DEVELOPING A MODERN AGRONOMY FOR SAINFOIN

(*Onobrychis viciifolia* Scop.)

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A thesis submitted in partial fulfilment of the University's requirements for the Degree of Doctor of Philosophy

September 2006

Coventry University
in association with
Royal Agricultural College

DECLARATION

The work within this thesis is based on the author's independent study at the Royal Agricultural College, Cirencester, under the supervision of G. P. F. Lane, Prof. W. P. Davies and Dr. R. N. Baines. The author is responsible for the experimental work, the results and the conclusions in this thesis. All assistance and advice received from colleagues has been acknowledged.

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September 2006

ABSTRACT

Exceptional animal performance and non-bloating characteristics make sainfoin a potentially valuable forage crop. Research information in the UK is very limited and the crop is in severe decline. The requirement to produce home grown protein and EU policy on fertiliser reduction has created a climate for legume developments. Yields of up to 15 t DM ha^{-1} in recent studies have illustrated that sainfoin has good potential and presented the case for developing a modern agronomy for the crop.

Effects of sowing depth and seed pod on emergence were examined under greenhouse conditions. Sainfoin can be sown at 1-4 cm depths as either hulled or dehulled seed without significant differences. Seedpods seemed to check emergence at 6 cm depth, but to assist emergence from surface placement. Sowing date and autumn management was also investigated in field conditions. April to July sowings gave similar yields over three years averaging about 8 t DM ha^{-1}. May sowing yielded up to 9.8 t DM ha^{-1} when established in favourable conditions. Autumn management had less effect than anticipated and early autumn cutting only reduced yield in the 3rd year. This study also explored mixtures of sainfoin with meadow fescue or tetraploid perennial ryegrass direct sown or undersown in spring barley. Undersowing reduced yields of sainfoin-grass mixtures in the establishment and 2nd years, but not in the 3rd year. Average yields of sainfoin and sainfoin-grass mixtures over three years were again, about 8 t DM ha^{-1}. Tetraploid perennial ryegrass cv. Condesa seemed more compatible with sainfoin than meadow fescue cv. Lifara in this study. An assessment of eight varieties over two years gave no significant yield differences, with an average of about 12 t DM ha^{-1} in the 2nd year. However, cv. Sombourne, uniquely, showed a quicker regrowth after harvest in the 2nd year.

A competition study to explore in detail the interaction between sainfoin and meadow fescue or perennial ryegrass was conducted in plastic containers. Root competition had more effect than shoot competition on competitive ability. Intraspecific competition of tetraploid perennial ryegrass was greater than interspecific competition. Intraspecific competition of meadow fescue and sainfoin was less than interspecific competition. Sainfoin grown with meadow fescue cv. Rossa was more competitive than with tetraploid perennial ryegrass cv. Condesa at a 1:2 ratio.

The study concluded that sainfoin is capable of giving moderate yields of high quality forage over two to three year periods in UK conditions. Strategies to enhance its persistency need to be better developed.

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ACKNOWLEDGEMENTS

I wish to express my gratitude and appreciation to Gerry Lane, my supervisor, for his guidance, advice and supervision throughout the period of study. I also would like to thank his patience and constant encouragement during this study.

I also wish to thank Prof. Paul Davies, for his supervision and encouragement throughout the study. All advice from him was critical and constructive. I also would like to thank Dr. Richard Baines who guided me through the competition study.

I also want to thank Sylvia de Oliviera, who helped me analyse plant samples, and staffs in the Rural Skill Centre and College Farm, who helped me prepare experimental plots. I am also grateful to Robin Hill, a sainfoin enthusiast, Chairman of Cotswold Seed Ltd, for his support to this study. I also thank many Chinese students at RAC, who helped me sowing and harvesting over three years.

The Royal Agricultural College provided facilities and support for my study. Thank you, RAC! I am always a RAC man! Many RAC staffs also helped and concerned my study directly and indirectly, thank you all!

The 100 Club of Royal Agricultural College, the Forage Legumes Special Interest Group of the British Grassland Society, and Cotswold Seed Ltd funded this study.

Finally I would like to thank my wife and daughter, the supporters behind forever.

Chapter One

General Introduction

> *"Sainfoin is something of an agricultural paradox; from the point of view of animal nutrition it seems to be the most desirable of all forage legume plants; from an agronomic point of view it is an undesirable plant because it doesn't grow very well."*
>
> Dr. J. E. Sheehy 1982

1.1 Sainfoin in the UK

Sainfoin (*Onobrychis viciifolia* Scop.), also known as St Foin, cock's head, holy grass, esparcette or French grass has been cropped for hundreds years in many parts of the world (Piper, 1914; Bland, 1971; Frame, Charlton & Laidlaw, 1998), including Asia, Europe and North America. It was first cultivated in southern France in 1582 and its culture described in 1629, following which it spread over Europe (Piper, 1914). It was introduced to North America in 1900 (Goplen, Richards & Moyer, 1991) where it is grown in western USA and Canada (Miller & Hoveland, 1995). Today, particularly in Eastern Europe, Italy, Spain, Iran and Turkey (Delgado, Andros, Sin & Ochoa, 2005; FAO, 2006), sainfoin is still being cropped. It seems especially popular in Turkey, where about 93,000 ha were reportedly grown in 1998. As Sheehy (1982) described above, sainfoin appears to be a difficult crop. As a leguminous forage crop with high nutrient value, it is favoured by animals; but its cultivation is not well understood and its potential advantages such as non-bloating have not yet been fully exploited.

Sainfoin became a traditional crop in the UK. It was cropped in the 17th, 18th, 19th and early 20th century in many areas of Britain, including the south and southeast of England, south Wales, north to the Humber and west to the river Severn. It was often linked with the chalky land of southern England and the low rainfall areas of East Anglia (Robinson, 1937; Bland, 1971). Its first introduction to the UK is not clear, but was reported in the 17th century. Hartlib (1652) recorded sainfoin as follows:

> *'I have seen it sown in divers places here in England, especially in Cobbam-park in Kent, about 4 miles from Gravesend; where it hath thriven extraordinary well upon along chalky banks, where nothing else would grow; and indeed much dry barren land is most proper for it or clover-grass and when the other grasses and plants are destroyed by the parching heat of the sun'*

The book "The English Improver Improved" published in 1652 also recorded sainfoin, *'there is thousand thousands of acres in England'*. Jethro Tull (1733) recorded sainfoin in his book "Horse Hoeing Husbandry" and Arthur Young (1813) described sainfoin in 'General View of the Agriculture of Oxfordshire' as well. Davies (1815) recorded that sainfoin was in large-scale production on the limestone soils of the Vale of Glamorgan in the early 19th century. Stephens (1819) stated that sainfoin proved a most useful and reliable forage on the calcareous soils of the southern counties of England. At the Royal Agricultural College, Cirencester, a sainfoin leaf was sculptured on the tower of the main building, which was built in 1845; and sainfoin was described as well in the Journal of Agricultural Students Gazette (1882-86) edited by students at the Royal Agricultural College. Rees (1928) commented on the spread of sainfoin in south Wales and recorded twenty-six farms in Glamorgan growing it during the late 1920s.

Sainfoin has been a minor crop and not recorded separately in British agricultural statistics. There is no record of the exact area cropped in the past, but there is no doubt that it was considered a superb forage no matter what status it has today. Jethro Tull (1733) indicated that sainfoin was a forage well known for its "wholesomeness". Arthur Young (1813) appraised sainfoin

> *"The merit of Oxfordshire farming is more conspicuous on account of sainfoin on all soils that are proper on it."*

Since the 1920s, sainfoin has experienced a constant decline (Bland, 1971; Sheldrick & Thomson, 1982). It is recorded that about 150 tonnes seeds were sold every year in late 1950s, enough for 2023-2428 hectares (Hill, 1997). In the late 1970s only approximately 150 hectares were cropped (Sheehy & Popple, 1981). A figure from NIAB showed that there were only 5 tonnes seed sold in 1982-1983 (Aldrich, 1984), sufficient for about 50 hectares.

A general discussion on sainfoin was held at the Grassland Research Institute, Hurley, in 1982 where a pamphlet, "The Future of Sainfoin in British Agriculture" was produced. Since then very little attention seems to have been paid to its development, although there are a number of farmers still interested in cropping sainfoin today, but without more modern agronomic recommendations. Doyle, Thomson and Sheehy (1983) did an economic assessment of sainfoin's potential future in British agriculture. It was estimated that sainfoin could potentially be grown on 950,000 ha in England and Wales, but it was unlikely to exceed 20,000 ha in the near future. To be more widely grown, it was suggested that the sainfoin yield needed to be increased by 35%, to about 11.5 t DM ha^{-1}. Under experimental conditions, yields of about 14-16 t DM ha^{-1} have been achieved (Sheehy, Minchin & McNeill, 1984; Lane & Koivisto, 1998) in the UK, which indicate the possibility of achieving 12 t DM ha^{-1} yield in practical farming.

Today, sainfoin has become rare in the UK, being grown by only a few farmers. Hutchinson (1965) suggested that the cause of its decline might have been due in part to its poor response to the changing requirements and circumstance of British agriculture. Hill (1997) further explained that this may also have been due to the availability of cheap nitrogen fertiliser, improved varieties of perennial ryegrass from the 1950s, and the expansion and dominance of autumn cereal cropping from the 1960s. Rochon, Doyle, Greef, Hopkins, Molle, Sitzia, Scholefield & Smith (2004) also pointed out that the decline of forage legumes in Europe was in part due to the availability of cheap inorganic nitrogen fertiliser and the expansion of production based on it since the early 1970s. Borreani, Peiretti and Tabacco (2003) explained its decline in Italy as a result of agricultural structural changes, and the gradual disappearance of livestock farms in hilly areas. Newman (1997) stated that the virtual disappearance of sainfoin was because of the end of the use of hard working draught horses, for which it was a major feed. However, agronomic problems may be the main cause of decline since sainfoin is reported to be of low yield, low persistence and poor regrowth after the 1st cut, compared to lucerne (*Medicago sativa*) (e.g. Green, 1967; Sims, Muir & Carleton, 1968; Doyle *et al.*, 1983; Kallenbach, Matches & Mahan, 1996).

1.2 Forage Quality and Animal Performance

Sainfoin has the useful combined characteristics of being non-bloating and palatable with high crude protein (CP) and a high voluntary intake leading to good animal performance (Spedding & Diekmahns, 1972; Sheldrick & Thomson, 1982; Beever, Dhanoa, Losada, Evans, Cammell, & France, 1986; Karnezos, Matches & Brown, 1994).

The most favourable feature that sainfoin possesses is probably the presence of condensed tannins (CT) in its leaves. An important feature of condensed tannins is that it helps minimise the degradation of protein in the rumen, thereby resulting in less fermentation and a good absorption of amino acids in the small intestine (Jones & Mangan, 1977; Waghorn, Jones, Shelton & McNabb, 1990). As a result, it does not cause ruminant animals to bloat (Sheldrick & Thomson, 1982; Beever *et al.*, 1986). Adding fresh sainfoin forage as a supplement to lucerne-based pasture helped to prevent bloat in cattle (McMahon, Majak, McAllister, Hall, Jones, Popp & Cheng, 1999). Condensed tannins have also been shown to limit proteolysis during ensiling

(Albrecht & Muck, 1991; Salawu, Acamovic, Stewart, Hvelplund & Weisbjerg, 1999). It can also reduce nematode parasites in sheep (Athanasiadou, Kyriazakis, Jackson & Coop, 2000). Sainfoin's crude protein content ranges from 17-25%, depending on growth stages and it has a low content of cell walls (Spedding & Diekmahns, 1972). Voluntary intake is also high, 20-40% more than grasses with the same digestibility. Osbourn, Thomson & Terry (1966) reported that the voluntary intake of sainfoin was higher than red clover (*Trifolium pratense*) and lucerne; the voluntary intake of sainfoin and red clover and lucerne being respectively 83 g kg^{-1} $W^{0.73}$ /24 hour, 74 g kg^{-1} $W^{0.73}$ /24 hour and 63.5 g kg^{-1} $W^{0.73}$ /24 hour. Sainfoin D-values range from 57% to 68% (Spedding & Diekmahns, 1972; Sheldrick, Newman & Roberts, 1995; Zarb, 2000). Thomson (1976) also reported that sainfoin had higher voluntary intake than lucerne, red clover, S24 and S22 rye grasses.

Sainfoin was often fed to heavy working horses and sick animals in the past. Jethro Tull (1762) described animals' preference for sainfoin hay in his book of Tull's Husbandry as follows:

> *"…has kept a team of working store-horse, round the year, fat without corn, and when tried with beans and oats, mixed with chaff, refused it for the hay. The same fattened some sheep in the winter in a pen, with only it and water; they throve faster than other sheep at the same time fed with peas and oats."*

This has since been testified by a series of more recent trials. A study conducted at the Grassland Research Institute at Hurley in 1980s showed that lambs fed sainfoin gained more live weight than those fed lucerne or perennial ryegrass (*Lolium perenne*). Live weight gains of lambs fed with perennial ryegrass, lucerne and sainfoin were 193 gram day^{-1}, 257 gram day^{-1} and 288 gram day^{-1} respectively (Sheldrick & Thomson, 1982). Karnezos, Matches & Brown (1994) also found lamb production by sainfoin and lucerne (822 kg lamb ha^{-1} and 795 kg lamb ha^{-1}

respectively) was higher than wheatgrass (*Agropyron elongatum)* and wheatgrass-sainfoin (533 kg lamb ha^{-1} and 658 kg lamb ha^{-1} respectively). Hart and Sahlu (1993) in an earlier study found that yearling Angora goats grazing sainfoin gained more weight and produced more mohair than goats grazing lucerne. Ulyatt (1981) assessed the feeding value of several forage species and found that sainfoin was 61% better than perennial ryegrass, and ranked second only to clover for promoting lamb growth. Feeding trials in Montana University in the USA indicated that sainfoin was equivalent to lucerne in a pig diet, and pasture trials showed that cattle and sheep preferred sainfoin to other leguminous forage (Cash, Bowman & Ditterline, 1993). Recent research in New Zealand showed than condensed tannins increased wool growth (Min, Fernandez, Barry, McNabb & Kemp*,* 2001).

1.3 Sainfoin in Sustainable Agriculture

Forage legumes are of importance in agriculture. Through the symbiotic association with *Rhizobium spp* they can convert atmospheric nitrogen into protein and enrich the nitrogen content of soil (e.g. Frame *et al.,* 1998; Newman, 1997). They also can enable high rates of livestock production through high voluntary intakes and the high net energy values of the forage (Thomson, 1976; 1977). In legume-grass pastures nitrogen fixed by legumes can be transferred to grass by grazing livestock and by the decomposition of dead legume plants (Sprent, 1996; Evers, 2006), and thus reduce pasture's demand for inorganic nitrogen fertiliser. This can significantly benefit, therefore, low-input agricultural systems. For example, the productivity of white clover (*Trifolium repens*) -grass pasture without inorganic nitrogen fertiliser has been assessed as similar to that of grass pasture with 200 kg ha^{-1} of inorganic nitrogen fertiliser (Davies & Hopkins, 1996), and the UK livestock industry annually benefits by an estimated £300 million pounds, from the conversion of inorganic nitrogen fertiliser based grass pasture to clover-based system without inorganic nitrogen fertiliser (Doyle & Bevan, 1996). Thus forage legumes and legume-based pastures can make a substantial contribution to sustainable agriculture (Davies & Hopkins, 1996; Sprent & Mannetje, 1996).

Following the rise of environmental concerns about agriculture from the European Union, policies have been formulated to reduce the use of nitrogen fertiliser in farmland to minimize nitrogen leaching to ground water (Directive 91/676/EEC, 1991). This policy further encourages the increased use of forage legumes in agriculture. Recent research shows that nitrogen losses are influenced more by the quantity of nitrogen circulating in the grassland system rather than by its source. However, losses in most nitrogen-based pastures are greater than in those based on white clover (Cuttle, Hallard, Daniel & Scurlock, 1992).

In the UK, since the crisis of Bovine Spongiform Encephalopathy (BSE) which resulted in a ban on the use of bovine animal proteins (meal and bone meal), animal feeding based on forage has been encouraged (Wilkins & Jones, 2000). Reductions in the use of nitrogen fertiliser, and the ban on the use of recycled ruminant animal proteins in feed, demands more vegetable protein-use and low-input sources in particular. The characteristics of high crude protein content and nitrogen fixation of particular legumes could help to fulfil these requirements (Chadd, Davies & Koivisto, 2002).

1.4 Botanical Characteristics of Sainfoin

Sainfoin is an erect or sub-erect plant and grows to a height of 40-100 cm or more (Robinson, 1937; Thomson, 1951b; Thomas & Davies, 1964; Bland, 1971; Spedding & Diekmahns, 1972; Frame *et al.*, 1998). It has many hollow stems, which develop from basal buds in a branched crown. The leaves are pinnate and have 10- 28 leaflets, borne in pairs on long petioles and with a terminal leaflet. The stipules are broad and pointed. The leaflet is thicker and has a higher weight compared with lucerne (Sheehy & Popple, 1981). The inflorescences are racemes on axillary stalks with about 80 pink flowers. The species is cross-pollinated. The seed is kidney-shaped and contained in a pod with a brown colour or dark brown if old. The 1000-seed weight of hulled and dehulled seed is about 20 g and 15 g respectively, greater than most other perennial leguminous seeds. The hulled seed is about 4.5 mm long and 3 mm broad. The root system consists of a deep taproot, and some main branches and many lateral roots.

Figure 1.1 Sainfoin plant

Sainfoin belongs to the genus of *Onobrychis*. Both *Onobrychis viciifolia* Scop. and *Onobrychis sativa* Lam. are used in the literature. Sainfoin was recognized and divided into two types-Common Sainfoin and Giant Sainfoin. Thomson (1938) studied the development of these two types in the establishment year and detailed them as follows:

Common or Single-cut Sainfoin (*O. sativa* var. *communis* (Ahlefed)) This type is long-lived and less vigorous in the establishment year, and reaches yield peak in third year. It does not form stem and flowers in the establishment year, and also does not form of stems and flowers after being cut in the subsequent year. The stands can generally survive 20-40 years, but some records say the plant can live 100 years (Piper, 1914).

Giant or Double-cut Sainfoin (*O. sativa* var. *bifera* Hort.) This type is short-lived, but grows rapidly compared with the common type. Its yield peaks in the year following sowing, and it lasts about two years. It sends up stems, forms flowers and sets seed in the establishment year. It may have sexual reproduction again with the formation of new stem material after cut in the establishment year.

However, some authorities (e. g. Frame *et al.,* 1998; Sheehy & Popple, 1981) in the UK have used *O. viciifolia,* and this name has been adopted for the purpose of this thesis.

1.5 Sainfoin Establishment

1.5.1 Climate and Soil Requirements

Sainfoin is adapted to a wide range of climatic conditions e.g. in Europe, North America, Asia, Australia and New Zealand, to neutral and alkaline soils of pH 6 or above, and also to dryland and irrigated areas, similar to lucerne. In the UK, it has been always linked with calcareous chalky or limestone soil and where it has been reported as growing well (Hartlib, 1652; Bland, 1971; Frame *et al.*, 1998). Sainfoin is not tolerant of waterlogging and the soil needs to be well drained (Sheldrick *et al.*, 1995). Studies conducted at the Grassland Research Institute, Hurley, showed that a thin and patchy sainfoin sward was encountered on clay with pH below 6, and that there were failures on alluvial sand with pH below 5 in the Thames Valley (Bland, 1971). Light or medium soil with pH 6 or above without waterlogging seems, therefore, to be preferred for sainfoin (Bland, 1971; Sheldrick *et al.*, 1995; Frame *et al.,* 1998).

1.5.2 Seed and Sowing

Sainfoin seed is generally bigger than other leguminous seed. Sainfoin seed size has been related to cotyledon area, seedling vigour and seedling growth in a number of studies. Sainfoin cotyledon area is highly correlated with seed size (Lin, 1963; {cited by Cooper & Fransen, 1974}). Cotyledons contribute 100% of total seedling photosynthate when the first leaf is unfolded, 54% when the first leaf is fully expanded and 18% when the second leaf is unfolded (Cooper & Fransen, 1974). Seed size appears to be of little importance with regard to seedling vigour except for samples with1000 seed weights below 19.5 g (Carleton & Cooper, 1974). Plants established from large seed had higher nitrogen fixation at most harvest dates up to 84 days (Dennis & Ditterline, 1996). It would appear, therefore, that selecting well-matured large seed is important for seedling growth and leads to quicker seedling emergence, more nodules and higher nitrogen fixation rates (Cash & Ditterline, 1996).

Sainfoin seed can be sown in hulled (with pod) or dehulled (without pod) forms. Dehulled seed is reported to have better germination than hulled seed (Finlayson, 1906; Zade, 1933 {cited by Bland, 1971}; Percival, 1936; Wiesner, Carleton & Cooper, 1968), but Chen (1992) reported that there was no significant difference in emergence in the field between hulled and dehulled seed. The seeds should be drilled or broadcast to a depth of 1.5-3 cm according to traditional experience (Sheldrick *et al*., 1995; Frame *et al*., 1998). Canadian experience suggested that the optimum depth was not more than 2 cm (Goplen *et al*., 1991) but Chinese experience suggested 4-5 cm (Chen, 1992). These different recommendations for optimum sowing depth probably reflect differences in soil texture and moisture availability.

Optimum seed rate was reported to be 80-120 kg ha^{-1} for hulled seed and 40-50 kg ha^{-1} for dehulled seed, to establish 70-150 plants m^{-2}and maintain stands at 50-60 plant m^{-2} according to traditional experience (Sheldrick *et al*., 1995; Frame *et al*., 1998). To help reduce weed ingression 4-6 kg ha^{-1} of meadow fescue or 1-2 kg ha^{-1} timothy seed was often added in the UK (Sheldrick *et al*., 1982). Density trials conducted in a greenhouse at the Grassland Research Institute, Hurley, indicated that 100 plants m^{-2} produced the maximum sainfoin yield in the establishment year and suggested a optimum seed rate of about 62.5 kg ha^{-1} assuming 80% germination (Sheehy *et al.*, 1984). However, 13-20 kg ha^{-1} for dryland hay and pasture and 34-40 kg ha^{-1} for irrigated hay were recommended under western Canadian conditions (Goplen *et al.*, 1991). A 140 kg ha^{-1} seed rate with 25 cm row spacing was suggested from a study in the former Yugoslavia (Cupina, 1999).

In the UK, sainfoin sowing normally took place between April and July when soil was warm enough for rapid seed germination and moisture abundant for seed absorption. Early spring sowing allows the crop a longer vegetative growing period to develop strong roots and shoots, and possibly ever to give a harvest in the establishment year. There is little evidence to show that April and May sowing results, however in better establishment compared with June and July sowing (Bland, 1971). Since sainfoin produces little vegetation in the establishment year, it was sometimes undersown in barley to bring in some financial return (Frame *et al.*, 1998).

The current practice of establishing legume-grass leys in August/September after a winter cereal harvest could be problematic for sainfoin, but spring sowing could result in poor yields in the establishment year. There is clearly a need to further investigate optimum strategies for establishing sainfoin in the context of modern mixed farming systems.

1.5.3 Seeds Mixtures

Legume-grass mixtures have many advantages over monocultures. Mixture yields are generally higher than that of the constituent grasses alone because it can achieve more efficient light utilization (Brougham, 1958), and the fixed nitrogen of legumes can be transferred to the grass (Sprent, 1996). It can also reduce weed encroachment and erosion and adds to stand longevity compared to monoculture (Droslom & Smith, 1976). Mixtures can also improve forage quality, such as *in vitro* dry matter digestibility (IVDMD), crude protein and Neutral Detergent Fibre (NDF) (Baylor, 1974; Sleugh, *et al.,* 2000). Mixtures can improve the seasonal distribution of forage, extend the peak of seasonal growth, and increase total production (Sleugh, Moore, George & Brummer, 2000). However, several studies have concluded that lucerne-grass mixtures offer little yield advantage over lucerne monocultures (Wilisie, 1949; Mooso & Wedin, 1990).

Sainfoin mixtures appear to have more production potential than monocultures. This has been testified in a number of studies. Traditionally sainfoin was sown with a reportedly non-aggressive companion grass, such as meadow fescue (*Festuca pratensis*) or timothy (*Phleum pratense* L.) and the addition of white clover to a sainfoin-grass mixture has also been suggested (Bland, 1971; Sheldrick *et al.,* 1995; Frame *et al.*, 1998).

Other companion crops were also tried in some countries. Sainfoin mixed respectively with Kentucky bluegrass (*Poa pratensis* L), red fescue (*Festuca rubra* L), black medic (*Medicago lupulina* L), ladino clover (*Trifolium repens*), birdsfoot trefoil (*Lotus corniculatus* L) and white clover (*Trifolium repens* L) were studied in Montana, USA over four years (Cooper, 1972). The birdsfoot trefoil-sainfoin mixture was most compatible and productive, whereas the ladino clover and white clover showed too much competitiveness. Sainfoin also grew well with Russian wild rye (*Psathyrostachys juncea*) in southwest Canada on dryland and with crested wheatgrass (*Agropyron desertorum*) (Hanna, Kozub & Smoliak, 1977; Kilcher, 1982). A study on sainfoin sown alone and mixed with tall wheatgrass (*Agropyron elongatum*), crested wheatgrass (*Agropyron desertorum*), and smooth brome grass (*Bromus inermis*) was carried out in Turkey and the results showed that sainfoin-grass mixtures had higher yields than sainfoin monoculture (Sengul, 2003). A study on sainfoin-lucerne mixture was also conducted in Canada, but here the composition shifted to lucerne dominance especially when they were drilled together (Jefferson, Lawrence, Irvine & Kielly, 1994).

There are relatively few sainfoin varieties in the UK. Well known British cultivars are cvs. Cotswold Common, Hampshire Common, Sombourne, Hampshire Giant and English Giant (Spedding & Diekmahns, 1972; Frame *et al.*, 1998). Comparative studies of yields are relatively few (Spedding & Diekmanhns, 1972; Sheehy *et al.*, 1984; Koivisto & Lane, 2001).

In the UK, sainfoin has been cropped for hay traditionally and sainfoin-grass mixtures have not been well studied. To re-establish sainfoin into modern grassland systems, a study of the selection of companion grasses, and how sainfoin interacts with them, is needed. More information is also needed on the relative yields and persistency of available sainfoin cultivars under the UK conditions.

1.5.4 Nitrogen Fixation

Sainfoin was generally reported to be insufficient in fixing nitrogen and has sometimes shown nitrogen deficiency symptoms in inoculated plants (Koter, 1965a; Sims, Muir & Carleton, 1968; Burton & Curley, 1968; Meyer, 1975). It can be cross-inoculated by *Rhizobium* species from sweetvetch (*Hedysarum sp*), crownvetch (*Coranilla sp*), purple and white prairie clover (*Dalea purpurea* and *Dalea candida*) (Burton & Curley, 1968; Peter, 2004). The true amount of fixed nitrogen is still unclear since the measurement of nitrogen fixation remains difficult (Larue & Patterson, 1981; Witty & Minchin, 1988). However an acetylene based estimate of sainfoin nitrogen fixation indicated about 146 kg ha^{-1} $year^{-1}$ (Sheehy & McNeill, 1988), compared to a range of 0-455 kg ha^{-1} $year^{-1}$ in grass-white clover swards (Whitehead, 1995).

Nitrate nitrogen is known to reduce nodulation as well as nitrogen fixation of legumes (Hartwig & Nosberger, 1996). Koter (1965b) found that low levels of inorganic nitrogen stimulated nitrogen fixation in sainfoin, but that high levels hindered it. Inoculated sainfoin with nitrate produced 20 to 30% more forage than inoculated sainfoin without nitrate, and a yield increase from nitrogen fertilizer was also reported by Sims *et al.* (1968), Meyer (1975) and Smoliak and Hanna (1975). This effect can also be observed in other forage legumes (Allos & Bartholomew, 1959; Hoglund, 1973; Peter, 2004). Hume (1985) found that the relative growth rate and nitrogen accumulation rate of inoculated sainfoin seedlings without added inorganic nitrogen were lower than seedlings provided with 210 mg $litre^{-1}$ of nitrate nitrogen, under greenhouse conditions. Although there was good nodulation activity and high nodule weight compared to other legumes, the application of 35 mg $litre^{-1}$ of nitrate nitrogen to inoculated sainfoin seedlings appeared to substitute for, rather than supplement, nitrogen fixation. However, Sheehy and McNeill (1988) found that there was no significant difference between the dry matter yield of sainfoin with or without nitrogen fertilizer application. Badoux' (1965) trial with giant sainfoin in Switzerland supported Sheehy and McNeill's result. He found that yield was not increased by the application of nitrogen fertiliser; on the contrary, there was a 4% reduction after a 90 kg ha^{-1} $year^{-1}$ treatment. Krall and Delaney (1982) found that the nitrogen fixation of sainfoin was superior to that of lucerne, and that sainfoin out yielded the lucerne.

Nitrogen fixation is linked with energy use (Schubert & Ryle, 1980). The reported insufficient nitrogen fixation of sainfoin may also be associated with energy supply. Sheehy and Popple (1981) found that sainfoin required gross photosynthesis of 258 kg CH_2O ha^{-1} day^{-1} compared to the 234.3 kg CH_2O ha^{-1} day^{-1} which lucerne required.

The energy requirement, in terms of respiratory CO_2 production, for sainfoin was 20 mol CO_2 per 1 mol N_2, but for lucerne and red clover it was 10 mol CO_2 per 1 mol N_2. The differences between sainfoin and lucerne in energy requirement may be due to their different leaf area indices (LAI). The LAI of lucerne is typically twice that of sainfoin, and sainfoin may, therefore, have less capacity to intercept sunlight and assimilate carbon. This may result in insufficient nitrogen fixation (Sheehy & Popple, 1981). Sainfoin translocated 9% of its photosynthate to the roots compared with 3% for lucerne (Sheehy & Popple, 1981). This may explain why sainfoin has good nodulation activity and a higher nodule weight compared to other legumes. Krall and Delaney (1982) found in a box study that sainfoin taproots contained 23.8% non-structural carbohydrate (NSC) and that of lucerne contained 33.9% NSC at the mature seed stage (Stage 9, Appendix1) for three year old plants, and sainfoin roots contained an average of 1.8 g plant^{-1} NSC, while alfalfa contained 6.9 g plant^{-1}. They considered that the lower NSC of sainfoin may be caused by greater nodulation and forage yield compared with lucerne, in contrast with the study of Sheehy and Popple (1981) and Hume (1985).The differences of NSC between the roots of sainfoin and lucerne may be because of their different LAIs.

The relationship between sainfoin *Rhizobium* spp. and soil nitrogen supply is likely to be an important area for future research.

1.6 Sainfoin Management

1.6.1 Weed Control

Sainfoin is usually considered to be a non-aggressive crop with slow regrowth after cutting, requiring it to be established with minimum competition from weeds. Weeds can have a crucial effect on sainfoin production in the establishment year. In a crop of sainfoin grown without herbicides, weeds yielded 98% of total yield from the first cut in the establishment year (Moyer, 1985).

Traditionally the addition of meadow fescue or timothy to sainfoin was a means to avoid weed ingress. Alternatively, undersowing sainfoin in spring barley may also suppress weeds during establishment. In the UK, weeds in sainfoin crops sown in the spring are mainly broad-leaved species and in the autumn chickweed *(Stellaria media)* is often severe. MCPA [a.i. 4-(4-Chloro-2-methyl-phenoxy) acetic acid] + MCPB [a.i. 4-(4-Chloro-2-methyl-phenoxy) butyric acid] has been applied in practice at the 1st trifoliate leaf stage of sainfoin (Stage 0, Appendix 1) to successfully control most spring germinating broad-leaved weeds, such as cleavers (*Galium aparine*), fat hen (*Chenopodium album*), groundsel (*Senecio vulgaris*) and red dead-nettle (*Lamium purpureum*), and carbetamide [(R)$^{-1}$-(ethylcarbamoyl) ethyl carbanilate] has been applied in winter successfully to maintain sainfoin swards free from grass weeds and chickweed (Sheldrick & Thomson, 1982; Frame *et al.*, 1998). The use of MCPA+MCPB for sainfoin seedlings was also recommended by Waddington (1978) and Moyer (1985), when broadleaved weed control was described as “fair” and it caused the least damage. Alternatively, Stewart (1968) recommended bromoxynil [3, 5 dibromo-4-hydroxybenzonitrile], which gave excellent control of broadleaved weeds over three years. Benefin[N-butyl-N-ethyl-alpha, alpha, alpha-trifluoro-, 6-dinitro-p-toluidine] controlled most broadleaved weeds and grasses. 2-4-DB [a.i. 4-(2, 4-dichlorophenoxy) butyric acid] is reported to have caused moderate damage to sainfoin initially, but achieved fair to good control of weeds and left no yield reduction (Waddington, 1978). However, in contrast, Stewart (1968) reported that 2-4-DB was ineffective for weed control in sainfoin.

1.6.2 Pests and Diseases

Sainfoin is reported to be relatively free from serious pest and disease problems compared with other legumes (Goplen *et al.*, 1991; Frame *et al.*, 1998). In the UK, crown rot (*Sclerotinia trifoliorum*), powdery mildew (*Erysiphe trifolii*), verticillium wilt (*Verticillium albo-atrum*), sainfoin rust (*Uromyces onobrychidis*), chocolate spot (*Botrytis cinerea*) and leaf spot caused by fungi such as *Ramularia onobrychildis, Septoria orobina, Aschochyta onobrychidis* and *Pleospora herbarum*, have all been found (Hughes, 1945; 1949; Sheldrick *et al.*, 1995; Frame *et al.*, 1998). *Ascochyta fabae* blight on sainfoin has been found in Iran and Turkey (Sharifnabi, 1996; Eken, 2003). Sainfoin rust (*Uromyces onobrychidis*) was also reported in Iran (Sharifnabi, 1995).

Sainfoin is tolerant to pea and bean weevil (*Sitona lineatus.*) (Wallace, 1968; Goplen *et al.*, 1991; Morrill, Ditterline & Cash, 1998). Some other root feeding insects were also found, such as *Sitona scissifrons* in Montana, USA (Wallace, 1968). A number of insects damaging seed production have also been found, including sainfoin bruchid (*Bruchidius unicolor*) in the USA, sainfoin midge (*Contarinia onobrychidis*), sainfoin seed chalcid (*Eurytoma onobrychidis*), *Perrisia onobrychidis, Apion pisi* L., *Odontothrips intermedius, Otiorhynchus ligustici, Phasgonophora sulcata* and *Meligithes erythropus* (Wallace, 1968; Goplen *et al.*, 1991; Morrill *et al.*, 1998). Root-knot nematode *(Meloidogyne* spp.) has been found on sainfoin in the USA (Gray, Wofford & Griffin, 1986) and Gray and Wofford (1993) reported that stem nematode (*Ditylenchus dipsaci*) had been shown to attack sainfoin, but only under greenhouse conditions.

1.6.3 Fertiliser

There are no specific recommendations for sainfoin in the UK. Bland (1971) reported that it responded well to farmyard manure, phosphate and potash but that the optimum amounts of application had not been studied. However, a tentative recommendation was suggested according to experience, soil analysis and analogy with other legumes (Bland, 1971; Sheldrick & Thomson, 1982) (Table 1.1).

Table 2.1 Provisionally recommended rates of nutrient for sainfoin.

	Seeding Year (total)			Production Years (per cut)	
	N	P_2O_5	K_2O	P_2O_5	K_2O
Soil Index	kg ha^{-1}				
0	25	100	125	100	120
1	--	75	75	80	100
2	--	50	30	50	60
Over 2	---	--	--	40	30

Table 1.2 Nutrients extracted annually from the soil by lucerne and sainfoin crops.

	Nutrient extracted kg ha^{-1}						
	P	K	Ca	Mg	Na	Cl	N
lucerne	66	430	355	76	13	82	-415
sainfoin	100	280	152	27.5	2	62	-465
	Fertiliser equivalent kg ha^{-1}						
	P_2O_5	K_2O	$CaCO_3$	NO_3			
lucerne	154	520	888	-1838			
sainfoin	229	337	380	-2059			

Whitehead (1966, 1969) and Sheehy *et al.* (1984) tested the nutrients extracted from soil by lucerne and sainfoin and converted them into fertiliser equivalents (Table 1.2). Sainfoin required more P_2O_5 and NO_3 than lucerne, but less K_2O and $CaCO_3$.

Sainfoin's response to nitrogen has been discussed in section 1.5.4. Nitrogen increased sainfoin yield and may affect its regrowth and stand persistence (Koter, 1965; Sims *et al.*, 1968; Jesen & Sharp; 1968; Meyer, 1975; Hume, 1985). However, a few reports have showed that there was no difference between fixed nitrogen and inorganic nitrogen (Badoux, 1965; Sheehy & McNeill, 1988).

Sainfoin's response to phosphate and potash has seldom been reported. A study on the response of sainfoin, lucerne and red clover to phosphate was conducted in dryland and irrigated areas in Montana, USA. Sainfoin yield was not increased by phosphorus, but lucerne and red clover were (Roath & Graham, 1968). Analysis of sainfoin and lucerne hays for phosphorus content showed that there was no significant difference between sainfoin and lucerne. Meyer (1975) found that P_2O_5 and K_2O, either alone or in combination with nitrogen, had very little effect on sainfoin's productivity, recovery or stand persistence. However, Shan, Singh, Kachroo and Khanday (1991) found that added P_2O_5 increased sainfoin yield.

1.6.4 Defoliation

Sainfoin in the UK was traditionally used mainly as a hay crop, but it can be cut for silage as well (Bland, 1971; Sheldrick *et al.*, 1995). Sainfoin aftermath was used for grazing, and light grazing only in the late autumn was suggested to allow the crop time to replenish root reserves (Sheldrick *et al.*, 1995). In Canada, a sainfoin-grass mixture was reported to be grazed or cut which lasted for five years in dryland conditions (Goplen *et al.*, 1991). A study in the southern Great Plains in the USA showed that light or medium grazing at bud (Stage 3-4, Appendix 1) or flowering stage (Stage 5-6, Appendix 1) may be suitable under irrigation (Mowrey & Matches, 1991).

Traditionally cutting has normally taken place at the bud (Stage 3-4, Appendix 1) to mid flowering stage (Stage 5-6, Appendix 1) for the first cut, which can provide about 70% of the total annual yield. Canadian experience showed that regrowth was better if a cut was taken at bud or early flowering stage (Stage 5, Appendix 1), but that yield is higher when the first harvest is at a more mature stage (Goplen *et al.*, 1991). Furthermore, sainfoin cutting for hay between the 75% and 100% bloom (Stage 6,

Appendix 1) stage can reportedly achieve the best yields and highest yields of nutrients, without appreciable loss of quality, since sainfoin retains its leaves longer than alfalfa. Protein, lignification and fibre content do not vary significantly between early, medium and late bloom (Stage 5-6, Appendix 1) (Koch, Dotzenko & Hinze, 1972; Goplen *et al.*, 1991; Mowrey & Matches, 1991).

Sainfoin regrowth is slow, and allowing enough time to replenish root reserves is important to maintain its persistence and longevity. The behaviour and preference of sainfoin is similar to that of lucerne in many respects. The recommended interval between cuts for lucerne is about 6 weeks, and it uses the root reserve in the first three weeks. In the second three weeks the root reserves are restored to the former level (Jones, 1955; Aldrich, 1984). Since the regrowth of sainfoin is slower than lucerne, the second and third cuts may be taken at intervals of about 7 weeks after the previous cut. The slow regrowth of sainfoin compared with lucerne may be due to essential differences in the root reserves. Carbohydrate in sainfoin at the bloom stage (Stage 6, Appendix 1) was 10% lower than that in lucerne (Cooper, 1968). As described in section 1.5.4, this seemed to have been also confirmed the study of Krall and Delaney (1982).

Autumn management appears to be crucial for sainfoin, as for lucerne, and stands to benefit from an autumn rest (Sheldrick *et al.*, 1995; Frame *et al.*, 1998). Final defoliation should probably be taking place when no further regrowth is likely (Jone, 1955; Mowrey & Matches, 1991; Frame *et al.*, 1998). However, no definitive work has been completed on sainfoin to verify this practice.

1.7 Seed Production

Robinson (1937) stated that in Hampshire sainfoin seed was not harvested for 5 or 6 years, to obtain seeds from long-lived plants. Seed yield was about 448 kg ha^{-1} of dehulled seed. Canadian experience showed that 500-900 kg ha^{-1} of cleaned seed can be obtained. And more than 1100 kg ha^{-1} have been achieved from cvs. Melrose and Nova (Goplen *et al.*, 1991). Sainfoin seeds ripen from the base of flower spike toward the top, and basal seeds shatter from the plants before the upper seeds are ripe. The crops are cut, therefore, when the basal seeds become brown.

1.8 Comparison of Perennial Forage Legumes

Sainfoin has several advantages over other perennial forage legumes (Table 1.3). The biggest advantage, as for birdsfoot trefoil, over other perennial forage legume is the presence of condensed tannins, which reduces the likelihood of ruminant animals bloating and helps reduce the degradation of protein in the rumen. The deep taproot of sainfoin, as for lucerne, improves its drought resistance. The disadvantages of sainfoin compared with white clover and lucerne are a lack of persistence and not being tolerant to frequent cuts. Sainfoin needs a higher seed rate than other perennial legumes and this may result in higher seed costs. Its yield is also lower than lucerne and red clover.

Table 1.3 Comparison of perennial forage legumes.

Legume	Drought tolerance §	Persistence §	Seed rate & cost kg ha^{-1} ‡	DM yield ton ha^{-1} ‡	Tolerance of Frequent cutting §	Relative maturity §	CP Range †	Bloat †	Nitrogen fixation kg ha^{-1} year^{-1} †
Sainfoin	H ‡	L ‡	rate:100 £200	8-12 ‡	L	Early ‡	17-25% ‡	No ‡	146¶†
Lucerne	H	H	rate:20 £64	10-18	M	Early	17-24%	Yes	78-225
White clover	L	H	rate:4	8-12	H	Early-medium	23.8-26.2%	Yes	100-400
Red clover	L	L	rate:13 £35	9-18	M	Medium-late	17.5-25%	Yes	67-225
Birdsfoot trefoil	M	M	rate:12	5-8*	H	Late	17-19%	No	49-168

L=low, M=moderate, H=high
§ Hall (2005)
†Sheaffer, Mathison, Martin, Rabas & Ford (2003)
‡Spedding & Diekmahns, 1972; Sheldrick *et al.*, 1995; Frame *et al.*, 1998
†¶† Estimated by Sheehy & McNeill, 1988
*Stands for yield of birdsfoot trefoil+grass.

