial is not surprising. Changes in soil characteristics may take several years in Mediterranean conditions (Smith, 2014). There were no differences recorded for active C and glomalin, both of which

may contribute to the formation of soil aggregates (Smith, 2014; Verhulst et al., 2010; Wright et al., 2007). Soil biological measurements that included enzymatic activity, functional diversity and diversity indices did not show clear and obvious trends and no differences were recorded ($p > 0.05$). Due to dry conditions, oversown species did not establish well and subsequently could not form dense and diverse root systems. Lucerne and *Lolium* spp. dominated all treatments regardless if it was oversown or manifested as weeds (Chapter 4). This led to treatments that were possibly too similar and the oversown fractions were overshadowed by the dominant species. Due to the absence of clear trends, it is recommended that soil characteristics be monitored over an extended period of time to obtain a better understanding of impact of lucerne-based multiple species pastures on aggregate stability (objective 1), soil C dynamics (objective 2) and soil microorganism activity and diversity (objective 3).

5.5 References

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Chapter 6

Conclusion and Recommendations

6.1 Synopsis

The Mediterranean climate of the southern Cape of South Africa is conducive to the cultivation of wheat (*Triticum aestivum*), barley (*Hordeum vulgare*), canola (*Brassica napus*) and lupins (*Lupinus* spp.) in crop rotation systems (Smith, 2014). Lucerne (*Medicago sativa* L.) swards are typically incorporated as pasture phases to form long-rotation cropping systems (van Heerden and Tainton, 1987). The cash crop phase as well as the pasture phase normally spans over a five to seven year period. Lucerne swards allow for diversification and the inclusion of livestock (Bell et al., 2014), is a high quality animal forage (Bouton, 2012), allows for the utilisation of out of season rainfall (Hayes et al., 2010), biologically fixes N (Carlsson and Huss-Danell, 2003), may improve soil structure (Durand, 1993), break disease cycles (Palm et al., 2014) and may improve subsequent cash crop yields (Peyraud et al., 2014). Despite all the possible advantages of utilising lucerne in long-rotation cropping systems, some farmers have considered excluding lucerne from their crop rotations. The main reasons for the exclusion of lucerne swards are low summer and winter herbage production due to moisture stress (van Heerden, 1976) and lucerne's natural winter dormancy (Malinowski et al., 2007). Low herbage production during these periods create considerable fodder flow deficits and make the management of fodder flow programmes challenging for farmers. Lucerne also has a poor reseeding ability due to autotoxicity. Thus, if a sward is damaged or plant population is reduced, the sward will not recover and may not be financially viable.

A study was conducted at Tygerhoek Research Farm to determine if the oversowing lucerne swards with annual winter growing crops, to create lucerne-based multiple species pastures, was a viable option for farmers to make the lucerne phase more sustainable. Sustainability was defined through the following three objectives.

6.1.1 The pasture must have the same or higher yield when compared to a pure lucerne sward

There was no interaction ($p < 0.05$) between the main factors regarding herbage production and therefore the main effect that refers to the species or species mixes oversown into the lucerne base is discussed.

No treatment was different from the control at any stage during the growing season from April to October. Low herbage production that farmers typically experience from early to mid-winter, June and July, was not abridged by lucerne based multiple species pastures. Various factors may have resulted in poor performance by oversown species. Drought conditions prevailed after oversowing in both seasons and was likely the largest contributor to poor establishment by oversown species in this study. MacLaren et al. (2019) also reported late rains in 2016 and 2017, which means that from at least 2016 onward, the performance of oversown species in lucerne-based multiple species pastures would likely have been disappointing due to poor rainfall after oversowing. Four consecutive failed seasons would be concerning for farmers and bring the viability of lucerne based multiple species pastures into question. Competition from the already established lucerne base also makes oversown species more vulnerable to drought conditions than the same species planted in monoculture (Bell et al., 2014; Harris et al., 2007). Lucerne will dry out the soil profile and has an established root system that competes fiercely with newly emerged seedlings. Due to poor initial establishment of the oversown species, it was challenging to determine what the possible effect of other factors may have been.

