THE ROLE OF GENOTYPE X ENVIRONMENT INTERACTIONS IN ANNUAL MEDICAGO SPP.

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Scope of Study: Annual medics (Medicago spp.) are selfseeded coolseason legumes. Originating from the Mediterranean basin, they present wide ranges of genetic material and adaptation. The potential of these pasture species for forage, seed production, and reseeding was investigated under the U.S. Southern Great Plains continental climate and in the medics' native habitat, Morocco. Eleven species of medics (23 accessions), two perennial alfalfas (M. sativa L.), and four clovers (Trifolium spp.) were included in the tests. The importance of genotype X environment (G X E) interactions in these species was evaluated under clean-tilled field conditions with a wide spectrum of environments. Biomass partitioning and root development were examined in these forage crops.

Findings and Conclusions: In the Southern Great Plains, forage and seed production of medics should come from spring planting (even though better establishment was obtained with fall plantings) until higher tolerance to cold than that found in the genotypes evaluated is developed. Production parameters were significantly influenced by genetic makeup and G X E interactions. Spring sown medics in Oklahoma or fall sown medics in Morocco produced forage yields equal to or higher than alfalfa and clovers. Forage of these species was good quality (high crude protein and low neutral and acid detergent fiber concentrations) at flowering. Establishment success positively influenced forage yield from spring plantings in Oklahoma. Certain species, e.g., M. scutellata, were able to produce an adequate soil seed bank in Oklahoma, and demonstrated good forage and seed potential. However, the hard seed of these species did not germinate well in the subsequent growing season. All species were suitable for forage and commercial seed production in Morocco when rainfall was sufficient. Small-seeded species, e.g., M. truncatula, were better seed producers than large seeded ones. Significant associations existed between medic seed characteristics. Annual medics did not partition their biomass among plant organs similarly over time. However, they developed similar rooting systems.

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PREFACE

Annual medics (<u>Medicago</u> spp.), a complex genetic material, present an enigma to the research in any of its aspects; i.e., phytosociology and gene dynamics. Throughout this study the author has been challenged with the diversity offered by, not only these species, but also by their associated biotic and abiotic factors that when gathered form an ecosystem in which genetic erosion might take place as a debt to pay for advanced technology. With limited knowledge, he recognized that much work must be accomplished to profit from the inestimable value of these forage species as has been done, still little, to their related perennials (alfalfa).

This contribution, though small, convinced the author that acclimatation in different ecosystems other than the native, which might appear obvious to some people, is possible. However, this could not have happened without appropriate guidance and continuous support. Dr. J. L. Caddel, the major adviser, provided continuous advice, help, and constructive critisism that made the author so grateful. Dr. L. M. Rommann provided all the indispensible moral support and therefore, he is urged to accept full gratitude and respect. The author wishes to express his appreciation to Drs. G. W. Horn, E. G. Krenzer, and R. L. Westerman for their valuable advice and time while serving as advisory committee members, and to Drs. E. L. Smith, and S. Christiansen for their important counsels. Dr. C. M. Francis, from Western Australia, was an excellent source of information and reviewed the draft. Thus,

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This work is dedicated to the author's parents, wife Najia, and daughters Nora and Samah for their enormous sacrifice and forgivness while he was neglecting their rights in the course of this graduate study.

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CHAPTER I

GENERAL INTRODUCTION

Oklahoma, located in the Southern Great Plains of the U.S., has several million hectares of rangeland which could be managed as a sustainable perennial grass production system. Forage production in these areas is relatively limited by low quality as biomass consists primarily of grasses. On the other hand in wheat growing areas, farmers realize currently small profits from grain production due to high costs of energy, nitrogen fertilizer, and low wheat prices.

To a certain extent, similar problems are encountered in North Africa, but in different ways. Rangeland in Morocco occupies 70% of the total land but is characterized as having very low productivity. Total digestible nutrients produced are about 120 forage units (equivalent to 120 kg of a barley with 90% dry matter) per hectare in these lands according to Moroccan Ministry of Agriculture reports. Temporary pasture, used on a rotational basis with cereal production and frequently called "weedy fallow", is botanically composed of Compositae and Crussiferae species, and these plants dominate the presence of potentially more benifecial native legumes. These unimproved pastures constitute about 27% of cultivated land. Crop residues, e.g., straw and stubble, are also fed to animals with minor, if any, protein supplementation. Underutilization of the potential forage and cereal production resources has created a situation where animal and cereal

production does not meet the nation's needs. Cereals for grain produce a relatively low yield, about 1000 kg/ha, and are associated with ill-devised cropping systems, low soil fertility, and frequent droughts.

Selected N-fixing legumes may improve quality and quantity of forage in rangeland. They may transform poor temporary pastures into productive forage areas and may ameliorate soil nitrogen levels, and improve grain yields. Annual legumes may also be grown with cereals. The legumes would be grazed after the cereal crop is removed or grazed together as emergency measures during droughts. Consequently, the integrated farming systems based on animals and crops, principally cereals, which are extensively used in North Africa, may be improved. Animal production can be stabilized and augmented, and farmers can be more self-sufficient and may have better returns.

Arrowleaf clover (<u>Trifolium vesiculosum</u> Savi) is an important pasture legume in Oklahoma and covers about one million hectares (L.M., Rommann, Forage Extension Specialist, Oklahoma State University, Stillwater, OK, personal communication). However, it is frequently difficult to establish. Sowing annual legumes for grazing as temporary pasture has only recently begun in Morocco, and it is yet to be initiated in rangeland.

Annual medics (<u>Medicago</u> spp.) and clovers (<u>Trifolium</u> spp.) may be a solution in both rangeland and cereal zones. Most medics and clovers are cool-season plants, nitrogen-fixing, and selfseeding. Their high level of prolificity, hardseededness, and tolerance to grazing pressure make their use economical and, potentially, widely adopted.