1.9 Conclusions

Research information on sainfoin is very limited. However based on published literatures, some of the advantages and disadvantages of the crop be deduced (Table 1.4).

Table 1.4 Advantages and disadvantages of sainfoin.

Advantage	Disadvantage
• Drought tolerant	• Lack of persistence
• High CP	• Low yield
• Non-bloating	• Uncompetitive with grasses
• High voluntary intake	• High seed rate and cost
• Good animal performance	• Insufficient nitrogen fixation

The high voluntary intake, enhanced animal performance, and non-bloating characteristics make sainfoin a valuable forage crop. However, a review available information reveals that our understanding and knowledge of sainfoin biology and agronomy is too little to make full use of these advantages. Following the increased demand for home-grown protein and increasing interest in the development of sustainable agricultural systems, it seems that there is a pressing need to understand more about sainfoin.

The major agronomic problems and uncertainties concerning sainfoin revealed in the literature reviewed can be summarised as follows:

- Low yield and lack of persistency
- Lack of information about available varieties
- Lack of understanding about the nature of competition between companion grasses and sainfoin
- Uncertainty about optimum establishment strategies
- Uncertainty about optimum cutting intervals and autumn management strategy
- Little information about *Rhizobium* spp. and nitrogen fertiliser interactions

2.0 Aims and Outlines

The overall aim of this thesis, therefore, is to address and explore some of the problems identified in the review of literature and summarised in 1.9 previously, and to establish and confirm modern husbandry guidelines for the establishment and maintenance of stands of sainfoin and sainfoin/grass mixtures.

The objectives are:

- To evaluate the varieties of sainfoin currently available
- To optimise plant establishment
- To optimise stand longevity in sainfoin and sainfoin-grass crops

- To apply a crop competition model to study the interaction between sainfoin and companion species

The outline of the thesis is as follows:

- Chapter one is the general introduction, which reviews the history of sainfoin in the UK and previous research.
- Chapter two aims to study the effects of sowing depth and the presence or absence of a seed pod on seedling emergence and vigour.
- Chapter three aims to investigate the impacts of sowing date on the establishment, growth and production of sainfoin, and also to explore the effect of autumn management (early or late cutting) on subsequent growth and yield.
- Chapter four aims to study the impact of direct sowing or undersowing in spring barley on the establishment, growth, production and persistence of sainfoin and sainfoin-grass in various seeds mixtures.
- Chapter five aims to evaluate and assess the yield potential of sainfoin cultivars available.
- Chapter six aims to study the nature of competition between sainfoin and two-grass species- meadow fescue and tetraploid perennial ryegrass.
- Chapter seven provides discussion of the main findings and conclusions from the study.

Chapter Two

The Effects of Sowing Depth and Seed Pod on Emergence

Abstract

The effects of sowing depth and seed pod on seedling emergence, emergence speed and seedling vigour (as seedling height) were studied with three sainfoin varieties under greenhouse conditions. Hulled (within pod) and dehulled (without pod) seed was sown at 0, 1, 2, 4 and 6 cm depths. Hulled seed sown at 6 cm depth and dehulled seed sown at 0 cm depth severely reduced emergence rates. Seed sown at 6 cm depth and hulled seed sown at surface level affected the speed of emergence, and subsequent seedling height.

2.1 Introduction

Sowing seeds at optimum depth can help to meet their germination requirement for soil moisture and reduce subsequent seedling death from dehydration. Optimum sowing depth varies with crops, the characteristics of seeds and soil texture and soil conditions (Arnott, 1969; Pratley, 1988). Small seed normally has less food reserve than large seed and should, therefore, be sown at a shallower depth. Conversely, large sized seed may be sown at a greater depth (Pratley, 1988). Deep sowing reduces emergence and seedling vigour (Arnott, 1969; Ries & Hoffman, 1995). Andrews, Douglas, Jones, Milburn, Porter, and McKenzie (1997) found that cool-season grasses with greater seed mass were associated with increased seedling emergence at 1 and 3 cm sowing depths, and that the larger grass seedlings had an increased coleoptile and mesocotyl widths, resulting in greater shoot strength to penetrate the soil. The type of emergence and soil type also affects optimum sowing depth. Epigeal emergence (the emergence of cotyledons above the surface of the ground after germination) such as sainfoin and soybean, normally requires more shallow depth than hypogeal emergence (where the cotyledons remain below the surface of the ground after germination) such as pea and cereal crops (Miller & Stritzke, 1995). Seed sown in light soil requires more shallow depth than in heavy soil because light soil provides less resistance to the seedling emergence (Pratley, 1988).

Some forage legumes are difficult to establish compared with many other crops, because their seeds are relatively small, and this necessitates sowing at shallow depths where the seed is vulnerable to soil moisture deficits (Sheaffer, 1989). Seedling death from dehydration, nearer the soil surface, can occur once the seed germinates and radicle emergence occurs in dry conditions.

Sainfoin seed is larger than most perennial forage legume seeds and is normally contained within a seedpod. Traditional experience suggests an optimum sowing depth of 15-30 mm (Sheldrick *et al.*, 1995; Frame *et al.*, 1998), which is shallower than the 20-50 mm suggested by Canadian and Chinese experience (Goplen *et al.*, 1991; Chen, 1992) and deeper than the 12-15 mm suggested for lucerne (Barnes, Miller & Nelson 1995; Frame *et al.*, 1998). There was no report to be found on sainfoin emergence problems. Sainfoin can be sown as hulled or dehulled seed in practice. However, removing the seed pod adds to the cost of the seed. A number of studies on germination showed that dehulled sainfoin seed had better germination than hulled (Finlayson, 1906; Zade, 1933; Percival, 1936; Wiesner *et al*, 1968), and that this may affect emergence. But Chen (1992) reported that there was no difference in emergence between hulled and dehulled seed in the field.

The objectives of this study were to investigate the effects of sowing depth and the presence or absence of a seed pod on seedling emergence, and also to study these effects on the speed of emergence and seedling heights of different varieties of sainfoin.

2.2 Materials and Methods

This study was conducted in a greenhouse at the Royal Agricultural College, Cirencester, in March 2003. This experiment was a 5 x 3 x 2 factorial design with three replications. Five levels of sowing depth, three varieties and two seed treatments were studied. The treatments were:

Sowing depth (SD): 0 cm, 1 cm, 2 cm, 4 cm and 6 cm,

Varieties (V): Cotswold Common, Perly and 'Commercial'

Seed treatment (ST): hulled and dehulled.

Seeds were sown in plastic pots (14.5 cm height and 13.5 cm radius). 20 seeds were sown in each pot. The substrate used was John Innes No 4 compost. Compost was filled into pots to a constant depth and patted three times. Seeds were then sown onto the surface of the compost, and covered according to the sowing depths and firmed with three pats afterwards.

After sowing, the pots were placed on a platform in the greenhouse and watered thoroughly. The pots were exposed to natural light in daytime and the temperature controlled to 20±2°C. Pots were watered regularly to keep compost moist.

Emergence of seedlings was noted and counted every day. The experiment lasted 27 days, prior to seedling harvest.

Data was first processed to test variance through Genstat 7 (Payne, Murray, Harding, Baird, Soutar & Lane, 2003) and then multiple comparisons of treatment means was performed by applying the Least Significant Difference (LSD) test. When the main effects involved interactions, the main effects were compared in the interactions as treating main effects alone could lead to misinterpretation (Clewer & Scarisbrick, 2001). Only first order of interactions will be considered in this study.

2.3 Results

2.3.1 Seed Weight

Prior to sowing, the thousand seed weight and germination of the three varieties were measured and tested respectively to determine the variance between the three varieties, which may have had an effect on emergence. Cotswold Common had a greater seed weight ($P<0.05$) than both Perly and 'Commercial' (Table 2.1 & 2.2). The weight of hulled seed was obviously greater ($P<0.001$) than that of the dehulled (Table 2.1 & 2.2). Germination rate was 85.6% on average, with no significant differences between varieties (Table 2.1).

Table 2.1 Analysis of variance of 1000 seed weight and germination of three tested varieties.

	d.f.	000' seed weight (M.S)	Germination (M.S)
Variety	2	1.15 *	101.2
Seed treatment	1	168.72 ***	
Residual	12(8) ‡	0.12	9.1

*, *** Significant at the 0.05 and 0.001 level of probability respectively.
‡ Figure in bracket indicates d.f of germination.

Table 2.2 Comparison of 1000 seed weight of three varieties.

Variety	000' seed weight (gram)
Cotswold Common	20.26a
Perly	19.59b
'Commercial'	19.44b
LSD (0.05)	0.44
Seed treatment	
Hulled	22.82a
Dehulled	16.7b
LSD (0.05)	0.36

Values followed by the same letter are not significantly different.

2.3.2 Seedling Emergence

2.3.2.1 Emergence Pattern

Data on seedling emergence was collected over a period of 27 days and is summarised in Figure 2.1-2.3. All three varieties started emerging on day 4 after sowing (Figure 2.1). Perly and 'Commercial' appeared to show a quicker emergence trend than Cotswold Common before Day 16. However, the emergence rate became similar after day 16. Dehulled seeds also showed a trend of quicker emergence than hulled seed before day 7 and then became similar (Figure 2.2). 1 cm and 2 cm depths generally showed quick emergence and 6 cm showed a slower emergence (Figure 2.3).

Figure 2.1 Patterns of emergence of three varieties of sainfoin over 27 days.

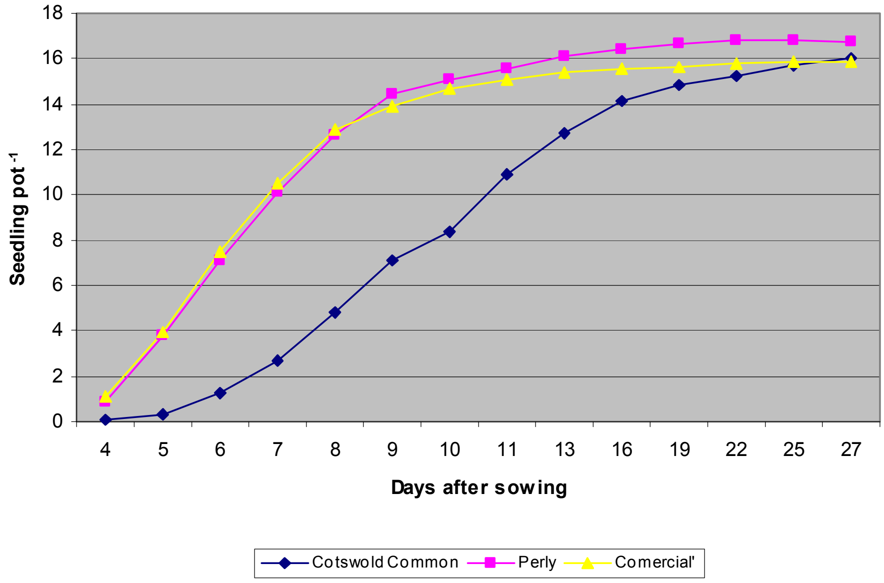


Figure 2.2 Patterns of emergence of hulled and dehulled sainfoin seed over 27 days.

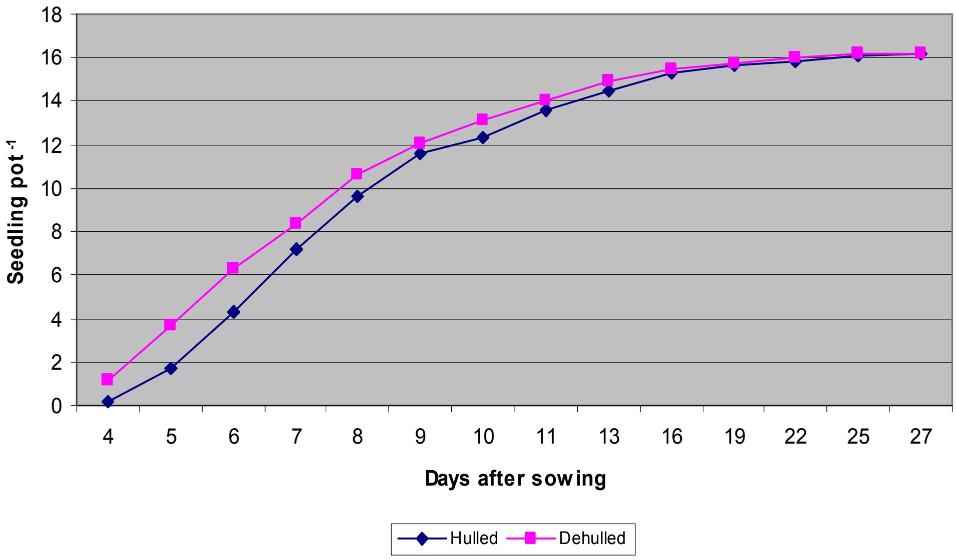
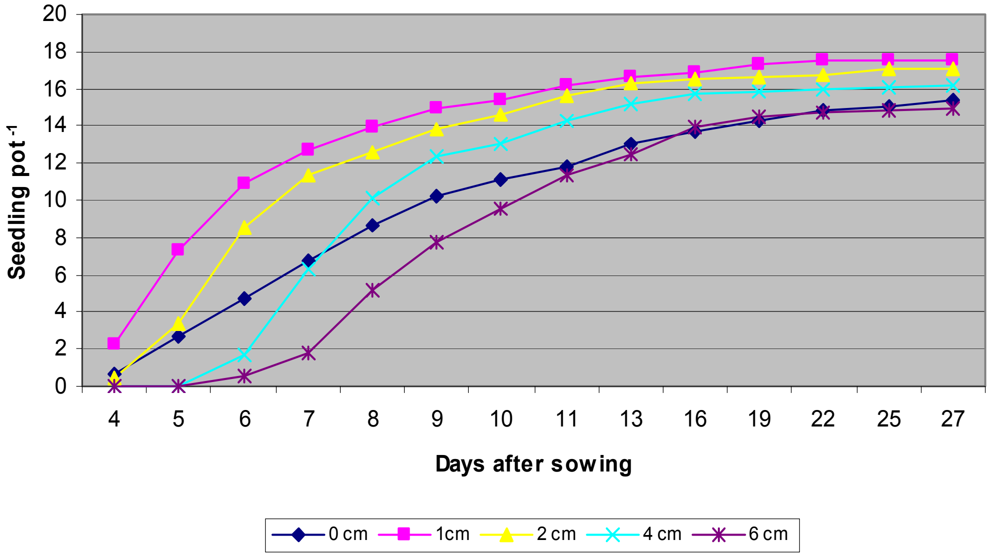


Figure 2.3 Patterns of emergence of sainfoin sown at different depths over 27days.



2.3.2.2 Emergence on Day 8 and Day 27

To investigate the rapidity of emergence, data for day 8 was analysed. Emergence rate on day 27 was analysed to observe the final treatment effects.

Table 2.3 Analysis of variance of seedling emergence by day 8 and day 27.

		Day 8	Day 27
	d.f.	M.S	
Sowing Depth (SD)	4	219.06 ***	25.07***
Variety (V)	2	611.7 ***	4.15
Seed treatment (ST)	1	14.8	0.03
SD× ST	4	40.83 ***	15.45***
SD×V	8	20.51**	1.86
V× ST	2	14.14	5.95
SD × V × ST	8	23.5	6.19
Residual	60(1)‡ (3)¶	6.4	2.6

, * Significant at the0.01 and 0.001 level of probability respectively.
‡ Figure in bracket indicates missing value on day 8.
¶ Figure in bracket indicates missing value on day 27.

Sowing depth interacted with seed treatment (P<0.001) and varieties (P<0.01) by day 8 (Table 2.3). Hulled seeds sown at 4 cm and 6 cm depths and dehulled seeds sown at 0 cm and 6 cm had a slower emergence by day 8, compared to other sowing depths (Table 2.4). Hulled seeds sown at 0 cm depth had quicker emergence than dehulled seeds sown at 0 cm depth, and hulled seeds sown at 4 cm depth had slower emergence than dehulled seeds at 4 cm depth. Perly and ‘Commercial’ had quicker emergence generally than Cotswold Common by day 8 (Table 2.5). However, there was interaction between Cotswold Common and Perly at 0 cm, which indicated there was no significant difference in emergence between these two varieties by day 8.

Table 2.4 Effect of depth x seed treatment on seedling emergence by days 8 and 27.

	Day 8		Day 27	
Depth	Hulled	Dehulled	Hulled	Dehulled
	seedling pot^{-1}			
0 cm	10.33 cd	5.94 f	16.89 ab	14.0 c
1 cm	13.33 ab	14.56 a	17.33 ab	17.78 a
2 cm	11.78 bc	13.44 ab	17.17 ab	17.67 a
4 cm	8.44 de	11.78 bc	16.56 ab	16.47 ab
6 cm	4.11 f	6.33 ef	13.78 c	16.0 b
LSD (0.05)	2.34		1.52	

Values followed by the same letter are not significantly different.

Table 2.5 Interaction of depth x variety on seedling emergence by day 8.

	Cotswold Common	Perly	‘Commercial’
Depth		seedling pot^{-1}	
0 cm	5.67 gh	8.08 efg	10.67 de
1 cm	7.83 efg	17.17 a	16.83 ab
2 cm	5.50 gh	16.33 ab	16.00 ab
4 cm	4.33 h	14.00 bc	12.00 cd
6 cm	0.67 i	6.17 fgh	8.83 ef
LSD (0.05)		2.91	

Values followed by the same letter are not significantly different.

Sowing depth affected ($P<0.001$) emergence rate and interacted ($P<0.001$) with seed treatment by day 27 (Table 2.3). The emergence rates of hulled seeds sown at 0, 1, 2 and 4 cm and dehulled seed sown at 1, 2 and 4 cm by day 27 were similar (Table 2.4). Dehulled seeds sown at 6 cm also had similar emergence rate as hulled seeds sown at 0, 1, 2 and 4 cm and dehulled seed at 4 cm. There was an interaction at 0 cm and 6 cm depths. Hulled seeds sown at 0 cm had greater emergence rate than dehulled seeds at 0 cm by day 27, and hulled seed sown at 6 cm had lower emergence rate than dehulled seeds at 6 cm.

2.3.3 Seedling Height

Sowing depth ($P<0.001$) and variety ($P<0.001$) both had effects on seedling height, but seed treatment did not (Table 2.6). There was also a significant interaction ($P<0.05$) between sowing depth and seed treatment.

Table 2.6 Analysis of variance of seedling height by day 27.

	d.f.	M.S
Sowing Depth (D)	4	5.0 ***
Seed treatment (ST)	1	0.05
Variety (V)	2	56.94 ***
SD × ST	4	1.56 *
V × ST	2	0.66
SD × V	8	0.93
SD × V × ST	8	0.31
Residual	60	0.58

*, *** Significant at the 0.05 and 0.001 level of probability respectively.

Table 2.7 Comparison of seedling height (cm) between varieties by day 27.

Cotswold Common	6.80 c
Perly	7.99 b
'Commercial'	9.55 a
LSD (0.05)	0.39

Values followed by the same letter are not significantly different

Table 2.8 Interaction of depth x seed treatment on seedling height (cm) by day 27.

Depth	Hulled	Dehulled
0 cm	8.41 ab	7.34 c
1 cm	8.62 ab	8.69 ab
2 cm	8.37 ab	8.70 a
4 cm	7.97 bc	8.29 ab
6 cm	7.29 c	7.41 c
LSD (0.05)	0.72	

Values followed by the same letter are not significantly different.

'Commercial' had the greatest height, and Cotswold Common the lowest (Table 2.7). Sowing at 6 cm using either hulled or dehulled seed reduced seedling height (Table 2.8). Dehulled seeds sown at 0 cm also showed reduced seedling height.

2.4 Discussion

Both hulled and dehulled seeds had quicker seedling emergence at 1 and 2 cm depths by day 8; hulled and dehulled seeds at the 6 cm depth and dehulled at 0 cm had slower emergence. Differences between hulled and dehulled seeds at 0 and 4 cm depths indicated that the seedpod affected emergence speed. At 0 cm depth, the seedpod may have protected seed from dehydration, and the hulled seed was less likely to become dehydrated than dehulled seed; as a result, hulled seed may have had quicker seedling emergence than dehulled at 0 cm. At 4 cm depth, seedpod seems to have delayed the emergence of hulled seed. This may be because the radicle and cotyledons in hulled seed took longer to break through the pod compared to dehulled seed at 4 cm depth. This did not happen at 6 cm depth; it may be because here sowing depth became the main factor affecting emergence speed.

Perly and 'Commercial' emerged more rapidly overall than Cotswold Common. Seed size seems not to have had effect on emergence speed. The differences between varieties may be due to genetic differences in seedling vigour apart from seed size (Shibles & MacDonald, 1962; Cooper & Qualls, 1968).

Seed treatment did not have any effect on seedling emergence at 1, 2 and 4 cm depths by day 27, which is in accordance with Chen's result (1992), but did have an effect at 0 and 6 cm depths. Hulled seed had better emergence than dehulled seed at 0 cm, and dehulled seed was better than hulled seed at 6 cm depth. Differences on the compost surface (0 cm) between hulled and dehulled could have been caused by seedpod, which may help keep the hulled seed moister compared to the dehulled, and also by surface seed placement, which created a different environment to those seeds covered with compost. At 6 cm depth, the pod may have had a negative effect on emergence, resulting in the lower emergence from hulled seed compared to dehulled seed. This may be because radicle and cotyledon in hulled seed took longer to break through the pod. Previous reports found that dehulled seed had better germination capacity than hulled seed (Finlayson, 1906; Zade, 1933) since empty pods and partially developed seeds were screened off from dehulled seeds, and the hard seed percentage was decreased. In this experiment, there were no emergence differences between hulled and dehulled seed at 1, 2 and 4 cm depths. This may be because pods were removed by hand since machinery could not be used for such a small amount of seed. As a result, the hard seed may not have been scarified. In practice, dehulling hard seed by machine may scarify it and this may increase its germination. Furthermore, peeling the seedpods may also screen out the dead and partially developed seeds. Hulled seed at 0, 1, 2 and 4 cm depths and dehulled seed at 1, 2 and 4 cm achieved better emergence than other depths. This was in accordance with other reports in the literature (Goplen *et al*, 1991; Chen, 1992; Sheldrick *et al.*, 1995).

Since this study was conducted in greenhouse conditions using a compost substrate the results may well be different from the field environment. Surface placement (0 cm) in practice may run a greater risk of dehydration. However, surface placement of hulled seed, which is a common occurrence when broadcasting, showed better emergence than dehulled seed.

Seedlings from some hulled seed were seen to emerge with parts of the seed pod still covering the cotyledons. This may have been due to loose compost in the pots.

2.5 Conclusions

- Sainfoin seed can be sown at 1-4 cm depths as either hulled or dehulled seed, and this did not cause significant differences in the optimum greenhouse compost based study described. Hulled seed sown at 0 cm and dehulled seed at 6 cm depth also achieved similar emergence rates as seeds sown at 1-4 cm depths.

- Hulled seed sown at 6 cm and dehulled seed at 0 cm depth reduced the emergence rate. Seedpods seemed to check emergence at 6 cm depth, but to assist emergence at 0 cm.
- Seed size seems not to have affected emergence speed and rate, or the seedling height.
- Perly and ‘Commercial’ demonstrated quicker emergence than Cotswold Common. Deep sowing (6 cm) either as hulled or dehulled seeds delayed emergence, so did surface placement (0 cm) of dehulled seed
- In this study the ‘Commercial’ seed batch showed greatest vigour, as reflected in seedling height, among the varieties tested.

Chapter Three

The Effects of Sowing Date and Autumn Management on Sainfoin Establishment, Regrowth and Yield

Abstract

Sainfoin was sown from April to September to investigate effects of sowing date on establishment, regrowth and production over three years. Plots were divided into early cut and late cut in autumn to study the effects of autumn management. April to July sowings achieved about 8 t DM ha^{-1} average yield over three years. August and September sowings gave about 4.8 t DM ha^{-1}. Early autumn cut reduced sainfoin yield in the 3rd year.

3.1 Introduction

3.1.1 Sowing Time for Legumes

Time of sowing is important for legumes in order to establish a satisfactory stand. Species and environment have the main effect on the optimum time of sowing (Miller & Stritzke, 1995) so that seeds can obtain enough moisture and experience favourable temperatures for germination and seedling growth. In addition, suitable moisture and temperature also improve *Rhizobium spp* survival, and the infection of legume roots (Frame *et al.,* 1998). For grass-legume swards, sowing in spring (March -- May) and late summer (August -- mid-August) is often recommended in the UK (Frame, 2000; Sheldrick, 2000). Sowing white clover is recommended for spring, to give the best establishment (Frame *et al.*, 1998). Red clover sown in the early season can also reportedly give higher herbage production in the establishment year and the following year (Frame, Harkess & Hunt, 1976b). It is also recommended to sow lucerne in the spring or late summer (Frame *et al.,* 1998).

Studies on sowing date for sainfoin have not been found in the UK. Traditional timings for sainfoin sowing were recommended between April and July (Bland, 1971; Sheldrick *et al.*, 1995). Little evidence showed that April and May sowing gave better establishment compared with June and July sowing (Bland, 1971). It may be because June and July sowing risks possible shortage of moisture resulting in poor emergence in dry years. However, according to Sheldrick (2000), late summer sowing may give rise to fewer problems with weed invasion, apart from chickweed. Late summer sowing of legume-grass leys after winter cereal harvest, has become popular in recent years.

3.1.2 Autumn Management

Legume growth and regrowth depends on carbohydrates stored in the taproot and crown; the higher level of root reserves, the more vigorous its regrowth (Bosworh, 2006). Sainfoin cutting interval could best be about seven weeks on the basis of experience and comparison with lucerne since its regrowth is slower than lucerne, where the recommended cutting interval is six weeks (Johnson, 1984). During the late summer and early autumn, leguminous plants start to restore root reserves (Frame *et al.*, 1998; Bosworh, 2006), which makes the time of autumn cutting potentially very important. The last harvest should take place at a time that allows enough time for perennial leguminous plants to build up root reserves before growth stops (Sheldrick *et al.*, 1995; Frame *et al.*, 1998).

No research information has been found on the ideal cutting regime for sainfoin. Optimum sowing dates, cutting interval, the ideal autumn management and its effect on subsequent performance is still not clear and justifies further investigation .

3.1.3 Objectives

The primary objective was to investigate the impacts of sowing date on the establishment, growth and yield of sainfoin. The secondary objective was to explore the effect of autumn management (early or late cutting) on subsequent growth and yield.

3.2 Materials and Methods

3.2.1 Experiment Site

The experimental site was located at Piggery Field, Coates Manor Farm, the Royal Agricultural College, Cirencester, UK (51° 42'N, 02° 01'W; 135 m ASL). The soil is Sherborne series Cotswold Brash. This is a shallow, stony, well-drained clay loam (Findlay, 1984; Conway, 2006). Soil depth is < 30 cm deep over limestone rock. Stone content is about 10% - 30%. The trial took place between 2003- 2005.

Table 3.1 Meteorological data at Cirencester.

	Precipitation(mm)				Mean air temperature(°C)			
	2003	2004	2005	10 year mean	2003	2004	2005	10 year mean
Jan	72.6	99.4	33.5		4.4	5.1	4.6	
Feb	24.8	32.1	22.8		3.2	4.9	3.7	
Mar	30.6	65.5	71.8		6.8	6.5	6.8	
Apr	46.5	76.6	54.7	60.8	9.6	9.5	9.3	8.3
May	58.5	57.5	45.3	52.6	12.4	13.3	11.9	11.8
Jun	69.4	43.2	36.3	58.2	16.2	16.6	15.8	14.1
Jul	80.8	47.1	32.3	39.3	17.3	17.2	17	16.8
Aug	10.2	114	34.5	63	19.6	17.6	17	16.8
Sep	12.5	48.8	36	70.5	15	15.2	14.7	13.3
Oct	43.2	135.1	90.2		8.9	10.6	12.2	
Nov	10.4	37.7	70.1		7.8	7.5	5.9	
Dec	84.2	52.1	67.9		5	4.7	3.7	

3.2.2 Experimental Design

This was a split plot design in a randomised block with three replications. Six sowing dates constituted the main treatments, and early autumn and late autumn cuts made up the sub-treatments. The plot size was 2×4 m.

Main treatments: April, May, June, July, August and September sowings

Sub-treatments: early autumn cut and late autumn cut

3.2.3 Field Preparation

The previous crop was a perennial ryegrass, red clover and white clover sward. The crop was sprayed off with an application of glyphosate [N- (phosphonomethyl) glycine], and then the site was ploughed after two weeks when the plants totally died.

The area was power-harrowed before sowing. Soil was sampled in the spring of 2003 and analyzed in the laboratory. The soil pH was 7.5 and the nutrient levels of phosphate, potash and magnesium were all at index 3 (MAFF, 2002).

3.2.4 Establishment

This trial was established in 2003 and repeated in 2004. The variety chosen for this trial was Cotswold Common. The main reasons for choosing this variety were because it is the most available landrace with considerable stocks. It also performed reasonably well in an earlier variety study at the Royal Agricultural College (Koivisto & Lane, 2001). The sowing dates are shown in Table 3.2. Sowing was at a seed rate of 90 kg ha^{-1} of hulled seed to target 150 plant m^{-1}. Seeds germination rate was at 88%. Seeds were broadcast by hand and then raked into soil, to about 1.0 cm deep and rolled straight away.

Table 3.2 Sowing dates of 2003 and 2004 establishments.

	Sowing date					
2003	Apr 17	May 15	Jun 20	Jul 16	Aug 15	Sep 16
2004	Apr 15	May 12	Jun 15	Jul 14	Aug 14	Sep 16

3.2.5 Management

"Bellmac Plus" (MCPA + MCPB) (United Phosphorous) was used to control broadleaved weeds post emergence. "Carbetamex" (Makhteshim) was used for grass weeds and chickweed. MCPA + MCPB was applied after the 1st trifoliate leaf (Stage 0, Appendix 1). Carbetamex was applied in December of each year to control grass weeds and chickweed.

Phosphorus (Triple Superphosphate, 44% P_2O_5) and Potassium (60% K_2O) were applied after harvesting in accordance with MAFF (2002).

Table 3.3 Fertilizer application chart (kg ha^{-1}) on sowing date trial after harvest.

	Establishment year	2nd & 3rd years		
		1st harvest	2nd harvest	3rd harvest
P_2O_5	30	20	-	-
K_2O	-	30	40	20

3.2.6 Population Measurement

A 25 x 25 cm quadrat was used to measure plant populations, and two determinations were made in each subplot. In the establishment years, the population of the April to July sowing was measured after harvest in August and the August and September sowing made in Mid-October. In subsequent years the measurements were made after the 1st harvest.

3.2.7 Sampling and Harvest

Before harvesting, two 50 × 50 cm quadrat areas were sampled at random from each plot to measure yield. For the 2003 establishment, the weed yield was also measured in the establishment year.

Harvest was carried out immediately after sampling with a BCS 610 Motormower, with plants cut at about 5 cm. At the 2003 establishment, a harvest was taken from the April and May sowings in August. The April, May, June and July sowings were sub-divided into subplots in September (early autumn cut vs. late autumn cut) of 2×2 m and an early autumn cut taken. For the 2004 establishment, only one harvest (early autumn cut) was taken in September. During the second year, three harvests were taken, the first in May when sainfoin was at half flowering (Stage 5-6, Appendix 1), and the second in July. For the third harvest, early autumn cuts were taken in September and late autumn cuts in November. The interval between harvests was about seven weeks (Table 3.4). The trial established in 2003 lasted for three years, and the one established in 2004 lasted two years until the project ended.

Table 3.4 Calendar of harvesting of sowing date trials.

Establishment Year	1st year harvests		2nd year harvests				3rd year harvests			
	1st	Early autumn cut	1st	2nd	3rd (autumn cut) Early	3rd (autumn cut) Late	1st	2nd	3rd (autumn cut) Early	3rd (autumn cut) Late
2003	Aug12	Sep 30	May 30	Jul 19	Sep 16	Nov 1	May 29	Jul13	Sep 12	-
2004	-	Sep 16	May 29	Jul13	Sep 12	-	-	-	-	-

3.2.8 Laboratory Analysis

DM yield samples were taken back to the laboratory after sampling, and dried in the oven at 100±2°C over 24 hours to obtain dry matter. After weighing the samples for dry weight, they were milled in a Glen Creston mill with a 0.75 mm screen and then stored in sealed plastic bags for crude protein and Neutral Detergent Fiber (NDF) analysis. Crude protein was analyzed by the Kjeldahl method (Gerhardt[1] No. KJTH and and MV1). Neutral Detergent Fiber was analyzed in FiberBags method (C. Gerhardt UK Ltd, Brackley Northants, UK).

3.2.9 Statistical Analysis

Methods used were described as in 2.2.

3.3 Results

3.3.1Establishment Year Crops

Table 3.5 Analysis of variance of sainfoin DM yield in the establishment year.

		2003 establishment			2004 establishment	
		1st harvest	2nd harvest	Annual Total		
Source of variation	d.f.	M.S			d.f	M.S
Sowing date	1 (3) ‡	0.08	3.54***	8.8***	2	1.1
Residual	2 (6)	0.08	0.02	0.05	4	0.2

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively.
‡ Numbers in the brackets stands for the d.f of the 2nd harvest in 2003 establishment.

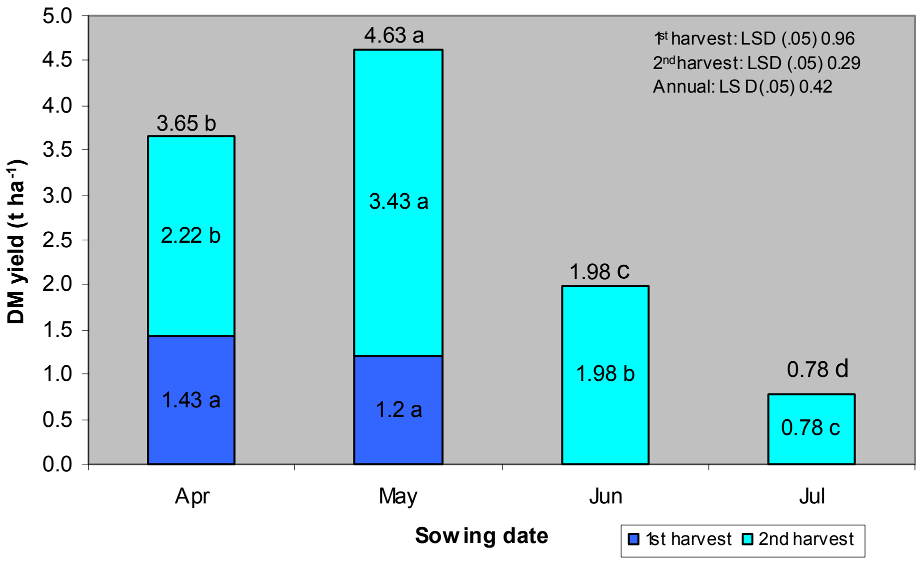
[1] Gerhardt is a reference of the manufacture of the digestion and distillation equipment used in this method. C. Gerhardt UK Ltd., Avonbury Court, Country Road, Brackley, UK NN13 7AX.

3.3.1.1. 2003 Establishment

In 2003, the April and May sowings gave two harvests (August and September) and the June and July sowings gave one harvest (September). The 2nd harvest in September was the early autumn cut, when the plots were divided into sub-plots, 'early autumn cut' and 'late autumn cut'.

Sowing date had a significant effect on sainfoin total yield (Table 3.5). There was no significant difference at the 1st harvest, but only at the 2nd harvest ($P<0.001$). The May sowing had the greatest annual yield, 4.63 t DM ha^{-1}, and the July sowing had the lowest, 0.78 t DM ha^{-1} (Figure 3.1).

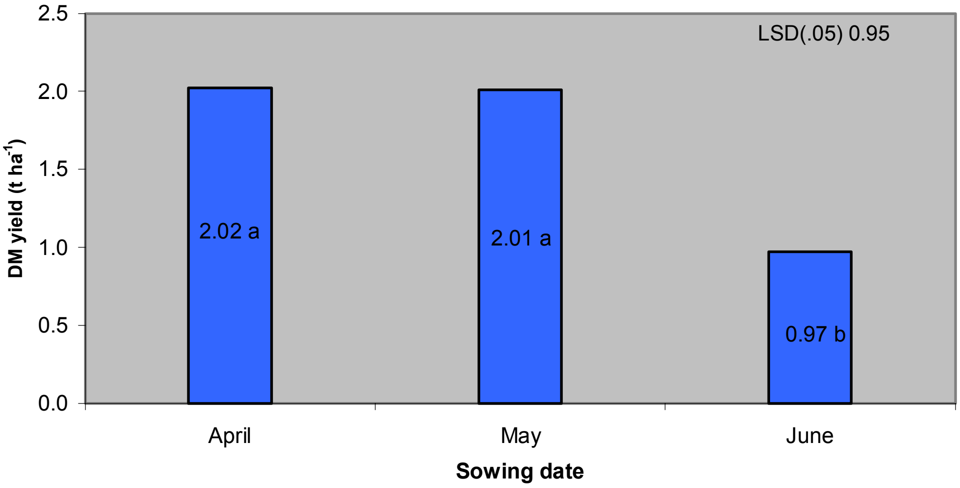
Figure 3.1 Sainfoin DM yield for 2003 establishment in the establishment year. Values at the top of the bars are total yield. Values followed by the same letter are not significantly different.



3.3.1.2. 2004 Establishment

The 2004 establishment only yielded one harvest from the April, May and June sowings. The April and May sowings had similar yields, and were higher than the June sowing (Figure 3.2).

Figure 3.2 Sainfoin DM yield for 2004 establishment in the establishment year. Values followed by the same letter are not significantly different.



3.3.3 Second Year Crops

Sowing date had significant effects (P<0.001) on sainfoin total DM yields for both the 2003 and 2004 establishments in the 2nd year (Table 3.6). Autumn management also had a significant effect (P<0.001) on yield of the 2003 establishment in the 2nd year. However, the effect of autumn management did not show in the 2nd year for the 2004 establishment.

Table 3.6 Analysis of variance of sainfoin DM yields in the 2nd year.

		2003 Establishment			
		1st harvest	2nd harvest	3rd harvest	Total
Source of variation	d.f	M.S			
Sowing date (SD)	5	66.89***	0.69*	0.13*	83.89***
Autumn management (AM)	1	0.5	0.23*	19***	21.04***
SD × AM	5	0.23	0.03	0.14	0.34
Residual	22	1.11	0.09	0.03	2.7

			2004 Establishment			
			1st harvest	2nd harvest	3rd harvest	Total
Source of variation	d.f 1st	d.f 2nd, 3rd, Total	M.S			
Sowing date (SD)	4	5	5.84*	0.6***	0.32**	49.04***
Autumn management (AM)	1	1	0.09	0.01	0.17	1.08
SD × AM	4	5	1.45	0.03	0.04	1.69
Residual	18	18(4) ‡	1.89	0.06	0.07	1.15

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively.
‡ Numbers in the brackets indicate the number of the missing values.

3.3.3.1 Sowing Date

For the 2003 establishment there were no significant differences in DM yield between the April to July sowings (Figure 3.3). The August and September sowings yielded significantly less. The August and September sowings also yielded significantly less than all other dates for the 2004 establishment, and the September sowing had no measurable harvest in the establishment year (Figure 3.5). For the 2004 establishment the April sowing out yielded all other sowing dates annually.

The yield of the 1st harvest accounted for 66.8% of the annual yield of the 2003 establishment and 65.8% of that for the 2004 establishment; the 2nd accounted for 22.6% and 19.4% respectively and the 3rd for 10.6% and 14.8% respectively (Figure 3.3 & 3.5).

Figure 3.3 Sainfoin DM yield for the 2003 establishment in the 2nd year. Values on the top of the bars are total yield. Values followed by the same letter are not significantly different.

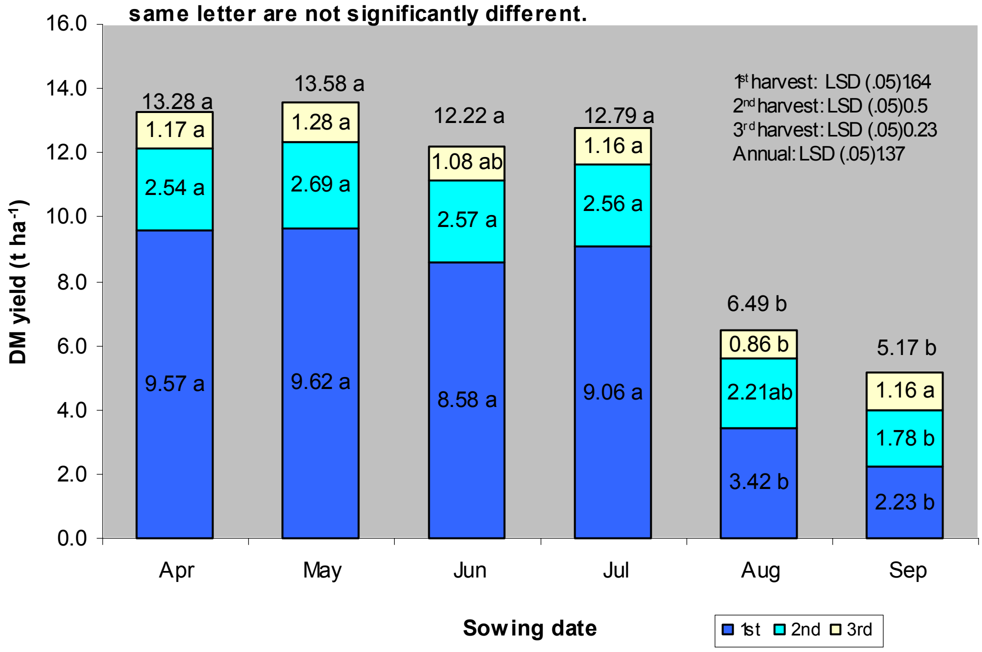


Figure 3.4 The 3rd block of sowing date of the 2003 establishment (photos taken in April 26 2004).