Allelopathic chemicals secreted by the lucerne may have been a contributing factor to poor establishment of some of the oversown species, but there could not be distinguished between the impact of allelopathy and competition at such an early stage. The effects of allelopathy on oversown species may be better understood if the trial is replicated in years with normal rainfall distribution.

Poor initial establishment affected the performance of the oversown fraction throughout the growing season, however, some oversown species and mixes showed potential to reduce fodder flow deficits, especially in late winter (August). When long-term average rainfall is considered, oversowing is viable from late April to early May. Oversowing from late April to early May is likely too late for oversown species to make a meaningful contribution to pasture herbage production in mid-winter as a result of plants that are still relatively small. The oversown species are likely to only be large enough to significantly contribute to herbage production from August onward. The maximum benefit of lucerne-based multiple species pastures will be realised if the oversown fraction can contribute to the lucerne fraction, rather than replace it (van der Colf et al., 2015). Yield penalties may be expected for both the lucerne base and oversown species (Purser, 1981; van Heerden, 1990), however, the combined production of these two fractions may however be higher than in pure lucerne swards.

It was not possible to distinguish between oversown and volunteer ryegrass and all ryegrass was considered part of the oversown fraction in treatments where ryegrass was oversown (ryegrass-clover mix, diverse mix 2 and Westerwolds ryegrass). As a result, the oversown fraction was higher and weed

fractions were lower in ryegrass containing treatments when compared to all other treatments. In lucerne swards, it seems that high ryegrass fractions replaced the lucerne base rather than complement it (Brand, 1996; van Heerden, 1990) and similar observations were made in this study. Black oats (*Avena strigosa* L.), forage barley (*Hordeum vulgare*) and diverse mix 1 (*Avena strigosa* L., *Raphanus sativus* L., *Vicia villosa*) showed some resilience toward ryegrass invasion and have the potential to complement rather than compete against the lucerne base. Oversowing or intercropping, as it is known in Australia, of small grains has shown potential to increase farm productivity (Humphries et al., 2004). Poor establishment does, however, make it challenging to comprehend what the potential of the small grain containing treatments were and further research in South African conditions is required. Treatments that did not contain ryegrass as an oversown species may also have been suppressed, at least to some degree, by volunteer ryegrass (weeds), as volunteer ryegrass likely competed for already limited resources. The growing season and morphology of some oversown species, like clovers and medics may also have inhibited them from making a reasonable contribution to herbage production due to directly competing with the lucerne base.

In Spring, October, no treatment was different from the control, although herbage production of all treatments peaked and followed a similar pattern as described by Purser (1981) and van Heerden (1990). The trends observed in spring regarding the oversown fraction contribution and weed invasion were similar to those described for late winter. Grazing management may however have had an impact on the performance of the oversown fractions. Grazing management was tailored to the lucerne base (Oberholzer et al., 1993) and it is possible that it had a negative effect on herbage production of some of the oversown fractions in spring (Dove et al., 2015). Of the small grain treatments, diverse mix 1 and black oats had an increased oversown fraction contribution while the forage barley oversown fraction decreased. This was likely due to some of the forage barley plants that had already started to enter into the reproductive phase as opposed to diverse mix 1 and black oats that was still mainly in the vegetative phase when late winter grazing commenced. This may also explain why some forage barley plants (still vegetative) recovered after grazing (Bell et al., 2014), while those that had already reached the reproductive stage did not (Radcliffe et al., 2012).

Following the growing season, in late spring, the canola treatment produced more herbage than the control ($p < 0.05$). This could however not be linked to any specific fraction contribution as all fractions were similar to that of the control ($p > 0.05$). By mid-summer (January), the control yielded the highest herbage ($p > 0.05$). Some of the ryegrass treatments tended to produce less herbage than the control in the subsequent autumn, although differences were not significant. This may indicate that herbage production can be lower if there is competition for limited resources in the previous growing season.

The three ryegrass-containing treatments also had the three lowest total herbage yields, however, they were not different from the control ($p > 0.05$). Only one data measurement was taken in season two due to dry conditions that led to crop failure of oversown species. The oversown fraction initially followed a similar fraction contribution as in season one before falling victim to drought conditions.