Primary objectives of this research were to study the effects of environmental factors and environment X genotype interactions on

phenology, survival, and forage and seed production of 11 annual medic species. They were grown in non-irrigated, clean-tilled conditions under continental and Mediterranean climates. Biomass partitioning and rooting were also evaluated in these species. A better understanding of these factors will allow forage agronomists and coworkers to be in a better position to study adaptability of annual legume species for Oklahoma and Morocco. Hopefully application of such findings will lead to improved temporary pastures, higher grain yields, and more productive rangelands.

This report is divided into three related subjects for convenience. Literature review, materials and methods, and results are presented separately for each subject. Extensive literature citations are listed so they can be used by coworkers, particularly in North Africa where medics are being given serious attention.

CHAPTER II

GENOTYPE X ENVIRONMENT INTERACTIONS AND STABILITY PARAMETERS IN ANNUAL MEDICAGO SPP.

Abstract

Medicago spp. for emergence, cold survival, forage yield, and quality. The study included fall and spring plantings under a continental climate of the Southern Great Plains of the U.S. and fall plantings under a Mediterranean climate of Morocco. Seven locations with different soil types, precipitations, and temperature regimes, when combined with years and planting seasons, constituted 19 environments for the test. Soil reaction and 'nitrogen supply' were also investigated in the U.S. Plant materials included 23 annual Medicago spp. accessions, two selected grazing strains of perennial alfalfa (M. sativa L.), and four clovers (Trifolium spp.). Two hundred viable seeds (scarified) were planted in single rows 2 m long and 1 m apart. Each experiment was a randomized complete block with four replications. The study was conducted between 1983 and 1987 under clean-tilled field conditions.

Emergence percentage differed among environments for most entries besides the existance of genotype x environment (G X E) interactions. Differences observed among genotypes in emergence and seed size indicate that the same seeding rate on all annual Medicago spp. will result in

different stand densities.

Cold survival was affected by G X E interactions in certain situations when snow covered medics in certain environments. Annual Medicago species were much more sensitive to cold than alfalfas and clovers. However, M. arabica, M. blancheana, M. minima, and M. polymorpha possessed some tolerance when they were covered with snow. Winter kill was the limiting factor for fall sown annual medic production in a continental climate.

Forage yield was not related to nitrogen supply source. Fall planting during two years in Oklahoma resulted in winter kill of all annual Medicago except in the first year when snow covered the experiment at Stillwater, where M. polymorpha, M. arabica, and M. minima produced 21, 13, and 2 g/m of dry matter (DM), respectively, compared to 98 g DM/m obtained in M. sativa and 81 g DM/m in T. vesiculosum. Genotype, environment, and G X E interactions had significant effects on yield. The best producing genotypes in spring were M. polymorpha (109 g/m) at Stillwater, M. scutellata Sava (72 g/m) at Perkins and Haskell (125 g/m), M. truncatula Borung at Sidi el Aydi (285 g/m), M. truncatula Paraggio (329 g/m) at Jemaa Shaim, and M. scutellata Robinson (606 g/m) at Tessaout. Forage yield was dependant on establishment success in Oklahoma with spring planting but not in Morocco.

Forage quality, estimated from crude protein (CP), neutral detergent fiber (NDF), and acid detergent fiber (ADF) concentrations, and leaf:stem ratio, was generally high in annual $\underline{\text{Medicago}}$ and comparable to that of $\underline{\text{M}}$. $\underline{\text{sativa}}$ and $\underline{\text{Trifolium}}$ spp. at flowering. Genotype X environment interactions were present only for CP. Mean CP

was 21.7 and 21.2%, respectively at Perkins and Tessaout with no difference among genotypes. Entries which contained the highest CP content at Stillwater, Haskell, Sidi el Aydi, and Jemaa Shaim were M. minima (24.7%), M. rugosa Pargosa (23.4%), M. sativa (27.4%), and M. truncatula Jemalong (25.4%), respectively. Mean NDF was 33.4% in Oklahoma. M. truncatula Cyprus contained the highest NDF with 36.7% comparatively to 27.3% in M. sativa in Morocco. Mean ADF was 22.8% with little differences among genotypes. M. tornata developed the highest leaf:stem ratio with 3:1 in Morocco.

Key words: Trifolium, annual medic, alfalfa, clover, emergence, cold, forage yield, nutritive value, Morocco, Oklahoma.

Literature Review

Introduction

The genus <u>Medicago</u> consists of one shrub, 21 herbaceous perennials, and 34 annual species (62). The annual species are known as medics or "burr clovers", and originated in the Mediterranean region (31, 53). Their taxonomy is complex and a species may frequently have one or more synonyms (42). Reported longevity of these species varies according to different authors. Spotted burr clover (<u>M</u>. <u>arabica</u> (L.) Huds.) and black medic (<u>M</u>. <u>lupulina</u> L.) were considered annuals by Andrew et al. (5) and Heyn (53); however, Sinskaya (80) reported they could be biennial or perennial under some conditions. Many intermediate forms between annual medic species occur naturally and demonstrate the presence of interspecific crossing as in the case between strand medic (<u>M</u>. <u>littoralis</u> Rhode), barrel medic (<u>M</u>. <u>truncatula</u> Gaertn.), and disk medic (<u>M</u>. <u>tornata</u> (L.) Mill.) (43).

Medics were introduced into Australia as contaminating weed seed in the 19th century (18). Due to the similarity of Australian ecosystems to their native habitat, annual medics, along with subclover (<u>Trifolium subterraneum</u> L.), have become indispensable components of the major Australian "ley-farming" system. Wolfe (91) defines this system to be the succession in years of a cereal crop after a self-regenerating forage legume. Medics may be interseeded with grasses to improve forage quality by increasing crude protein content (66).