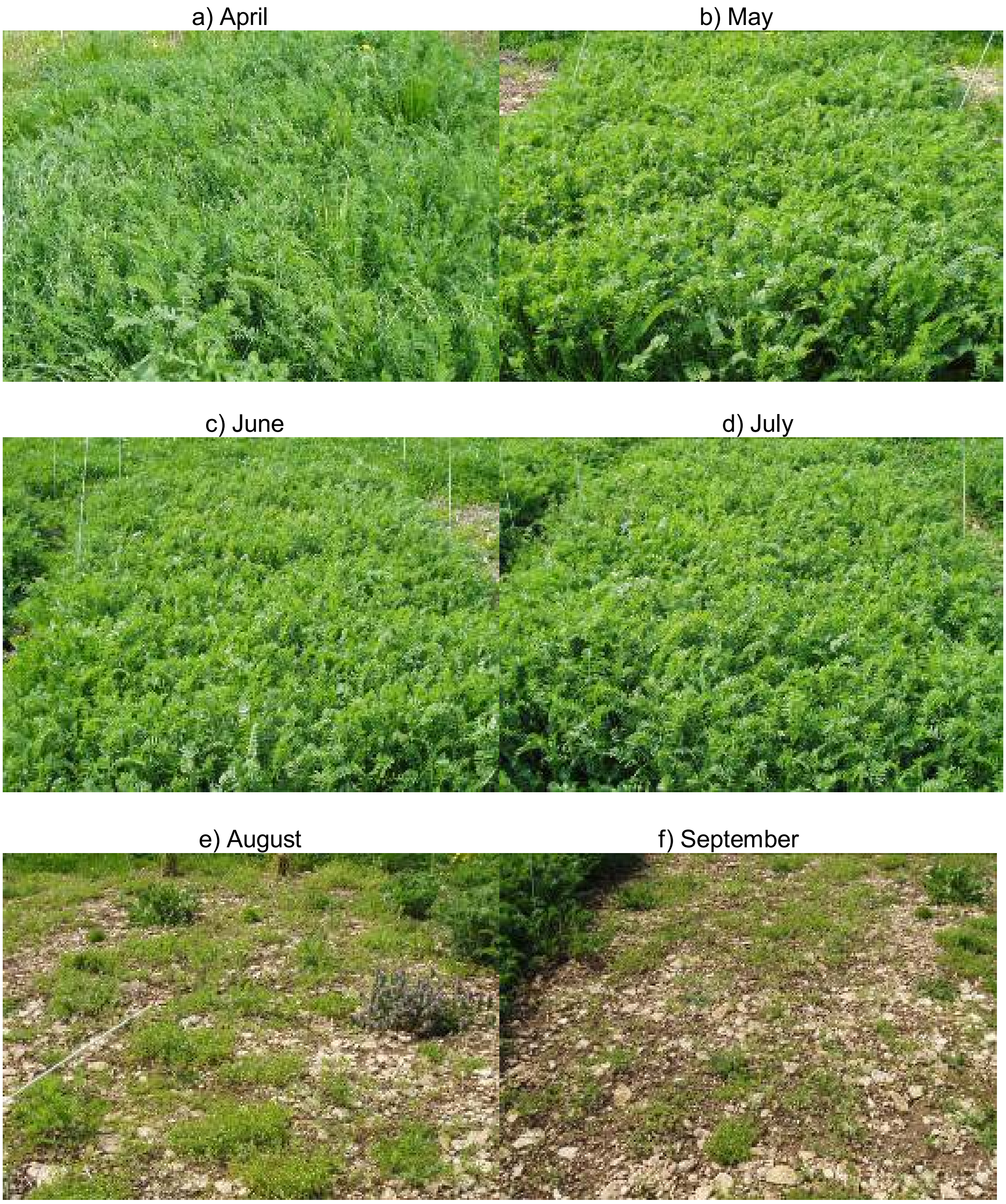
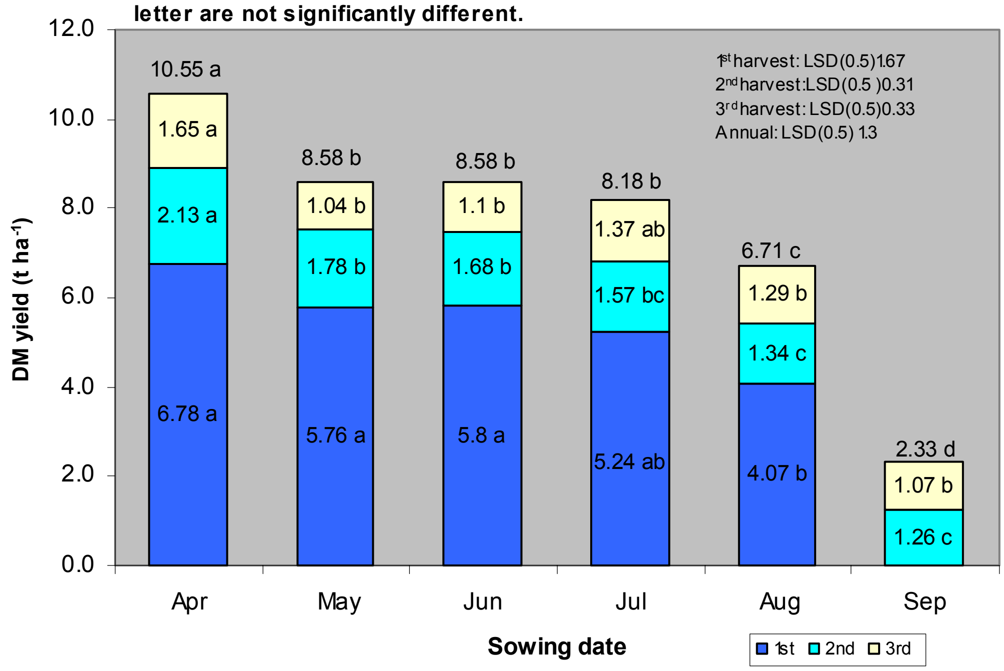


Figure 3.5 Sainfoin DM yield for the 2004 establishment in the 2nd year. Values on the top of the bars are total yield. Values followed by the same letter are not significantly different.

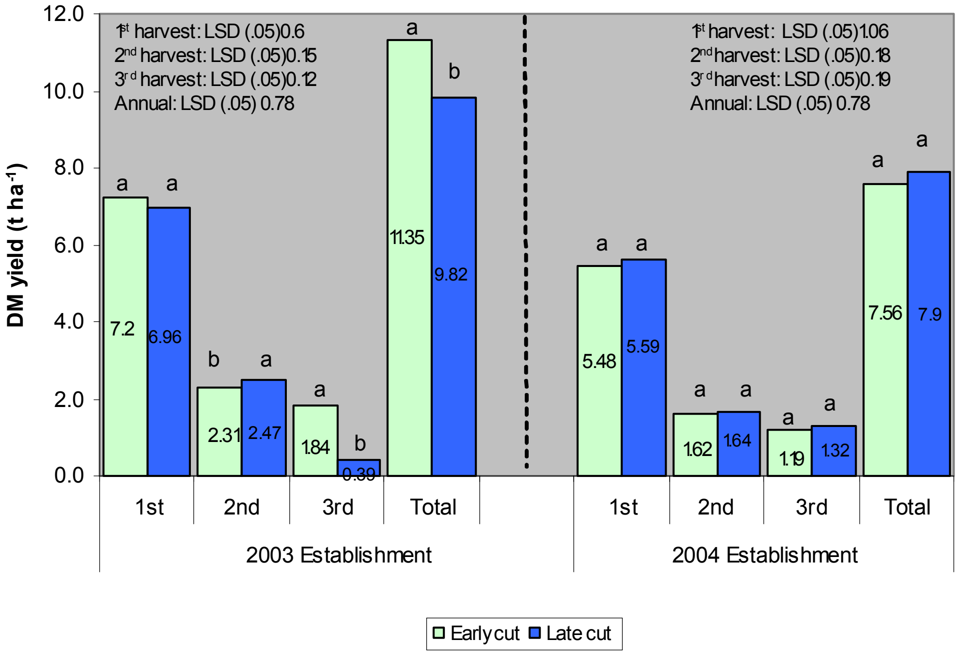


3.3.3.2 Autumn Management

The April to July sowings of 2003 establishment were given an early autumn cut in the establishment year, and for the 2004 establishment, the April to June sowings were given an early autumn cut in the establishment year.

Autumn management (early cut vs. late cut) in the establishment year only had a significant effect ($P<0.05$) at the 2nd harvest for the 2003 establishment in the 2nd year, but not for the 2004 establishment (Table 3.6) (Figure 3.6). Although for the 2003 establishment autumn management statistically also had significantly effects on annual yield and the 3rd harvest yield (Table 3.6) (Figure 3.6), the differences arose from different cutting of the 3rd harvest (early cutting and late cutting) rather than from autumn management in the establishment year. This was not the case for the 3rd harvest of the 2004 establishment; since it was the final year of the experiment the 3rd harvest was not split into early and late cuts (Figure 3.6).

Figure 3.6 Sainfoin DM yield of early autumn cut vs late autumn cut in the 2nd year. Bars followed by the same letter are not significantly different.



3.3.4. Third Year Crops

Data for the 3rd year crops only refers only to the 2003 establishment since the 2004 establishment experienced two growing seasons only during this study period.

3.3.4.1 Sowing Date

Sowing date had significant effects ($P<0.05$) on the yield of the 3rd harvest and on the total annual yield (Table 3.7). The August and September sowings had improved substantially on the previous year, but the September sowing again yielded significantly less than the May to July sowings (Figure 3.7). The yield of the 1st harvest accounted for 55.7% of total annual yield, and the 2nd accounted for 14.5% and the 3rd for 29.8% (Figure 3.7).

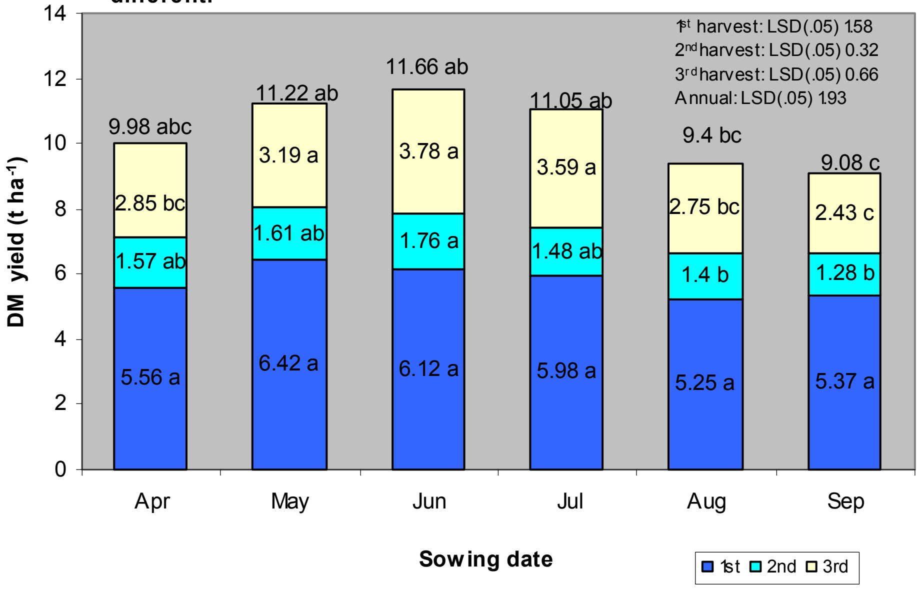
Table 3.7 Analysis of variance of sainfoin DM yield of 2003 establishment in the 3rd year.

		2003 establishment			
		1st	2nd	3rd	Total
Source of variation	d.f	M.S			
Sowing date (SD)	5	1.25	0.17	1.61*	8.03*
Autumn management (AM)	1	3.17	1.19***	1.11	11.91*
SD × AM	5	0.56	0.06	0.06	0.51
Residual	22 (4)‡	1.75	0.07	0.29	2.58

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively.

‡ Numbers in the brackets stands for the missing values at the 3rd harvest and at total yield.

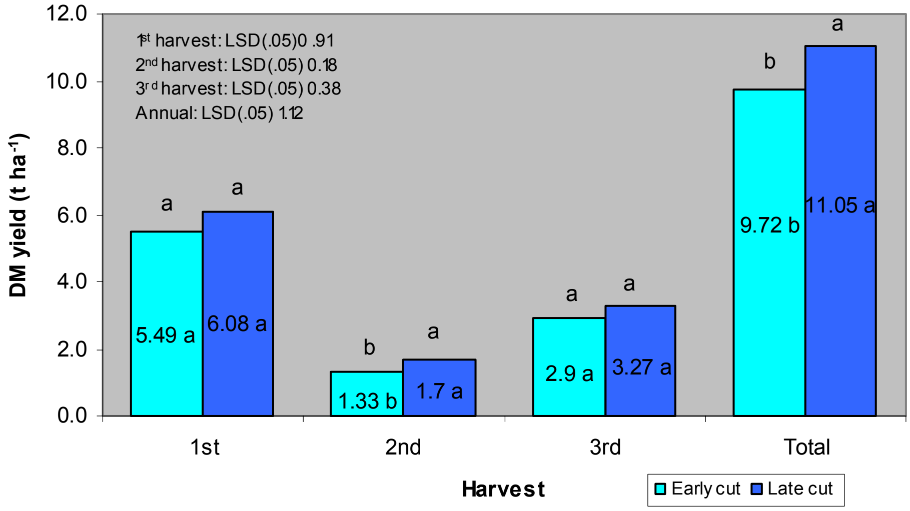
Figure 3.7 Sainfoin DM yield of 2003 establishment in the 3rd year. Comparison made within harvest. Values on the top of the bars are total yield. Values followed by the same letter are not significantly different.



3.3.4.2 Autumn Management

An early autumn cut in the previous season reduced the yield of the 2nd harvest (P<0.001) and the annual yield (P<0.05) (Table 3.7). The yield of late autumn cut plots was1.33 t DM ha^{-1} higher than early autumn cut for the whole season (Figure 3.8). The yield of late autumn cut plots was 0.37 t DM ha^{-1} higher than that of early autumn cut plots for the 2nd harvest.

Figure 3.8 Sainfoin DM yield of early autumn cut vs late autumn cut in the 3rd year. Values followed by the same letter are not significantly different.



3.3.5 Yield Distribution and Average Yield over Three Years

Sainfoin yield of the April to July sowings from the 2003 establishment peaked in the 2nd year, and then declined in the 3rd year (Figure 3.9). However, yields of the August and September sowings increased in the 3rd year. Over three growing seasons, the May sowing achieved the highest average yield, 9.83 t DM ha^{-1}, and the August and September sowings the lowest, 5.3 and 4.78 t DM ha^{-1} respectively (Figure 3.10). The April, June and July sowings had 8.48-8.93 t DM ha^{-1} yield with no significant differences.

Figure 3.9 Sainfoin DM yield of 2003 establishment over three years.

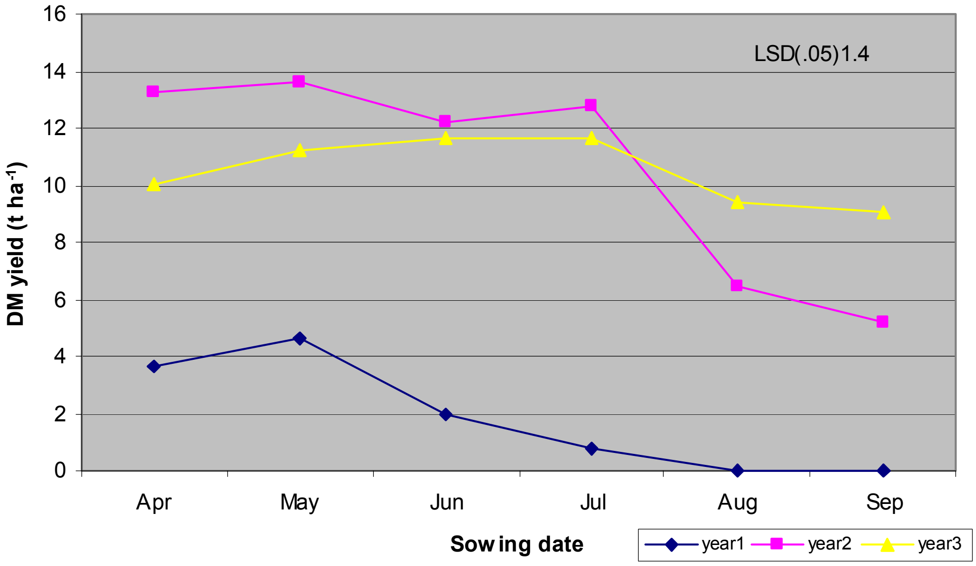
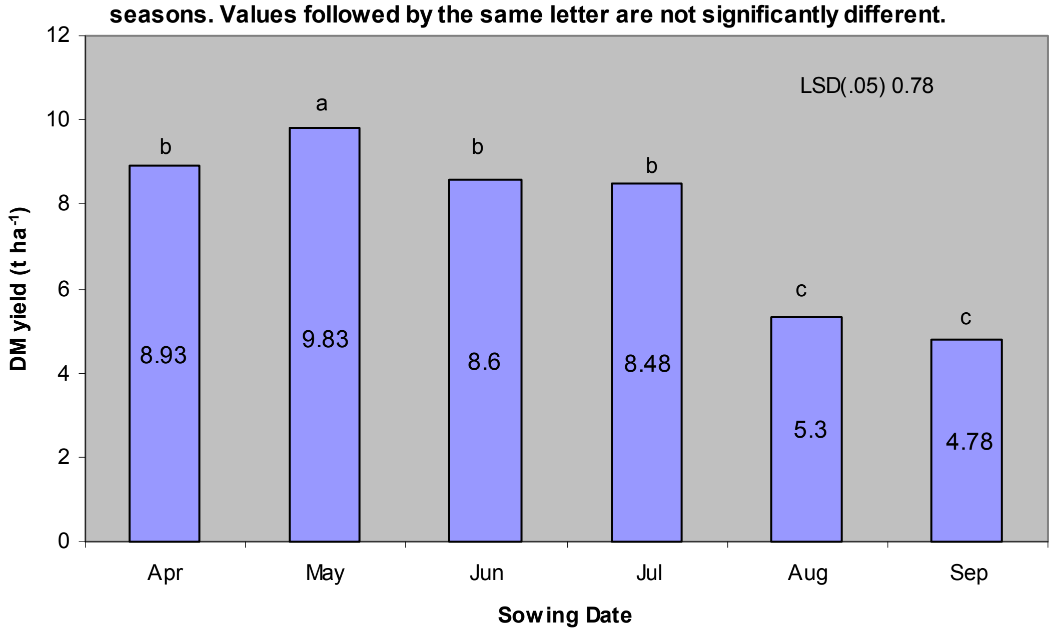
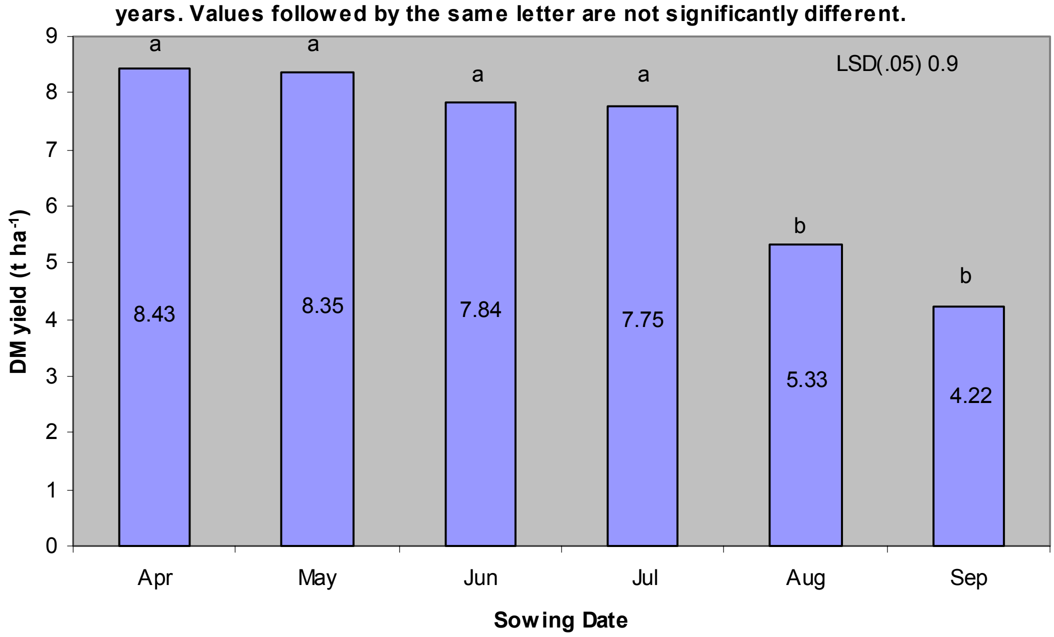


Figure 3.10 Sainfoin average yield of 2003 establishment over three growing seasons. Values followed by the same letter are not significantly different.



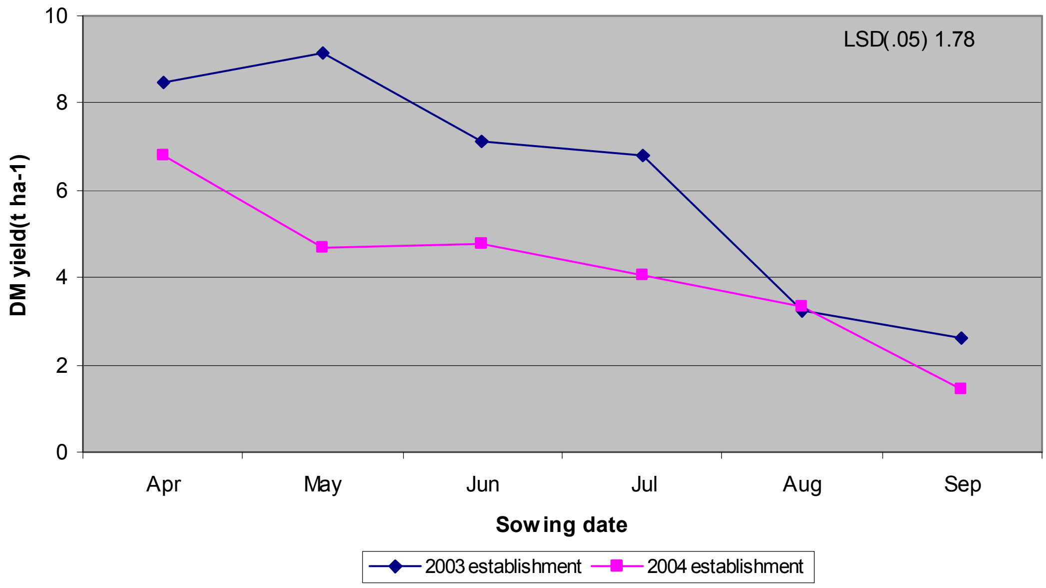
Combined the 2003 and 2004 establishments, the April to July sowings achieved 7.75-8.43 t DM ha^{-1} with no significant differences (Figure 3.11). The August and September sowings yielded 4.22-5.33 t DM ha^{-1}, which were lower than the April to July sowings.

Figure 3.11 Sainfoin average yield of 2003 and 2004 establishments over three years. Values followed by the same letter are not significantly different.



Two years' average yields of the May to July sowings of the 2003 establishment were higher than that of the 2004 establishment, about 2.33-4.43 t DM ha^{-1} higher (Figure 3.12). However, there were no yield differences between the April, August and September sowings of the 2003 establishment and that of the 2004 establishment.

Figure 3.12 Sainfoin two year's average yield of 2003 establishment vs. 2004 establishment over the establishment and 2nd years.



3.3.6 Plant Population

All sowings exhibited a substantial plant population decline over the experimental period (Figure 3.12 & 3.14). Sowing date had significant effects on plant populations both for the 2003 establishment, but only in the 2nd year for the 2004 establishments (Table 3.8). The autumn management only had a significant effect (P<0.01) on plant population of the 2003 establishment in the 3rd year.

Table 3.8 Analysis of variance of sainfoin plant population.

Source of variation	d.f	2003 establishment: Establishment year (M.S)	2nd year (M.S)	3rd year (M.S)
Sowing date (SD)	5 (5) ‡	8834*	24609***	1162**
Autumn management (AM)	1		676	880*
SD × AM	5		418	448
Residual	22 (10)	1774	872	196

Source of variation	d.f	2004 establishment: Establishment year (M.S)	2nd year (M.S)
Sowing date (SD)	5 (4)	3578	5251*
Autumn management (AM)	1		3127
SD × AM	5		1633
Residual	22 (8)	1648	1147

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively.
‡ Numbers in the brackets stands for the d.f of establishment year.

For the 2003 establishment the September sowing achieved the highest plant population in the establishment year, and then showed the most substantial decline (Figure 3.13). Sainfoin yield was negatively correlated with the plant population in the 2nd year (P=0.001, r^2= 0.82), indicating that plant size and plant numbers were in an inverse relationship (Figure 3.14). However, the linear relationship did not exist in the 3rd year (P=0.06, r^2=0.82). Early autumn cutting in the 2nd year reduced sainfoin plants in the 3rd year, compared to late autumn cutting (96.6 plant m^{-2} of late autumn cut plots and 86.7 plant m^{-2} of early autumn cut plots).

For the 2004 establishment, the May sowing achieved the lowest population in the establishment year (Figure 3.15). Linear relationship between sainfoin yield and plant population also did not exist in the 2nd year (P=0.19, r^2=0.01).

Figure 3.13 Sainfoin plant population of 2003 establishment.

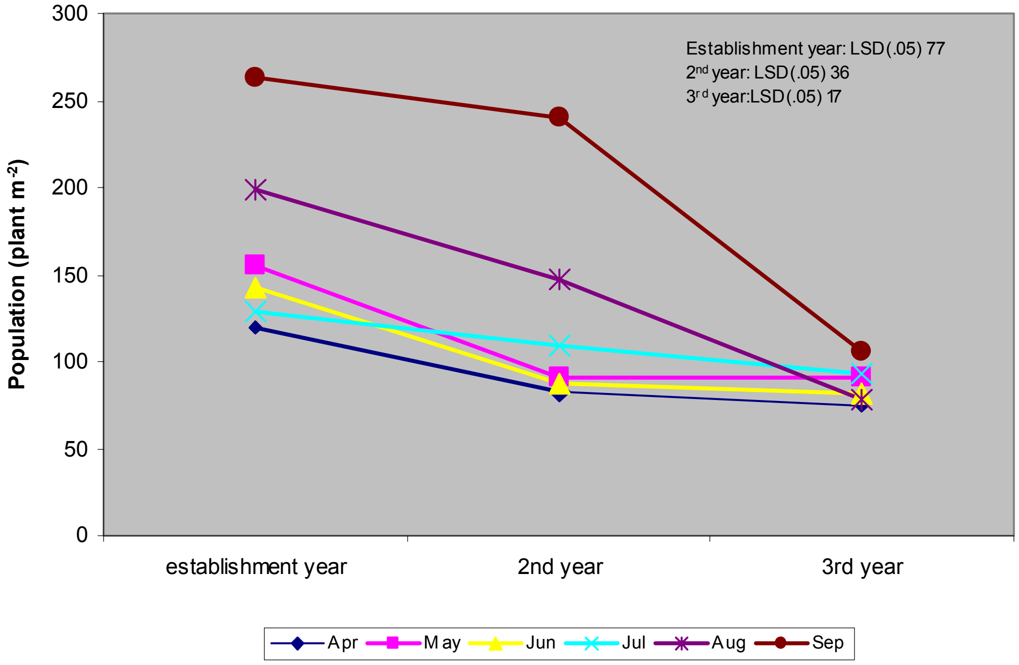


Figure 3.14 Relationship between sainfoin yield and population of 2003 establishment in the 2nd year.

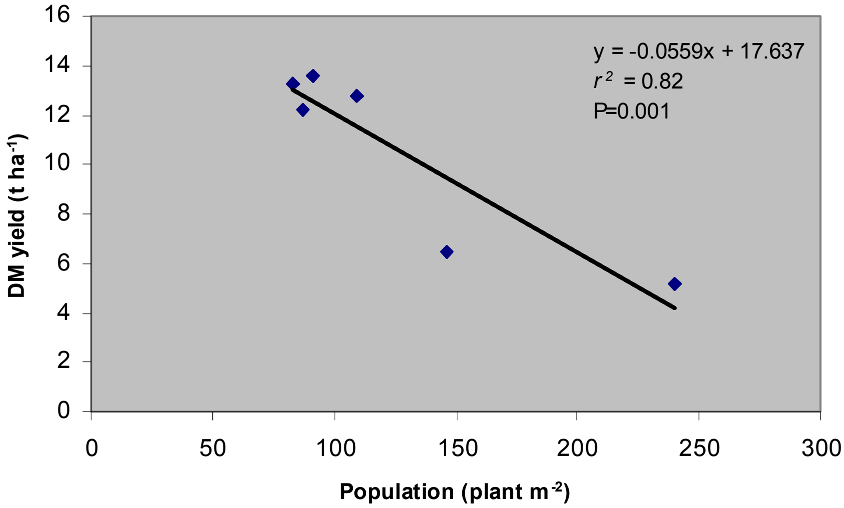
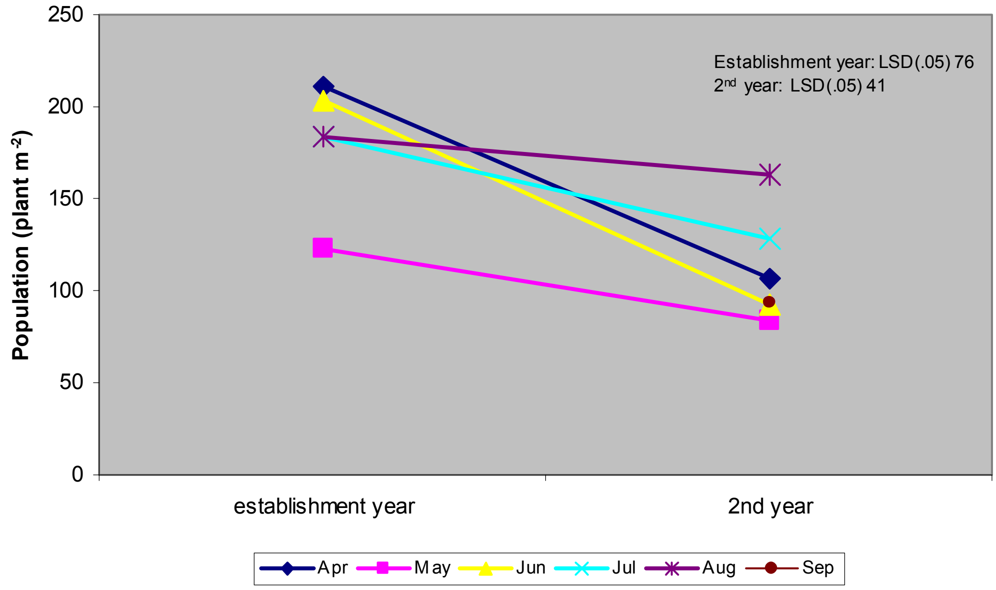


Figure 3.15 Sainfoin plant population for 2004 establishment.



3.3.7 Weed Ingression in the Establishment Year

The April and July sowings had more weeds than the May sowing, and there were no significant differences between the April, June and July sowings and between the May and June sowings in the establishment year (Figure 3.16). The July sowing had the highest weed content, of 50.8%, and the May sowing had the lowest, of 6%. The April and June sowings had a similar weed content, of 26.3% and 22.2% respectively. There was a negative relationship between sainfoin yield and weed content (P=0.04, r^2=0.74) at the 1st harvest of the 2003 establishment in the establishment year (Figure 3.17).

Figure 3.16 Sainfoin yield vs. weed yield of 2003 establishment at the 1st harvest in the establishment year.

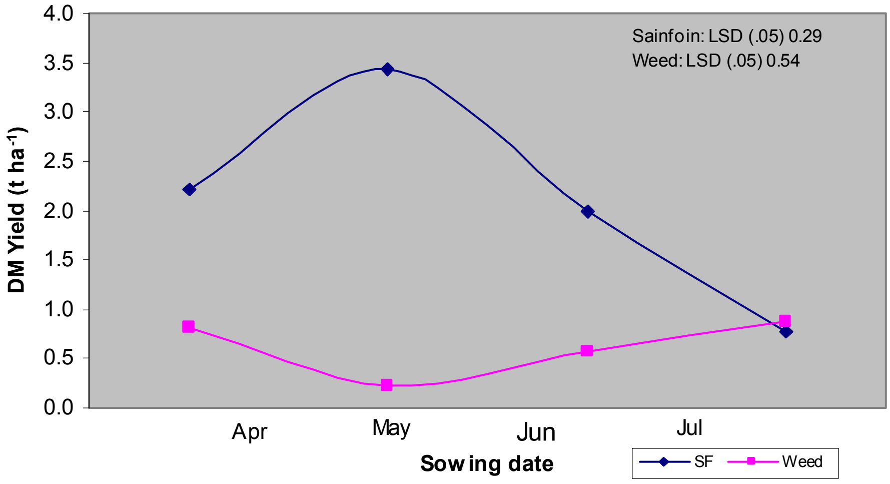
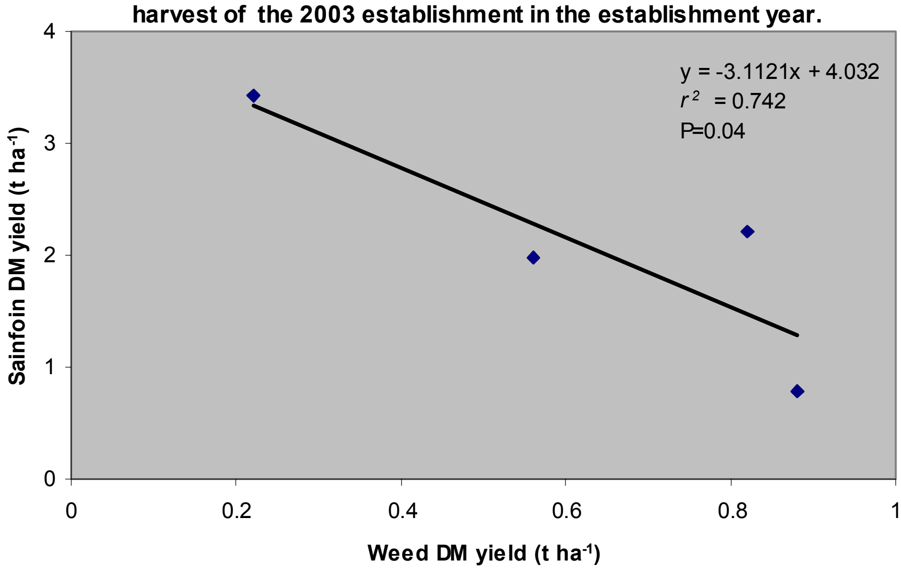


Figure 3.17 Relationship between sainfoin and weed of the 1st harvest of the 2003 establishment in the establishment year.



3.3.8 Forage Quality

Sainfoin of the 2003 establishment was at half flowering (Stage 5, Appendix 1) when cut at the 1st harvest in the 2nd year, while it was at mid–vegetative stage (Stage 1, Appendix 1) at the 2nd and 3rd harvests.

3.3.8.1 Crude Protein

Sowing date did not have any effect on crude protein content (Table 3.9) (Figure 3.18). However, crude protein content increased as the harvests progressed (Figure 3.19). Average crude protein content of early autumn cut plots was significantly increased from 149.8 g kg^{-1} DM at the 1st harvest, to 188.9 g kg^{-1} DM at the 2nd harvest and 230.1g kg^{-1} DM at the 3rd harvest. The annual average crude protein content was 170.3 g kg^{-1} DM. Since the 3rd harvests of the early autumn cut and late autumn cut were taken at different times, only data for the 1st and 2nd harvests was compared for the effect of the autumn management. The Autumn management had no effect on the crude protein content, and the crude protein of the early autumn cut at the first two harvests was 160.3 g kg^{-1} DM and the late cut was 165 g kg^{-1} DM with no significant difference(t_{stat}=-1.86<$t_{(5,\ 5\%)}$=2.57). Although plots of the early autumn cut and the late autumn cut at the 3rd harvest were taken at two different times, the crude protein content was similar (t_{stat}=-4.4<$t_{(5,\ 5\%)}$ =2.57).

Table 3.9 Analysis of variance of sainfoin CP in the 2nd year.

		Harvest		
		1st	2nd	3rd
Source of variation	d.f	M.S		
Sowing date (SD)	5	36	15.7	115
Residual	6	45.1	4.3	167.4

Figure 3.18 Sainfoin CP of early autumn cut plots in the 2nd year. Values followed by the same letter are not significantly different.

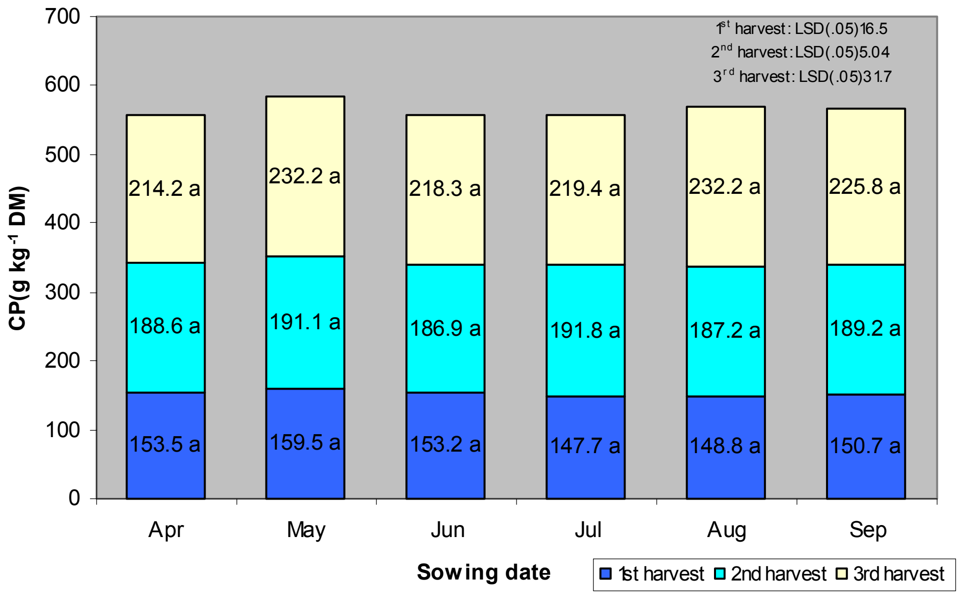


Figure 3.19 Sainfoin CP of early autumn cut plots in the 2nd year. Values followed by the same letter are not significantly different(P<0.001).

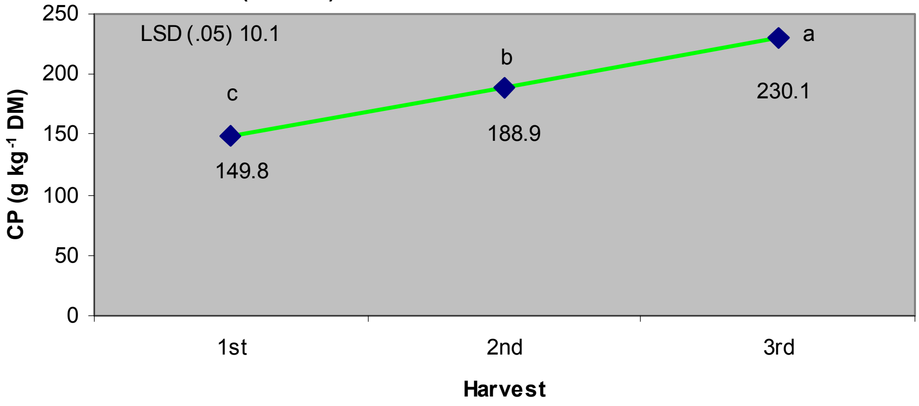


Table 3.10 CP yield of the 2003 establishment in the 2nd year.

April	May	June	July	August	September	LSD(.05)
2.2 ab	2.35 a	2.03 b	2.1 b	1.12 c	0.95 c	0.2

Values followed by the same letter are not significantly different.

Crude protein yield varied (P< 0.001) as sowing dates changed (Appendix 3.3). The August and September sowings had the lower crude protein yield (Table 3.10). The May sowing had the highest crude protein yield apart from no difference with the April sowing.

3.3.8.2 Neutral Detergent Fibre

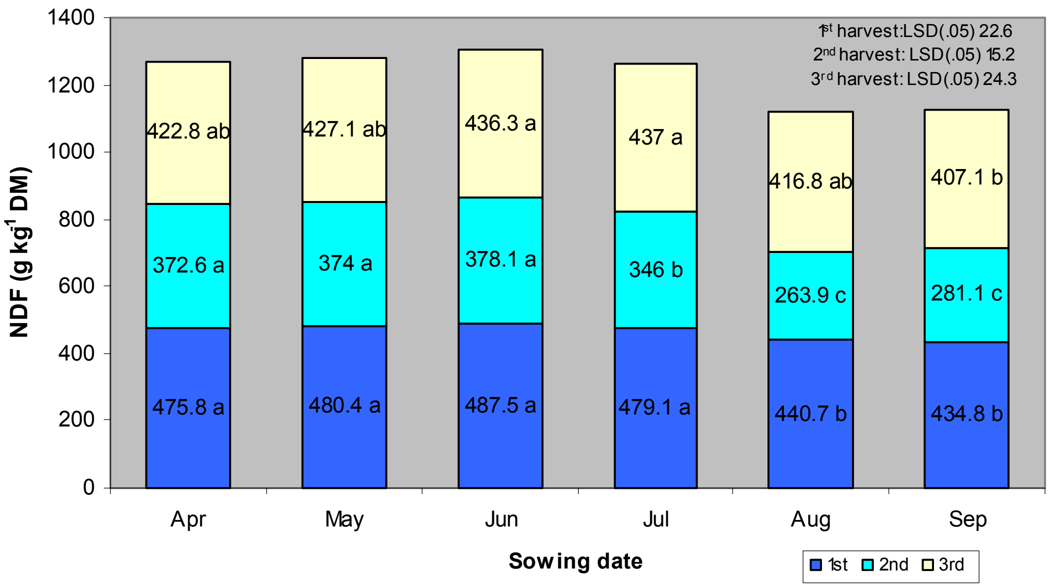
Sowing date had significant effects on NDF at the 1st (P<0.01) and the 2nd harvests (P<0.001), but not at the 3rd harvest (Table 3.11). At the 1st harvest, the April to July sowings had significantly higher NDF content than August and September sowings (Figure 3.20). At the 2nd harvest, the April to June sowings had significantly higher NDF content than the July to the September sowings, and the July sowing was higher than the August and September sowings. At the 3rd harvest, the September sowing had lower NDF content than other sowings.