Other factors to consider were the lucerne cultivar used, dormancy class and age of the lucerne base. The lucerne cultivar used for this trial was L70 with a dormancy class of 7. It is possible that lucerne a more dormant lucerne cultivar will be less competitive and lead to better performance by oversown species (Humphries et al., 2004). The trial was conducted in year two and three of the lucerne phase and plant populations may have been too high for oversown species to establish well as a result of competition from the lucerne base. It is likely that oversown species will perform better and have a higher chance of increasing total herbage production in older lucerne swards (Egan and Ransom, 1996; Harris et al., 2008). The successful establishment of oversown species is vital to ensure the success of lucerne-based multiple species pastures. Long-term rainfall averages suggest that oversowing will likely only be viable from late April to early May and onward. It is thus unlikely that lucerne-based multiple species pastures will reduce the fodder flow deficit in early winter. There is potential for lucerne-based multiple species pastures to make meaningful contribution in late winter.

6.1.2 Pasture quality must not be compromised by the inclusion of winter annual crops.

The effect of twelve oversown species and mixes on pasture quality was determined in later winter and spring in the southern Cape. For this study, pasture quality was defined according to ME, TDN, CP, NDF and ADF. Interaction was recorded between the season and oversown species or mixes for all of the quality parameters.

Various factors within a lucerne-based multiple species pasture may affect pasture quality (Scholtz et al., 2009). In this study, the species composition, physiological stage and weed invasion were the main determining factors regarding pasture quality. Drought conditions after oversowing resulted in poor performance from oversown species, except for ryegrass. The lucerne base and ryegrass (either as an oversown species or as a weed) were the dominant species and dominated the effects of the other oversown species. This led to pasture composition from the various treatments that were relatively similar. While there were differences recorded between treatments for quality parameters, no treatment could be identified to be of superior or inferior grazing quality. Grazing management ensured that all treatments were of a relatively high quality due to plants being in a young physiological stage.

High values were recorded for CP (Avci et al., 2018; Brand, 1996; Karayilanli and Ayhan, 2016), TDN (Karayilanli and Ayhan, 2016) and ME (Karayilanli and Ayhan 2016). Acid-detergent fibre (Brand, 1996; Scholtz et al., 2009) and NDF (Avci et al., 2018; Brand, 1996; Karayilanli and Ayhan, 2016; Scholtz et al., 2009) were low. The young physiological stage of the lucerne base and ryegrass due to grazing management (Machado et al., 2007) likely resulted in high CP, TDN and ME (Humphries, 2012) and low NDF and ADF (Deak et al., 2009). A contributing factor toward high CP was that the lucerne base was still relatively young, as CP has been known to decline in older lucerne swards (Coruh and Tan, 2008; Testa et al., 2011). The relatively low NDF range can be linked to the high legume fractions from the lucerne base. Increased legume fractions typically result in reduced NDF and increased grass fractions will increase NDF (Deak et al., 2009). Although species diversity may lead to low levels of ADF (Deak et al., 2009), the low ADF in this study was likely due to the lucerne base that was in a young physiological stage (Karayilanli and Ayhan, 2016).

6.1.3 The soil quality must improve or at least maintained by the end of the lucerne phase

There was no interaction ($p < 0.05$) between the main factors for soil physical, chemical or biological parameters and therefore the main effect that refers to the species or species mixes oversown into the lucerne base is discussed.

There were no differences recorded for any soil physical, biological or chemical parameters measured between treatments throughout the course of this study. The soil physical and chemical parameters measured included aggregate stability and active C. Aggregate stability was similar to values previously recorded on Tygerhoek Research Farm in older lucerne swards. This may indicate that aggregate stability will reach an equilibrium of around 52 to 55% on Tygerhoek Research Farm and higher inputs of organic matter may be needed to further increase aggregate stability. Aggregate stability was higher than values recorded for annual pastures in the Western Cape and this was likely due to reduced soil disturbance in perennial pastures. Active C in the current study was lower than values previously recorded on Tygerhoek Research Farm. This was likely due to a combination of seasonal variation (Haynes, 2005) as active C reflects short term changes in soil (Smith, 2014). The soil biological parameters measured included glomalin, microbial diversity and enzyme activities. There were no clear trends for any of the biological parameters.