Genetic material has been exchanged between Mediterranean countries and Australia (48). Selected annual medic cultivars from Australia, where most cultivar development and medic management reasearch has been

conducted, are now being reintroduced for extensive use in North Africa (5, 9, 47) and the Middle East (25, 74, 90).

Annual Medic Ecology

Ecology of annual medics as a group is of important interest. They are adapted in many environments (27) and are encountered in most bioclimatic stages from the humid to the Saharian (61, 69). Francis (43) collected \underline{M} . $\underline{littoralis}$ in Libyan areas of average annual rainfall as low as 100 mm and \underline{M} . $\underline{laciniata}$ (L.) Willd. in regions with 50 mm or less. Distribution of species depends, in addition to rainfall, on other determining factors such as altitude, soil texture, pH, and salinity (1).

In the description of registered cultivars in Australia, Barnard (11) and Mackay (64) reported that some medic species such as M. scutellata Mill. and M. rugosa Desr. are well adapted to clay soils, while others like M. littoralis and M. tornata grow on loamy and sandy soils. However, some species such as M. truncatula and M. polymorpha L. are adapted over a wide range of soils. The presence of many ecotypes within each species enables it to occupy a large spectrum of environments according to Abdelguerfi (1).

Andrew (4) reported that dry matter of annual medic production often diminished with increased soil acidity. However, M. littoralis and M. tornata were found to be somewhat tolerant to acid soils as opposed to M. rugosa cv. 'Paragosa', which has preference for alkaline soils (64). Ewing (41) recently discovered some lines of M. murex Willd. growing well in sandy soils at pH 5.0 in Sardinia, Italy.

Cultural Practices in Annual Medics

When medics are not sown with wheat (<u>Triticum aestivum L.</u>), Bakhtri (10) indicated that optimal medic seeding rates are 10 kg/ha. However, Cocks (25) demonstrated that dry matter increased with increased density. A seeding rate of 50 kg/ha produced the highest forage yield in three annual medics, <u>M. rigidula L., M. noeana L., and M. truncatula.</u> He also mentioned that species x seeding rate interactions were present.

Seeding depth was investigated by Derkaoui (36) on a clay soil. He found that optimal seeding depth was 3-4 cm with little effects of seed size on seedling density. However, he also found that plants of M. scutellata cv. 'Robinson' and M. truncatula cv. 'Jemalong' emerged from depths of 10 cm. Similar results were obtained in Syria (ICARDA research, unpublished data).

Annual medics require more P, K, and Ca than subclover (47). Rudd (76) indicated that pasture responses to superphosphate applications were unlikely when soil phosphorus exceeded 30 ppm by the Bray and Kurtz No. 1 extraction procedure. His results showed that optimal P rates were 32-38 kg/ha in sandy soils and higher in heavier-textured soils. These phosphorus values appeared to be lower than those reported by Dahmane and Graham (33). Excess aluminum reduced plant growth and decreased concentration of P and Ca in annual medics and perennial alfalfa (M. sativa L.) (5).

Burr medic (M. polymorpha), arrowleaf clover (\underline{T} . vesiculosum Savi), and subclover were reported to require 500 degree-days above the base temperature (10°C) to produce 6000 kg/ha of dry matter from fall planting in New Zealand (56). Clarkson and Russell (21) found that

flower initiation in medics was accelerated, except in \underline{M} . scutellata, by a long photoperiod (>= 18 hours) or vernalization at 1oC over three weeks. Therefore, flower initiation was reached more rapidly for winter than for summer plantings in Australia. However, they indicated from another study (23) that flowering stage was hastened by mean daily temperatures above 8.5oC but less than 26.5oC. It was also reported that fall planting of alfalfa resulted in higher number of seedlings/m2 than spring planting in Oklahoma (39).

Rhizobium Compatibility and Nitrogen Fixation in Medics

Although effective nitrogen-fixing bacteria (Rhizobium meliloti L.) for annual medics are well distributed throughout many soils, some medics have specific requirements and need artificial inoculation with proper Rhizobium when sown in new environments (17). Artificial inoculation is necessary when Rhizobium is absent in soils (8), and when competition exists between effective and non-effective strains as reported by Pinto and Vincent (72) on M. truncatula. Specificity of symbiosis between host and bacterium was substanciated with the findings of Brockwell (16). Cocks (25) noted that inoculation expanded adaptability and stability of medic commercial cultivars.

Andrew (4) demonstrated that strains of <u>Rhizobium</u> which nodulate medics were extremely sensitive to pH in pure culture, and maximum nodulation occurred at pH 6.0. He also reported <u>R</u>. <u>meliloti</u> tolerated acidity less than did <u>R</u>. <u>trifolii</u> L. However, Howieson and Ewing (55) collected some strains of <u>R</u>. <u>meliloti</u> in Sardinia, Italy, which were more saprophitically competitive and markedly superior in colonizing an

acid loamy sand (pH 5.0) than the Australian commercial inocula for commercial medic cultivars. Nodulation was better established with these strains on \underline{M} . polymorpha and \underline{M} . murex than on \underline{M} . littoralis, \underline{M} . truncatula, and \underline{M} . tornata.

The amount of nitrogen fixed is influenced by plant species and growing conditions. Smith and Baltensperger (81) found M. lupulina and M. truncatula fixed more nitrogen than did M. polymorpha. Hopmans et al. (54) indicated that, under controlled environments and with acetylene reduction techniques, the ratio of moles of N fixed to moles acetylene reduced was similar for M. truncatula and T. subterraneum with a 1:3 value. Perennial M. falcata L. was more efficient in reducing acetylene than annual medics according to Rumbaugh and Johnson (77).