Table 3.11 Analysis of variance of sainfoin NDF in the 2nd year.

		Harvest		
		1st	2nd	3rd
Source of variation	d.f		M.S	
Sowing date (SD)	5	1022**	5150***	266
Residual	5	77	35	89

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively.

Figure 3.20 Sainfoin NDF in the 2nd year. Values followed by the same letter are not significantly different.



NDF content also varied as the season progressed (Figure 3.21). The annual NDF content was 424.3 g kg^{-1} DM. Only the data of the 1st and 2nd harvests were compared for the effect of autumn management. Autumn management had no effect on NDF content, and NDF of the early autumn cut at the first two harvests was 427.2 g kg^{-1} DM and the late cut was 426.7 g kg^{-1} DM (t_{stat}=-0.17< $t_{(5,\ 5\%)}$=2.57). Not surprisingly the late autumn cut plots at the 3rd harvest had higher NDF content compared to plots of the early autumn cut at the 3rd harvest (Figure 3.22).

Figure 3.21 Sainfoin NDF of early autumn cut of 2003 establishment in the 2nd year. Values followed by the same letter are not significantly different(P<0.001).

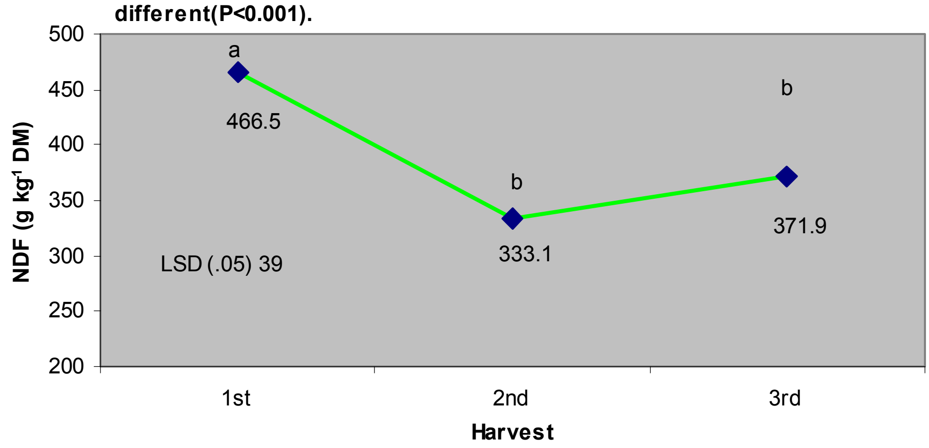
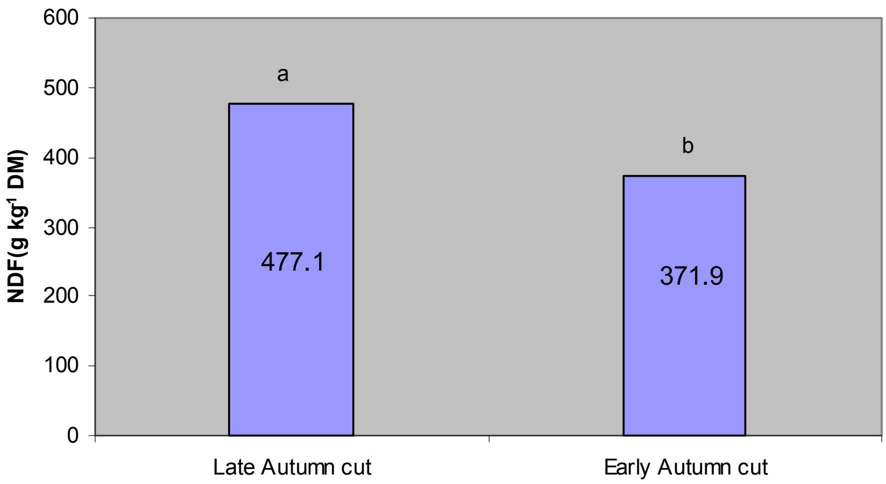


Figure 3.22 NDF of early autumn cut vs. late autumn cut at the 3rd harvest in the 2nd year(t_{stat} =19.3 > $t_{(5,\ 5\%)}$ =2.57). Bars followed by the same letter are not significantly different.



3.4 Discussion

3.4.1 Sowing Date and Yield

Three year's average sainfoin yields from 2003 and 2004 establishments showed that the sowing date generally affected yield. The April to July sowings achieved 7.75-8.43 t DM ha^{-1} with no significant differences. This was significantly higher than the August and September sowings, which yielded 4.22 and 5.33 t DM ha^{-1}. These results are in accordance with traditional recommendations for sainfoin sowing dates, which are between April and July (Bland, 1971; Spedding & Diekmahns, 1972). However, sainfoin yields and crop performance between the April and July sowings varied between these two different establishment years. Average yields over three years from the 2003 establishment showed that the May sowing achieved a significantly higher yield (9.83 t DM ha^{-1}) than the April, June and July sowings (8.48-8.93 t DM ha^{-1}). Average yields over two years from the 2004 establishment showed that April sowing had a greater yield (6.8 t DM ha^{-1}) than the May to July sowings (4.07-4.77 t DM ha^{-1}). These results were in partial agreement with Bland (1972) who also suggested that the April and May sowings may achieve better establishment than the June and July sowings (according to the weather conditions). Furthermore, the April and May sowings can give two harvests in the establishment year, whereas the June and July sowings only give one. The August and September sowings gave no measurable production in the establishment years from either the 2003 or 2004 establishment. Although no other studies on sowing date were found, the results of this study was in agreement with traditional recommendations and are also similar to the results of sowing date studies on red clover (Frame *et al*., 1976b).

Sainfoin yield over three years peaked in the 2nd year and then declined in the 3rd year. This trend was similar to the results of the trial at the Grassland Research Institute in 1956 (Spedding & Diekmahns, 1972). The April to July sowings in the establishment year for the 2003 establishment achieved 0.78–4.63 t DM ha^{-1} yield. The May sowing of 2003 establishment in the establishment year yielded 4.63 t DM ha^{-1}, which is similar to the result obtained in Aberystwyth by the Institute of Grassland and Environmental Research, of 4.7 t DM ha^{-1} (Fychan & Jones, 1997). The April to July sowings in the 2nd year for the 2003 establishment produced 12.85–14.45 t DM ha^{-1} yield, and 9.55-11.5 t DM ha^{-1} yield in the 3rd year. These results were similar to the results in the literature (Spedding & Diekmahns, 1972; Sheehy &

Popple, 1981; Goplen *et al.*, 1991; Koivisto & Lane, 2001). However the yields of the April to July sowings in the 2nd year of 2004 establishment were only 8.18–10.55 t DM ha^{-1}, which was lower than the 2003 establishment. This may have been due to the dry conditions in June and July of 2004 compared to 2003 (rainfall 26.2 mm and 33.7 mm lower in 2004 respectively) and wet weather and low temperature in August (rainfall 103.8 mm higher and temperature 2 °C lower in 2004) which led to delayed emergence and increased weed competition.

3.4.2 Autumn Management

Although no research information was available on the autumn management of sainfoin, leaving enough time for other perennial legumes (such as lucerne and red clover) to accumulate root reserves in the autumn is an important farming practice (Grandfield, 1935; Jones, 1955; Bosworh, 2006). Jones (1955) studied autumn management of lucerne in a series of experiments. Spaced plants were cut in August, September and November in the 1st harvest year, and root weights were taken on April 8 of the following year. He found that November cutting gave higher root weights and bigger root diameters than August and September cuts. He also found that September cut plants yielded less than August and October cut plants, from data extracted from 64 fields sown with different lucerne varieties over a five year period.

In this study, autumn management in the establishment year appeared to improve yield in the 2nd year for the 2003 establishment but not for the 2004 establishment. This was due in fact to the reduced yield of the late cut taken in November 2003, when excessive loss of leaves resulted in a poor recovery of the plant material. The late autumn cut treatment out yielded the early autumn cut treatment in the 3rd year, giving an extra yield of 1.33 t DM ha^{-1} (9.72 t DM ha^{-1}compared with 11.05 t DM ha^{-1}).

Exact timing of autumn cutting of sainfoin stands is clearly a topic which justifies more detailed study. An early cut at the end of August, or the very beginning of September, may be preferable to maintain better yield and quality.

3.4.3 Weed Ingression

Weed ingression was a significant feature of these plots. Weed content among the April and July sowings at the 1st harvest in the establishment year varied. The May sowing (May 15th) had the lowest weed content, at only 6% whilst the July sowing (July 16th) had the highest weed content, at 50.8%, and the April (April 17th) and June (June 20th) sowings had the similar weed content, at 26.3% and 22.2% respectively. The weed content of the April sowing was similar to the reported results of Fychan and Jones (1997) (24.9% weed, established on May 1st 1997). These results indicate that a Mid-May sowing could minimise weed ingression. A negative linear relationship between sainfoin yield and weed mass at the 1st harvest in the 2003 establishment emphasises the importance of adequate broad leafed weed control. MCPA + MCPB application in this study was effective to control most of the broadleaved weeds.

3.4.4 Plant Population

120-156 plant m^{-2} from the 2003 establishment was achieved in the establishment year, which met the suggestion of 70-150 plant m^{-2} from the Grassland Research Institute studies (1982). This declined to 75-106 plant m^{-2} in the 3rd year. Roughly 123 -211 plant m^{-2} from the 2004 establishment was achieved in the establishment year, similar to the results of Fychan and Jones (1997). This dropped to 84-163 plant m^{-2}. For the 2003 establishment there was a negative linear relationship in the 2nd year between sainfoin yield and plant population, indicating that the August and September sowings had a large number of small plants with lower yields, compared to lower population from the April to July sowings with higher individual plant yields. However, there was no linear relationship between sainfoin yield and the plant population in the 3rd year, nor for the 2004 establishment in the 2nd year. The nonlinear relationship between yield and population may be explained by the law of constant final yield (Harper, 1977) which states that yield is independent of population at some stages when the plant population exists above a certain level.

3.4.5 Forage Quality

The crude protein content increased as the season progressed. The 1st harvest had 149.8 g kg^{-1} DM and the 2nd harvest 188.9 g kg^{-1} DM and the 3rd harvest 230.1 g kg^{-1} DM. This is because the leaf percentage increased as the season progressed. Fagan and Rees (1930) reported leaf percentage increased from 43 % in May 26 at the 1st growth, to 65% in July 12 and 90% August 11 at regrowth. Also after the 1st cut, sainfoin did not form stems, and this decreased stem percentage (Bland, 1970).

Neutral Detergent Fibre declined following the seasonal progress. The 1st harvest of early autumn cut treatments had higher Neutral Detergent Fibre, 466.5 g kg^{-1} DM, compared to 331 and 371.9 g kg^{-1} DM at the 2nd and the 3rd harvests. The highest Neutral Detergent Fibre in the 1st harvest is due to the reproductive period (Stage 5, Appendix 1) while the 2nd and 3rd harvests occurred in the vegetative period (Stage 1, Appendix 1). The August and September sowings were less fibrous than the April to July sowings at the 1st and 2nd harvests. This may be because plants of the August and September sowings did not have enough favourable weather period to develop, and this delayed growth in the subsequent year. By the time of the 1st harvest, the August and September sowings were at early bud (Stage 3, Appendix 1) while the April to July sowings were at early flowering (Stage 5, Appendix 1).

Autumn management did not affect crude protein and Neutral Detergent Fibre content in subsequent years. However, the late autumn cutting decreased sainfoin forage quality at the 3rd harvest because of loss of leaf.

3.5 Conclusions

- The April to July sowings gave similar yields over three years of, about 8 t DM ha^{-1}. However, this varied depending on weather conditions at establishment. The May sowing yielded up to 9.8 t DM ha^{-1} if established in favourable weather conditions. April appeared the most reliable sowing date, with good soil moisture for germination and emergence.
- August and September proved to be unsuitable sowing dates and produced no measurable yield in the establishment year and less in subsequent years, compared to the April to July sowings. This implies that sainfoin would not be suitable for sowing after harvesting winter cereals.
- When the 3rd harvest took place in late autumn (November) this led to a lower yield and an increase of fibre in the forage, compared to the early autumn cut due to leaf loss.
- Early autumn cutting reduced sainfoin yield in the 3rd year.
- Sowing date had no effects on sainfoin forage quality between the April and July sowings.
- Autumn management did not affect sainfoin forage quality in subsequent years.

Chapter Four

The Effects of Direct Sowing and Undersowing in Spring Barley on Sainfoin and Sainfoin-Grass Mixtures

Abstract

Sainfoin grown with meadow fescue (cv.Lifara) or perennial ryegrass (cv. Condesa) in two seeds mixtures under direct sowing and undersowing in spring barley were studied over a three years period. 2/3 sainfoin-1/3 meadow fescue gave the highest average yield (9.07 t DM ha^{-1}) over three years. Other mixtures yielded similar to sainfoin. Undersowing reduced the yields of sainfoin-grass mixtures in the establishment and 2nd years. Sainfoin appeared to more compatible with perennial ryegrass cv. Condesa than with meadow fescue cv. Lifara.

4.1 Introduction

4.1.1 Sainfoin-Grass Mixtures

Sainfoin is traditionally sown as a forage crop for hay, normally with a less aggressive companion grass, such as meadow fescue or timothy to suppress weeds (Bland, 1971; Sheldrick *et al.*, 1995; Frame *et al.*, 1998). Sainfoin was also used in pasture for grazing (Goplen *et al*., 1991). Lambs were traditionally grazed on the aftermath. Legumes grown with grasses in mixtures provide advantages for forage production over grasses grown alone. However, research information on sainfoin-grass mixtures in the UK is very limited.

A five-year study on mixtures of sainfoin-grass was reported in dryland conditions in Canada (Goplen *et al*., 1991). Five-year average yields of mixtures of sainfoin-Russian wild rye, sainfoin-crested wheatgrass and sainfoin-pubescent wheatgrass were 5.78 t DM ha^{-1}, 5.21 t DM ha^{-1} and 4.84 t DM ha^{-1} respectively, and the average legume percentages were 61%, 48% and 39% respectively. A five-year study on grazing with sheep on a sub-irrigated site at Lethbridge, Canada, showed that sainfoin yielded as much as lucerne (Goplen *et al*., 1991). A study in the southern Great Plains in the USA showed that light or medium grazing at bud or flowering stage may be suitable under irrigation (Mowrey & Matches, 1991).

Sainfoin grown with Kentucky bluegrass and red fescue (*Festuca rubra*) was studied in Montana, USA over a four year period (Cooper, 1972). There were no yield differences between them. Sainfoin also grew well with Russian wild rye in southwest Canada on dryland and with crested wheatgrass (Hanna *et al.,* 1977; Kilcher, 1982). A study on sainfoin sown alone and mixed with tall wheatgrass, crested wheatgrass, and smooth brome grass was carried out in Turkey. The results showed that sainfoin-grass mixtures had higher yields than sainfoin monoculture (Sengul, 2003).

Sainfoin grown with other legumes was also tested in a few studies. Sainfoin grown with ladino clover, birdsfoot trefoil and white clover was studied in Montana, USA over a four year period (Cooper, 1972). Sainfoin-birdsfoot trefoil was most compatible and productive, whereas the ladino clover and white clover showed too much competitiveness. A study on a sainfoin-lucerne mixture was also conducted in Canada and the results showed the composition shifted to lucerne dominance, especially when they were drilled together (Jefferson *et al.*, 1994). In recent years sainfoin grown with lucerne for grazing has been studied to make use of the condensed tannins in sainfoin to reduce bloating (McMahon *et al.*, 1999).

Little Information on sainfoin-grass mixtures in the UK is available. A four-year study on sainfoin-grass by the Grassland Research Institute in 1950s showed that sainfoin grew better with meadow fescue and timothy than with cocksfoot in terms of forage production and that sainfoin monoculture had better production than sainfoin-grass mixtures (Spedding & Diekmahns, 1972). NIAB (2000) reported on a trial conducted for Cotswold Seeds Ltd which involved two varieties of sainfoin and mixtures with perennial ryegrass, meadow fescue, timothy and cocksfoot (*Dactylis glomerata*). Sainfoin (cv. Emyr) with cocksfoot (cv. Prairial) yielded best (over 15 t DM ha^{-1}), but not significantly better than sainfoin sown alone.

Undersowing legumes in a cereal crop is a method which has been widely used for forage establishment (Miller & Stritzke, 1995; Frame *et al.*, 1998; Odhiambo & Bomke, 2001). Sainfoin undersown in a cereal crop was traditionally practised in the UK. This gives some production in the establishment year. Sainfoin undersown in rye and wheat-vetch (Maksimenko, 1973) and in barley (Popov, 1979) was tested in the former Soviet Union. Undersowing reduced weed invasion in the establishment year but pure stands suffered from weed competition. A study on sainfoin undersown in silage maize was also conducted in Slovak Republic (Jamriska, 2002).

4.1.2 Objectives

The objectives of this experiment was,

- to explore the impact of direct sowing and undersowing on the establishment, growth, production and persistence of sainfoin-grass mixtures,
- secondly, to investigate the compatibility of sainfoin with meadow fescue or tetraploid perennial ryegrass,
- and thirdly, to examine the effect of combinations of different seed rates in mixtures on forage yields.

4.2 Materials and Methods

4.2.1 Experimental Site

The site is described as in 3.2.1.

4.2.2 Experiment Design

The experimental design was a split plot arrangement of a randomised block design with three replications. Direct sowing and undersowing constituted the main plots, and the monocultures of sainfoin, meadow fescue, perennial ryegrass and their mixtures made up the sub-plots. Sainfoin and grasses were mixed in two ratios. 1/3 sainfoin monoculture seed rate and 2/3 grass monoculture seed rate made up one mixture, and 2/3 sainfoin monoculture seed rate and 1/3 grass monoculture seed rate made up another mixture. The size of plots was 2 m by 4 m.

Treatments were as follows:

Main treatments:

- Direct sowing
- Undersowing in spring barley

Sub-treatments:

- Sainfoin (SF) cv. Cotswold Common,
- Meadow fescue (MF) cv. Lifara,
- Tetraploid perennial ryegrass (PRG) cv. Condesa,
- 1/3 MF and 2/3 SF,
- 2/3 MF and 1/3 SF,
- 1/3 PRG and 2/3 SF, and
- 2/3 PRG and 1/3 SF.

4.2.3 Field Preparation

As described in 3.2.3.

4.2.4 Establishment

Barley was drilled on April 21st 2003 at a seed rate of 120 kg ha^{-1}. Sainfoin, meadow fescue, perennial ryegrass and their mixtures were sown in May 21st when the barley was about at 3 leaves stage (Zadoks 1.3, Zadoks, Chang, & Konzak, 1974). Some fine soil was added in the seeds to ease broadcasting. Seeds were broadcast by hand, and raked into the soil to about 1.0 cm deep, and then rolled. Sainfoin was sown at a seed rate of 90 kg ha^{-1}; meadow fescue at 22 kg ha^{-1}; perennial ryegrass at 29 kg ha^{-1}, mixtures were weighed out to supply 1/3 or 2/3 of the monoculture seed rates.

4.2.5 Management

MCPA + MCPB was used to control broadleaved weeds and applied after the 1st trifoliate stage of sainfoin (Stage 0, Appendix 1)

Fertiliser was broadcast by hand. Nitrogen was also applied in the spring in the 2nd and 3rd year. Phosphate and potash were applied in accordance with MAFF (2002). 30 kg ha^{-1} of P_2O_5 was given to all plots on August 27, 2003 after barley and direct sown plants were cut (Table 4.1). 50 kg ha^{-1} of nitrogen was given in April in the 2nd and 3rd years; 20 kg ha^{-1} of P_2O_5 and 30 kg ha^{-1} of K_2O were given after the 1st harvests in the 2nd and 3rd years; 40 kg ha^{-1} of K_2O and 20 kg ha^{-1} of K_2O were given after the 2nd and 3rd harvests in the 2nd and 3rd years respectively.

Table 4.1 Fertilizer application.

		N	P_2O_5	K_2O
		kg ha^{-1}		
Establishment year		-	30	-
	1st	50	20	30
2nd year	2nd	-	-	40
	3rd	-	-	20
	1st	50	20	30
3rd year	2nd	-	-	40
	3rd	-	-	20

4.2.6 Plant Population Measurement

A 25 x 25 cm quadrat was used to measure plant population and two determinations were made in each subplot. In the establishment year, determination was made after barley was cut. In the 2nd and 3rd years, determinations were made in early June after the 1st harvests.

4.2.7 Sampling and Harvest

One harvest was taken from the direct sown subplots and barley was harvested on August 8, 2003. No harvest was taken from undersown plots in the establishment year, as there was little growth present. In the 2nd year, three harvests were taken on May 26-27, July 17-18 and October 15 respectively. In the 3rd year, three harvests were taken on May 29, July 13 and September 12. However, monocultures of meadow fescue and perennial ryegrass gave only two harvests in the 2nd and one harvest in the 3rd year.

4.2.8 Laboratory Analysis

Samples were taken back to the laboratory after sampling, hand-separated into sainfoin, grass and weed fraction, and then dried in the oven at 100±2°C over 24 hours to determine dry matter yields. Crude protein and Neutral Detergent Fiber was analyzed by the methods described as in 3.2.8.

4.2.9 Statistical Analysis

Statistical analysis methods used were described as in 2.2.

4.3 Results

4.3.1 DM Yield

4.3.1.1 Direct Sowing and Undersowing

Undersowing in spring barley generally reduced the annual average yields of sainfoin, meadow fescue, perennial ryegrass and mixtures in the establishment and the 2nd years ($P<0.001$) (Table 4.2). However, undersown were comparable with direct sown plots in the 3rd year (Table 4.3).

Undersown crops did not produce any yield in the establishment year, but direct sown crops produced 1.8 t DM ha^{-1} yield (Table 4.3). However, the spring barley gave 4.8 t DM ha^{-1} grain yield. Undersown crops achieved 10.31 t DM ha^{-1} average yield in the 2nd year, which was 1.91 t DM ha^{-1} lower than direct sowing (Table 4.3). There were also significant reductions in the yields of the 1st ($P<0.001$) and 2nd ($P<0.05$) harvests in the 2nd year (Table 4.2 & 4.3). Direct sown and undersown crops yielded 7.7 t DM ha^{-1} and 7.9 t DM ha^{-1} respectively with no significant yield difference in the 3rd year (Table 4.2 & 4.3). There was a significant difference between direct sown and undersown crops at the 2nd harvest in the 3rd year. Direct sown crops achieved 7.24 t DM ha^{-1} mean yield over three years, which was significantly higher (Appendix 4.1 & 4.2) than the 6.07 t DM ha^{-1} of undersown crops.

Table 4.2 Analysis of variance of sainfoin-grass DM yield of direct sowing and undersowing over three years.

	d.f	Establish, year	2nd year harvests				3rd harvests			
			1st	2nd	3rd	Total	1st	2nd	3rd	Total
		M.S								
Sowing treatment (ST)	1		19.7***	0.87*	0.06	38.1***	0.28	0.07*	0.1	0.26
Mixtures(M)	6	0.48	16.8***	2.47***	13.6***	81.6***	15.5***	2.21***	3.13***	37.5***
ST x M	1		0.97	0.08	0.1	1.73	0.49	0.02	0.04	0.57
Residual	26 (12)	0.21	1.14	0.07	0.13	1.6	0.47	0.01	0.04	0.5

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively;
d.f values in bracket stand for the d.f of the establishment year.

Table 4.3 DM yield of direct sown and undersown crops over three years.

		1st harvest	2nd harvest	3rd harvest	Annual
		t DM ha^{-1}			
Establishment year	Direct sowing	1.8	N.A	N.A	1.8
	Undersowing	N.A	N.A	N.A	N.A
2nd year	Direct sowing	8.11 a	1.98 a	2.13 b	12.22 a
	Undersowing	6.74 b	1.69 b	2.05 b	10.48 b
	LSD(.05)	0.68	0.17	0.23	1.5
3rd year	Direct sowing	5.85 a	0.81 b	1.01 a	7.7 a
	Undersowing	6.01 a	0.89 a	1.0 a	7.9 a
	LSD(.05)	0.43	0.07	0.13	0.47

Values followed by the same letter are not significantly different.
N.A= Not applicable.

4.3.1.2 Sainfoin-Grass Mixtures

There were significant differences between sainfoin mixtures in the 2nd (P<0.001) and 3rd years (P<0.001) (Table 4.3). Perennial ryegrass was out yielded by sainfoin and the mixtures except 1/3 sainfoin-2/3 meadow fescue in the establishment year (Figure 4.1). Sainfoin monoculture and sainfoin-grass mixtures out yielded meadow fescue and perennial ryegrass monocultures in the 2nd year (Figure 4.3). Sainfoin-meadow fescue mixtures yielded higher than sainfoin monoculture, and sainfoin-perennial ryegrass mixtures yielded as well as sainfoin monoculture in the 2nd year (Figure 4.3). The first cut of sainfoin and the mixtures constituted 60-65% of annual yield; for meadow fescue and perennial ryegrass the figure was 82-85%. Sainfoin-meadow fescue mixtures yielded the highest, and meadow fescue and perennial ryegrass yielded lowest in the 3rd year (Figure 4.4). Sainfoin yielded similar to sainfoin-perennial grass mixtures in the 3rd year. The 1st harvest gave 65-78% of the annual yield.

Figure 4.1 Comparison of DM yield of monocultures of sainfoin, meadow fescue, perennial ryegrass and their mixtures in direct sowing in the establishment year.Values followed by the same letter are not significantly different.

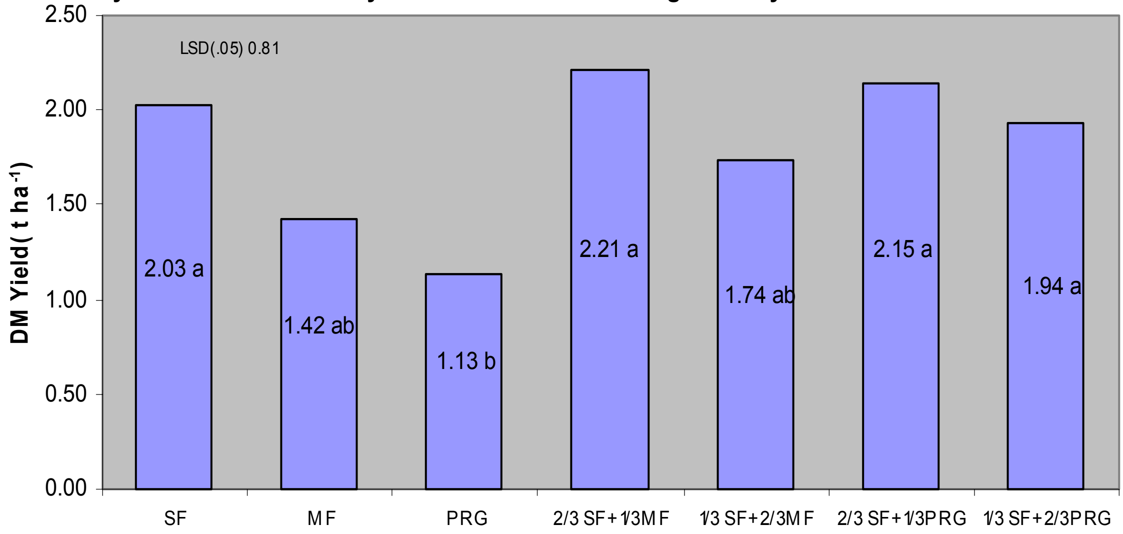


Figure 4.2 The 3rd block of direct sowing (photos taken in April 26 2004)

a) SF

b) MF

c) PRG

d) 2/3SF+1/3MF

e) 1/3SF +2/3MF

f) 2/3SF+ 1/3 PRG

g)1/3SF+2/3PRG

Figure 4.3 Yield of sainfoin, meadow fescue, perennial ryegrass and mixtures in the 2nd year. Values in the top of bars stands for annual yield. Values followed by the same letter are not significantly different.

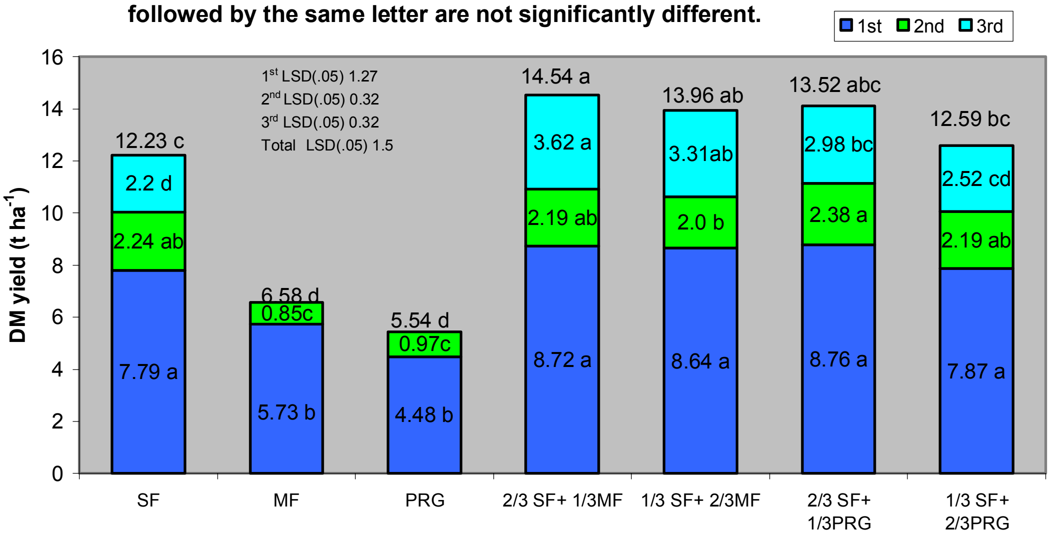
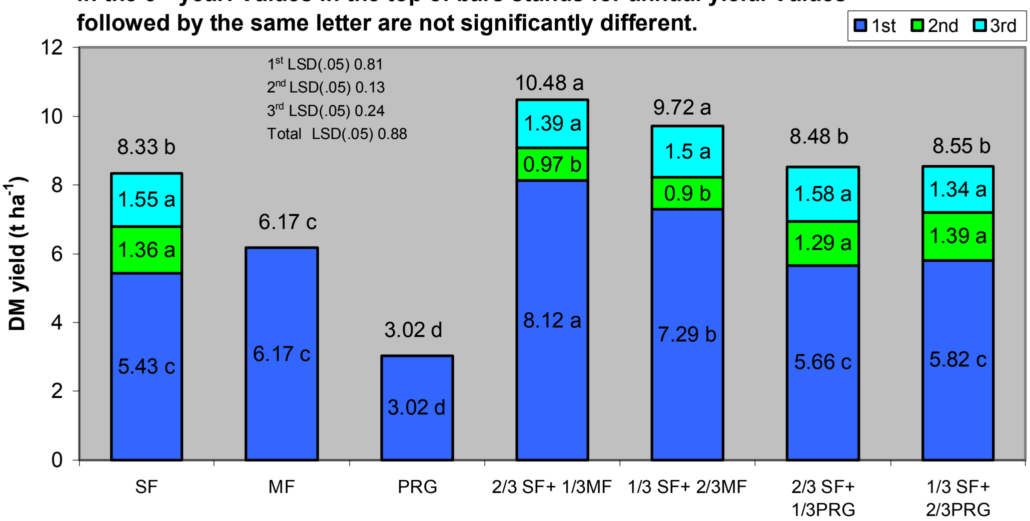


Figure 4.4 Yields of sainfoin, meadow fescue, perennial ryegrass and mixtures in the 3rd year. Values in the top of bars stands for annual yield. Values followed by the same letter are not significantly different.



Over the three year period of the trial, 2/3 sainfoin-1/3 meadow fescue was the most consistent and highest yielding mixture (Appendix 4-3 & Figure 4.5). The yield of monocultures and sainfoin-grass-mixtures peaked in the 2nd year and declined in the 3rd year (Figure 4.6).

Figure 4.5 Three-year's mean yield of sainfoin,meadow fescue, perennial ryegrass and their mixtures. Bars with the same letter are not significantly different.

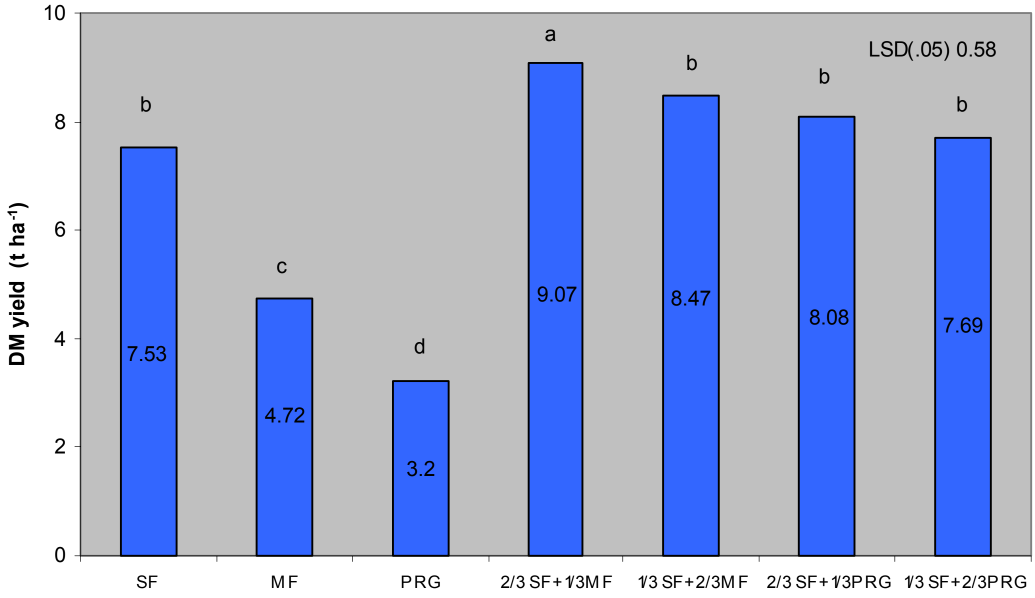
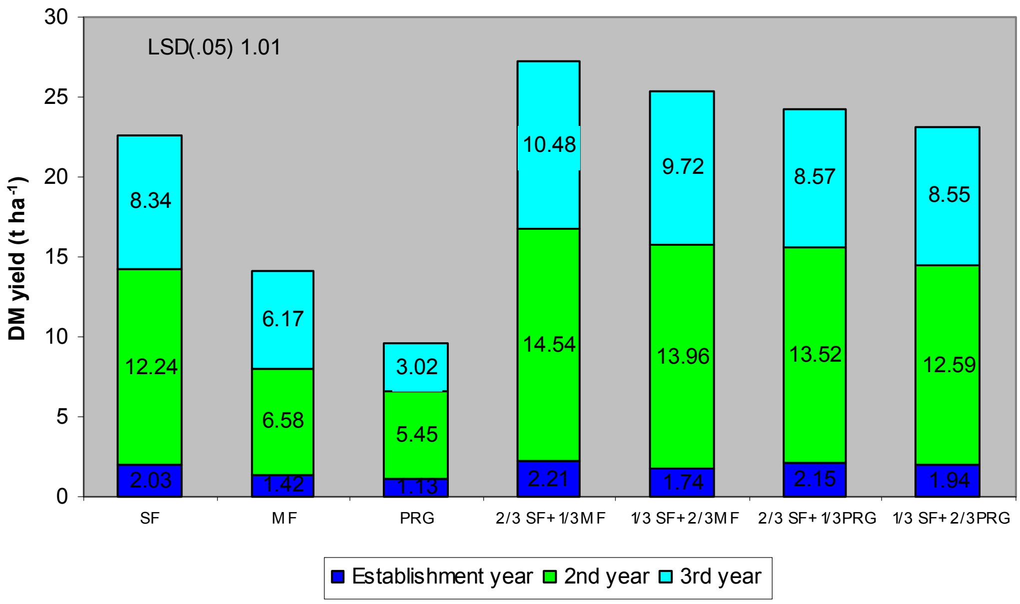


Figure 4.6 Yield distribution of sainfoin, meadow fescue, perennial ryegrass and their mixtures over three years. LSD appropriate for within and between years.



4.3.2 Sainfoin Proportion in Mixtures

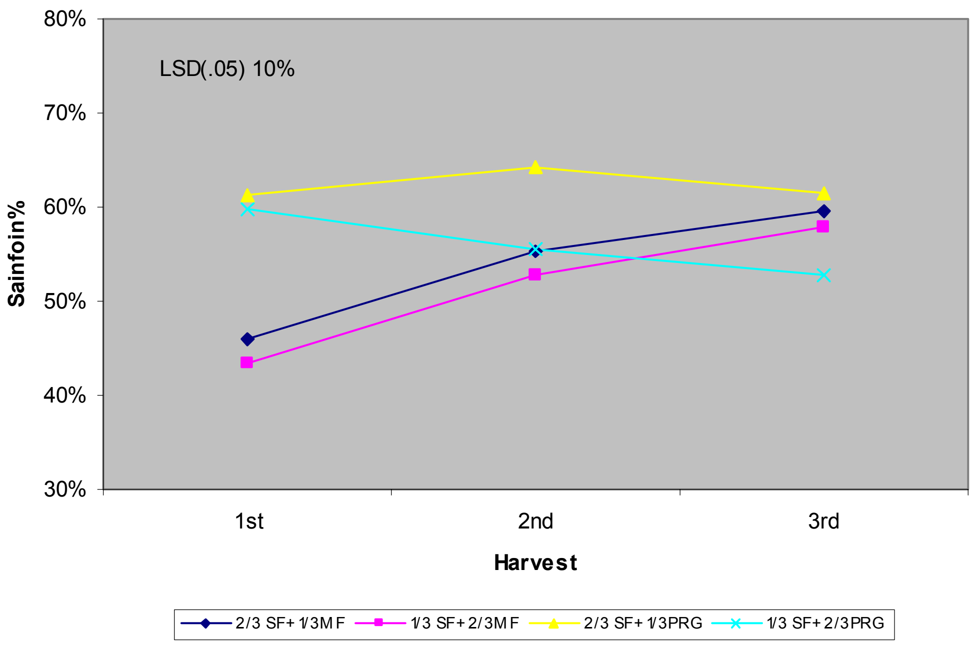
The sainfoin proportion differed in mixtures and between years (Table 4.4). There were no significant differences in sainfoin proportion between sainfoin-meadow fescue mixtures at all harvests (Figure 4.7). However, the sainfoin proportion in the sainfoin-meadow fescue mixtures increased as the season progressed. Sainfoin proportions in sainfoin-meadow fescue mixtures were 57.8 and 59.5% at the 3rd harvest, higher than 43.4 and 46% at the 1st harvest and 52.8 and 55.4% at the 2nd harvest. There were also no significant differences in sainfoin proportion between sainfoin-perennial ryegrass mixtures at all harvests, or between harvests (Figure 4.7).

Table 4.4 Analysis of variance of sainfoin proportion in sainfoin-grass mixtures of direct sowing over harvests and growing seasons.

	d.f	M.S
Mixtures (M)	3	0.02 *
Harvests (H)	2	0.01
M x H	6	0.008
Residual	22	0.003
Mixture (M)	3	0.03
Year (Y)	2	0.08 ***
M x Y	6	0.06***
Residual	22	0.009

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

Figure 4.7 Sainfoin percentage in mixtures. Datas are the average of the 2nd and 3rd years. LSD appropriate for within and between harvests.



Growing season had an effect ($P<0.001$) on the sainfoin proportion in mixtures, and sainfoin proportion in mixtures also significantly interacted ($P<0.001$) with growing seasons (Table 4.4). Sainfoin proportion in direct sown sainfoin-meadow fescue mixtures constantly declined over the three years' period (Figure 4.8). Sainfoin proportion in sainfoin-perennial ryegrass mixtures increased in the 2nd year and then kept stable in the 3rd year (Figure 4.8). Sainfoin proportion in undersown sainfoin-grass mixtures in the 2nd and 3rd year showed a similar trend to direct sown crops (Figure 4.9)

Figure 4.8 Sainfoin forage proportion in mixtures in direct sowing over three growing seasons . LSD appropriate for within and between years.