Due to drought conditions, the establishment of oversown species was poor. This led to little return of organic matter, both above and below ground, to the soil from oversown species. The lucerne base and ryegrass, both as an oversown species and weed, dominated the pasture at the time of sampling. Treatments were thus very similar in terms of herbage composition at the time of sampling. The

oversown species that were present, did not constitute a large enough part of herbage production to meaningfully influence soil biological parameters. Soil characteristics may also take several years to change in Mediterranean conditions (Smith, 2014) and should be monitored over several seasons to be fully understood. If oversown species make up a larger fraction of herbage production, different results may be obtained over time.

6.2 General conclusion

The sustainability of the lucerne phase in long-rotation cropping systems was neither improved nor reduced through oversowing the lucerne base with annual winter growing crops according to the criteria set out for this study. Therefore, no treatment can be excluded, however some treatments showed more potential than others.

Drought conditions after oversowing resulted in poor establishment of oversown species and was the most determining factor regarding herbage production as it led to poor performance of oversown species. Despite of this black oats, forage barley and diverse mix 1 showed the most potential to improve herbage production, especially in late winter. Ryegrass, both as an oversown species and weed, competed fiercely with lucerne and oversown species and replaces the lucerne base, rather than complements it. Stooling rye and all treatments that consisted of only brassicas, clovers and medics showed little potential in this trial. Viable oversowing dates according to the long-term average rainfall will likely enable oversown species to contribute to herbage production in late winter when lucerne production is still suppressed due to its winter dormancy. Data could only be collected over a single full growing season and research over an extended period of time is needed to make conclusive findings.

Poor establishment of oversown species and dominance from the lucerne base and ryegrass, both oversown and as a weed also impacted forage and soil quality. Grazing management allowed for a high-quality pasture in winter and spring. It was however not possible to determine if any treatment reduced or improved pasture quality as treatment were likely too similar at the time of sampling. Dominance from the lucerne base and ryegrass, both oversown and as a weed also resulted in similar organic matter returns to the soil both in quality and quantity. The effect of the oversown fraction on soil quality was subsequently overshadowed.

6.3 Limitations

Drought conditions in both years of this study was the single most limiting factor as it resulted in poor establishment of oversown species in both years. Poor establishment has a knock-on effect that continues throughout the growing season and dilutes the effects that oversown species may have had on herbage production, pasture and soil quality. Due to drought conditions, data could also only be collected over a single full growing season. Lucerne-based multiple species pastures should be monitored over an extended period of time to fully understand how herbage production, pasture quality and soil quality will be affected.

Due to management constraints, it was not practically viable to apply different grazing regimes to different treatments. It is possible that different grazing regimes may result in better performance by some of the oversown species.

There was also a buffer zone around each block that could not be fenced off. These buffer zones were not of the same size due to the shape of the paddock where the lucerne base was established in 2017. To compensate for this, it took sheep different amount of time to graze the pasture to an approximate height of 50 mm. The increased amount of time spent in blocks with larger buffer zone would enable animals to select for more palatable forage within that block.

6.4 Future research

Further research is required to determine how oversown species will establish in years of normal rainfall distribution and how this will affect herbage production, pasture and soil quality. Other factors that could not be monitored in this trial include the effect of the lucerne cultivar, sward persistence, lucerne dormancy class, the allelopathic effect on oversown species, the effect of different seeding rates of oversown species, grazing management or a combination of these factors and requires further investigation in South African conditions. If there is better performance by oversown species and they make larger contributions toward herbage production, the effects on pasture quality should also be monitored. Due to the absence of clear trends regarding soil quality, it is recommended that soil characteristics be monitored over an extended period of time to obtain a better understanding of the impact of lucerne-based multiple species pastures on aggregate stability, soil C dynamics, soil microorganism activity and microorganism diversity.

6.5 References

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