Residual nitrogen after medic depends on crop management, and particularly stocking rate (27). Cocks (26) reported that over 300 kg/ha/year of nitrogen was fixed under field conditions in Syria, with $\underline{\mathsf{M}}$. rigidula fixing more nitrogen than $\underline{\mathsf{M}}$. truncatula or $\underline{\mathsf{M}}$. rotata $\underline{\mathsf{L}}$. Due to residual nitrogen from medics, total dry matter production, grain yield, and nitrogen content of wheat were all enhanced according to Scott (79).

Frost Tolerance in Annual Medics

Hanson and Barnes (51) reported most of the annual medic species lack winterhardiness in comparison to perennial alfalfa. However, large differences in frost tolerance among medics species were observed by Cocks (26) and Francis (44). They also reported that \underline{M} . $\underline{rigidula}$, \underline{M} . \underline{rotata} , and \underline{M} . \underline{noeana} were more frost-tolerant than any Australian cultivar when temperatures reached 0 \hat{C} .

Variety x location interactions were present for winterhardiness according to Dexter (37) in several crops. He also reported that polyploid varieties were more winter hardy than their related diploids, and upright cultivars were generally less tolerant to frost than those with a more branching habit. Cold damage was shown by Walton (88) to be amplified by frost occurrence, therefore, a tolerant cultivar may not be so when temperatures become unusually lower than expected.

Pests and Diseases on Medics

M. minima (L.) Mill., M. polymorpha, and M. truncatula were all suitable hosts for both blue-green aphid (Acyrthosiphon kondoi Shinji) and spotted alfalfa aphid (Therioaphis maculata Buckton), and in some cases forage production was curtailed by 50% due to damage caused by these insects (63). Pea aphid (A. pisum Harris) (60), clover root curculio (Sitonia hispidula Fabr.) (27), alfalfa weavil (Hypera postica Gyllenhal) (12), redlegged earthmite (Halotydeus destructor Tucker) (60), and lucerne flea (Sminthurus viridis L.) (19) were reported to attack medics and reduce their production and persistence. Bretag and Kollmorgen (15) found some Fusarium spp. to cause root rot in medic species, which might be controlled by soil incorporation of metalaxyl. Variability in level of pest resistance strongly indicates that cultivar improvement can be made. New cultivars, highly resistant to aphids, were recently released and are currently in use, e.g., M. truncatula cv. 'Paraggio' and 'Parabinga' (32).

Forage Production in Annual Medics

Data on forage production of medic species vary in the literature. Aitken (3) found that annual medics were similar to subclover in growth habit, rate of development, and forage production. Radwan et al. (74), when evaluating Australian commercial cultivars, observed large variation in plant vigor, date of flowering, nodulation, and forage production. Some cultivars produced less than local M. polymorpha which produced 4,400 kg/ha, compared to 2,500 kg/ha from M. truncatula cv. Jemalong and M. scutellata cv. 'Snail'. These findings concur with other data gathered in Syria, where M. rigidula outyielded Jemalong (25). Crawford (30) obtained a maximum of 28 kg/ha/day of dry matter over 98 days in South Australia from M. arabica, M. polymorpha, and M. truncatula.

Severe water stress (22) as well as waterlogging (45) impairs forage yield in annual medics. Large differences in tolerance to waterlogging were noted among species, and in some instances among lines within the same species. The most tolerant to waterlogging were \underline{M} . $\underline{polymorpha}$, \underline{M} . $\underline{arabica}$, and \underline{M} . $\underline{intertexta}$ L., the least tolerant were \underline{M} . $\underline{scutellata}$, \underline{M} . \underline{minima} , and \underline{M} . $\underline{tornata}$ (45).

Rumbaugh and Johnson (77) investigated the feasibility of using annual medics as reseeding pasture legumes at two sites in northern Utah. Their results from 584 accessions representing 34 species planted in spring showed that most of the species were easily established and grew more rapidly than did perennial \underline{M} . $\underline{falcata}$. However, none of the 34 species reproduced to initiate a soil seed bank at the drier location of the two sites. Medicago laciniata (L.) Mill., M. lupulina, M.

<u>murex</u>, and \underline{M} . <u>muricoleptis</u> Tin. excelled in the number of seedlings produced by natural reseeding in the fall of the first year at their second location. However, only \underline{M} . <u>lupulina</u> produced abundant seedlings during the second year following seeding. This species also had superior ground cover.

Medics appear to be best adapted to semi-arid regions with mild temperatures and adequate winter precipitiation (18). Under the continental climate of Oklahoma with cold winters, Denman et al. (34) and Kneebone (59) reported that annual medic and clover species have fair to poor performance compared to the excellent performance of alfalfa and vetch (Vicia dasycarpa Tonore). M. arabica, M. lupulina, M. minima, M. orbicularis L., and M. polymorpha occur naturally in the flora of Oklahoma (89). They were probably introduced as forages or weeds and have spread into restricted micro-environments.

Association between forage and seed yields have been studied for medics by Ceccarelli and Somaroo (20). They indicated that dry matter and seed yield behave to a large extent as independent agronomic variables in \underline{M} . aculeata \underline{L} . and \underline{M} . rigidula, and that development of cultivars combining high seed yield and adequate dry matter should be possible. Crawford (29) noted that positive associations between forage production and seed yield were present in a study of 206 lines of \underline{M} . truncatula collected in 15 countries.

Forage Quality of Annual Medics

Forage quality of annual medics has been investigated in only a limited number of cases. Radcliffe and Cochrane (73) found dry matter digestibility of barrel medic (\underline{M} . $\underline{truncatula}$) declined by 10-12

percentage points from the vegetative stage to flowering. It was still 60% digestible at flowering; however, it decreased to 30% at senescence. They also indicated that barrel medic digestibility was comparable to that of subclover. Snail medic (\underline{M} . scutellata) had higher digestibility than barrel medic throughout the growth cycle. The digestibility of snail medic was 78% and 50% at the vegetative stage and at the end of the maturation stage, respectively (58).