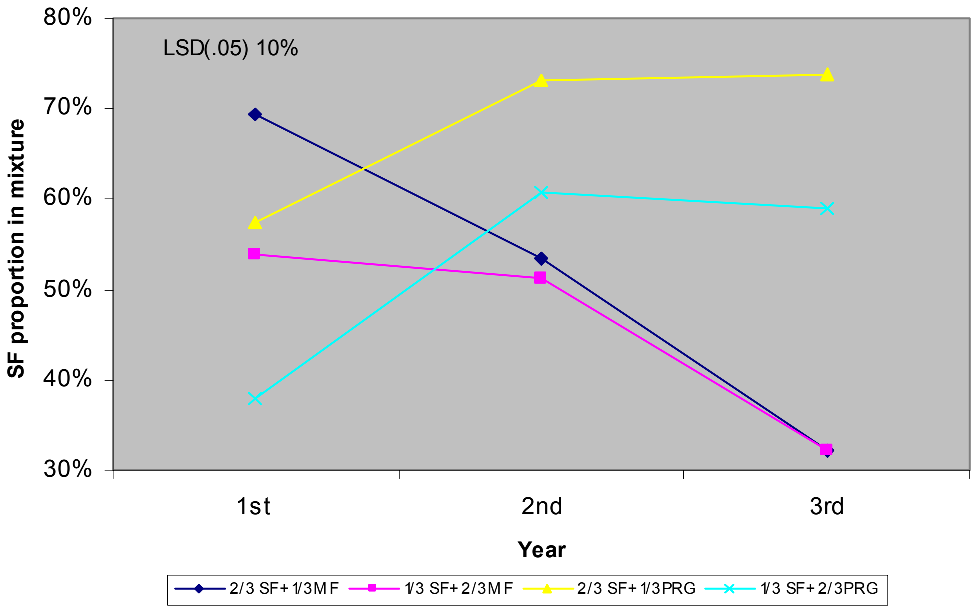
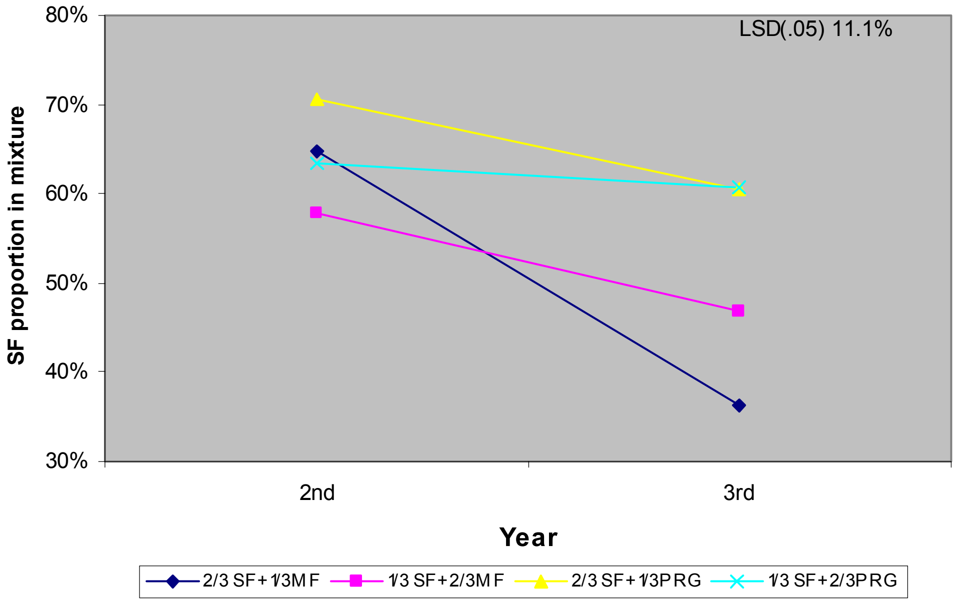


Figure 4.9 Sainfoin forage proportion in mixtures in undersowing over two growing seasons . LSD appropriate for within and between years.



4.3.3 Plant Population

Sainfoin-grass mixtures in direct sowing and undersowing achieved 87.5 and 98.7 plant m^{-2} respectively in the establishment year with no difference (t_{stat} = -2.36 < $t_{critical\ (5,\ 5\%)}$ = 2.08). Sainfoin population in the mixture of 2/3 sainfoin-1/3 meadow fescue showed a sharp decline in the 2nd year (Appendix 4.4 & Table 4.5).

Table 4.5 Sainfoin population in sainfoin-grass mixtures over three years.

	1st year	2nd year	3rd year
2/3 SF + 1/3 MF	73.1 a	48 b	33 bc
1/3 SF + 2/3 MF	37.9 a	37.7 a	32 a
2/3 SF + 1/3 PRG	56 a	48 a	44 a
1/3 SF + 2/3 PRG	36.2 a	35.5 a	25.3 a
LSD(.05)		16.9	

Values followed by the same letter are not significantly different.
Comparison made within mixtures.

4.3.4 Forage Quality

There were significant differences between crude protein (P<0.001) and Neutral Detergent Fibre (P<0.001) in sainfoin-grass mixtures. Crude protein and Neutral Detergent Fibre both interacted (P<0.001) with harvests (Table 4.6).

Table 4.6 Analysis of variance of CP and NDF concentration of sainfoin-grass mixtures in the 2nd year.

		CP	NDF
	d.f	M.S	
Mixtures (M)	6	15004***	31349***
Harvests (H)	2	1195***	99431***
M x H	12	2453***	5561***
Residual	21	9.1	8.2

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

Over the three harvests in the 2nd year, sainfoin and 2/3 sainfoin-1/3 perennial ryegrass had the highest crude protein, and meadow fescue and perennial ryegrass had the lowest crude protein (Figure 4.10). Crude protein varied between harvests in the 2nd year (Table 4.7). Crude protein concentration of sainfoin, meadow fescue and sainfoin-meadow fescue mixtures increased as the season progressed (Table 4.7). Perennial ryegrass monoculture declined in the 2nd harvest. For the sainfoin-perennial ryegrass mixtures, crude protein in the 3rd harvest was at the highest level, and the 1st harvest was lowest. Sainfoin and Mixtures achieved higher crude protein yields than grasses, but 1/3 sainfoin-2/3 meadow fescue yielded lower than other mixtures (Appendix 4-5 & Table 4.7)

Figure 4.10 CP of sainfoin, meadow fescue, perennial ryegrass and their mixtures over three harvests in the 2nd year. Bars followed by the same letter are not significantly different.

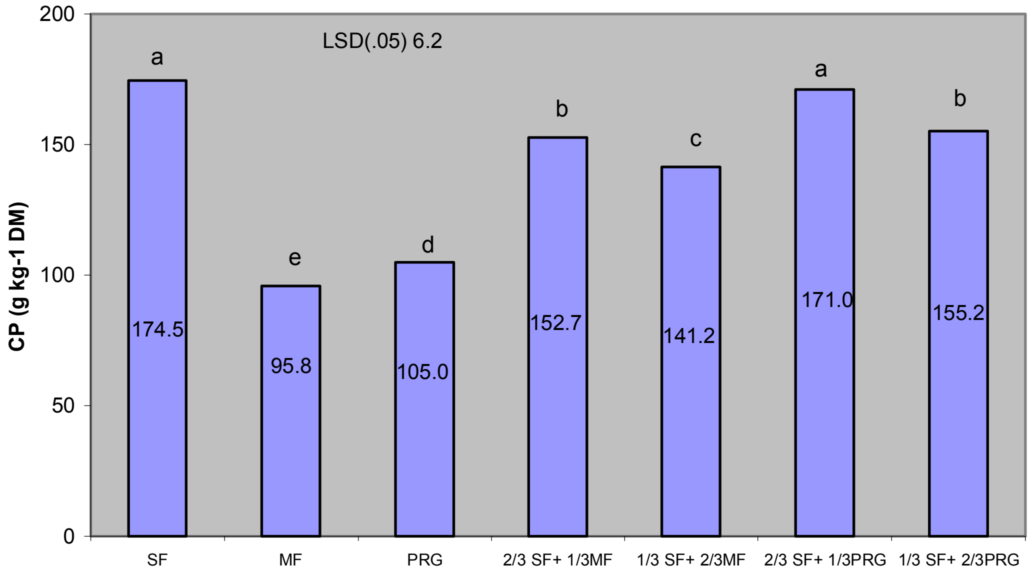


Table 4.7 CP content and yields of sainfoin, meadow fescue, perennial ryegrass and their mixtures in the 2nd year. Comparisons made between harvests.

	Harvest			CP yield
	1st	2nd	3rd	
	g kg^{-1} DM			t ha^{-1}
SF	158.9 c	196.4 b	207 a	2.13 a
MF	94.7 b	102.7 a	N.A	0.63 d
PRG	111.3 a	78.3 b	N.A	0.58 d
2/3 SF+ 1/3MF	148 c	166.8 a	155.4 b	2.23 a
1/3 SF+ 2/3MF	130 b	159 a	160.3 a	1.51b
2/3 SF+ 1/3PRG	171.9 b	155.4 c	181 a	2.17 a
1/3 SF+ 2/3PRG	152.4 b	143.2 c	172.8 a	1.94 a
LSD(.05)		6.3		0.37

Values followed by the same letter are not significantly different.

Meadow fescue was most fibrous, and sainfoin and perennial ryegrass were least fibrous (Figure 4.11). Neutral Detergent Fibre also varied between the three harvests in the 2nd year. For sainfoin and meadow fescue, forage in the 2nd harvest was least fibrous (Table 4.8). For perennial ryegrass, forage in the 2nd harvest was more fibrous than the 1st harvest. For the mixtures, forage in the 1st harvest was most fibrous and the 2nd harvest was least fibrous.

Figure 4.11 NDF of sainfoin, meadow fescue, perennial ryegrass and their mixtures over three harvests in the 2nd year. Bars followed by the same letter are not significantly different.

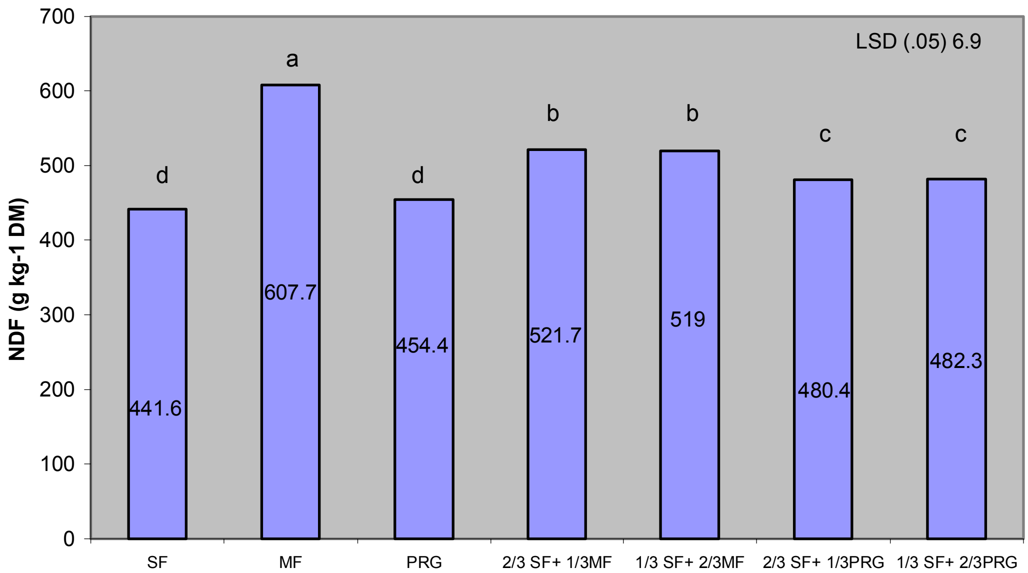


Table 4.8 NDF of sainfoin, meadow fescue, perennial ryegrass and their mixtures in the 2nd year. Comparisons made between harvests.

	Harvest 1st	Harvest 2nd	Harvest 3rd	LSD(.05)
	g kg^{-1} DM			
SF	450.1 a	399.3 b	455.7 a	
MF	622 a	512.7 b	N.A	
PRG	446.2 b	485.9 a	N.A	
2/3 SF+ 1/3MF	536.1 a	466.3 c	520.9 b	6.0
1/3 SF+ 2/3MF	540 a	469.3 c	494.3 b	
2/3 SF+ 1/3PRG	488.2 a	453.2 c	481.8 b	
1/3 SF+ 2/3PRG	491.4 a	459.7 c	475.8 b	

Values followed by the same letter are not significantly different.

There were no significant differences in average crude protein and Neutral Detergent Fibre between the direct sown and undersown crops in the 2nd year, but they both interacted (P<0.05) with mixtures (Table 4.10 & 4.11). Direct sown meadow fescue alone had higher crude protein and Neutral Detergent Fibre concentration than the undersown.

Table 4.9 Analysis of variance of average CP and NDF in the 2nd year.

	d.f	CP (M.S)	NDF (M.S)
Sowing treatment (ST)	1	24.1	4.24
Mixtures(M)	6	3794.1 ***	12414 ***
ST x M	6	54.3*	61.1*
Residual	13	16.5	20.7

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

Table 4.10 Comparison of average CP and NDF under direct sowing and undersowing in the 2nd year.

	CP Direct sowing		CP Undersowing		NDF Direct sowing		NDF Undersowing	
	g kg^{-1} DM							
SF	174.9	a	174.1	a	437.2	a	446	a
MF	100.3	a	91.2	b	615.2	a	600.3	b
PRG	97.8	a	112.3	a	452.5	a	456.3	a
2/3 SF+ 1/3MF	153.8	a	151.6	a	523.2	a	520.3	a
1/3 SF+ 2/3MF	140.6	a	141.8	a	520.3	a	517.8	a
2/3 SF+ 1/3PRG	169.5	a	172.6	a	482.1	a	478.7	a
1/3 SF+ 2/3PRG	152	a	158.5	a	479.5	a	485.2	a
LSD(.05)	8.7				9.8			

Values followed by the same letter are not significantly different.

4.4 Discussion

4.4.1 Direct Sowing vs. Undersowing

Undersowing in spring barley suppressed the growth of sainfoin, meadow fescue, perennial ryegrass and their mixtures in the establishment year and reduced the DM yield in the establishment year and 2nd year. Direct sown crops gave an average yield of 1.8 t DM ha^{-1} in the establishment year and 12.22 t DM ha^{-1} in the 2nd year, but undersown crops produced no yield except 4.8 t DM ha^{-1} grain yield of spring barley in the establishment year and 10.31 t DM ha^{-1} in the 2nd year. This is due to the effect of spring barley, which suppressed weed germination and growth, but also competed with sainfoin, meadow fescue, perennial rye grass and their mixtures for light, moisture and nutrients in the establishment phase, and thus negatively affected the growth and production in the establishment and 2nd years. However, undersowing had no effect in the 3rd year and yields were comparable with direct sown crops. Although there was no research information available comparing direct sowing approaches and the undersowing of sainfoin, there were many other studies such as with lucerne undersown in oats in northeast USA, and red clover with barley in Turkey. These agreed with the results in section 4.3 (Sheaffer, Barnes & Marten, 1988; Hall, Curran, Werner & Marshall, 1995).

4.4.2 Sainfoin, Meadow Fescue, Perennial Ryegrass and Their Mixtures

The 2/3 sainfoin-1/3 meadow fescue mixtures yielded highest with a three-year mean of 9.07 t DM ha^{-1}; other mixtures and sainfoin alone yielded 7.53-8.47 t DM ha^{-1} with no significant differences. Meadow fescue and perennial ryegrass yielded just 4.72 t DM ha^{-1} and 3.2 t DM ha^{-1}, much lower than sainfoin and mixtures. This agreed with many other studies, that legumes complement grasses by increasing total seasonal yield and protein concentration of the forage. These results are also in agreement with some previous results. Dubbs (1968) conducted sainfoin-grass studies over three years. Eski sainfoin grew with ten grass species, such as basin wildrye (*Elymus cinereus Scrib. And Merr.)* and green needlegrass(*Stipa viridula trin.*) etc, yielding 3.3 -4.5 t DM ha^{-1}. Sainfoin alone yielded similar to the mixtures in most cases. The results of Goplen (1991) showed that sainfoin alone yielded higher than when grown with Russian wildrye, crested wheatgrass and pubescent wheatgrass. The results of Cooper (1972) and Sengul (2003) did not show any yield differences between sainfoin and sainfoin-grass mixtures over four and three years' period. NIAB's (2000) results also showed that sainfoin (cvs. Emyr and Cotswold Common) yielded similar to mixtures with Timothy, cocksfoot and meadow fescue, but more than sainfoin with perennial ryegrass (cv. Elgon). Cooper's (1972) results gave yields of sainfoin and sainfoin-grass mixtures over four years, similar to results obtained in this study. However Sengul's (2003) results over three years only provided a yield of 4.5 t DM ha^{-1}.

4.4.3 Sainfoin Proportion in Sainfoin-Grass Mixtures

The average sainfoin content in direct sown sainfoin-meadow fescue mixtures declined significantly, from 61.6% in the establishment year to 32.2% in the 3rd year. The may probably be due to the early heading nature of cv. Lifara meadow fescue, which have been more competitive than expected in the spring. Over the same period the sainfoin content in sainfoin-perennial ryegrass mixtures increased from 44.7% in the establishment year to 66.5% in the 3rd year. The reason for the increase of sainfoin content in sainfoin–perennial ryegrass is not clear. It may perhaps be due to:

a) The low precipitation in August and September in 2003, which was 10.2 mm and 12.5 mm, compared to 63 mm and 70.5 mm in previous ten year mean, which limited perennial ryegrass growth, and the dry period in June and July in 2004 (26.2mm and 33.7 mm lower) compared to 2003,

b) The fast growth of sainfoin in the spring, which competed with cv. Condesa perennial ryegrass for light, and
c) the lower tillering, compared to diploid perennial ryegrass, and late heading of Condesa tetraploid perennial ryegrass (NIAB, 2004).

4.4.4 Forage Quality

Annual crude protein concentration of sainfoin was 174.5 g kg^{-1} DM, and it increased as the season progressed. This may be due to the leaf percentage increase (Fagan & Rees, 1930). Meadow fescue showed the same trend as sainfoin, an increase in crude protein concentration as the season progressed. This may be due to two reasons. Firstly when cv. Lifara meadow fescue was cut at the 1st harvest, it was already quite mature and in the heading stage. Secondly, according to Minson, Harris, Raymond, and Milford (1964) the leaf blades of meadow fescue declines from about 58% on May 6 to 15% on May 27, and then increases to about 23% on June 9 and 26% on June 16. Annual crude protein concentration of perennial ryegrass was 105 g kg^{-1} DM, and this declined during the season. This is also probably because of the leaf blades percentage decline. Terry and Tillery (1964) found that leaf blade of perennial ryegrass was constantly declining from 70% on April 27 to 18% on May 29, and then to 11% on June 11. Crude protein concentration of the 2/3 sainfoin-1/3 meadow fescue and 1/3 sainfoin-2/3 meadow fescue were lower than that of the 2/3 sainfoin-1/3 perennial ryegrass and 1/3 sainfoin-2/3 perennial ryegrass. This may be due, in part, to the sainfoin percentage of sainfoin-meadow fescue being lower than that of sainfoin-perennial ryegrass at the 1st harvest in the 2nd year.

Meadow fescue was most fibrous, and sainfoin and perennial ryegrass were the least fibrous. Sainfoin-meadow fescue mixtures were more fibrous than the sainfoin-perennial ryegrass.

There were no significant differences in crude protein or Neutral Detergent Fibre between the direct sown and undersown crops, apart from meadow fescue alone. Direct sown meadow fescue had a higher concentration of crude protein and Neutral Detergent Fibre than that which was undersown.

4.5 Conclusions

- Undersowing negatively affected the production of sainfoin, meadow fescue, perennial ryegrass and their mixtures in the 1st and 2nd years compared to direct sowing, but this effect did not extend to the 3rd year.
- 2/3 sainfoin-1/3 meadow fescue was the most productive mixture, producing an average of 9.07 t DM ha^{-1} over the three year study period. However, meadow fescue cv Lifara was mature and at the heading stage at the 1st harvest, and this decreased crude protein concentration and increased Neutral Detergent Fibre.
- Sainfoin complements the production of meadow fescue and tetraploid perennial ryegrass in mixtures. Sainfoin monoculture and mixtures all had similar production levels, apart from the high yielding 2/3 sainfoin-1/3 meadow fescue mixture.
- Yields of both sainfoin and sainfoin-grass mixtures declined from the 2nd to the 3rd growing season.
- Sainfoin monoculture contained more crude protein than most sainfoin–grass mixtures.
- Sainfoin-perennial ryegrass mixtures contained more crude protein than the sainfoin-meadow fescue.
- Tetraploid perennial ryegrass cv. Condesa seemed more compatible to sainfoin than meadow fescue cv. Lifara over the three years period of this study.

Chapter Five

Variety Comparison

Abstract

Eight cultivars were tested in 2004-2005. An average yield of 11.9 t DM ha^{-1} was achieved, with no yield differences between cultivars in the 1st full harvest year. Cvs. Makedonka and Gan appeared to Giant type. Cv. Sombourne showed a quicker regrowth than other cultivars after harvest in the 2nd year.

5.1 Introduction

Well-known cultivars in the UK are Cotswold Common, Hampshire Common, Sombourne, Hampshire Giant and English Giant (Sheldrick *et al.*, 1995; Frame *et al.*, 1998). Cotswold Common has been reported to yield 11-15 t DM ha^{-1} (Spedding & Diekmahns, 1972; Sheehy *et al.*, 1984; Koivisto & Lane, 2001). Sombourne is a hybrid of Hampshire Common and Hampshire Giant. There are other bred cultivars in the USA and Canada. The cvs. Eski, Remont and Renumex, developed in the USA were originally from Turkey and Iran, and the cvs. Melrose and Nova were bred in Canada from Russian introductions (Cash, 1993). There are also cultivars in other countries, such as cvs. Zeus and Vala in Italy (Frame *et al.*, 1998), cv. Makedonka in Yugoslavia (Cupina, 1999), cv. Perly in Switzerland, cv. Fakir in France and cv. Emyr in Hungary (Koivisto & Lane, 2001). There are also two bred cultivars in P.R.China, cvs. Great wall 1 and Mengnong (Wu & Wang, 1990; Chen, 1992). Recently a new variety 'Shoshone[2]' was released in the USA with better forage production than Remont under both dryland and irrigated sites in Wyoming and Montana. A variety trial conducted in Montana, USA during 1975-1983, showed that cvs. Eski, Melrose and Remont yielded 11, 10.4 and 8.3 t DM ha^{-1} in irrigated areas, and 4.2, 3.8 and 3.2 t DM ha^{-1} on dryland (Cash, 1993). However Goplen *et al*. (1991) found that Nova yielded 7% more DM than Melrose and 15% more than Eski and Remont. A trial at Colesborne in 1943 showed Cotswold Common to be more uniform and persistent

[2] Shoshone is a new variety released by the College of Agriculture, Agricultural Experiment Stations at the University of Wyoming and Montana State University, and by the United States Department of Agriculture, Natural Resources Conservation Service. It was developed by Dr. Fred A. Gray, Professor of Plant Pathology-Nematology, Department of Plant Sciences, College of Agriculture, University of Wyoming, Laramie, fagray@uwyo.edu. More information at www.wyseedcert.com/Shoshonereleasewpic.rtf (Accessed at November 3 2005).

than other cultivars, and a later trial at the Grassland Research Institute, also showed Cotswold Common to be more persistent than cultivars from former East Germany, France, Russia and Turkey (Bland, 1971). A comparison of six cultivars, G35, Remont, Cotswold Common, Melrose, Eski and Pola, conducted in a greenhouse at Massey University, New Zealand, showed that Cotswold Common yielded less than the others (Dehabadi, 1993). A trial on cvs. Emyr, Cotswold Common, Sombourne and Nova carried out at the Royal Agricultural College, Cirencester in 1998 showed that cvs. Emyr, Cotswold Common and Sombourne all had greater annual DM yields than Nova (Koivisto & Lane, 1998).

Overall only a few bred sainfoin cultivars have ever been developed. Research effort on sainfoin breeding is still very small, compared to other legumes, such as lucerne, white clover etc. Selecting sainfoin cultivars with improved yield potential and persistence is important. The objective of this study was to assess the yield potential of sainfoin cultivars available. The only criterion for selection for this study was the availability of seed.

5.2 Materials and Methods

5.2.1 Experiment Site

The experimental site was described as in 3.2.1.

5.2.2 Experiment Design

This was a randomised block design with three replications. There were eight sainfoin cultivars tested in the trial, cvs. Cotswold Common, Sombourne, Perly, 'Commercial', WY-942-94, Makedonka, Ning, and Gan. The plot size was 1m by 2m.

5.2.3 Field Preparation

Field preparation was described as in 3.2.3.

5.2.4 Establishment

This trial was established on May 12, 2004, a previous trial established on another site in 2003 having failed. The seed rates of cultivars were calculated to achieve a target of 150 plant m^{-2}. According to the germination rate and 1000 seed weight, appropriate seed rates were calculated as follows: Cotswold Common 90 kg ha^{-1}, Sombourne 97 kg ha^{-1}, Perly 88 kg ha^{-1}, 'Commercial' 123 kg ha^{-1}, WY-942-94 166 kg ha^{-1}, Makedonka 145 kg ha^{-1}, Ning 103 kg ha^{-1}, and Gan 98 kg ha^{-1}. Seeds were broadcast by hand and then raked into soil to about 1.0 cm deep and rolled.

5.2.5 Management

MCPA + MCPB were used to control broadleaved weeds. Carbetamex was used for grass weeds and chickweed. MCPA + MCPB was applied after the 1st trifoliate leaf stage (Stage 0, Appendix 1). Two applications of MCPA + MCPB were applied to try to kill a serious infestation with black nightshade (*Solanum nigrum*). This was not entirely successful. Carbetamex was applied in December 2004 to kill grass weeds and chickweed.

Phosphorous and potassium were applied after harvesting by hand broadcast (Table 5.1) in accordance with MAFF (2002).

Table 5.1 Fertilizer application chart on sowing date trial.

	Establishment year	2nd year		
		1st harvest	2nd harvest	3rd harvest
	kg ha^{-1}			
P_2O_5	30	20	-	-
K_2O	-	30	40	20

5.2.6 Population Measurement

A 25 x 25 cm quadrat was used to facilitate plant population density assessment, and two determinations were made in each subplot. In the establishment year a determination was made in July. In the 2nd year the determinations were made after the 1st harvests.

5.2.7 Sampling and Harvest

Two quadrats (0.5×0.5m) samples were taken from each subplot to measure yield just before harvesting. Harvesting was carried out immediately after sampling. Cutting was performed by BCS 610 Motormower. Plants were cut at about 5 cm high. There was relatively little production in the establishment year and plants were trimmed off on November 1, 2004. Three harvests were taken in the following season on May 29, July 13, and September 12, 2005.

5.2.8 Statistical Analysis

As described in 2.2.

5.3 Results and Discussion

5.3.1 Yield

Yields were relatively poor in the establishment year because of broadleaved weeds, especially black nightshade. A drought in May 2004 resulted in delayed sainfoin emergence, which led to the postponement of the spray of MCPA+MCPB, and broadleaved weeds established quickly and suppressed the sainfoin seedlings. The trial was given a trim in November, and yielded an average of 0.43 t DM ha^{-1} (Table 5.3). All cultivars showed a quick growth in the 2nd year and yielded between 10.67-12.54 t DM ha^{-1}(Table 5.3). There were no significant yield differences between cultivars in the 2nd year (Table 5.2). However, there were yield differences at the individual harvests.

The yield of Cotswold Common in the trial was similar to the yield in the experiment of sowing date and autumn management (see Chapter 3). It was also in agreement with the results from Grassland Research Institute (Spedding & Diekmahns, 1972) and from a more recent trial in Cirencester (Koivisto & Lane, 2001). Sombourne also yielded similar to the trial of Koivisto and Lane (2001). Makedonka and Gan showed characteristics of the Giant type, and may perhaps be a Giant type. Makedonka and Gan flowered and were tall and vigorous in the establishment year. They formed stems and flowers after the 1st harvest in the 2nd year. Sombourne also formed a few stems and flowers after the 1st harvest in the 2nd year, and also showed a quicker regrowth than other cultivars after harvest.

Table 5.2 Analysis of variance of population and DM yields in the establishment and the 2nd years.

		Population		DM yield				
		Establish-ment year	2nd year	Establishm-ent year	2nd year harvests 1st	2nd	3rd	Total
	d.f	M.S						
Cultivar	7	811	118.8	0.08	1.21	0.45	0.59	1.1
Residual	14	1486	208.3	0.05	1.27	0.33	0.11	0.8

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively.

Table 5.3 Comparison of DM yields of different cultivars in the establishment and 2nd years.

	Establishment year	2nd year 1st harvest	2nd harvest	3rd harvest	Total
	t DM ha^{-1}				
Cotswold Common	0.12 b	8.11 a	2.67 ab	1.6 b	12.54 a
Sombourne	0.4 ab	6.07 b	3.43 a	2.77 a	12.3 a
Perly	0.55 a	7.07 ab	2.77 ab	2.2 a	12 a
'Commercial'	0.57 a	7.23 ab	2.2 b	2.27 a	11.67 a
WY-942-94	0.35 ab	6.77 ab	2.27 b	1.6 b	10.67 a
Makedonka	0.66 a	6.87 ab	2.8 ab	2.7 a	12.37 a
Ning	0.37 ab	7.33 ab	2.47 ab	2.3 a	12.07 a
Gan	0.44 ab	6.3 ab	2.83 ab	2.47 a	11.63 a
LSD(.05)	0.4	1.99	1.0	0.58	2.43

Values followed by the same letter are not significantly different.

5.3.2 Plant Populations

There were no significant differences in plant population for both growing seasons (Table 5.2). Cultivars achieved an average of 107 plant m^{-2} in the establishment year, lower than the targeted 150 plant m^{-2} mainly because of weed problems (Table 5.4). The average population declined to 62 plant m^{-2} in the 2^{nd} year. Only 57.9% of plants survived, but this was in accordance with the suggested population (Hill, 1997; Sheldrick & Thomson, 1982).There were no linear relationship between yield and plant population in the establishment year (P=0.36, r^2=0.32) or in the 2^{nd} year (P=0.35, r^2=0.03).

Table 5.4 Plant Population in the establishment and the 2^{nd} years.

	Cotswold Common	Sombourne	Perly	Commercial	WY-942-94	Makedonka	Ning	Gan	LSD (.05)
					plant m^{-2}				
Establishment year	101.3	114.7	125.3	125.3	77.3	114.7	98.7	96	67.5
2^{nd} year	64	61.3	50.7	56	62.8	72	64	64	25.7

5.4 Conclusions

This two year study was not enough fully to assess the yield potential and persistence of the tested cultivars. A minimum of four years would probably be needed, longer if the persistence of the sainfoin population with time were to be more fully assessed, in addition to early yield performance. However, from two year's results it is still possible to conclude as follows:

- The cultivars yielded an average yield of 11.9 t DM ha^{-1} with no yield differences in the 2^{nd} year.
- Cv. Makedonka and Gan exhibited characteristics of Giant type.
- Cv. Sombourne also showed some characteristics of Giant type, and a quicker regrowth than other cultivars after harvest in the 2^{nd} year.

Chapter Six

Competition between Sainfoin, Perennial Ryegrass and Meadow Fescue

Abstract

Competition between sainfoin and meadow fescue or perennial ryegrass was conducted in a plastic container over two years. Root and shoot systems of sainfoin and grasses was separated to investigate the root, shoot and full competition between sainfoin and grass. Root competition had more effect than shoot competition. Full competition increased grass yields in mixtures and decreased sainfoin yield. Intraspecific competition of perennial ryegrass was greater than interspecific competition, but intraspecific competition of sainfoin and meadow fescue was less than the interspecific competition. Sainfoin grown with meadow fescue at a 1:2 ratio was more competitive than with perennial ryegrass.

6.1 Introduction

This chapter explores the nature of competition between sainfoin and two grass species, meadow fescue and perennial ryegrass, in terms of the yield of species in mixtures and in pure stands and plant survival.

6.1.1 Competition

Competition between species, including grassland species interactions and intercropping systems, is one of the most important subjects in ecology (Hamilton, 1996). Legume-grass mixtures play a very important role in British grassland, especially important in the success of white clover-ryegrass swards, which is largely attributed to their morphology and clover nitrogen fixation. However, the success of legume-grass swards is not easy to fully understand, since keeping the balance between the two species is difficult in agricultural practice. Studying the basis of competition between legumes and grasses is very important for legume-grass based grassland systems. The competition between white clover and ryegrass and between lucerne and grass has been already studied in some experiments (Grieshaber-Otto, 1984; Baines, 1988; Faurie, Soussana & Sinoquet, 1996). A few studies have explored sainfoin and common grass species (Dubbs, 1968), but none have attempted to separate above- and below-ground competition.

Interactions between species exist between shoots and roots. Above- and below-ground competition has been frequently compared (e.g. Donald, 1958; Snaydon, 1971). Below ground competition was reported to have more severe competition than above ground (Donald, 1958; Wilson, 1988). This is generally agreed to be due to root competition starting earlier than shoot competition, and before the shoots are well developed to cause mutual competition (Milthorpe, 1961). Shoots only compete for one resource, light, while roots compete for a wide range of soil resources, water and many mineral nutrients (Casper & Jackson, 1997). Wilson (1988) analysed 23 previous studies on shoot and root competition, and in 68% of cases the root competition had a greater impact on growth than shoot competition. Wilson and Tilman (1991) also found in an experiment that root competition was more important than shoot competition, particularly at lower nitrogen levels.

In most of the early competition studies, species were grown in monocultures and mixtures (e.g. De Wit, 1960), i.e. with no competition and with full competition. In this study, the method of competition study applied was developed by Snaydon (1979), based on the methods developed and modified by Donald (1958), De Wit (1960) and Schreiber *et al.* (1967). Two species are normally grown in monocultures and mixtures at a single overall density, whereas the proportions of species in mixtures can be varied, and shoots and roots of the two species are separated respectively (Figure 6.4) to give full, root, shoot and no competition.

6.1.2 Theoretical Concepts

The data from competition studies is often complicated to present and interpret. To quantify competition and facilitate data presentation, competition indices and graphic presentations were proposed by many researchers (e.g. De Wit, 1960; De Wit & Van Den Bergh, 1965; Snaydon & Satorre, 1979; 1989). Competition indices and graphic presentations (e.g. the replacement diagram proposed by De Wit 1960) can help to interpret such data and facilitate presentation and interpretation of results. Many competition indices have been proposed in the literature. Snaydon and Satorre (1979) have summarized them into three categories namely ‘competitive ability’, ‘resource complementarity’ and ‘competitive severity’.

In the following sections, W_{ii} stands for the weight of species i in pure stands, and W_{jj} for the weight of species j in pure stands. W_{ij} stands for the weight of species i when grown with species j, and W_{ji} for the weight of species j when grown with species i.

6.1.2.1 Competitive Ability

Competitive ability can be considered as the ability of species to gain the advantage over one another for common resources (Snaydon & Satorre, 1979). The measures used are Relative Crowding Coefficient (RCC), Relative Replacement Rate (RRR) and Aggressivity (De Wit & Gourdriaan, 1974; De Wit & Van den Bergh 1965; McGilchrist & Trenbath 1971).

Relative Crowding Coefficient RCC (De Wit & Gourdriaan, 1974) is calculated from the performance of each species grown in mixtures and separately. The greater RCC value the greater competitive ability of the species in question (Harper, 1977; Williams & McCarthy, 2001).

$$\text{RCC} = \left(\frac{W_{ij}}{W_{ji}} \bigg/ \frac{W_{ii}}{W_{jj}} \right)$$

Relative Replacement Rate (De Wit & Van den Bergh 1965). Relative Yield (RY, see next section) of species *i* at harvest 1 divided by the RY of species *j* at harvest 1 divided by harvest 2. If the RRR > 1, species i is more competitive than species j; if the RRR < 1, species i is less competitive than species j (De Wit & Van Den Bergh, 1965; Tow & Lazenby, 2001).

$$\text{RRR} = \left(\frac{W_{ij}}{W_{ji}} \bigg/ \frac{W_{ii}}{W_{jj}} \right)^1 \Bigg/ \left(\frac{W_{ij}}{W_{ji}} \bigg/ \frac{W_{ii}}{W_{jj}} \right)^2$$

Note that 1 and 2 stand for successive harvest periods.

Aggressivity (McGilchrist & Trenbath, 1971). The values for Aggressivity are between +1 and -1. An aggressivity value of 0.2 for example means that one species has increased by 10% and the other in mixture reduced by 10%, compared to pure stands.

$$\text{Aggressivity} = \frac{1}{2}\left(\frac{W_{ij}}{W_{ii}} - \frac{W_{ji}}{W_{jj}} \right)$$

6.1.2.2 Resource Complementarity

Resource Complementarity considers the performance of mixtures compared to the component species grown in pure stands.

Relative Yield (RY) as defined by De Wit (1960) measures yield in mixtures divided by the yield in monoculture. If the RY = 1, it means that one species does the same in mixture against the other as in pure stand against its own species; if the RY > 1, it means that one species does better in mixtures against another than in pure stand and intraspecific competition is greater than interspecific competition. if the RY < 1, it means that one species does better in a pure stand than in mixtures, and intraspecific competition is less than interspecific competition.

RY = $W_{ij} \big/ W_{ii}$

Relative Yield Total (RYT) and Land Equivalent Ratio (LER) The sum of the RY values of two species in a mixture gives Relative Yield Total (RYT). If RYT = 1, the two species compete for the same resources; if RYT < 1, the two species are mutually antagonistic; if RYT > 1, there is avoidance of competition to some extent and there is a yield advantage in the mixture (Snaydon & Satorre, 1979; Williams & McCarthy, 2001; Silvertown & Charlesworth, 2001). However Williams & McCarthy (2001) argued that RYT had its limitation, and that RY should be taken into account when looking at RYT. Land Equivalent Ratio (LER) was proposed by Willey and Osiru (1972) and is identical to RYT.

RYT = $\dfrac{W_{ij}}{W_{ii}} + \dfrac{W_{ji}}{W_{jj}}$

LER = $\dfrac{W_{ij}}{W_{ii}} + \dfrac{W_{ji}}{W_{jj}}$

6.1.2.3 Competition Severity

Snaydon and Satorre (1979) proposed Competitive Severity (CS), which is defined as the reduction in competition for limiting resources by individual plants, which is caused by sharing resources with neighbouring individuals. A CS value of 0 means no competition between species. A CS value of 1 means a tenfold decrease in plant size for the species.

CS = $\log_{10}\left(\dfrac{W_i}{W_{ii}}\right)$ (for pure stand)

CS = $\log_{10}\left(\dfrac{W_i}{W_{ij}}\right)$ (for mixtures)

W_i stands for the weight per plant of species i as spaced plant, and W_j stands for the weight per plant of species j as spaced plant.

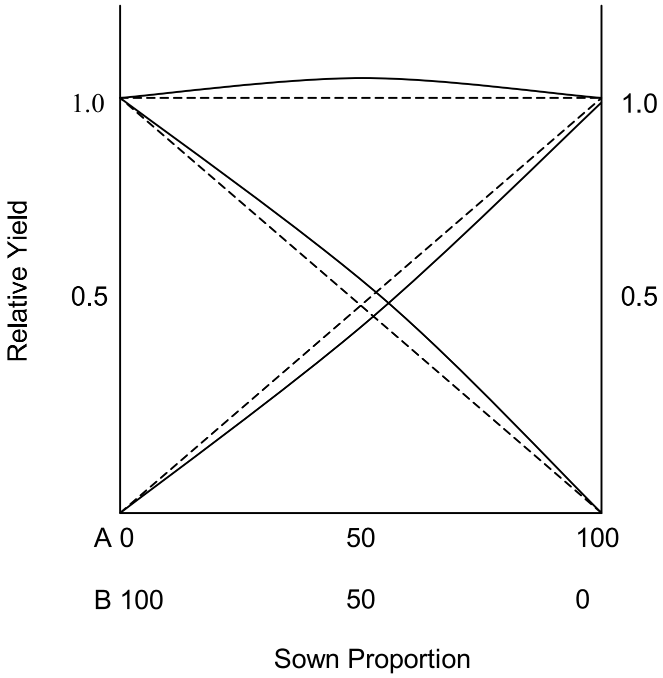
6.1.2.4 Plant Survival

Competition between species has effects not only on the performance of the species such as growth and yield but, as a consequence, plant survival may also be affected (Silvertown & Charlesworth 2001). So survival rate can be also used to measure competition. Survival rate as a competition index has also been used in previous studies (e.g. Grieshaber-Otto, 1984; Baines, 1988).

6.1.2.5 Graphic Presentations

Competition data can be presented graphically through the use of replacement diagrams (De Wit 1960) (Figure 6.1) and bivariate diagrams (Figure 6.2). In the replacement diagram, RY indices can be plotted against the original proportions. In the bivariate diagram, the yield of one species can be plotted against that of the other using log scale axes. Replacement and bivariate diagrams have been used in many studies and their use is explained as follows (De Wit & Van Den Bergh, 1965; Pearce & Gilliver, 1979; Grieshaber-Otto, 1984; Baines, 1988).

Figure 6.1 Replacement Diagram (De Wit, 1960). RY of two components are plotted against the proportions of the components in the mixture.

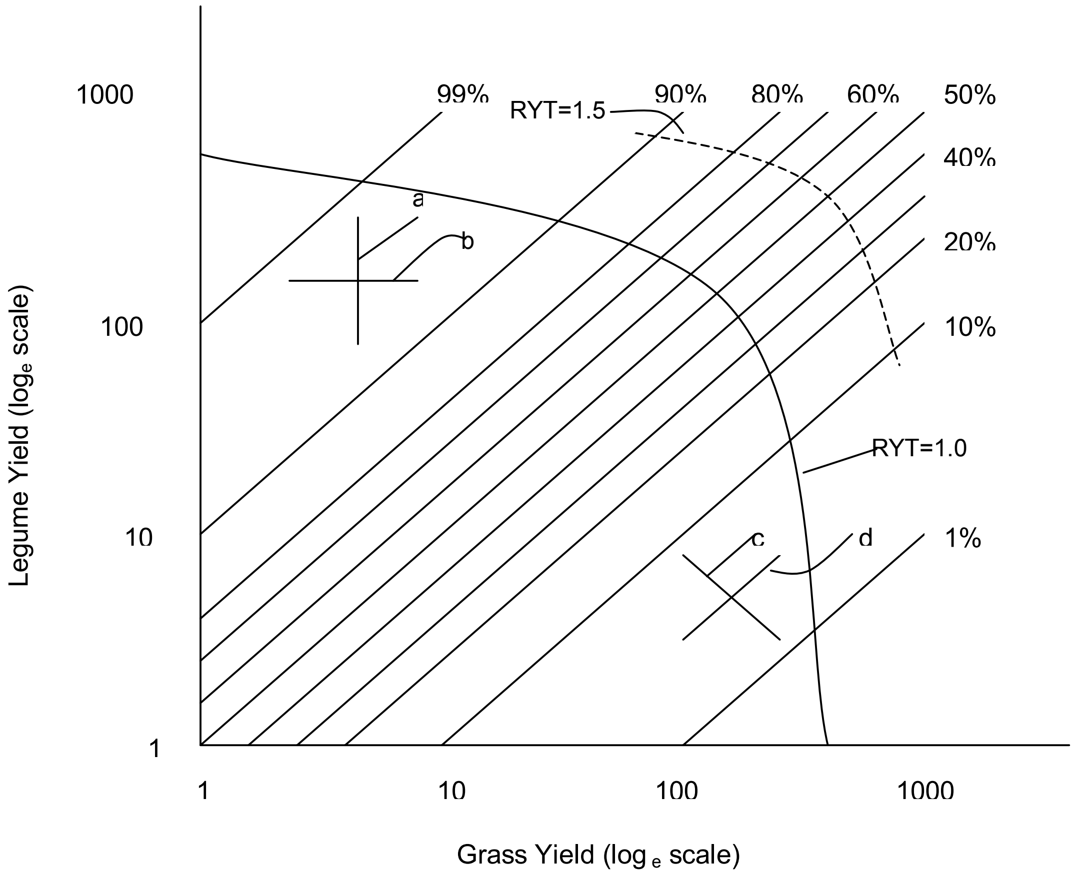


In Figure 6.1, two components in a mixture are plotted against their original proportion. When RY value is above the horizontal line that joins the two RY values of 1, it indicates greater interspecific competitive ability. When the RY value is below the line, it indicates less interspecific competitive ability (Snaydon & Satorre, 1979; Williams & McCarthy, 2001). When individual component yields are above a line 0% and 100% for each species it indicates that the species in question is exploring more resource compared to pure stands while a yield below the line indicates that less resource is used compared to pure stands.

The bivariate diagram (Figure 6.2) was further developed by Snaydon and Satorre (1989). The two axes were transformed into $\log_e$. Transforming into a $\log_e$-$\log_e$ diagram has several advantages for presenting raw data, and shows the indices of competition such as aggressivity, RYT and competitive severity.

Firstly, it displays the yield of both components, indicating the RYT of the mixture and showing the resource complementarity, e.g. the distance from the curvilinear RYT =1.0. Secondly, the distance of actual value from the expected value (diagonal line – the proportion of component) measures the competitive ability of the species.

Figure 6.2 A bivariate diagram of legume yield against grass, using $\log_e$ transformation. The percentage of legume forms a logit scale. The bars indicate the LSDs for (a) legume yield, (b) grass yield, (c) legume proportion and (d) RYT.



6.1.3 Study Aim

The aim of the present study is to explore the interaction between sainfoin and meadow fescue, and between sainfoin and perennial ryegrass, by partitioning the root and shoot systems (effectively to group shoot, root and full competition) and by varying the ratio of grass to sainfoin.

6.2 Materials and Methods

6.2.1 Experiment Design

This was a replacement experiment of 2^4 factorial with two replicates. The four factors were root competition, shoot competition, grass species and sainfoin-grass ratio. The treatments were as follows.

- ± Root: roots of sainfoin and grass were associated and not associated
- ± Shoot: shoots of sainfoin and grass were associated and not associated
- Grass species: meadow fescue or perennial ryegrass
- Grass ratios: sainfoin established at a 2:1 or a 1:2 ratio with grass[3]

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Figure 6.3 The separation of root and shoot to group root, shoot, full and nil competition.

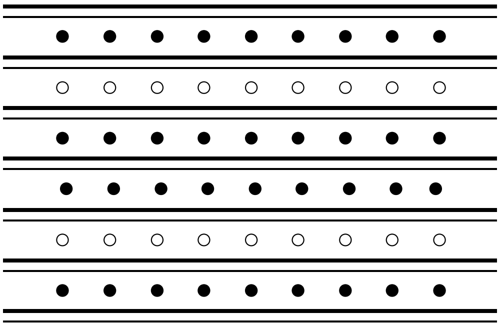
	+ Root	- Root
+ Shoot	Full competition	+ Shoot competition
- Shoot	Root competition	Nil competition

The combinations of root and shoot partition are shown in Figure 6.3. The separation of roots and shoots in the containers created root, shoot, full and nil competition.

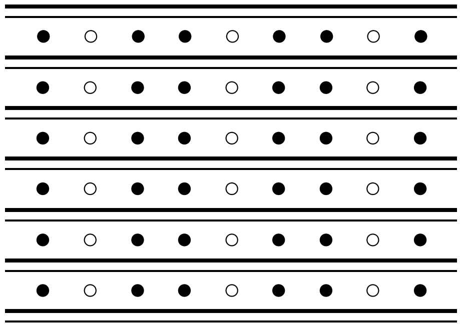
The sainfoin variety was cv. Cotswold Common. The meadow fescue variety was cv. Rossa, and the tetraploid perennial ryegrass variety was cv. Condesa. Below ground was partitioned by MDF (medium density fibreboard) into equal rows, to give equal environment and avoid the movement of water and soil nutrients between rows. Above ground was equally separated by white plastic film, which was reflective and non-transparent, and this reduced any light loss caused by the film separation. Above ground partitions were placed in a north-south direction to minimize shading, while below ground was either at right angles or to parallel to above ground (Figure 6.4).

[3] All the ratios of 2:1 or 1: 2 in refer to sainfoin **:** grass (either meadow fescue or perennial ryegrass).

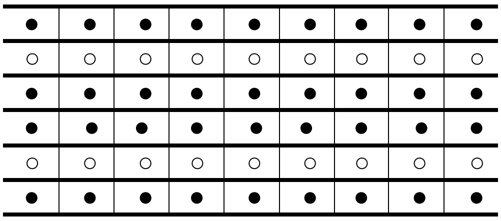
Figure 6.4 The arrangement of partitions of below-ground and above-ground, ● stands for sainfoin, ○ stands for grass, ——— above ground separation, ▬▬▬ below ground separation.



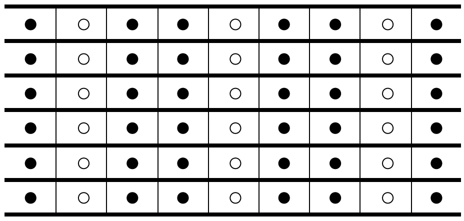
(a) Nil competition, plants separated from below-ground to above-ground.



(b) Full competition, plants separated from below-ground to above-ground.



(c) Shoot competition, below-ground separated by row and above-ground by column.



(d) Root competition, below-ground separated by row and above-ground by column.

6.2.2 Experimental Management

6.2.2.1 Container Preparation

Plastic containers of 0.55 m × 0.36 m × 0.30 m were sited in the Piggery Field, Coates Manor Farm without shading. Containers were partitioned by MDF and then filled with soil from Piggery Field (Sherborne series Cotswold Brash, soil P and K indices 3). Soil was well mixed before filling into containers to limit variation between containers (and treatments). Meteorological data in 2004- 2005 was described as in 3.2.1 and data in 2006 was in Appendix 6-5.

6.2.2.2 Establishment

Both sainfoin and grasses were seeded in plastic cells on May 20 2004 under greenhouse conditions and then seedlings were transplanted to plastic containers in the field on July 1 2004. All treatments were maintained at the same density of 273 plants m^{-2} (54 plants per container in six rows and nine plants per row). After transplanting, the seedlings were watered and left to establish. After one week, seedlings were clipped to about 5 cm high, and then separated above ground by plastic film 0.40 m high as indicated in Figure 6.4.

6.2.2.3 Management

Containers were regularly watered to avoid drying out by estimating the soil moisture deficit, and by observing surface drying. One harvest was taken in the establishment year on November 1 2004, and then containers were left in the field over the winter without the aboveground film partition (giving no shoot separation in the winter period). Dead plants were counted and replaced the following spring on April 10 2005, and the film was replaced to partition shoots again. Four harvests were taken in the second year, on May 28, July 13, August 28, and November 1 2005 respectively. The interval between the first three harvests was about 7 weeks. The 4th harvest was taken in November at the end of the season, when there was not much further growth, and then the containers were left in the field over winter without film partition. Plant survival and loss was calculated again in the spring of 2006.

30 kg ha $^{-1}$ P_2O_5 was applied in 2004 after harvest; 50 kg ha $^{-1}$ was applied in April 2004; 20 kg ha $^{-1}$ P_2O_5 and 30 K_2O kg ha $^{-1}$ was applied in June after the 1st harvest; 30 K_2O kg ha^{-1} after the 2nd harvest. At each harvest, sainfoin and grass components were separated and then dried in the oven at 100±2°C over 24 hours, and the dry matter determined.

6.2.3 Data Analysis and Presentation

Dry matter yield, plant survival and sainfoin/grass proportion in mixture was analysed. Relative Yield and Relative Crowding Coefficients were used to measure the competitive ability of sainfoin and grasses. Relative Yield Total was used to look at Resource Complementarity. Bar charts in $\log_e$ scale and bivariate diagrams (Figure 1 & 2) are used to present data.

The precondition of an analysis of variance is that the data meets the assumptions of independence of errors, normality, homogeneity of variance and additivity (Sokal & Rohlf, 1995; Clewer & Scarisbrick, 2001). Otherwise, the data needs to be transformed to other scales to conform to these assumptions. Biological experimental data sometimes does not follow a normal distribution and often need to be transformed (Bland & Altman, 1996). In this study several transformations were applied to yield, plant survival and sainfoin proportion before analysis of variance was carried out. Logarithm transformation was applied to sainfoin and grass yields; square root of arcsine transformation was applied to plant survival; and a logit transformation was applied to sainfoin proportions (Snedecor & Cochran, 1980).

Analysis of variance was carried out with Genstat 7 (Payne *et al.,* 2003). Multiple comparisons of treatment means was performed by applying the Least Significant Difference test. The transformed data was presented in a back-transformed format (Sokal & Rohlf, 1995). For the logarithm transformed data, the log-transformed means were back-transformed (anti-logged) to geometric means, and then multiple comparisons were performed by comparing the ratio of two means to the back-transformed LSD value, which was called the Least Significant Ratio (LSRs) in this case. An anti-log of the difference of two means was undertaken, the resulting difference becoming a ratio (e.g. $\log_e A - \log_e B = \log_e (A/B)$).

Only main effects and first order of interaction are considered in this study. When main effects were significant and also involved interactions, the main effects are discussed in association with the interactions as considering main effects alone could perhaps lead to misinterpretation (Clewer & Scarisbrick, 2001).

Survival data was assessed throughout the study, but only yield data for the 2nd year is presented as the 1st year was considered as an establishment year.

6.3 Results

6.3.1 The Survival of Sainfoin and Grasses

6.3.1.1 Sainfoin

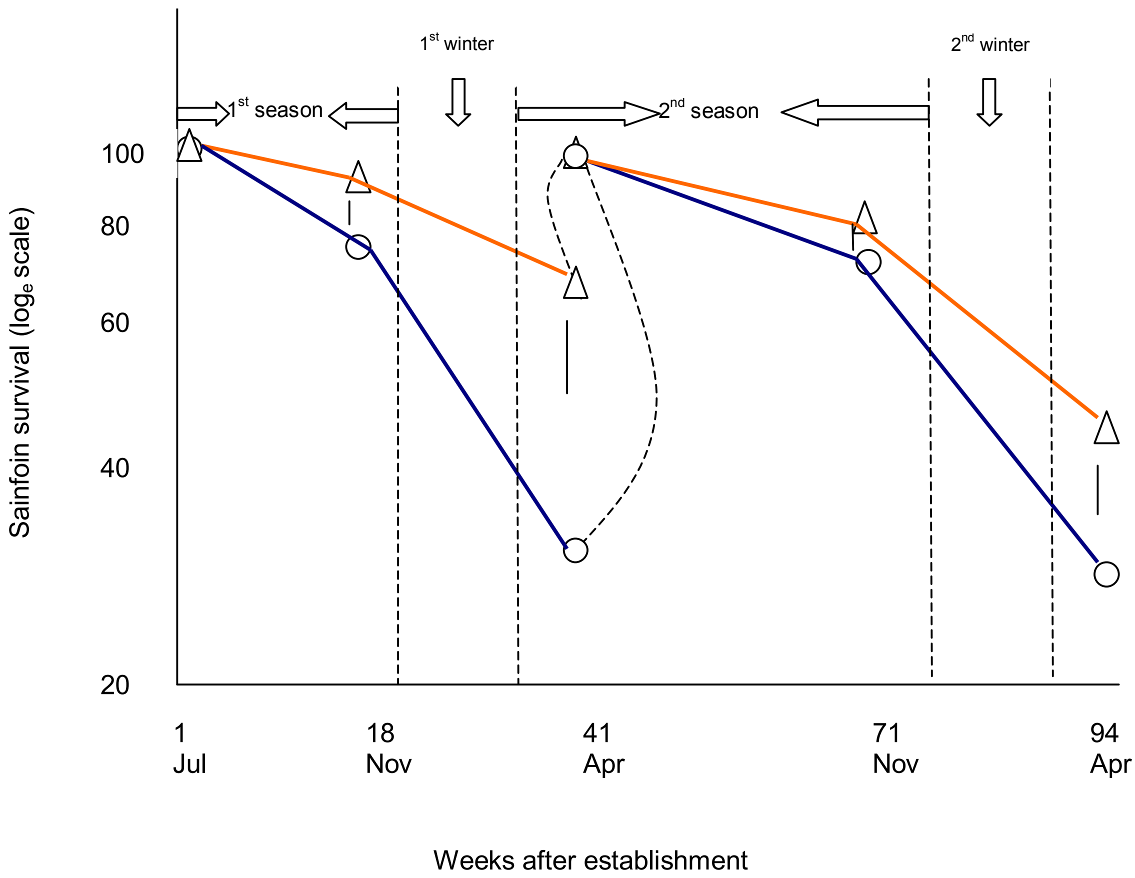
Sainfoin plant survival decreased in both growing seasons and over both winters (Figure 6.5). Increasing the ratio of grasses significantly reduced sainfoin survival during both growing seasons ($P < 0.001$ and $P < 0.01$) (Appendix 6-1). The species of grass, root and shoot competition generally did not have an effect on sainfoin survival except for shoot competition ($P<0.01$) affecting sainfoin survival in the 2nd growing season, and grass species ($P<0.05$) affecting survival over the 1st winter (Appendix 6-1) (Table 6.1).

Sainfoin established at a 1:2 ratio with grass had a lower survival rate than when at a 2:1 ratio with grass at the end of both growing seasons (Figure 6.5). By the end of 1st growing season, 93.6% sainfoin plant survived when established at a 2:1 ratio with grass but only 75% survived when established at a 1:2 (Figure 6.5). The same trend was observed at the end of 2nd year, 85.4% of sainfoin plants survived when in a 2:1 ratio but only 70.7% survived when grown at a 1:2 ratio.

Over the winters of 2004 and 2005 sainfoin survival was reduced compared to the end of the previous season. After the 1st winter, 66.2% sainfoin plants survived when grown at a 2:1 ratio with grass, but only 26.3% survived when grown at a 1:2 ratio.

After the 2nd winter, 48.8% sainfoin plants survived when grown at a 2:1 ratio but only 25.6% survived when grown at a 1:2 ratio.

Figure 6.5 Sainfoin survival (arcsine√ %) in association with grass ratio at the end of 1st and 2nd season and in the following springs. Δ indicates 2:1 ratio with grass and O indicates 1:2 ratio. Bars indicated LSD(.05). At week 41 plant survived over the winter were counted and dead plants were replaced.



Grass species only affected sainfoin survival over the winter of the 1st growing season (Table 6.1). Sainfoin grown with meadow fescue had a 38.5% survival rate, which was lower than the 53.4% survival rate when grown with perennial ryegrass. Shoot competition affected ($P<0.01$) sainfoin survival in the 2nd growing season when 70.4% sainfoin plants survived, lower than 85.7% when grown without shoot competition (Table 6.2).

Table 6.1 Effect of grass species on sainfoin survival (arcsine √ %). Values in percentage are back-transformed. Values in bracket are transformed data of arcsine of square root.

		MF	PRG	LSD (.05)
Grass species	1st growing season	89.3%(1.237) a	81.5%(1.126) a	(0.1231)
	1st winter	38.5%(0.669) b	53.4%(0.819) a	(0.1314)
	2nd growing season	78.4%(1.087) a	78.8%(1.092) a	(0.1106)
	2nd winter	35.3%(0.636) a	38.2%(0.666) a	(0.138)

Values followed by the same letter are not significantly different.

Table 6.2 Effect of shoot competition on sainfoin survival (arcsine √ %). Values in percentage are back-transformed. Values in bracket are transformed data of arcsine of square root.

		Full competition	Nil competition	LSD (.05)
Shoot	1st growing season	82.4%(1.138) a	88.6%(1.226) a	(0.1231)
	1st winter	43.1%(0.716) a	48.8%(0.773) a	(0.1314)
	2nd growing season	70.4%(0.996) b	85.7%(1.183) a	(0.1106)
	2nd winter	31.6%(0.597) a	42.0%(0.705) a	(0.138)

Values followed by the same letter are not significantly different.

6.3.1.2 Grasses

The survival rate of grasses was generally not affected by ratio, root and shoot competition in either growing season or over the winters (Appendix 6-1). There were no significant differences in survival between meadow fescue and perennial ryegrass, except a small significant difference at the end of the 1st growing season (P< 0.05) (Appendix 6-1) (Table 6.3). 99.8% of meadow fescue survived, which was higher than the 98.5% of perennial ryegrass. This would not be considered significant agronomically.

Table 6.3 Survival of meadow fescue and perennial ryegrass (arcsine √ %). Values in percentage are back-transformed. LSD and values in bracket are transformed data of arcsine of square root.

		MF	PRG	LSD(.05)
Species	1st growing season	99.8%(1.534) a	98.5%(1.446) b	(0.0838)
	1st winter	96.3%(1.378) a	95.0%(1.346) a	(0.1098)
	2nd growing season	93%(1.302) a	93.9%(1.321) a	(0.0895)
	2nd winter	72.6%(1.02) a	73.6%(1.031) a	(0.1315)

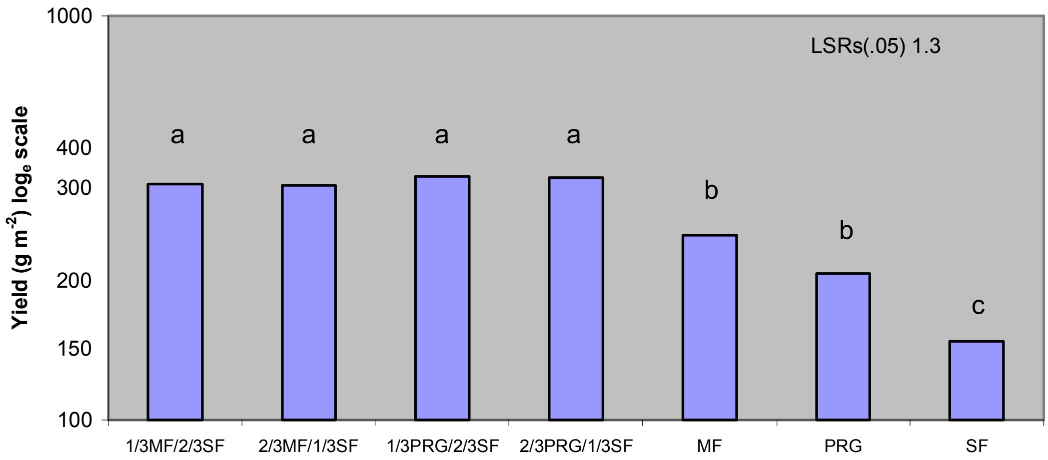
Values followed by the same letter are not significantly different.

6.3.2 Annual Yields

6.3.2.1 Mixture vs. Pure Stand

All mixtures outyielded pure stands of sainfoin, meadow fescue and perennial ryegrass (Figure 6.6). Meadow fescue and perennial ryegrass also out yielded sainfoin, but with no significant difference between the grasses.

Figure 6.6 Annual DM yields of mixtures and pure stands. Values are geometric means. LSRs are presented. Bars with the same letter are not significant.

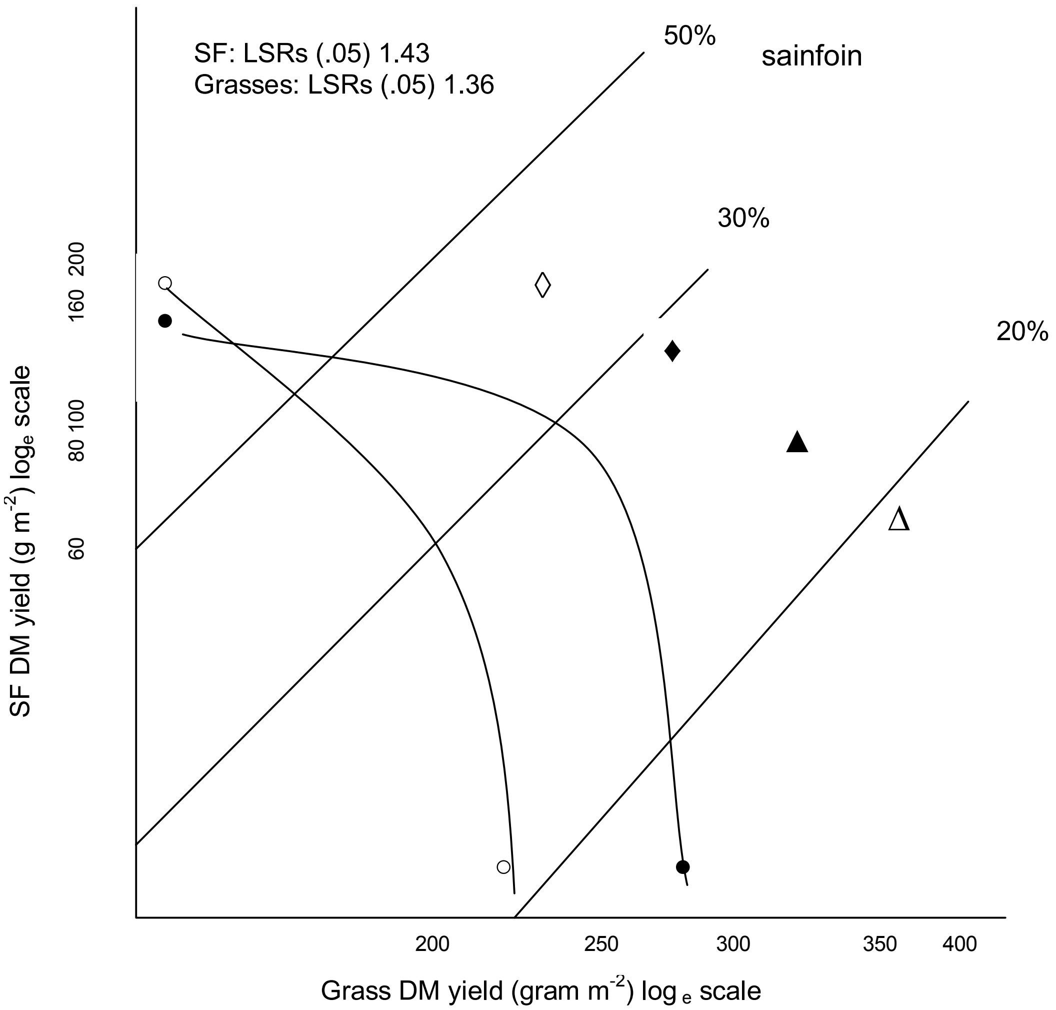


6.3.2.2 Sainfoin Yields

Sainfoin annual yield was affected by the grass ratio ($P<0.001$) and root competition ($P<0.001$), and grass ratio interacted with grass species ($P<0.05$) (Appendix 6-2). However, grass species and shoot competition had no effects on sainfoin annual yield.

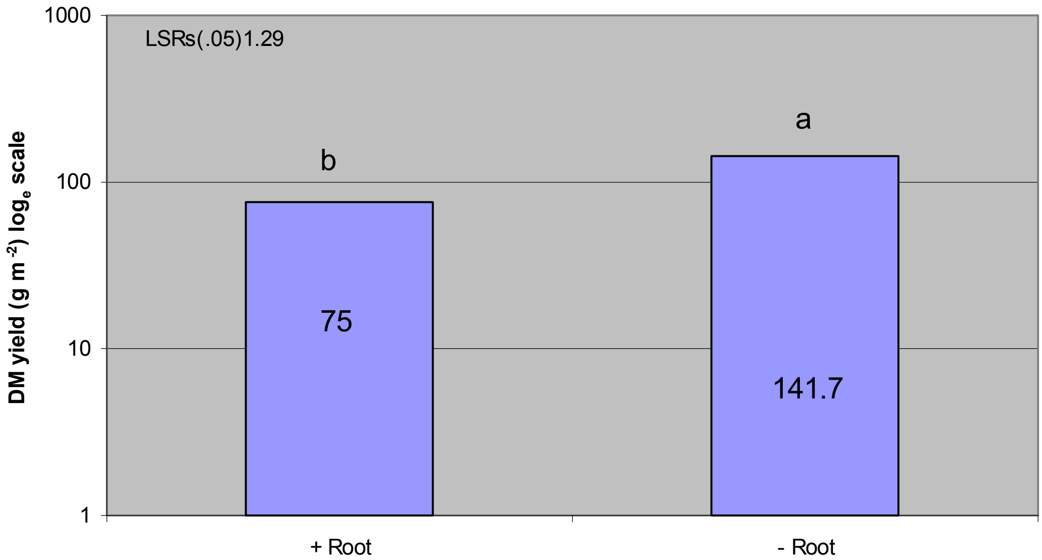
The annual total yield showed that increased grass ratios reduced sainfoin yield when grown with either meadow fescue or perennial ryegrass (Figure 6.7). However, there was a significant interaction. Sainfoin grown at a 2:1 ratio with perennial ryegrass yielded more than when grown with meadow fescue at the same ratio, but sainfoin grown at a 1:2 ratio with perennial ryegrass yielded less than when grown with meadow fescue (Figure 6.7).

Figure 6.7 Effect of grass species and ratio on the annual yield (g m^{-2} log$_e$ scale) of sainfoin and grass when sainfoin and grasses grown alone (MF●, PRG ○), in 2:1 and 1:2 ratio with grass (SF: MF 2:1 ♦, SF: MF 1:2 ▲, SF: PRG 2:1 ◊, SF: PRG 1:2 Δ). LSRs are presented.



Root competition significantly reduced sainfoin annual yield (Figure 6.8). Without root competition (141.7 g m^{-2}) the yield of sainfoin was nearly twice the yield of root competition (75 g m^{-2}).

Figure 6.8 Effects of root competition on sainfoin yield. Values are geometric means and LSRs are presented. Bars with the same letter are not significantly different.



6.3.2.3 Grass Yields

There was no yield difference between meadow fescue and perennial ryegrass, and shoot competition had no effect on grass annual yield (Appendix 6-2). Grass ratio ($P<0.05$) and root competition ($P<0.001$) did have effects on grass yield, and they also interacted ($P<0.05$).

Table 6.4 Interaction of ratio x root on grass yield (g m^{-2}). Values are geometric means and LSRs are presented.

SF: Grass	+ Root	- Root	LSRs
2:1 ratio	362.5 a	168.9 b	1.37
1:2 ratio	361.4 a	303.4 a	

Values followed by the same letter are not significantly different.

Root competition increased the annual grass yield at 2:1 ratio, compared to nil root competition, but had no effect at 1:2 ratio (Table 6.4). Grass species had no effects on annual grass yield when in root competition. However, the 1:2 ratio yielded higher than 2:1 ratio when there was no root competition annually.

Figure 6.9 Pictures of competition study.

a) after transplanting

b) May 28 2005

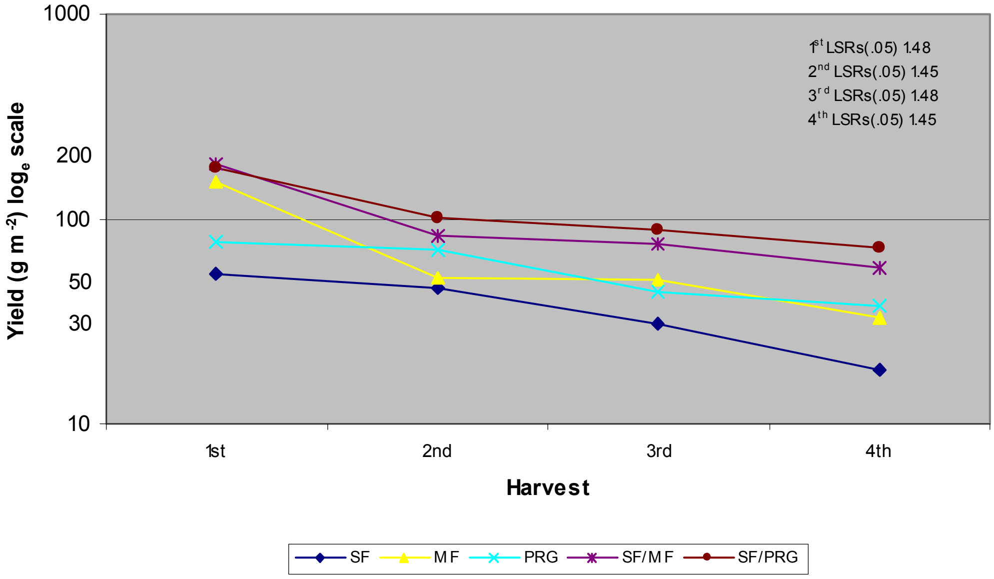
c) competition between sainfoin and meadow fescue

6.3.3 Seasonality of Yield

6.3.3.1 Seasonal Yield Distribution

Grass species had an effect on mixtures and pure stand throughout the season, except at the 3rd harvest ($P<0.05$) (Appendix 6-2). There was no significant yield difference between the mixture of sainfoin-meadow fescue and the mixture of sainfoin-perennial ryegrass (Figure 6.10). However, both mixtures with meadow fescue and perennial ryegrass yielded higher than the pure stand of sainfoin, meadow fescue and perennial ryegrass except for meadow fescue at the 1st harvest and perennial ryegrass at the 2nd harvest. The yield of sainfoin pure stand was lower than that of meadow fescue and perennial ryegrass, except at the 2nd harvest. Meadow fescue yielded higher than perennial ryegrass at the 1st harvest, but was lower yielding at the 2nd harvest, and there were no differences at the 3rd and 4th harvests.

Figure 6.10 Yield distribution of mixtures and pure stands. Values are geometric means. LSRs are presented.



6.3.3.2 Sainfoin Yields

Effects of Grass Species and Ratios

Grass species had no effect on the annual yield of sainfoin in the 2nd year. However, it did have effects on sainfoin yield at the 1st ($P<0.05$), 2nd ($P<0.05$) and 3rd ($P<0.01$) harvests (Appendix 6-2). There were interactions between grass species × ratio at these three harvests and between grass species × root at the 1st and 3rd harvests.

Table 6.5 The interaction of grass species x ratios on sainfoin DM yields (g m^{-2}). Values are geometric means and LSRs are presented.

Harvest		2:1 ratio	1:2 ratio	LSRs
1st	MF	29.67 b	29.05 b	1.21
	PRG	60.01 a	31.66 b	
2nd	MF	42.06 a	28.91 a	1.49
	PRG	43.03 a	16.02 b	
3rd	MF	30.57 a	19.41 b	1.32
	PRG	28.22 a	11.98 c	
4th	MF	14.59 b	8.59 c	1.47
	PRG	22.87 a	5.76 d	

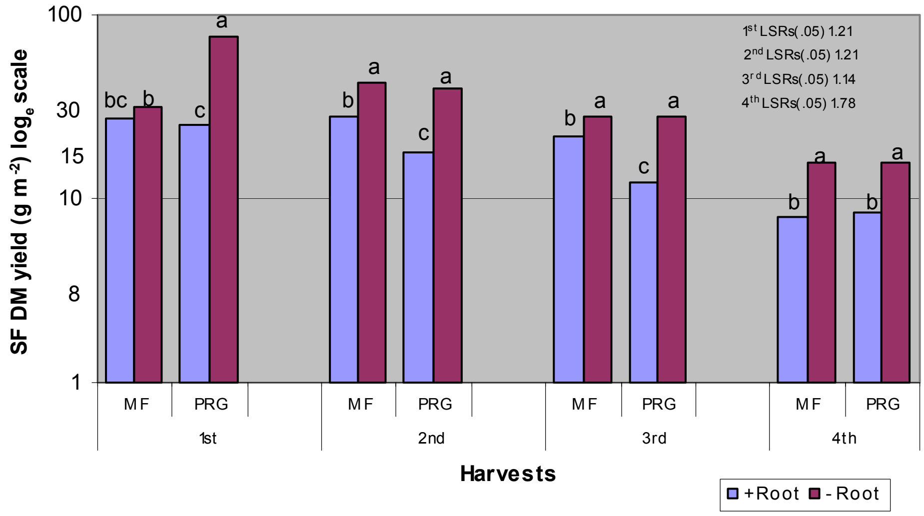
Values followed by the same letter are not significantly different.

Yields of sainfoin grown at 2:1 ratio with either perennial ryegrass or meadow fescue were higher than that grown at 1:2 ratio, apart from sainfoin grown with meadow fescue at the 1st harvest (Table 6.5).

Effects of Root Association

In general root competition significantly reduced sainfoin yield (Appendix 6-2). However, it interacted with grass species at the 1st ($P<0.01$) and 3rd ($P<0.01$) harvests. The yield of sainfoin in nil root competition was consistently higher than that when in root competition throughout the growing season except at the 1st harvest with meadow fescue (Figure 6.11).

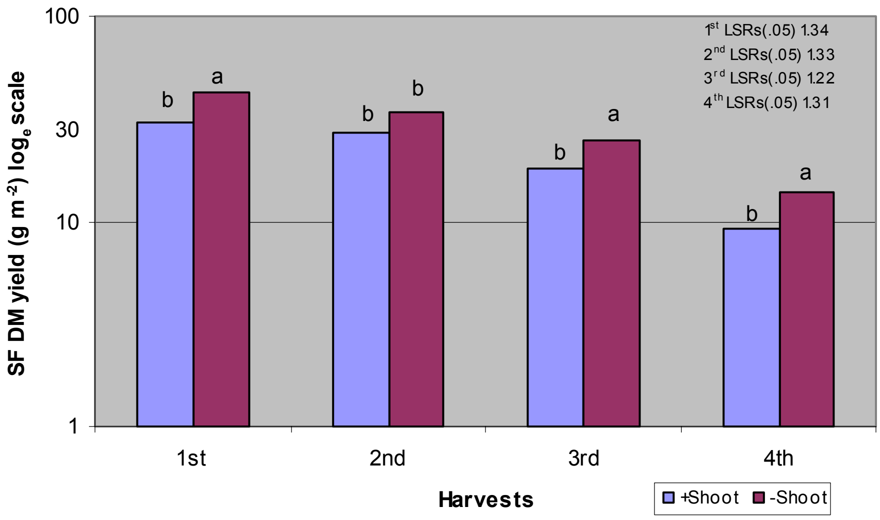
Figure 6.11 Interaction of grass species x root on sainfoin yield. Values are geometric means and LSRs are presented. Bars within harvest with the same letter are not significantly different.



Effects of Shoot Association

Shoot association had no significant effect on the annual yield, but had effects at the 1st ($P<0.05$) and 3rd harvests ($P<0.01$) (Appendix 6-2). Shoot competition generally reduced sainfoin yield, compared to nil shoot competition through the season except at the 2nd harvest (Figure 6.12).

Figure 6.12 The effect of shoot association on sainfoin yield in the 2nd year. Values are geometric means and LSRs are presented. Bars with the same letter within harvest are not significantly different.

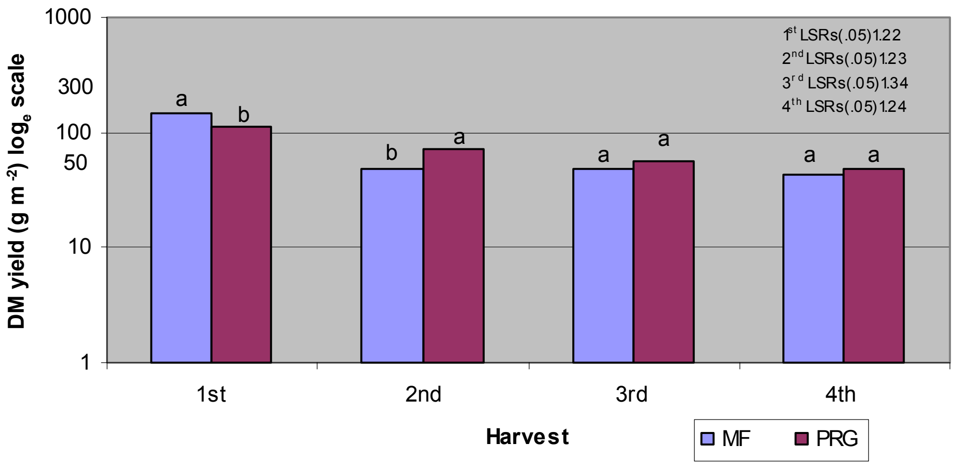


6.3.3.3 Grass Yields

Effects of Grass Species

There was no significant difference between meadow fescue and perennial ryegrass annually (Appendix 6-2). There were, however, significant differences at the 1st ($P< 0.05$) and 2nd ($P< 0.01$) harvests. At the 1st harvest the meadow fescue yield was higher than perennial ryegrass, but this was reversed at the 2nd harvest (Figure 6.13). There were no yield differences between meadow fescue and perennial ryegrass at the 3rd and 4th harvests.

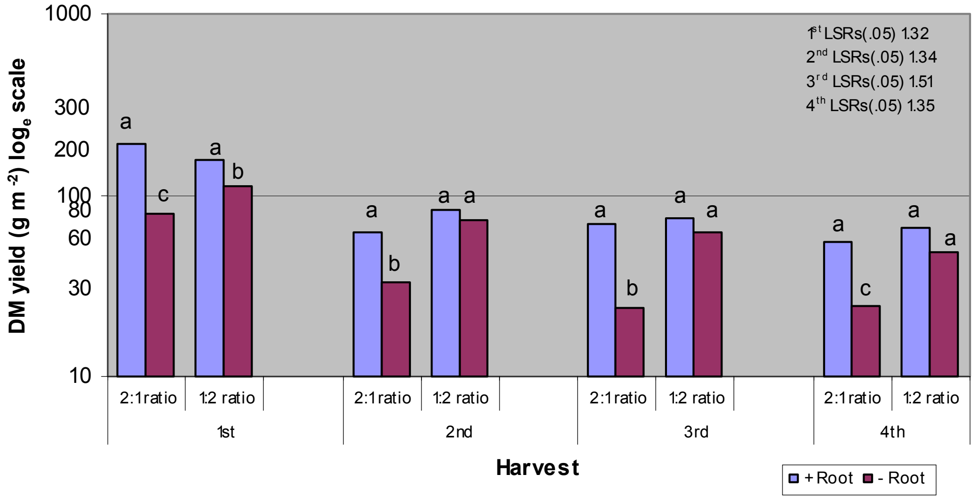
Figure 6.13 Grass yield. Values are geometric means and LSRs are presented. Bars within harvest with the same letter are not significantly different.



Effects of Ratio and Root

The sainfoin-grass ratios and roots significantly affected grass yield, and the ratio and root interacted throughout the season except that there was no significant difference between ratios at the 1st harvest (Appendix 6-2). Root competition increased grass yield at the 2:1 ratio, but had no effect on grass yield at the 1:2 ratio except for at the 1st harvest (Figure 6.14).

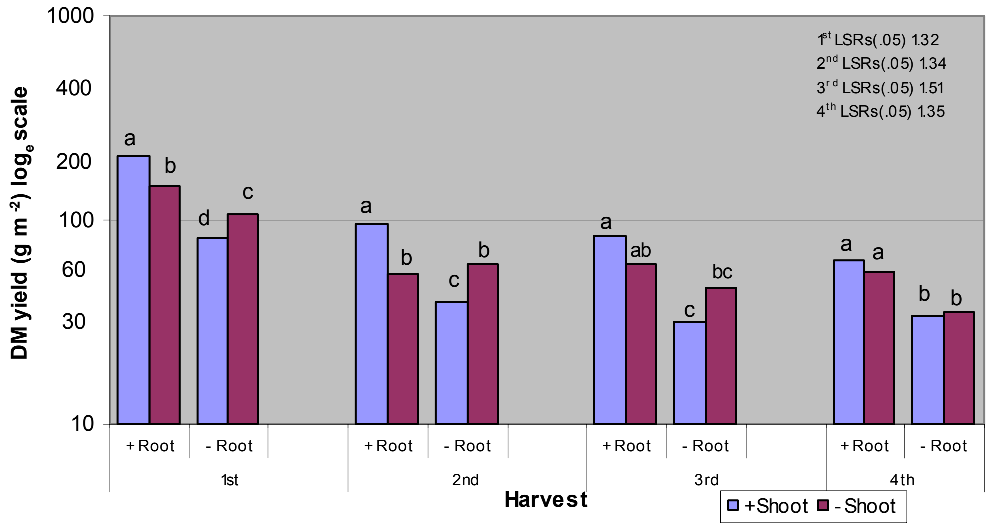
Figure 6.14 Interaction of ratio x root on grass yield . Values are geometric means and LSRs are presented. Bars within harvest with the same letter are not significantly different.



Effects of Shoot and Root Association

Shoot competition did not have an effect on grass yield (Appendix 6-2). The shoots interacted with the roots throughout the season, except for the 4th harvest (Appendix 6-2). Full competition had higher yield than nil competition and shoot competition at all the harvests (Figure 6.15).

Figure 6.15 The interaction of root x shoot on grass yield. Values are geometric means. LSRs are presented. Bars within harvest with the same letter are not significant.



6.3.4 Sainfoin Proportion in Mixtures

The sainfoin proportion in the mixtures peaked at the 2nd harvest, and then declined as the season progressed (Figure 6.16 & 6.17).

Grass species had a substantial effect on sainfoin proportion at the 2nd ($P<0.05$) and 3rd ($P<0.01$) harvests (Appendix 6-4). Ratio also had an effect on sainfoin proportion at the 2nd ($P<0.05$), 3rd ($P<0.01$) and 4th ($P<0.05$) harvests (Appendix 6-4). Root and shoot competition had strong effects on the sainfoin proportion ($P<0.001$) at all four harvests (Appendix 6-4).