Haying of annual medics is sometimes practiced instead of grazing. Barrel medic hay when harvested at flowering stage was similar to a good quality alfalfa hay with 65% digestible dry matter (35). Vercoe and Pearce (87) estimated medic hay intake by mature ewes to be 604 ± 83 g of organic matter/day. The most suitable stage for hay cutting appeared to be flowering in terms of nutritive efficiency that maximizes animal production per unit area(7)

Annual medics are rich in crude protein (CP). The content of CP was reported to decrease from the vegetative stage to the end of the growth cycle. Crude protein concentration of \underline{M} . scutellata hay was 22% according to Jones and McLeod (58), compared to \underline{M} . truncatula hay (17%) harvested at the same stage (35, 73). Mohamed and Kovacs (67) studied 28 ecotypes and 12 cultivars of \underline{M} edicago and showed that annual species contain more crude protein and saponin but less methionine than perennials.

Neutral detergent fiber (NDF), acid detergent fiber (ADF), and lignin contents were all negatively correlated with crude protein content and to in vitro dry matter digestibility (IVDMD) in annual medic species (7). Correlation coefficients between CP and NDF, and between CP and ADF were estimated to be -0.94 and -0.93, respectively. On the

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other hand, CP and digestibility appeared to be positively correlated (r=0.74), as was the reverse between intake and NDF (r=-0.74) (7). Belyea and Ricketts (14) reported that NDF and ADF were, respectively, 40% and 30% of total dry matter in alfalfa at 1/10 bloom stage, and 60% and 45% at full bloom. They predicted the corresponding net energy at these stages to be 1.43 and 0.99 MCal/kg DM. This suggests that NDF, ADF, and CP can be used to differentiate quality among medic species. Leaf:stem ratio is a criterion reported by Barnes and Gordon (13) to be a good indicator of alfalfa forage quality. This ratio was 2:1 and 1:2 at the vegetative and early seed stages, respectively. Higher leaf:stem ratio would indicate that digestibility would be greater because leaves are nore nutritious and more rapidly digested than stems.

Genotype X Environment Interactions

Studies to evaluate genotype X environment (G X E) interactions have been conducted for many crops, and particularly forage species.

"The significance of G X E lies in their impact on reliability of estimates for variance components, especially genetic variance as the basis for predicting genetic improvement in selection programs" (28). Therefore, G X E may be viewed as part of the source of random error of estimates, and breeding for improved yield over a broad spectrum of environments would have large G X E effects. Moll et al. (68) demonstrated that G X E in corn (Zea mays L.) grain yield, were largely accounted for by the lack of responsiveness to environmental variations and that improved populations were more responsive than original populations. Genotype X environment interactions vary from one trait to the other of the same plant, as it was so in sugarcane (Saccarum

officinarum L.) (83).

Several models have been presented to estimate G X E and stability analysis. Eberhart and Russell (38) and Perkins and Jinks (71) used linear regression to evaluate the performance and stability of cultivars across environments. Regression coefficient (bi) and residuals (s2di) were used as criteria for average responsiveness to environmental effects and stability, respectively (38), and by definition a stable variety would have b = 1 and $s^2d = 0$. Tan et al. (85) found that G X E interactions could be explained by the differences between linear responses estimated through regression in smooth bromegrass (Bromus inermis Leyss). However, use of linear regression has proved to present some limitations in certain instances, because it is based on mean values which quantify environment effects. Therefore, Gray (50) and Nguyen et al. (70) used coefficient of determination between the performance of individual genotypes and environment index, and ecovalence as additional stability indices. Values of regression coefficients higher than $1.0 \; (b > 1.0)$ for some cultivars of alfalfa corresponded to either better or lower yield than average response according to Taliafero et al. (84).

It appears from this review that much work on annual medics needs to be done, particularly under conditions other than Australia. Even there, genotype X environment interaction effects on winter and summer survival, phenology, and forage yield components of annual medics have not been clarified. Research on these pasture species in North Africa has been restricted to reporting their occurrence; however, research concerning genetic and agronomic evaluations has been initiated. Also, data are not currently available to explain the performance of these

species obtained under a continental climate.

The objectives of the present study were to evaluate the effects of genotype, G X E interactions, and environment on the following agronomic characteristics of annual medics: emergence, cold survival and growth habit, regrowth capacity, life cycle, dry matter production and forage chemical components, and to estimate stability parameters of these traits. A secondary objective was to compare the performance of medics to that of alfalfa and certain clovers, when grown under diverse environments.

Materials and Methods

Site Descriptions

The study was conducted in seven locations over four years, two in Oklahoma, U.S., and two in Morocco between 1983 and 1987. Oklahoma is dominated by a continental climate, while Morocco is under Mediterranean climate, the natural habitat of medics. Table 1 presents the distribution of average monthly temperature and rainfall at two sites: Stillwater in Oklahoma and Settat in Morocco. Each site, based on temperature and rainfall (2, 78), represents the corresponding macroclimate (40).

Climatic differences between Stillwater and Settat are large. Stillwater is much colder in winter than Settat. Temperatures attained -24°C at Stillwater but only -2.5°C at Settat in 1983. Rainfall is twice as high in Stillwater as in Settat, in addition to the near absence of rain from May to October in Settat. Most precipitation in Stillwater occurs during spring and summer, with some snow during fall and winter, while precipitation is concentrated as rain during winter months in Settat. Longevity is limited in Settat by the low amount of available water and drought in late spring and summer.