Perennial ryegrass reduced sainfoin proportion more than meadow fescue through the season except at the 1st harvest (Figure 6.16)(Table 6.6). Sainfoin proportion in the meadow fescue mixtures peaked at the 2nd harvest and then declined. 1:2 ratio decreased sainfoin proportion more than the 2:1 ratio except at the 1st harvest.

Figure 6.16 The effect of grass species and ratios on sainfoin proportion (logit). Values and LSD are presented in Table 6 below.

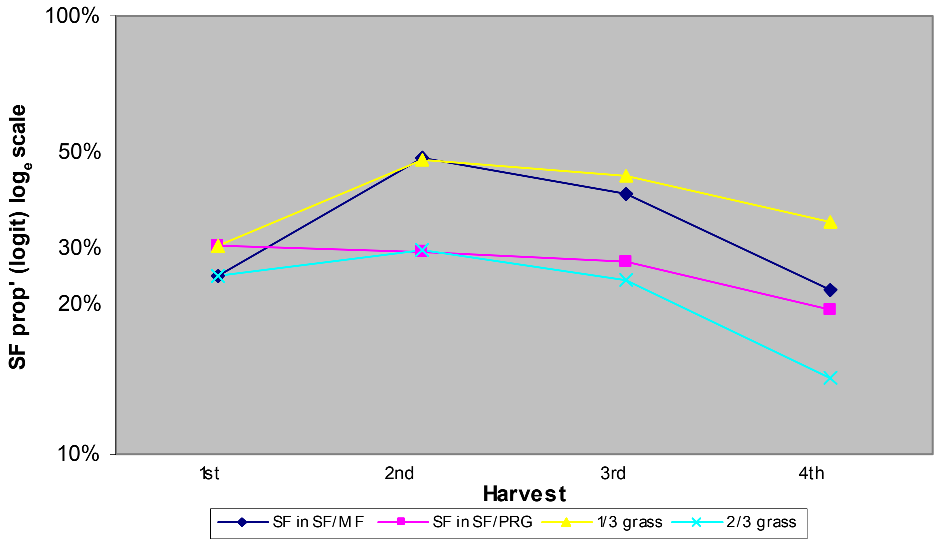


Table 6.6 The effect of grass species and ratios on sainfoin proportion (logit). Values in bracket and of LSD are logit transformed. Values in percentage are in inverse logit.

	1st harvest	2nd harvest	3rd harvest	4th harvest
SF in SF/MF	25.4%(-5.973) a	47.4%(-5.346) a	39.3%(-5.536) b	23.8%(-6.04) a
SF in SF/PRG	29.8%(-5.812) a	28.9%(-5.845) b	27.3%(-5.899) a	21.3%(-6.15) a
1/3 grass	29.7%(-5.816) a	47% (-5.356) b	43%(-5.445) b	34%(-5.68) a
2/3 grass	25.5%(-5.968) a	29.1%(-5.835) a	24.9%(-5.991) a	14.9%(-6.51) a
LSD (.05)	(0.2119)	(0.2649)	(0.1505)	(0.507)

Values followed by the same letter are not significantly different.

Root and shoot competition severely decreased sainfoin proportion, compared to nil competition (Figure 6.17) (Table 6.7). Root competition decreased sainfoin proportion 39.6%-46.7%, more than nil root competition over the growing season. Shoot competition decreased sainfoin proportion 24.1%-26.9% more than nil shoot competition.

Figure 6.17 The effect of root and shoot on sainfoin proportion (logit). Values and LSD are presented in Table 7 below.

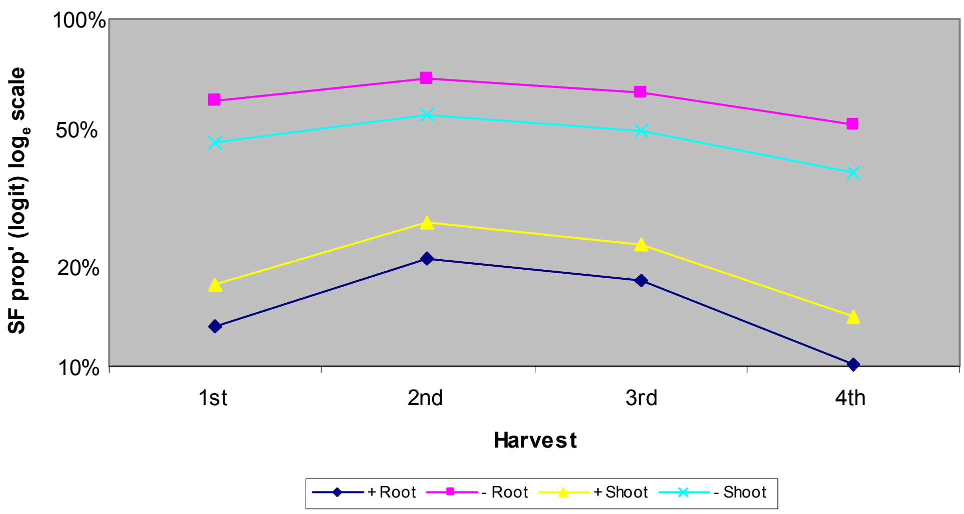


Table 6.7 The effect of root and shoot on sainfoin proportion (logit). Values in bracket and of LSD are logit transformed. Values in percentage are in inverse logit.

	1st harvest	2nd harvest	3rd harvest	4th harvest
+ Root	13.0% (-6.646) b	20.4%(-6.194) b	17.6%(-6.342) b	10.1% (-6.9) b
- Root	58.4% (-5.138) a	67.1%(-4.997) a	61.0%(-5.094) a	49.7%(-5.3) a
+ Shoot	17.2%(-6.366) b	25.9%(-5.953) b	22.4%(-6.098) b	14.0%(-6.57) b
- Shoot	44.1% (-5.419) a	52.8%(-5.239) a	47.9%(-5.337) a	36.1%(-5.62) a
LSD(.05)	(0.2119)	(0.2649)	(0.1505)	(0.507)

Values followed by the same letter are not significantly different.

6.3.5 Relative Yield

The annual Relative Yield indicated that ratio did not have any effect on the Relative Yields of either sainfoin or meadow fescue (Figure 6.18). The Relative Yield Total was above 1.0, which indicated that there was some avoidance of competition and that the species partially utilised different resources (De Wit & Van Den Bergh, 1965; Harper 1977).

Over the season the Relative Yield of meadow fescue was higher than that of sainfoin in either the 2:1 or 1:2 ratios, indicating that meadow fescue had greater competitive ability than sainfoin at the four harvests (Figure 6.19). However, the Relative Yield of meadow fescue varied greatly following the seasonal change. Relative Yield values of meadow fescue were above 1.0, except for the 2:1 meadow fescue at the 2nd and 3rd harvest, which indicated that intra-specific competition of meadow fescue was greater than inter-specific competition. Relative Yield value of sainfoin was below 1.0 and above 0.5 at the all four harvests, which indicated that inter-specific competition of meadow fescue was greater than intra-specific competition. Seasonal Relative Yield Total was all above 1.0 throughout the season, as was the annual Relative Yield Total.

Figure 6.18 Relative Yield and Relative Yield Total of sainfoin against meadow fescue in 2:1 and 1:2 ratios. ◙ stands for RYT.

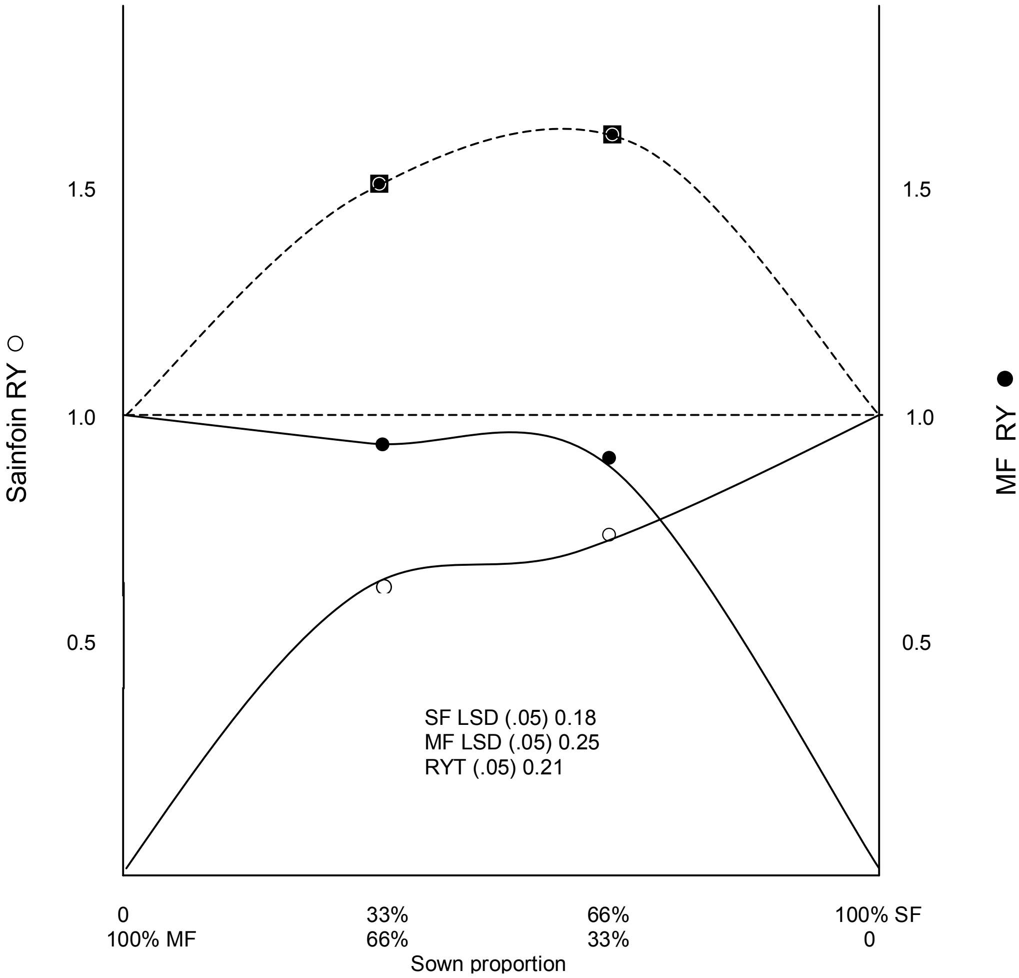
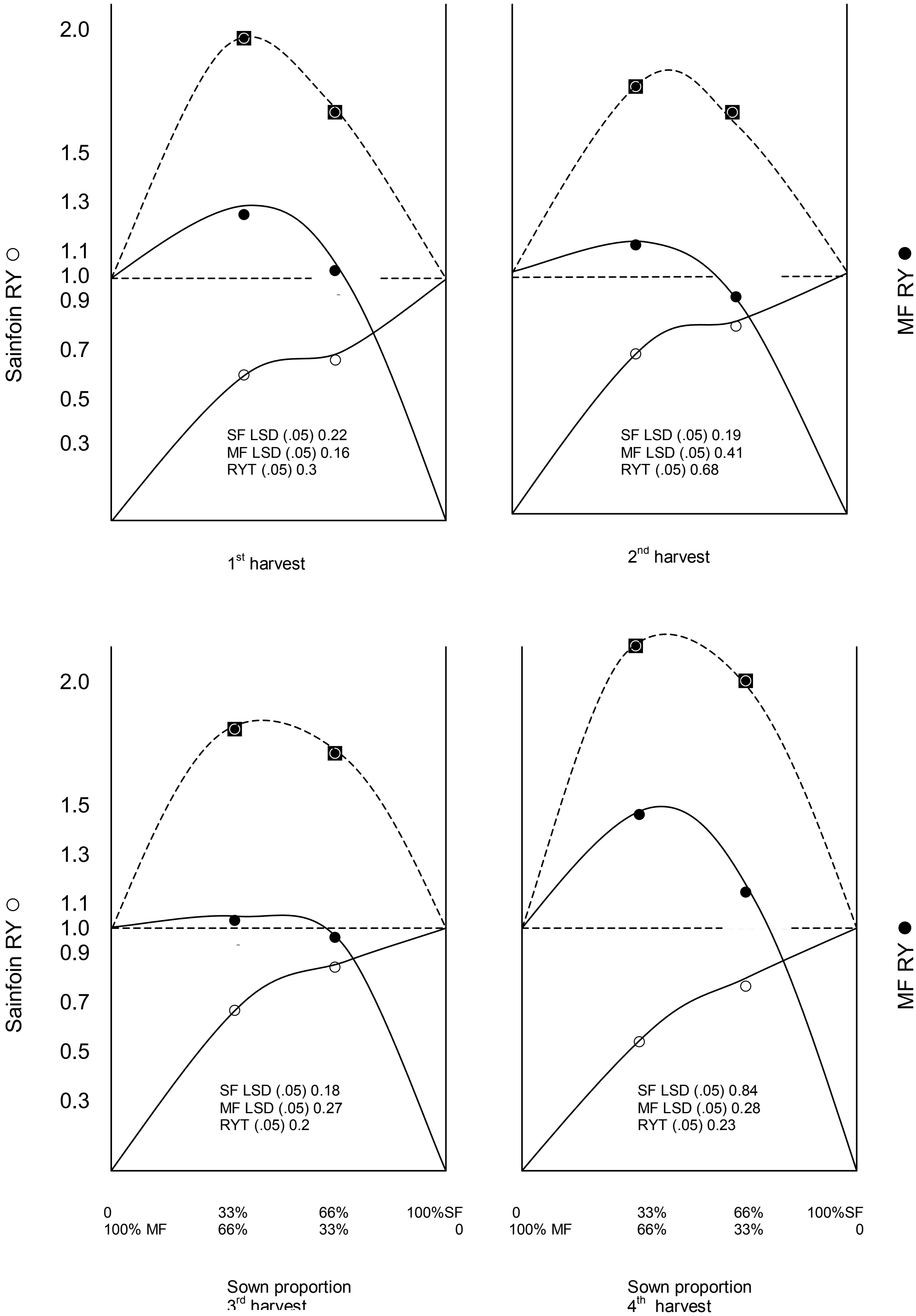


Figure 6.19 Relative Yield and Relative Yield Total of sainfoin against meadow fescue in 2:1 and 1:2 ratios throughout the season. ◙ stands for RYT.



Annual Relative Yield of perennial ryegrass in the 1:2 ratio was higher than that of sainfoin, showing greater competitive ability than sainfoin, but the Relative Yield of perennial ryegrass in the 2:1 ratio was similar to that of sainfoin (Figure 6.20). Relative Yield Total of both was also above 1.0.

Seasonal Relative Yield of perennial ryegrass was generally higher than that of sainfoin, showing greater competitive ability than sainfoin except for the 2:1 ratio of perennial ryegrass at the 4th harvest, which showed greater competitive ability of sainfoin than perennial ryegrass (Figure 6.21). Intra-specific competition of perennial ryegrass was greater than inter-specific competition throughout the season. Inter-specific competition of sainfoin was generally greater than intra-specific competition, but the 2:1 ratio of sainfoin also showed greater intra-specific competition than inter-specific competition at the 1st and 4th harvests.

The Relative Yield Total of sainfoin-perennial ryegrass was above 1.0 throughout the season (Figure 6.21), which indicated that they partially competed for the same limiting resources (De Wit & Van Den Bergh, 1965; Harper 1977) and showed a yield advantage over monocultures.

Figure 6.20 Relative Yield and Relative Yield Total of sainfoin against perennial ryegrass in 2:1 and 1:2 ratios. ◙ stands for RYT.

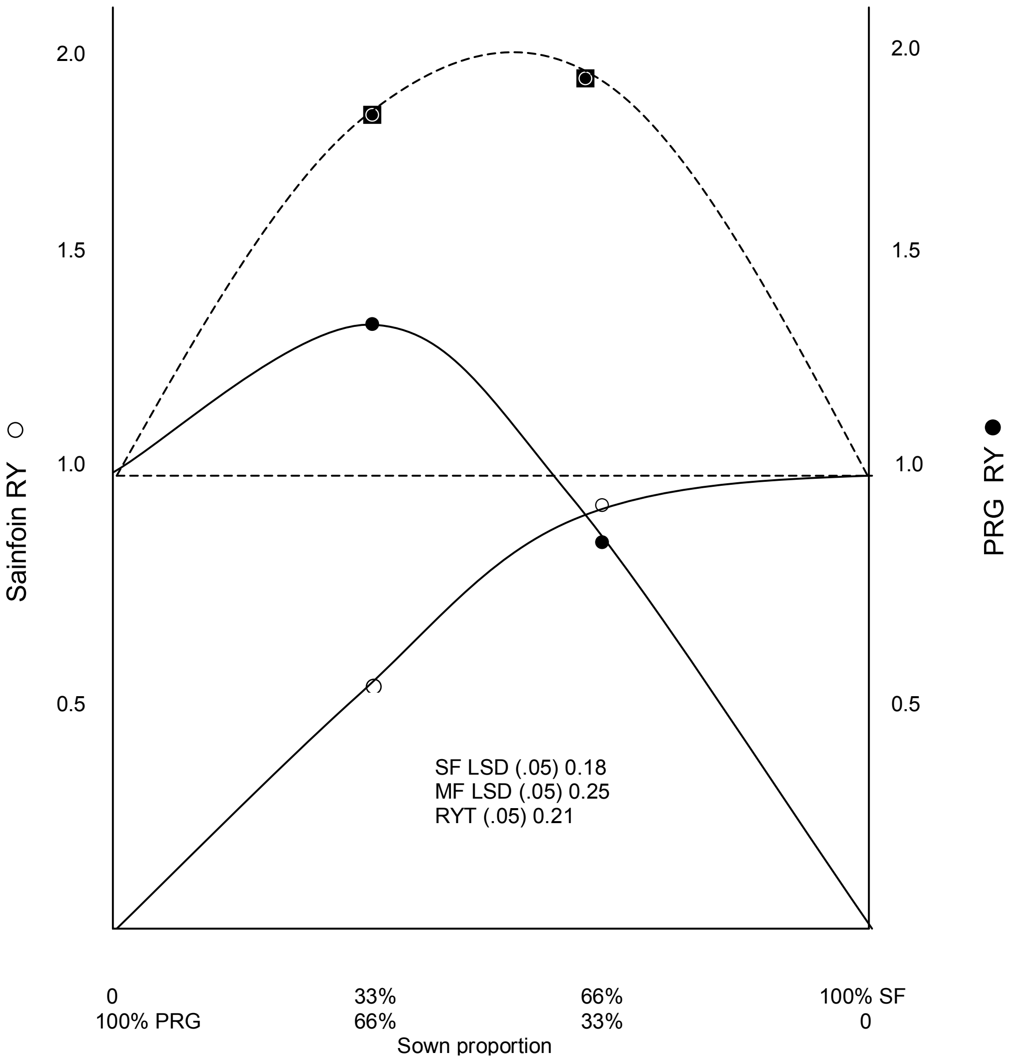
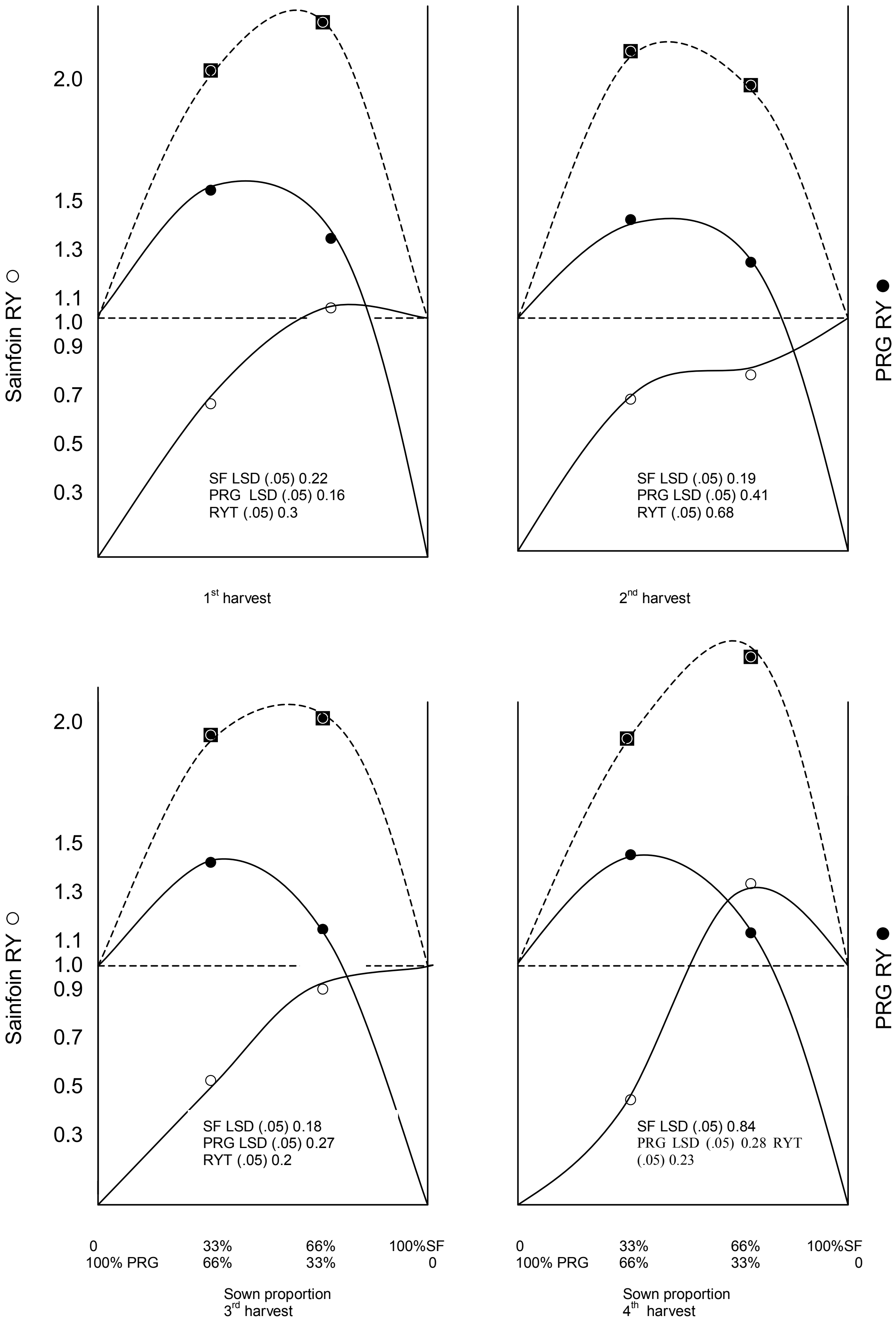


Figure 6.21 Relative Yield and Relative Yield Total of sainfoin against perennial ryegrass in 2:1 and 1:2 ratios throughout the season. ◙ stands for RYT.



6.3.6 Relative Crowding Coefficient

6.3.6.1 Effects of Root and Shoot on the Relative Crowding Coefficient

Root competition significantly (P<0.001, P<0.05) decreased the Relative Crowding Coefficient compared to nil root competition throughout the season (Appendix 6-3). Relative Crowding Coefficient in root competition was above 0.6, and in nil root competition was below 0.4 at all harvests (Figure 6.22). Shoot competition only had a small significant effect (P<0.05) on Relative Crowding Coefficient, compared to nil shoot competition, at the 1st harvest (Appendix 6-3). Relative Crowding Coefficient in shoot competition was greater than in nil shoot competition annually, and was also greater than nil shoot competition at the 1st harvest with no differences at the other harvests (Figure 6.23).

Figure 6.22 Root effect on Relative Crowding Coefficient. LSD is presented. Bars within harvests with the same letter are not significant.

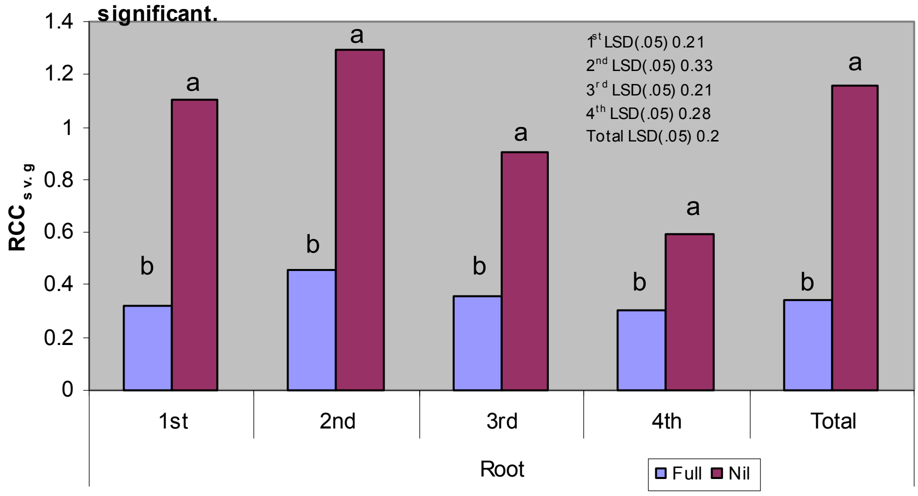
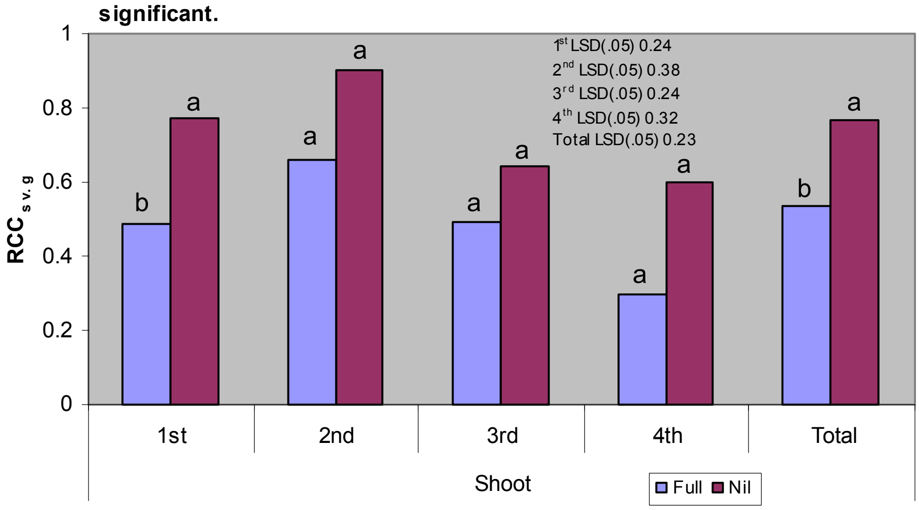


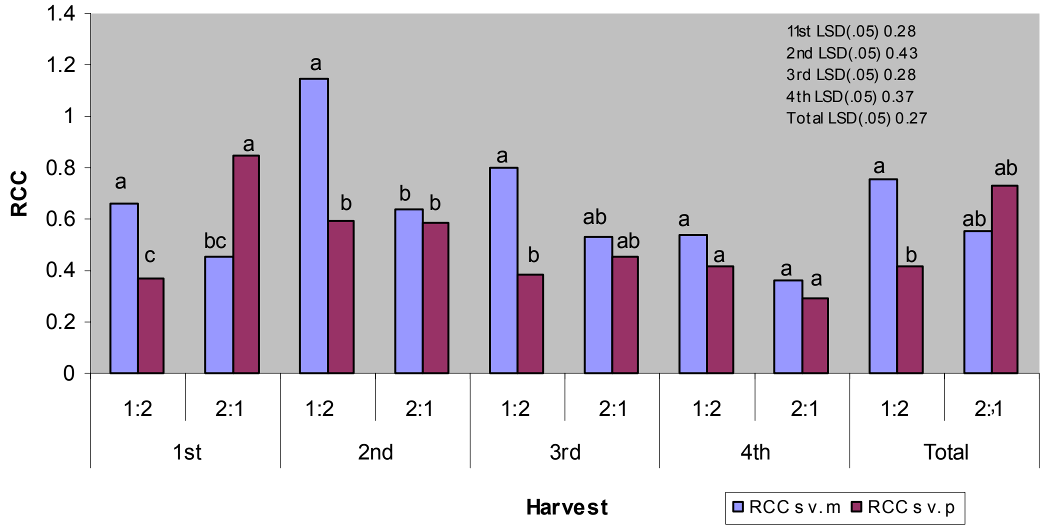
Figure 6.23 Shoot effect on Relative Crowding Coefficient. LSD is presented. Bars within harvest with the same letter are not significant.



6.3.6.2 Effects of Grass Species x Ratio on Relative Crowding Coefficient

There was an interaction (P<0.05) of grass species x ratio on Relative Crowding Coefficient (Appendix 6-4). Relative Crowding Coefficient for sainfoin-meadow fescue at the 1:2 ratio was greater than for sainfoin-perennial ryegrass annually, and also at the 1st, 2nd and 3rd harvests. But at the 2:1 ratio, the Relative Crowding Coefficient for sainfoin-meadow fescue was similar to sainfoin-perennial ryegrass annually, and also at the 1st harvest (Figure 6.24).

Figure 6.24 Relative Crowding Coefficient of sainfoin against meadow fescue and perennial ryegrass. LSRs are presented. Bars within harvest with the same letter are not significant.



6.4 Discussion

6.4.1 Sainfoin Survival and Proportion

Sainfoin survival generally declined through the two growing seasons and two winters. Greater grass ratios reduced sainfoin survival through the growing season and the winter. Sainfoin grown in the 2:1 ratios with grass had a better survival rate than in the 1:2 ratios. This may be because the increased grass ratio occupied more space above- and below ground, and competed for more resources, such as light, water and nutrients thus reducing the ability of sainfoin to gain more resources resulting in limitations on sainfoin plant development.

Over the winter of 2004, 27.4% (2:1 ratio with grass) and 48.7% (1:2 ratio with grass) of sainfoin plants died, compared to 6.4% and 25% respectively in the growing season. Over the winter of 2005, 36.6% (2:1 ratio with grass) and 45.1% (1:2 ratio with grass) of sainfoin died, compared to 14.6 % and 29.3% respectively in the growing season. More sainfoin plants died in the winter than in the growing season.

More plants in the 1:2 ratio with grass died in the winter, compared to the 2:1 ratios. This may well be because competition in the 1:2 ratio with grass affected the accumulation of sainfoin root reserves more than in the 2:1 ratios, and this resulted in sainfoin plants with less resistance to winter kill.

In the 2nd growing season and winter more sainfoin plants died, compared to the 1st growing season and winter. This may be because the 1st growing season only had one harvest and the 2nd growing season had four successive harvests, which possibly affected root reserve. In addition, the 2nd winter was much colder than the 1st, with the average temperature of the 1st winter (November-March) being 1 °C higher than the 2nd winter (section 3.21, Appendix 6-5)

Sainfoin proportion peaked at the 2nd harvest and then declined. Root competition reduced sainfoin proportion more than shoot competition. The change of sainfoin proportion is similar to the result also previously demonstrated for white clover-perennial ryegrass by Baines (1988).

6.4.2 Yield

6.4.2.1 Yield of Mixture and Pure Stand

Yields of pure stands of sainfoin were generally lower than mixtures of sainfoin-meadow fescue and sainfoin-perennial ryegrass, as well as pure stand of meadow fescue and perennial ryegrass. These results were similar to some previous field studies, which mixed sainfoin with crested wheatgrass, tall wheatgrass and smooth brome grass, and which indicated that the yield of sainfoin monoculture was lower than mixtures (Hanna *et al.,* 1977; Kilcher, 1982; Sengul, 2003). However, a four year field study on sainfoin mixtures with cocksfoot, timothy and meadow fescue at the Grassland Research Institute during 1952-1956 showed no differences between sainfoin and sainfoin-grass mixtures (Spedding & Diekmahns, 1972). In the study described in Chapter 4 on sainfoin-grass mixtures, the field data also showed that sainfoin and sainfoin-grass mixtures had similar yields. In this container study, pure stand sainfoin yielded lower than grasses. This result was in agreement with Hu and Jones' (2001) results on two tropical legumes and grasses in a pot study, which showed legume pure stands yielded less than pure grass stands. This may be because the partition created a micro-climate in containers, and this changed the growing environment of light, temperature and soil nutrient conditions compared to field condition. Theses results may be different, therefore, from the field trial to some extent. Kallenbach, Matches and Mahan (1996) reported that sainfoin production is low after a high temperature period, and that high temperature with defoliation leads to the inability of photosynthesis and carbohydrate reserves to support high metabolic rates during high temperature, resulting in the death of many sainfoin plants. In the containers, the temperature in the summer was about 5 °C higher than the field temperature. Furthermore, in the container study reported here the plants were irrigated during the growing season whereas the plants in the field trial reported in chapter 4 were not. This may have given an extra advantage to the grasses in this container competition study.

6.4.2.2 Yield of Sainfoin and Grass in Mixtures

Increased grass ratio generally reduced sainfoin yield in mixtures and increased grass yield in this study. This finding was in agreement with a competition study by Grieshaber-Otto (1984), who tested five different ratios of lucerne-perennial ryegrass and found that increased perennial ryegrass reduced Relative Yield of lucerne in the lucerne-perennial ryegrass mixtures. It was also in agreement with Hu and Jones' (2001) study between two tropical legumes and two tropical grasses. This may be because increased grass crowded out the space of sainfoin and gained more resources, including moisture.

Root competition reduced sainfoin annual yield in mixtures, and increased grass annual yield at the 1:2 ratio with sainfoin. Shoot competition did not affect annual yield of either sainfoin or grass. These results were in agreement with previous findings (Donald, 1958; King 1971; Snaydon & Baines, 1980; Snaydon & Harris 1981). Wilson (1988) summarised many competition studies, and also concluded that root competition is usually more important than shoot competition in determining competition balance, competition intensity and resource exploitation. Casper and Jackson (1997) explained that this was because root competition involved more resources (nutrients and water) than shoot competition (light). Grass benefited from root competition. This may be due to:

a) fine branched grass roots occupying more space underground and gaining advantage in obtaining nutrients and moisture
b) decomposition of the roots and nodules from dead sainfoin plants from the previous growing season and winter, supplying fixed nitrogen to grasses.

6.4.3 Resource Complementarity

Relative Yield Total of sainfoin-meadow fescue and sainfoin-perennial ryegrass was both more than 1.0, which indicated that sainfoin and grasses partially competed for the limiting resources and that the mixtures had yield advantages over monocultures. Some previous competition studies on legume-grass mixtures(Martin & Snaydon, 1982; Grieshaber-Otto, 1984; Wilson & Newman, 1987; Baines, 1988) also showed a Relative Yield Total above 1.0.

6.4.4 Competitive Ability

Relative Yield of sainfoin was annually below 1.0 both with meadow fescue and perennial ryegrass. It showed that interspecific competition of sainfoin against grass was greater than intraspecific competition. The Relative Yield of meadow fescue was annually below 1.0 as well, which showed that interspecific competition was greater than intraspecific competition. The Relative Yield of perennial ryegrass was above 1.0, which indicated that intraspecific competition was greater than interspecific competition. The Relative Yield of sainfoin was generally smaller than that of meadow fescue and perennial ryegrass. This indicated that sainfoin had less competitive ability than both meadow fescue and perennial ryegrass.

The Relative Crowding Coefficient (SF vs. MF or PRG) was reduced by root competition. This indicated that root competition reduced sainfoin's competitive ability, and benefited the meadow fescue and perennial ryegrass. Shoot competition generally had no effect on the Relative Crowding Coefficient except for a reduction at the 1st harvest. This may be related to the morphology of meadow fescue and perennial ryegrass in the spring, which was higher and relatively leafier than at other stages. Root competition was more intense than shoot competition in this study. This was in accordance with many previous studies (Snaydon, 1971; Wilson, 1988). The value of the Relative Crowding Coefficient of sainfoin against meadow fescue was greater than that of sainfoin against perennial ryegrass at the 1:2 ratio, which indicated that sainfoin grown with meadow fescue was more competitive than with perennial ryegrass. However, at the 2:1 ratio with grass, sainfoin had the same competitive ability against both meadow fescue and perennial ryegrass.

As this competition study was conducted in plastic containers, and the plants regularly watered and the shoots separated by white plastic film, this probably created a micro-climate and different soil conditions compared with the field. Temperature and soil moisture were both higher than for the field experiment, previously described in Chapter 4. As a result, pure stands of grasses in the containers gave four successive harvests in the 2nd year, compared with two harvests in the 2nd year and one in the 3rd year from the grasses grown in the field trial. It seems that higher temperatures also may have checked sainfoin growth. These are some of the limitations of this study. However, this study has revealed some interesting interactions between sainfoin and grasses, for instance, that root competition between sainfoin and grasses was more intense than shoot competition and that grasses also benefited from growing with sainfoin.

6.5 Conclusions

This study has shown the following:

- Sainfoin survival in mixtures was reduced by an increased grass ratio from 2:1 to 1:2.
- More sainfoin plants died in the winter than during the growing season.
- Greater grass ratio reduced sainfoin yields in mixtures.
- Yields of sainfoin-meadow fescue and sainfoin-perennial ryegrass mixtures were greater than that of the monocultures of sainfoin, meadow fescue and perennial ryegrass.
- Root competition had more effect than shoot competition on yield and competitive ability. Full competition generally increased the yields of grasses in mixtures and decreased that of sainfoin.
- Root competition increased grass yield at the 1:2 ratio with sainfoin, and reduced sainfoin yield.
- Intraspecific competition of tetraploid perennial ryegrass was greater than interspecific competition, and intraspecific competition of meadow fescue and sainfoin was less than interspecific competition.
- Sainfoin grown with meadow fescue cv. Rossa was more competitive than with tetraploid perennial ryegrass cv. Condesa at the 1:2 ratio with grass. This suggests that meadow fescue would be more likely to be a successful companion with sainfoin in a mixture than tetraploid perennial ryegrass.

Chapter Seven

General Discussion and Conclusions

7.1 Introduction

Sainfoin has been a traditional forage crop in the UK since the 17th century. The herbage has superior feeding value and gives rise to excellent animal performance (Spedding & Diekmahns, 1972; Sheldrick & Thomson, 1982). However, sainfoin has almost disappeared from British agriculture today. It has been nearly 25 years since a general discussion on sainfoin's future in British Agriculture was held at the Grassland Research Institute, Hurley, in 1982 (G.R.I, 1982). Since then almost no research on sainfoin has taken place in the UK. The biology and agronomy of sainfoin is still not well understood. However, policy changes to European agriculture are now creating an environment which allows and encourages greater development of legume-based pastures (Directive 91/676/EEC (1991); Rochon *et al.*, 2004). This policy change, together with the yield potential of the crop ascertained by this study, and the reported remarkable animal performance (e.g. Frame *et al.,* 1998) which it makes possible, demand greater attention and justify more studies into sainfoin.

In chapter 1(section 1.8) a series of advantages and disadvantages of the crop were set out (Table 1.4). The major agronomic problems and uncertainties identified from the review of literature were also identified. In this final chapter the extent to which these have been addressed in this study, will be critically discussed, together with a reflection on any shortcomings of the investigations and recommendations for future research.

7.2 Yield

Sainfoin is generally reported to have a lower yield than lucerne, of about 8-12 t DM ha^{-1} in Europe and 8-10 t DM ha^{-1} in the UK (Green, 1967; Frame *et al.*, 1998). Other reports suggest yield variations from as little as about 4 t DM ha^{-1} (Robinson, 1937; Goplen *et al.*, 1991; Fychan & Jones, 1997) in the establishment year or in unfavourable conditions to about 14-16 t DM ha^{-1} (Sheehy *et al.*, 1984; Lane &

Koivisto, 1998). In this study (Chapter 3) April and May sowings with cv. Cotswold Common achieved about 14.3 t DM ha^{-1} in the first harvest year. This implies that about 12 t DM ha^{-1} yield could be achieved in practice. According to Doyle *et al.* (1983) who did an economic assessment of the potential of sainfoin, if stable yield could be improved to 11.5 t DM ha^{-1}, it could increase the possibility of sainfoin being more widely grown. About 12.6 -14.5 t DM ha^{-1} of sainfoin–grass mixtures were encouragingly achieved in the first harvest year (Chapter 4).

2-2.4 t ha^{-1} crude protein yield was obtained from the April to July sowings in the first harvest year, but only 0.9-1.1 t DM ha^{-1} from the August and September sowings. The crude protein yield of the April-July sowings was similar to that reported for established red clover and lucerne (Doyle *et al.*, 1983).

7.3 Establishment Strategies

The basics of seed treatment and sowing depth were considered in Chapter 2, since these were judged to be potentially important factors which might bear significantly on the success of establishment (Zade, 1933; Wiesner *et al*, 1968; Arnott, 1969; Ries & Hoffman, 1995). Sainfoin seeds can be sown as hulled (in a seedpod) or as dehulled seeds. The study in Chapter 2 suggested that there is likely to be no difference in emergence between hulled or dehulled seeds at normal sowing depths (between1–4 cm). It also revealed that seeds sown on the surface established better as hulled seed, probably because the seedpod acted as a moisture reservoir. This implies that broadcasting hulled seeds may have an advantage over dehulled seed since broadcasting may leave some seeds on the soil surface. Sainfoin has been reported to have about 4-20% hard seeds (Spedding & Diekmahns, 1972), which can also affect germination and emergence. Seed scarification and germination benefits was not studied, but could probably be achieved during mechanical dehulling. A further study on the effects of hard seeds and the possible benefits of scarification on germination and emergence of sainfoin may be needed.

With all perennial forage legumes it is important to consider the average yield of the crop over all seasons. Whereas yields in the first full production year of up to 14.3 t DM ha^{-1} have been achieved, average yields over two or three seasons can be substantially below this (Fig 3.10). In recent years, UK livestock farmers have increasingly turned to establishing grass-legume leys after winter cereal harvest in July and August, in order to minimise the establishment period and to maximise the stocking density per unit of forage area. For ryegrass-clover leys this practice has proved reasonably satisfactory (e.g. Laidlaw & McBride 1992), but for sainfoin such a practice (of August sowing) could lead to substantial reductions in average yields. As expected, appropriate sowing date influences germination and emergence and establishment of good stands (Miller & Stritzke, 1995). The April and May sowings appeared the most suitable dates with an average yield of 8.4 t DM ha^{-1} for sainfoin, over three years of the present study. The June and July sowings gave only slightly lower yields, but the August and September sowings averaged only 5.33 and 4.22 t DM ha^{-1} respectively.

Undersowing in spring cereals could be one solution (Miller & Stritzke, 1995; Frame *et al.*, 1998; Odhiambo & Bomke, 2001) and in the study described in chapter 4 the yields were, by the second full production year (2005), not significantly different to the direct sown crops. The sainfoin in this study was undersown with the spring barley at Zadoks growth stage 1.3 (Zadoks *et al.*, 1974) and the cereal crop was taken for grain. Alternative strategies which may have improved the establishment of sainfoin and achieved a worthwhile forage yield, could have involved sowing sainfoin and spring cereal seed at the same time and/or taking the barley/sainfoin/grass mixtures as wholecrop forage. Undersowing in forage peas, as Koivisto (2002) did, could have been a further option although this would have restricted weed control to cultural methods only. Although undersowing does not appear particularly satisfactory from this study, it may, in practice, be the best option and provide a more worthwhile output during the establishment period with only a moderate depression in subsequent yield.

Sainfoin seed rates used in this study were 90 kg ha^{-1}. From Canadian experience (Goplen *et al.*, 1991) on seed rate (20-40 kg ha^{-1} drill) a lower seed rate may be possible. A future study on seed rates and seedling establishment could perhaps help to reduce seed costs, and should be explored.

7.4 Weed Control

Weed control is an important factor affecting the establishment and yield of any crop (Willard, 1951). Figure 3.15 (section 3.3.7) showed there was between 6%-51% of weeds in the herbage yield in the establishment year. In several situations during this study weed control became problematic. Black nightshade and chickweed severely invaded plots in the 2004 establishment of the sowing date and variety trials. MCPA + MCPB successfully controlled most broad-leaved weeds in 2003. In 2004, however, a dry period delayed the emergence of sainfoin until June and MCPA + MCPB was applied late and broadleaved weeds, especially black nightshade, were not satisfactorily controlled. Chickweed also occurred seriously in 2004. Carbetamex used in sainfoin pure sands successfully controlled this weed during the winter. Other weeds control options, such as bromoxynil (Stewart, 1968), was reported to give excellent control of broadleaved weeds causing only slight injury to sainfoin. Benefin also gave good control of most broadleaved weeds and grasses in Montana USA.

Mature sainfoin may be tolerant to glyphosate. An observation study was conducted during this study period and up to 4 l ha^{-1} Roundup Gold was applied to sainfoin (cv. Emyr) in the half-flowing stage and grass weeds were killed without apparent damage to the sainfoin. This could be an important and valuable attribute since it may offer opportunities for enhancement of poor stands of sainfoin by renovation or overseeding techniques.

7.5 Variety Choice

Cv. Cotswold Common along with another seven cultivars (Chapter 5) achieved an average annual yield of 11.9 t DM ha^{-1} in the first full harvest year. Surprisingly, there were no significant differences between varieties. Cv. Sombourne exhibited quicker regrowth than other cultivars after cutting. This could be important since sainfoin was frequently reported to be slow in recovery after cutting. In Chapter 5 and in a previous study (Koivisto & Lane, 2001) it was shown that cvs. Cotswold Common and Sombourne had similar annual production potential based on two year's trials. Further investigation of cv. Sombourne to study its seasonal yield distribution and persistency over several years, may be worthwhile.

7.6 Sainfoin-Grass Mixtures

Growing legumes and grasses in mixtures can achieve more efficient light utilization (Brougham, 1958), and the grass can benefit from the fixed nitrogen of legumes (Sprent, 1996). It can also help to reduce weed ingress (Droslom & Smith, 1976), improve forage quality (Baylor, 1974; Sleugh, Moore, George & Brummer, 2000), improve the seasonal distribution of forage, and increase total production (Sleugh *et al.,* 2000).

Over three years of field trials on sainfoin-grass mixtures (Chapter 4), sainfoin and tetraploid perennial ryegrass seemed to grow well together and sainfoin-meadow fescue appeared slightly less compatible. This may be due in part to the characteristics of the late heading cv. Condesa (NIAB, 2004) and early heading meadow fescue cv. Lifara, particularly at the first harvest. Furthermore, the seed rate of meadow fescue was a little more (about 7 kg ha^{-1}) than is normally advised. This result appears to contradict a traditional recommendation for meadow fescue as an ideal companion grass for sainfoin (e.g. Bland, 1971; Sheldrick *et al.,* 1995). It also contradicted the results from the competition study (Chapter 6) which suggested that meadow fescue would be more likely to be a successful companion than tetraploid perennial ryegrass. However, since the competition study was conducted in an artificially maintained environment and the field study in slightly different weather and soil conditions, it may not be valid for these two studies to be so directly compared.

In the field study (Chapter 4) straight sainfoin and sainfoin-grass mixtures in two ratios showed few differences in production over three years. However, it suggests that a late heading tetraploid perennial ryegrass (such as cv. Condesa) could be a suitable companion for sainfoin. The incorporation of sainfoin with a tetraploid perennial ryegrass-white clover mixture to help to reduce bloating could therefore be envisaged as, for example, McMahon *et al.* (1999) recent study on sainfoin-lucerne mixtures.

The competition (chapter 6) study gave yields of sainfoin monoculture lower than that of the mixtures and grasses, which were different from the results in the field study (Chapter 4). This may indicate the limitation of this study. The container and below- or aboveground divisions created a slightly different environment with higher temperatures and more moisture, which was different from the field. It is known that higher temperatures can lead to low sainfoin yield (Frame *et al.,* 1998) and grass yields almost certainly benefited from the periodic watering of the containers. However, a detailed study of the results shows the following:

- Root competition had more effects than shoot competition on yield and competitive ability as reflected in many other legume-grass studies (e.g. Donald, 1958; Wilson, 1988).
- Root competition and full competition generally increased the yields of grasses in mixtures and decreased that of sainfoin.
- Sainfoin grown with meadow fescue was more competitive than with the tetraploid perennial ryegrass, suggesting that meadow fescue overall was the less aggressive competitor.

7.7 Persistency

Sainfoin has been reported to be less persistent than lucerne (Frame *et al.*, 1998). Sainfoin yields in this study (Chapter 3 & 4) peaked in the second year (April-July sowings) and declined in the third year (between 5 - 30% decrease). Sainfoin plant populations also declined over the experimental period (Chapter 3, 4, 5). A three cut system with seven week intervals was applied.

Early autumn cutting reduced sainfoin population and yield in the third year. However, this study only examined production; the dynamics of carbohydrate in roots during the growing season and in the autumn is still not clear. The cutting system and autumn management applied in this study may have affected sainfoin persistency. Autumn management (early v. late cutting in order to facilitate the build-up of carbohydrate root reserves) did appear to have an effect on subsequent production, but the effect was not as marked as that reported by Jones (1955) for lucerne. A more detailed study on this factor alone over an extended period would also be beneficial for the information of future sainfoin growers.

No major pest or disease problems were found in this study, apart from sainfoin rust found in the 2nd year of 2004 establishment of the sowing date trial.

Sainfoin plant survival and yield in mixtures were both reduced by an increased grass ratio in the competition study described in Chapter 6. More sainfoin plants died during the winter than in the growing season (section 6.41) suggesting that competition was less important than winter kill, and emphasising the importance of any factor (such as autumn management) which is likely to have an effect on winter plant survival and subsequent vigour.

7.8 Response to *Rhizobium* Inoculation and Nitrogen

Sainfoin appears to be insufficient in fixing nitrogen and can show nitrogen deficiency symptoms in inoculated plants (Sims *et al.,* 1968; Burton & Curley, 1968; Meyer, 1975). Nitrogen fixation can be stimulated by low levels of inorganic nitrogen and checked by high levels (Koter, 1965b). For this reason a small amount (50 kg N ha^{-1}) was applied to the sainfoin - grass mixtures field trial described in chapter 4.

In this study no *Rhizobium* spp. innoculant was applied in any field trials. This was for two reasons; a) sainfoin plants grown previously had shown roots with apparently reasonable numbers of nodules and b) sainfoin was a commonly grown plant on Coates Manor Farm in the past and it was felt likely that suitable bacteria for nodulation would probably still survive in the soil. Two experiments designed to investigate the effectiveness of commercially available sainfoin inoculant strains and the effects of nitrogen supply on nodulation were in fact conducted in the laboratory and greenhouse during this study (Appendix 9.5 & 9.6). Both of these experiments became contaminated and failed however, and it was concluded that the laboratory conditions available were not suited to this type of work. The interaction of *Rhizobium* *spp* and nitrogen in soil on the nodulation and growth of sainfoin would be worth further study in a laboratory.

7.9 Overall Conclusions

- Sainfoin is capable of giving moderate yields of high quality forage over two to three year periods in UK conditions. Strategies to enhance its persistency need to be better developed.
- April or May sowing is preferable to sowing at any other time.
- Undersowing in spring cereals can be successful but may incur a yield penalty.
- The ideal sowing depth is between 1–4 cm and there is no particular value in sowing dehulled seed.
- The use of a species-specific inoculum should be investigated in suitable conditions.
- Weed control with MCPA + MCPB or carbetamide is adequate and glyphosate tolerance should be investigated further.
- There is little difference between the yields of the studied cultivars of sainfoin.
- Sainfoin mixtures with late heading tetraploid perennial ryegrass can both be successful.
- Autumn management of sainfoin appears to be less critical than for lucerne but needs more detailed investigation.
- Root competition between sainfoin and grasses was severe than shoot competition.
- Competition study showed that perennial ryegrass was more aggressive than meadow fescue.

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Appendices

9.1 Code and Definition of Sainfoin Morphological Stages

The stages are quantified by Kalu and Fick (1981) for alfalfa and have been modified by Borreani and Tabacco (2003) for sainfoin.

Code	Stage name	Stage definition
0	Rosette	No stems or floral buds
1	Mid-vegetative	Stem length ≤ 30cm; no buds or flowers
2	Late -vegetative	Stem length≥30cm; no buds or flowers
3	Early bud	1 to 2 nodes with buds; no flowers or seed pods
4	Late bud	> 2 nodes with buds; no flower or seed pods
5	Early flowering	One node with one open flower; no seed pods
6	Late flowering	≥ 2 nodes with one open flower; no seed pods
7	Early seed pod	1 to 3 nodes with green seed pods
8	Late seed pod	> 3 nodes with green seed pods
9	Ripe seed pod	Nodes with mostly brown mature seed pods

Kalu, B.A. and Fick, G.W. (1981). Quantifying morphological development of alfalfa for studies of herbage quality. *Crop Science*. 21:267-271

Borreani, G., Peiretti, P.G. and Tabacco, E. (2003). Evolution of yield and quality of sainfoin (*Onobrychis viciifolia* Scop.) in the spring growth cycle. *Agronomie.* 23:193-201.

9.2 Appendix to Chapter 3

Appendix 3-1 Analysis of variance of average DM yield over three year.

		2003 establishment	2003+2004 establishment
	d.f.	M.S	
Sowing Date	5	13.1***	9.4***
Residual	8 (2)	0.17	0.23

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.
d.f values in bracket stand for the missing value.

Appendix 3-2 Analysis of variance of average DM yield of the 2003 and 2004 establishments over two years.

	d.f	M.S
Sowing Date (SD)	5	27.8***
Establishment Year (EY)	1	37.3***
SD x EY	5	3.5 *
Residual	21(1)	1.1

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.
d.f values in bracket stand for the missing value.

Appendix 3-3 Analysis of variance of crude protein yield of the 2003 establishments in the 2nd year.

	d.f	M.S
Sowing Date	5	0.72***
Residual	5	0.01

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

9.3 Appendix to Chapter 4

Appendix 4-1 Analysis of variance of direct sowing and undersowing over three years.

	d.f.	M.S.
Sowing Treatment (ST)	1	6.09***
Year (Y)	2	167.1***
ST x Y	2	2.13***
Residual	10	0.06

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

Appendix 4-2 Yields of direct sowing and undersowing over three years.

	1st year	2nd year	3rd year	Average
Direct sowing	1.8	12.2 a	7.7 a	7.24 a
Undsowing	N.A	10.3 b	7.9 a	6.07 b
LSD(.05)		0.44		0.25

Values followed by the same letter are not significantly different.

Appendix 4-3 Analysis of variance of sainfoin-grass mixtures over three years.

	d.f.	M.S.
Mixture (M)	6	42***
Year (Y)	2	482.5***
M x Y	12	9.1***
Residual	40	0.4

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

Appendix 4-4 Analysis of variance of sainfoin population in mixtures over three years.

	d.f.	M.S.
Mixture (M)	3	708*
Year (Y)	2	694*
M x Y	6	257*
Residual	19 (3)	97

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

Appendix 4-5 Analysis of variance of crude protein yield of sainfoin-grass mixtures in the 2nd year.

	d.f	M.S
Sowing Date (SD)	6	1.1***
Residual	6	0.02

*, **, *** Significance at the 0.05, 0.01 and 0.001 level of probability respectively.

Appendix 4-6 Plant population of direct sowing and undersowing in the establishment year.

		Direct sowing	Undersowing
SF		74.7	92.8
MF		77.9	78.9
PRG		69.3	85.3
2/3SF +1/3MF	SF	62.9	83.2
	MF	50.1	41.6
1/3SF +2/3MF	SF	35.2	40.5
	MF	76.8	66.1
2/3SF +1/3 PRG	SF	46.9	65.1
	PRG	29.9	37.3
1/3SF +2/3 PRG	SF	36.1	36.3
	PRG	52.4	64.0

9.4 Appendix to Chapter 6

Appendix 6-1 Analysis of variance of survival (arcsine transformation) of sainfoin and grasses.

		the end of 1st year		over winter of 1st year		the end of 2nd year		over winter of 2nd year	
		Sainfoin	Grasses	Sainfoin	Grasses	Sainfoin	Grasses	Sainfoin	Grasses
Source of variation	d.f	Mean sqare							
Grass	1	0.1	0.06*	0.18*	0.01	0.00	0.003	0.007	0.00
Ratio	1	0.57***	0.001	1.36***	0.003	0.26**	0.02	0.48**	0.012
Root	1	0.01	0.002	0.01	0.02	0.06	0.04	0.014	0.05
shoot	1	0.06	0.00	0.03	0.01	0.28**	0.03	0.094	0.001
Grass.Ratio	1	0.03	0.00	0.1	0.03	0.001	0.00	0.00	0.035
Grass.Root	1	0.01	0.002	0.05	0.01	0.05	0.002	0.11	0.033
Ratio.Root	1	0.003	0.002	0.004	0.01	0.00	0.02	0.007	0.001
Grass.shoot	1	0.07	0.00	0.00	0.01	0.09	0.01	0.06	0.002
Ratio.shoot	1	0.001	0.01	0.01	0.01	0.00	0.03	0.00	0.097
Root.shoot	1	0.27	0.01	0.03	0.05	0.01	0.06	0.014	0.044
Grass.Ratio.Root	1	0.00	0.0002	0.03	0.02	0.02	0.004	0.007	0.054
Grass.Ratio.shoot	1	0.01	0.01	0.00	0.01	0.00	0.02	0.014	0.125
Grass.Root.shoot	1	0.11	0.00	0.04	0.01	0.04	0.03	0.014	0.002
Ratio.Root.shoot	1	0.2	0.00	0.13	0.01	0.004	0.005	0.023	0.007
Grass.Ratio.Root.shoot	1	0.03	0.01	0.12	0.02	0.04	0.06	0.00	0.065
Residual	16	0.03	0.01	0.03	0.02	0.02	0.01	0.033	0.03

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively

Appendix 6-2 Analysis of variance of yield ($\log_e$ transformation) of the 2nd year

Source of variation	d.f	1st harvest			2nd harvest			3rd harvest			4th harvest			Total yield		
		Sainfoin	Grass	Mixture	Sainfoin	Grass	Mixture	Sainfoin	Grass	Mixture	Sainfoin	Grass	Mixture	Sainfoin	Grass	Mixture
		Mean Square														
Grass (G)	1	1.26*	0.67*	0.33*	0.64*	1.29**	0.4*	0.64**	0.16	0.05	0.01	0.07	0.31*	0.02	0.002	0.02
Ratio (Ra)	1	0.88*	0.05	0.004	3.72***	2.34***	0.75*	3.44***	2.12*	0.27	7.3**	1.52***	0.32*	3.63***	0.68*	0.001
Root (Ro)	1	3.16***	3.11***	2.69**	3.0***	1.26**	0.87**	2.38***	3.07***	1.67***	3.56*	2.42***	2.32***	3.25***	1.76***	2.38***
Shoot (Sh)	1	0.86*	0.01	0.43*	0.42	0.06	0.99*	0.77**	0.01	0.68**	1.24	0.02	0.51*	0.49	0.001	1.0*
G×Ra	1	0.77*	0.09	0.003	0.75*	0.001	0.01	0.33*	0.01	0.003	1.46	0.03	0.08	0.53*	0.09	0.00
G×Ro	1	1.71**	0.002	0.04	0.28	0.003	0.02	0.68**	0.01	0.003	0.01	0.12	0.03	0.42	0.04	0.01
Ra×Ro	1	0.15	0.64*	0.51*	0.002	0.54*	0.35*	0.51*	1.59*	0.56*	0.22	0.56*	0.47*	0.05	0.69*	1.08***
G×Sh	1	0.09	0.09	0.21	0.24	0.003	0.01	0.003	0.1	0.06	0.54	0.44*	0.01	0.03	0.03	0.12
Ra×Sh	1	0.001	0.31	0.3	0.23	0.19	0.46*	0.001	0.02	0.22	0.13	0.01	0.24	0.01	0.09	0.85**
Ro×Sh	1	0.19	0.8*	0.01	0.001	2.1***	0.02	0.1	1.03*	0.05	0.06	0.07	0.1	0.02	0.5*	0.03
G×Ra×Ro	1	0.06	0.16	0.16	0.07	0.07	0.01	0.002	0.02	0.001	0.06	0.0	0.00	0.06	0.001	0.002
G×Ra×Sh	1	0.48	0.01	0.02	0.07	0.01	0.07	0.01	0.003	0.003	0.13	0.03	0.08	0.25	0.01	0.03
G×Ro×Sh	1	0.2	0.58	1.18	0.19	0.05	0.00	0.14	0.12	0.16	0.05	0.06	0.03	0.08	0.08	0.1
Ra×Ro×Sh	1	0.01	0.01	0.02	0.00	0.03	0.15	0.24	0.12	0.25	0.05	0.002	0.13	0.002	0.001	0.002
G×Ra×Ro×Sh	1	0.13	0.001	0.02	0.004	0.00	0.00	0.05	0.01	0.01	0.77	0.02	0.01	0.002	0.00	0.00
Residual	31	0.15	0.06	0.06	0.14	0.07	0.06	0.07	0.15	0.07	0.58	0.08	0.06	0.11	0.08	0.06

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively

Appendix 6-3 Analysis of variance of Relative Crowding Coefficient.

		1st	2nd	3rd	4th	Total
		Mean Square				
Grass (G)	1	0.01	0.55	0.36	0.05	0.04
Ratio (Ra)	1	0.11	0.4	0.06	0.13	0.02
Root (Ro)	1	3.22***	3.73***	1.6***	0.45*	3.5***
Shoot (Sh)	1	0.33*	0.24	0.1	0.36	0.22*
G×Ra	1	0.71**	0.37	0.18	0.01	0.4**
G×Ro	1	0.33*	0.01	0.002	0.03	0.08
Ra×Ro	1	0.08	0.01	0.17	0.16	0.23*
G×Sh	1	0.00	0.01	0.03	0.01	0.01
Ra×Sh	1	0.03	0.01	0.03	0.01	0.00
G×Ra×Ro	1	0.5	0.27	0.06	0.002	0.25
G×Ra×Sh	1	0.01	0.03	0.07	0.01	0.003
Residual	11	0.05	0.12	0.05	0.08	0.04

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively

Appendix 6-4 Analysis of variance of sainfoin proportion(logit), Relative Yield Total, Relative Yield of sainfoin and grasses

Source of variation	d.f	1st harvest SF prop'n	1st harvest SF RY	1st harvest Grass RY	1st harvest RYT	2nd harvest SF prop'n	2nd harvest SF RY	2nd harvest Grass RY	2nd harvest RYT	3rd harvest SF prop'n	3rd harvest SF RY	3rd harvest Grass RY	3rd harvest RYT	4th harvest SF prop'n	4th harvest SF RY	4th harvest Grass RY	4th harvest RYT	Total SF prop'n	Total SF RY	Total Grass RY	Total RYT
		Mean Square																			
Grass (G)	1	0.21	0.39**	0.64*	1.72	1.99**	0.07	13.52***	3.08	1.05***	0.04	0.63**	0.38*	0.1	0.51	0.02	0.16	0.08	0.04	0.38*	0.16
Ratio (Ra)	1	0.19	0.27*	0.00	0.29	1.83**	0.4**	0.61	3.18	2.38***	0.83***	0.45*	0.02	5.58**	2.45	0.62*	0.12	3.2***	0.71***	0.47*	0.12
Root (Ro)	1	18.2***	1.71***	4.8***	7.32***	11.47***	1.15***	9.7***	0.85	12.46***	1.37***	2.45***	4.18***	20.47***	0.67	3.26***	3.5***	11.7***	1.36***	0.82**	3.5***
Shoot (Sh)	1	7.17***	0.2*	0.25	3.07***	4.08***	0.24*	1.55**	5.23*	4.64***	0.21*	0.1	1.98***	7.35**	0.01	0.07	1.34**	2.9***	0.15*	0.09	1.34***
G×Ra	1	0.85**	0.42**	0.34	0.13	0.21	0.11	0.001	4.95*	0.07	0.09	0.02	0.001	0.53	0.84	0.03	0.17	0.43	0.21*	0.19	0.17
G×Ro	1	0.08	0.27*	0.001	0.34	0.83	0.04	4.48***	0.08	0.6**	0.32**	0.13	0.04	0.35	0.11	0.11	0.02	0.14	0.09	0.09	0.02
Ra×Ro	1	0.04	0.003	0.02	0.06	0.21	0.01	0.11	1.42	0.02	0.13*	0.15	0.01	0.24	0.05	0.004	0.01	0.78*	0.01	0.04	0.01
G×Sh	1	0.11	0.19	0.11	0.02	0.33	0.04	0.53	2.41	0.22*	0.02	0.08	0.32*	1.09	0.9	0.59*	0.01	0.004	0.05	0.01	0.01
Ra×Sh	1	0.06	0.03	0.02	0.05	0.04	0.01	0.33	0.28	0.41**	0.22*	0.16	0.01	0.72	1.24	0.18	0.02	0.85**	0.13*	0.23	0.02
Ro×Sh	1	0.13	0.04	1.18**	0.06	0.05	0.02	0.56	0.16	0.41**	0.02	0.89**	0.24	1.5	0.19	0.16	0.93**	1.14***	0.001	0.13	0.93**
G×Ra×Ro	1	0.17	0.02	0.21	0.1	0.01	0.01	0.35	3.45	0.004	0.02	0.03	0.01	0.32	0.31	0.01	0.01	0.08	0.001	0.04	0.01
G×Ra×Sh	1	0.31	0.14	0.02	0.42	0.09	0.01	0.17	0.03	0.01	0.02	0.05	0.11	0.01	0.86	0.001	0.17	0.13	0.09	0.06	0.17
G×Ro×Sh	1	0.17	0.5	1.19	3.11***	0.01	0.01	0.69	0.01	0.05	0.05	0.02	0.06	0.58	0.33	0.33	0.13	0.03	0.08	0.07	0.14
Ra×Ro×Sh	1	0.23	0.08	0.09	0.01	0.47	0.19	0.29	1.26	1.1	0.64	0.43	0.02	1.04	0.05	0.18	0.09	0.00	0.16	0.00	0.09
G×Ra×Ro×Sh	1	0.002	0.17	0.002	0.001	0.003	0.02	0.04	0.04	0.05	0.09	0.01	0.21	0.003	0.32	0.003	0.01	0.00	0.04	0.00	0.01
Residual	31	0.07	0.04	0.1	0.15	0.12	0.03	0.14	0.81	0.04	0.03	0.06	0.07	0.4	0.61	0.07	0.09	0.08	0.03	0.05	0.09

*, **, *** Significant at the 0.05, 0.01 and 0.001 level of probability respectively

Appendix 6-5 Meteorological data at Cirencester.

2006	Precipitation(mm)	Mean air temperature(°C)
Jan	20.2	3.9
Feb	32.3	2.7
Mar	79.6	4.8
Apr	26.7	8.6

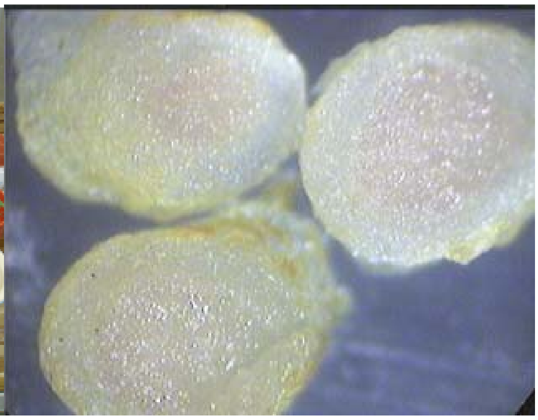
9.5 Inoculant Strains Trial of Sainfoin

Aim Evaluate five available inoculant strains.

Materials and Methods

This trial was conducted in greenhouse in Royal Agricultural College. Sainfoin cv. Common Cotswold and innoculant strains USDA3172, UMR6918, UMR6861, UMR6862, UMR6808 were used. Sainfoin seeds were inoculated and then sown into sand at 10 seeds per pot in three replicates. A control treatment was also set. Before sowing, sand was sterilized by autoclaving and seeds and pots by Sterilox. Pots were watered with deionised water when the sand was dry. Temperature was maintained at ±20°C.

Sainfoin seedlings were taken out of pots at day 42. Sand was washed away from roots. Nodules were counted and looked at effectiveness of nodulation. However, this trial failed because the controls were contaminated and it was abandoned (section 7.8, P146).



9.6 Sainfoin Inoculant Response to Nitrogen

Aim Examine sainfoin inoculant' response to different level of Nitrogen.

Materials and Methods

This trial was conducted in greenhouse in Royal Agricultural College. Sainfoin cv. Common Cotswold and innoculant strain UMR6918 was used. Sainfoin seeds were inoculated and then sown into sand at 10 seeds per pot in three replicates. Before sowing, sand was sterilized by autoclaving and seeds and pots by Sterilox. Pots were flushed with nitrogen solution in 0, 2.5, 5, 7.5, 10, 15 and 20 mg l^{-1} when the sand was dry. Nitrogen solution made from deionised water. Temperature was maintained at ±20°C.

Sainfoin seedlings were taken out of pots at day 42. Sand was washed away from roots. Nodules were counted and looked at effectiveness of nodulation. However, this trial failed because the controls were contaminated and it was abandoned (section 7.8, P 146).

9.7 Publications

9.7.1 Liu, Z. and Lane, G.P.F. (2005) The Effect of Sowing Date and Autumn Management on Sainfoin (*Onobrychis viciifolia*) Regrowth and Yield. In*, Proceedings of XX International Grassland Congress*. Ed by O'Mara, F.P. et al. Dunlin, June 2005. Wageningen Academic Publishers. 361

The Effect of Sowing Date and Autumn Management on Sainfoin *(Onobrychis viciifolia)* Regrowth and Yield

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Keywords: Sowing Date, Autumn Management, Regrowth and Yield

Introduction Sainfoin was a traditional leguminous crop in the UK. It was largely grown during the 17-19th century (Bland, 1971) due to its characteristics of palatability, non-bloating, high protein, high voluntary intake etc (Frame *et al*., 1998), but it has almost disappeared in recent years. With the rise of organic farming and the requirement for home grown protein, it seems to be the time for sainfoin to come back. The objective of this experiment is to explore the impact of sowing date and autumn management on sainfoin's growth and yield.

Materials and methods Six main treatments (sowing dates: April, May, June, July, August & September) and two sub treatments (autumn cut v non-cut) in a randomised block design with three replications on plots of 2m× 4m were established at the Royal Agricultural College, Cirencester, UK in 2003. The variety was a landrace, Cotswold Common. The seed rate was 90 kg/ha of hulled seed. Seed was broadcasted and then raked into soil up to about 0.5-1.0 cm deep and rolled. One harvest was obtained in August of 2003 and a further two in May and July of 2004. 30kg/ha of P_2O_5 were given after harvest in 2003 and 20kg/ha P_2O_5 and 30 K_2Okg/ha after first harvest and 40 K_2Okg/ha after second harvest in 2004. MCPA/MCPB was used to control broadleaved weed just at first compound leaf at trifoliate stage in establishment year and Carbetamax used to kill grass weeds and chickweed in January 2004. Autumn cuts were taken in September 2003.

Results Yields of first two harvests in 2004 are given in table1. There were significant differences between sowing dates. August and September sowing gave significantly

(P<0.001) reduced yields. Table 2 shows the dry matter yields of autumn cut v non cut treatments. There were no significant differences.

Table1 Dry Matter (t/ha) of first two harvests in 2004.

Treatment	1st harvest	2nd harvest	Total
April	9.15	2.62	11.76 a
May	9.68	2.82	12.50 a
June	8.53	2.67	11.20 a
July	8.77	2.72	11.49 a
August	3.42	2.21	5.63 b
September	2.23	1.78	4.01 b

Table 2 DM (t/ha) of Autumn cut v non-cut.

Treatment	Non-cut	Cut
April	11.76a	12.46a
May	12.50a	12.10a
June	11.20a	11.09a
July	11.49a	11.76a

Means with the same letter in the same row or column are not significantly different.

Conclusions One year's results show that sowing at any time between April and July can give similar yields in the following season. August and September sowing gave large reductions in forage yields in the following season. Autumn (September) cutting in the establishment year appeared to have little effect on yields in the following season.

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9.7.2 Liu, Z., Lane, G. P. F. and Davies, W. P. (2006) The Effects of Establishment Method on the Yield of Sainfoin (*Onobrychis viciifolia*) and Sainfoin-Grass Mixtures. In, *Proceedings of British Grassland Society 8th Research Conference*. 4th-6th September 2006. 9

THE EFFECTS OF ESTABLISHMENT METHOD ON THE YIELD OF SAINFOIN (*ONOBRYCHIS VICIIFOLIA*) AND SAINFOIN-GRASS MIXTURES

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INTRODUCTION

Sainfoin (*Onobrychis viciifolia)* is a valuable forage legume crop and was traditionally sown with meadow fescue *(Festuca pratensis)* or timothy (*Phleum pratense* L.). Today sainfoin has almost disappeared from British agriculture. In view of its valuable nutritional properties (high protein, condensed tannins and non-bloating), a few farmers still grow it, based on limited traditional experience. A study of sainfoin agronomy appeared necessary in order to preserve interest in the crop and to provide practical guidelines for farmers. The main objective of this trial was to explore the impact of direct sowing against undersowing on the establishment, production and persistence of sainfoin and sainfoin-grass mixtures. A further objective was to compare the efficacy of sainfoin mixtures with meadow fescue or tetraploid perennial ryegrass, in different proportions.

MATERIALS AND METHODS

This trial was conducted at Coates Manor Farm, Cirencester, UK on Sherbourrne series soil between 2003-2005. The design was a randomised block with split plots and three replications. Direct sowing or undersowing in spring barley constituted the main plots, and sainfoin (SF), meadow fescue (MF), and perennial ryegrass (PRG) sainfoin/meadow fescue and sainfoin/perennial ryegrass in 1/3 and 2/3 combinations made up the sub-plots. The trial was established in May 2003. 50 kg N ha^{-1} were applied to all plots in the spring of 2004 and 2005. One harvest was obtained from the direct sown treatments in 2003. Three harvests were taken in 2004 and 2005. The sainfoin variety was cv. Cotswold Common; meadow fescue was cv. Lifara and the tetraploid perennial ryegrass cv. Condesa.

RESULTS

Undersowing significantly reduced the yields of sainfoin, grasses and sainfoin-grass mixtures, compared to direct sowing over three years (Table 1). The effects of undersowing were evident in 2003 and 2004. In 2005 there was no significant difference in yield.

2/3sainfoin-1/3meadow fescue yielded more than all other treatments, (Table 1). Not surprisingly, meadow fescue and perennial ryegrass monocultures yielded significantly less than sainfoin and sainfoin-grass mixtures.

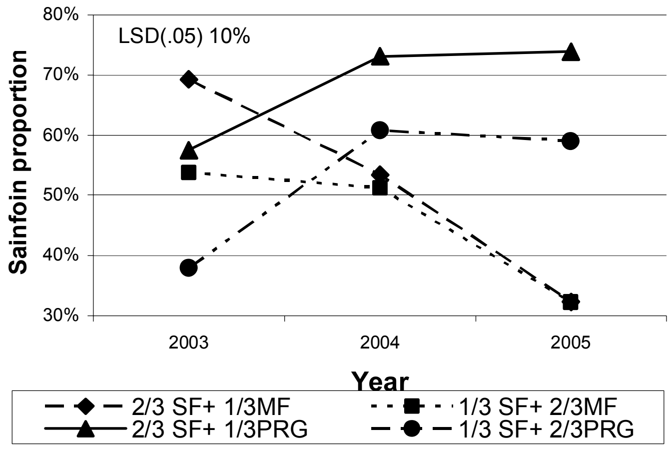
Table 1. Comparison of yields (t DM ha^{-1}) between direct sowing and undersowing, and between sainfoin/grass mixtures.

	2003 §	2004	2005	Average
Direct sowing	1.8	12.22 a	7.7 a	7.24 a
Undersowing	N.A	10.3 b	7.9 a	6.07 b
LSD (.05)		0.44		0.11
SF	2.03 a	12.23 c	8.33 b	7.53 b
MF	1.42 ab	6.58 d	6.17 c	4.72 c
PRG	1.13 b	5.45 d	3.02 d	3.2 d
2/3 SF+ 1/3MF	2.21 a	14.54 a	10.48 a	9.07 a
1/3 SF+ 2/3MF	1.74 ab	13.96 ab	9.72 b	8.47 b
2/3 SF+ 1/3PRG	2.15 a	13.52 abc	8.48 b	8.08 b
1/3 SF+ 2/3PRG	1.94 a	12.59 bc	8.55 b	7.69 b
LSD (.05)	0.81	1.5	0.88	0.58

Values within rows followed by the same letter are not significantly different.
§ Values in the establishment year are only the yields of direct sowing.

The average sainfoin content in direct sown sainfoin-meadow fescue mixtures declined significantly from 61.6% in 2003 to 32.2% in 2005 (Figure 1). Over the same period the average sainfoin content in sainfoin-perennial ryegrass mixtures increased from 44.7% in 2003 to 66.5% in 2005. Undersown mixtures showed the same trend. Undersowing had no significant effects on the sainfoin content of mixtures in any year.

Figure 1. Sainfoin proportion in direct sown mixtures over three growing seasons . LSD appropriate for within and between years.



DISCUSSION

Undersowing gave a yield disadvantage compared to direct sowing in 2003 and 2004. This was not unexpected and probably due to the competition from the barley for light, moisture and nutrients during establishment (Sheaffer *et al.*, 1988). Against this disadvantage could be set the proceeds of the spring barley crop for grain or wholecrop forage. The mixture containing 2/3 sainfoin and 1/3 meadow fescue consistently outyielded ($P<0.05$) straight sainfoin and all other mixtures. This is in agreement with Cooper's results (1972). The reason for the decline in the sainfoin content of sainfoin/meadow fescue mixtures is not clear but possibly due to the erect growth habit and early flowering of cv Lifara. Conversely, perennial ryegrass cv. Condesa, being a tetraploid, has lower tiller numbers compared to diploid varieties.

CONCLUSIONS

- Undersowing reduced the yield of sainfoin, perennial ryegrass, meadow fescue and sainfoin/grass mixtures, compared to direct sowing in the establishment and 2nd years, but had no effect in the 3rd year.
- 2/3sainfoin-1/3 meadow fescue gave a consistent yield advantage over straight sainfoin and all other sainfoin/grass mixtures, otherwise, sainfoin/grass mixtures yielded similar to sainfoin.
- Perennial ryegrass cv. Condesa was more compatible with sainfoin than meadow fescue cv. Lifara

ACKNOWLEDGEMENTS

Financial assistance from the Royal Agricultural College 100 Club, the British Grassland Society Forage Legumes Special Interest Group and Cotswold Seeds Ltd in support of this work is gratefully acknowledged.

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COMPETITION BETWEEN SAINFOIN, MEADOW FESCUE AND PERENNIAL RYEGRASS

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INTRODUCTION

Detailed research on competition between sainfoin *(Onobrychis viciifolia)* and grasses is very limited. Investigating the nature of competition between sainfoin and grass is important to optimise the performance of sainfoin/grass swards. The aim of this study was to explore the interaction between sainfoin and meadow fescue (*Festuca pratensis*), and between sainfoin and perennial ryegrass *(Lolium perenne)*, by partitioning the root and shoot systems (to achieve pure stands, root, shoot and full competition) and by varying the ratio of the grasses to sainfoin.

MATERIALS AND METHODS

This was a replacement experiment of 2^4 factorial with two replicates. The four factors were: root (±) × shoot (±) × grass (two species) × grass ratio (1/3 and 2/3). The trial was conducted in well drained plastic containers of 0.55 m × 0.36 m × 0.30 m at Coates Manor Farm, Cirencester, UK between 2004-2006. Sainfoin (SF. cv. Cotswold Common), meadow fescue (MF. cv. Rosso) and perennial ryegrass (PRG. cv. Condesa) were seeded in plastic cells in May 2004 and then transplanted to plastic containers in the field in July. Before transplanting took place the containers were subdivided with MDF (medium density fibreboard) to partition root competition, and then filled with soil. Each container was maintained at the same density of 273 plants m^{-2} (54 plants per box in six rows of nine plants). Shoot competition was achieved by opaque white plastic film erected North-South, to minimise inter-row shading. One harvest was taken in November 2004. Containers were left over the winter without film partition. Dead plants were replaced and film replaced in the following spring. Three further harvests were taken in 2005. Containers were regularly watered with mains tap water and suitable P_2O_5 and K_2O dressings applied after each harvest. ANOVA was completed using Genstat v7. Relative Crowding Coefficients (De Wit and Gourdriaan, 1974) were calculated to indicate competitive ability. Component and mixture yields and survival of individual species were also determined.

RESULTS

Grass species did not significantly affect sainfoin yield (table 1). However, an increase in the ratio of grass to sainfoin from 1/3 to 2/3 significantly ($P<0.05$) reduced the yield of sainfoin. Root competition significantly ($P<0.05$) reduced sainfoin yield but shoot competition did not.

Table 1. Annual sainfoin DM yield (g m^{-2}). Values are geometric means and Least Significant Ratio (LSR) is presented.

	MF	PRG	LSR (.05)
Grass spp.	100.4 a	105.8 a	
	1/3 grass	2/3 grass	
Ratio	144.3 a	73.6 b	1.29
	+	-	
Root	75 b	141.7 a	
Shoot	91.1 b	116.6 b	

Values within rows followed by the same letter are not significantly different.

There was no significant difference between meadow fescue and perennial ryegrass yields (Table 2). Root competition significantly ($P<0.05$) increased grass yield at the 1/3 grass: 2/3 sainfoin ratio, but had no effect at the 2/3 grass ratio. Without root competition, 2/3 grass yielded significantly ($P<0.05$) more than 1/3 grass. Full competition (+ Root + Shoot) significantly ($P<0.05$) increased grass yield compared to shoot competition only and nil competition.

Table 2. Annual grass DM yields (g m^{-2}). Values are geometric means and LSRs are presented.

	MF	PRG	LSRs (.05)
Grass	284.3 a	288.3 a	1.25
	+ Root	- Root	
1/3 grass	362.5 a	168.8 b	
2/3 grass	361.4 a	303.4 a	
	+ Root	- Root	1.37
+ Shoot	6.013 a	5.293c	
- Shoot	5.77 ab	5.551 b	

Values within rows followed by the same letter are not significantly different.

Relative Crowding Coefficient (RCC) was affected by root competition, shoot competition and grass ratios. The value of $RCC_{s\ v.\ m}$ was significantly ($P<0.05$) greater than that of $RCC_{s\ v.\ p}$ at 1/3 grass ratio, but there was no difference at the 2/3 grass ratio (Table 3). Root competition significantly ($P<0.05$) decreased the competitive ability of sainfoin and so did shoot competition.

Table 3. Relative Crowding Coefficient of sainfoin against grasses.

	$RCC_{s\ v.\ m}$	$RCC_{s\ v.\ p}$	LSD (.05)
1/3 grass	0.55 a	0.73 a	0.27
2/3 grass	0.76 a	0.42 b	0.27
	+ Root	- Root	
	0.34 b	1.15 a	0.20
RCC s v. g	+ Shoot	- Shoot	
	0.54 b	0.77 a	0.229

Values within rows followed by the same letter are not significantly different.
$RCC_{s\ v.\ m}$ refers to RCC of sainfoin against meadow fescue; $RCC_{s\ v.\ p}$ refers to sainfoin against perennial ryegrass; $RCC_{s.v.g.}$ refers to sainfoin against all grasses

DISCUSSION

Increasing grass ratios generally reduced sainfoin yield. This agrees with the results of Grieshaber-Otto (1984) who worked with *Medicago sativa*. Increasing the grass:sainfoin ratio probably resulted in increased competition for light and soil resources. Root competition reduced sainfoin yield but increased grass yield. Probably the root system of grasses occupied more space and there may have been some nitrogen transfer. Sainfoin grown with 2/3 meadow fescue was more competitive than when grown with 2/3 perennial ryegrass.

CONCLUSIONS

- Sainfoin yield was affected more by the quantity of the accompanying grasses rather than the species.
- Root competition reduced sainfoin yield but increased grass yield.
- With 2/3 meadow fescue (cv. Rossa) sainfoin was more competitive than with 2/3 perennial ryegrass (cv. Condesa).

ACKNOWLEDGEMENTS

Financial help is acknowledged from the RAC 100 Club, BGS Forage Legumes Group and Cotswold Seeds Ltd.

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