Ecological charactersitics of each location are presented in Table 2 (2, 82), from which it may be deduced that the study was conducted over a broad spectrum of environments. Experiments were repeated during two consecutive years (both fall and spring) at Haskell and Woodward, OK. Stillwater and Perkins, OK. were sites used during single years. The Moroccan sites of Sidi el Aydi and Tessaout were used

during two years, but Jemaa Shaim was used for only one year.

Planting Date

Fall and spring plantings were evaluated in Oklahoma since cold may be a limiting factor to growing medics. Growth conditions, particularly temperature and rainfall, are different between these two planting dates as is shown in Table 1. Plants sown in spring were subjected to warm temperatures, ample precipitation, and long daylengths without the cold temperature exposure through winter from fall plantings. Fall planting was the only planting time in Morocco since the lack of rainfall in summer months would certainly limit medic growth. All fall plantings were made in late October to early November, while spring plantings were established in late March.

Soil Reaction

The presence of acid soils in Oklahoma presented an opportunity to study the effects of these soils on medic adaptability. Two sites differing in pH were chosen at Haskell in the first year. One site had a pH of 4.7, while the other was limed to bring pH to the 6.8-7.0 range for both fall and spring plantings. All other sites in Oklahoma were limed to bring pH to the above range. Lime was not applied in Moroccan sites where pH was between 7.2 and 8.2.

Combination of site, year, planting date, and pH resulted in 19 environments (Table 3). Herein, the definition of an environment corresponds to that of Comstock and Moll (28) for macro-environment. Certain agronomic characters were not evaluated in all of these environments due to experiment termination prior to the measurement of

these characteristics. The number of environments with their designations will be presented separately for each character.

Genetic Material

Twenty-three different genotypes representing 11 annual medic species, two alfalfas, and four clover species were included in the study (Table 4). Because seed quantity was small, it was necessary to interchange certain entries between experiments so that same genetic material would be used across years. Four medic species were in very limited amounts and were studied separately at Stillwater for one year with both fall and spring plantings. The words genotype, accession, entry, and strain are used interchangeably in this context since commercial cultivars, breeding lines, and ecotypes were included.

Experimental Design

In the first year in Oklahoma, each experiment, except those conducted on the four medic species at Stillwater, was a split-plot arranged in a randomized complete block with four replications. The main plot factor (nitrogen supply) had three levels:

- I. No <u>Rhizobium</u> sp. inoculation of seed and no nitrogen fertilizer application. This level represented the control and helped determine if naturally-occurring soil <u>Rhizobia</u> effectively nodulate the species under investigation.
- II. Artificial <u>Rhizobium</u> inoculation of each seedlot without nitrogen fertilizer application. This was used to indicate whether inoculation was more beneficial than soil Rhizobia.
- III. No artificial inoculation, but nitrogen was applied at the rate

of 40 kg N/ha. This level was applied to avoid nitrogen deficiency if soil bacteria and artifical inoculation were ineffective. The rate was chosen according to Foury (42), who reported that 40 kg N/ha is adequate for alfalfa during the establishment year when nitrogen fixation is not at an optimal level. The fertilizer was applied at planting.

There were no agronomically important differences among 'nitrogen supply' levels at any of the three locations in fall or spring plantings of the first year. Consequently, in the subsequent years, each location had a randomized complete block design with four replications. The same design was adopted for the experiments that evaluated species in small quantity at Stillwater.

Conduct of Experiments

Seeds were tested for germination and entries possessing a high proportion of hard seed were mechanically scarified. Seeds were inoculated with Rhizobium bacteria provided by the Nitragin Company, Inc., Wisconsin. Annual medics received 'Medicago Spec. 1' inoculum; while alfalfa, crimson (T. incarnatum L.), red (T. pratense L.), subterranean, and arrowleaf clovers were inoculated with inocula 'A', 'R', 'B', 'WR', and 'O', respectively.

Seeds were hand-sown at the rate of 100 viable seeds/linear meter. Entries were sown in single rows 2 m long and 1 m apart (the experimental unit). Phosphorus was applied at the rate of 65 kg P_2O_5/ha . Soil tests indicated that K was sufficient in all sites except at Woodward, OK. This site was adjusted according to soil test recommendations for alfalfa (57). In experiments where the 'nitrogen

supply' factor was not investigated, 40 kg N/ha were applied at sowing in addition to commercial inoculation. All experiments were conducted under non-irrigated, clean-tilled conditions except at the Tessaout site which received two irrigations by flooding to overcome drought; one after planting and the second at the vegetative stage in fall, 1985. All plots were hand-weeded at least twice to avoid weed competition and were terminated when all annual medics died.

Observations and Measurements

Emerged seedlings were counted 30 days after planting. Percent of emergence was calculated on the basis of 200 seeds/plot. Cold damage was recorded in January for fall plantings in Oklahoma by grading rows from one to nine. Grade 1 represented a row without visual injury, while grade 9 meant that all leaves and stems were apparently dead. After the winter months, plants that recovered were observed. Growth habit was rated during the seedling count. Grade 1 corresponded to prostrate, grade 2 to semi-upright, and grade 3 to decumbent.

Forage was hand-harvested when in each experiment half of the accessions were at the flowering stage. The species did not reach the flowering at the same time. Green forage yield was weighed and dry matter determination samples of 500 g wet weight were taken and dried at 70°C until dry. Certain accessions were harvested twice in some experiments. Therefore, forage yield reported here includes the total dry yield produced. Leaf:stem ratio was measured on a dry weight basis after hand separation of leaves and stems in Moroccan experiments. Chemical analysis for crude protein, NDF, and ADF, were conducted on the first growth cycle according to standard methods of analysis (75, 86).

Statistical Analyses

Responses within each experiment were analyzed with analysis of variance for split-plot and randomized complete block designs (24). Gabriel (46) and Harvey (52) demonstrated that least square method and regular analysis of variance can be applied on qualitative characters. Mean comparisons among treatments were evaluated with the protected least significant difference (LSD) with P = 0.05. Combined analyses were conducted by using a "mixed" model; environments were random and plant accessions were fixed. Homogeneity of variances was tested with Bartlett's test (24, 49) and environments with similar variances were grouped to calculate combined analyses of variance from which environmental, genotypic, and G X E effects were examined. In the presence of G X E effects, data are presented for all genotypes in each environment because mean values over environments will not show changes in ranking genotypes from one environment to another.

Simple correlations and linear regression coefficients were calculated for forage dry matter yield (g/m) and emergence (plants/linear m) to determine if establishment success was a predictor for performance. All coefficients were calculated using the mean over replications in each environment or species.

Simple correlations were calculated among forage yield and forage quality estimators to detect any association existing between pairs of these parameters. Correlations were based on the means of replications for a genotype in an environment.

Stability analyses of genotypes were studied by regression methods for emergence percentage as outlined by Eberhart and Russell (38). An

environment index (I) was calculated by taking the mean of all cultivars grown at the ith environment and subtracting from this the mean of all genotypes included in the combined analysis in all environments. regression of index I, y = a + b I, on the mean of the parameter was obtained for each genotype. Regression coefficient (bi) and mean square for deviation from regression (sdi²) were calculated. Therefore, regression coefficient measures the relative increase in the response by increasing environment index. When a genotype has a b < 1, its responsiveness to environmental changes is below the average. On the other hand, a genotype with b = 1 represents average responsiveness to environmental differences, while this responsiveness becomes greater than the average if the genotype has a b > 1. Mean square for deviation from regression measures how well predicted response agrees with observed response and includes G X E interactions (38). Stability analysis was not performed for agronomic parameters other than emergence because of the limited number of environments where these traits were measured.

Results and Discussion

Emergence

Individual Experiment Analyses

Emergence is a primary component of forage yield of annual medics (90). Stand establishment of a medic pasture greatly depends on emergence. The analysis of variance for this parameter in each experiment is presented in Table 5. Replication effects did not differ significantly (P > 0.05) within any environment except at Sidi el Aydi and Tessaout during the 1985 plantings in Morocco. At Sidi el Aydi soil was heterogeneous and at Tessaout differences among replications were probably due to irrigation by flooding to allow germination. Irrigation was not uniform but followed a topographical gradient which corresponded to replication effects.

In Oklahoma, during the first year when the factor 'nitrogen supply' was included as a main plot treatment, effects of levels of this factor on percent emergence were not different (P > 0.05) (Table 5) in any environment except at Woodward and in the limed site at Haskell in the 1983 fall plantings. Differences were small though significant. At Woodward there was a slight increase in emergence in plots receiving Rhizobium inoculation compared to that obtained in plots fertilized with nitrogen (40% vs. 32% with LSD (P = 0.05) = 5.5%. At Haskell there was inconsistency in the response of genotypes due to the presence of 'nitrogen supply' X genotype interactions. These types of interactions also existed at Woodward when using a spring planting (Table 5). Differences in emergence among genotypes were significant in all

environments studied (Table 5), except at Woodward during the second year for both fall and spring plantings and at Tessaout in 1985. There was poor emergence at Woodward in the spring of 1985 due to wind erosion which displaced soil particles exposing the seeds on the soil surface. This resulted in poor contact between seeds and soil. Water erosion from irrigation by flooding also displaced seeds at Tessaout in the fall of 1985 and differences among genotypes could not be detected. Percent emergence was also very low during the second year in Morocco both at Sidi el Aydi and at Tessaout. Germination and emergence were impaired by insufficient rainfall after planting (20 mm of rain from Nov. 15 to Jan. 12 at Sidi el Aydi).

Combined Analysis and G X E Interactions

Estimations were made on 13 genotypes which were present in 12 environments. Data from environments having 'nitrogen supply' factor were averaged over levels of this factor as differences among effects of these levels were not significant (P > 0.05). Since significant differences among replications within each experiment were generally absent, the average of four single combined analyses was used to construct analysis of variance tables (as advised by Dr. Stroup, Professor of Statistics, Univ. of Nebraska, Lincoln, NE.) Each single combined analysis was based on three replications; i.e., combined analysis No 1 had replications 1, 2 and 3, while combined analysis No 2 had replications 2, 3 and 4, and so on. Mean squares from the combined analyses of variance No 1, 2, 3 and 4 were averaged in the end (Jackknife method (65)).

Woodward (spring 1985) and Sidi el Aydi and Tessaout (fall 1986)

were not included in the combined analysis as there was extremely poor emergence in most genotypes. Woodward (fall 1983) and (spring 1984) and the limed site at Haskell (fall 1983) plantings were also excluded due to the presence of significant differences among 'nitrogen supply' levels and/or genotype X 'nitrogen supply' interactions which did not allow averaging of data over main plot treatments. Perkins data (spring of 1985) was also not included in the combined analysis because of heterogeneity of variances.

Combined analysis of variance conducted on the remaining environments; StF83, StS84, HaF83, HaS84, H1S84, H1F84, H1S85, WoF84, PeF84, SiF85, JmF85, and TeF85, showed that environment, genotype, and genotype X environment interaction effects were highly significant (P < 0.01) (Table 6). The presence of G X E interactions indicated that responsiveness of different genotypes was not consistent from one environment to another. Therefore, ranking of genotypes for emergence changed over environments and no clear pattern was apparent for this establishment parameter among accessions, even over environments with homogeneous variances. Emergence percentages for each genotype and in each environment are exhibited in Tables 7 - 15 from which certain trends may be drawn.

Certain cultivars had either high or low emergence in most environments. Those ranking high were M. polymorpha Circle Valley, M. littoralis Harbinger, and M. sativa OK-83-Graze and Spredor II.

Genotypes ranking low were M. rugosa Sapo and Paraponto, M. tornata

Tornafield, T. incarnatum, and T. pratense. By looking at the extremes and means of the ranges of emergence percent (Tables 7-15), higher emergence percents were obtained in fall plantings than in spring

plantings in Oklahoma. This concurs with results of El Toumi (39) for alfalfa. Consequently, fall planting would be more desirable to ensure good stand establishment. No major differences were detected in emergence from one year to another in Oklahoma, but the first year in Morocco was far better than the second year and that was due to the lack of rain after planting in the second year. Fall plantings in Oklahoma were not different from fall plantings of 1985 in Morocco.

Genotype X environment (G X E) interactions were highly significant (P < 0.01) for emergence percent. Part of the reason may be attributed to the extreme contrasts both in environments as was shown by Comstock and Moll (28) and in the genotypes according to results obtained by Moll et al. (68) in corn. The significant effects of G X E interactions on Medicago spp. and Trifolium spp. emergence demonstrated strongly that recommendations for uniform seeding rates across various locations as presented by Bakhtri (10) might need to be reconsidered. Forage legumes plantings should be adjusted for seeding rate according to specific seed weight (reflected by the weight of 1,000 seeds), for percent emergence, and for the prevailing conditions where seed is planted when optimal stand density is desired.

Stability Analysis

Regression analysis for stability parameters showed that environmental linear effects and G X E (linear) were highly significant (P < 0.001) (Table 16). Differences among genotypes means, tested by genotype mean squares against non-linear pooled deviations, were significant (P < 0.001). Differences among regression coefficients of different cultivars, tested by G X E (linear) against the pooled

deviations and tested by genotype mean squares against G X E mean squares, were also significant. All regression coefficients were different from zero (P < 0.05). However, some bi were statistically different from 1 according to t tests (Table 17). Mean squares (s2di) for deviation from regression (Table 16), when tested against the pooled error, indicated that certain of them were significantly different from zero (Table 17).

Cultivars with bi not different from 1 (P > 0.05) were Circle Valley, Cyprus, Paraggio, and 'Mount Barker'. Their standard deviations (s^2di) of residuals about the regression line were 26.57, 0.00, and 0.00 and were statistically equal to zero. According to Eberhart and Russell (38), such cultivars were stable and had average responsiveness to environmental changes for emergence. Circle Valley was ranked high for emergence percentage in most environments included in the study. Borung and Yuchi had bi higher than unity and s^2 di not different from zero. Therefore, they were supposed to be stable and possess an above average emergence. Borung showed average and Yuchi had above average emergence and both were relatively stable. Harbinger and OK-83-Graze demonstrated regression coefficients higher than unity (bi > 1). Their s^2 di were different from zero. The two cultivars had above average emergence responsiveness to environmental differences, but OK-83-Graze appeared to be more unstable than Harbinger. In field conditions these two genotypes were ranked high in most environments. Paragosa, Tornafield, Jemalong, and crimson clover (\underline{T} . incarnatum) showed a bi < 1 and s^2 di not different (P > 0.05) from zero. Thus, their responsiveness would be less than average but they appeared to be stable. Paragosa and Jemalong were in fact medium emerging in many environments, while Tornafield and

crimson clover emerged poorly. Paraponto (bi = 0.60 and s^2 di = 116.68) would be, according to the linear model of Eberhart and Russell (38), the least desirable cultivar because its responsiveness to environmental changes was below average and its stability index (s^2 di) was different (P < 0.001) from zero. This cultivar was ranked low for emergence in most situations.

Results obtained in stability analyses confirmed the information obtained from individual experiment analyses for cultivars which had regression coefficients equal to or below unity (bi < 1). However, it was difficult to group cultivars which had a bi superior to unity since some of them, e.g. OK-83-Graze, had above average emergence percent, while others, e.g. Borung, had an average emergence percent. Mean squares for deviation (s^2 di) from the regression lines indicated that 10 cultivars were stable among 13 included in the analysis. However, in the combined analysis many of these stable cultivars, Paraggio and Cyprus, were more stable than others, e.g. Tornafield. In contrast, unstable cultivars, e.g. Paraponto, emerged poorly in most environments. Deviations observed between G X E interactions and stability parameters may have originated from the use of linear regression. Gray (50) and Nguyen et al. (70) demonstrated the limitations of the use of linear models for perennial grass forage yields. Taliafero et al. (84) indicated that cultivars of alfalfa with bi higher than unity had either better or lower forage yield than average response. A wide spectrum of environments and genotypes was reported by Comstock and Moll (28) to inflate G X E interactions which were considered as part of the lack of responsiveness of genotypes to environment changes. This may be another way to explain these deviations.

Emergence appeared to be heavily influenced with environmental changes and G X E interactions. The genotypic effects were present but minor. Assuming good seed quality, improvement in emergence percentage can be realistically achieved with good seedbed preparation, adequate soil moisture, and pest and disease control. Relationships between this establishment parameter and cultivar performance are discussed later in the forage section.

Cold Survival

Tolerance to cold was investigated only in Oklahoma for fall plantings during two years. Cold damage was not apparent in the spring plantings from Oklahoma nor in the fall plantings from Morocco. The lowest temperatures recorded in January and March were -24 °C and -4 °C at Stillwater, OK. in 1983, respectively. It was -1 °C in January at Sidi el Aydi, Morocco in 1985. Forage growth rate was limited with low temperatures during winter months in Morocco.

Individual Environment Analyses

The analysis of variance for each environment is presented in Table 18. Differences among replications were not significant in any environment. When 'nitrogen supply' was included as main plot treatments in the first year of the test, differences among effects of this factor levels were not significant (P > 0.05). Interactions of genotype X 'nitrogen supply' were also absent. However, effects associated with genotypes were significant in all environments, except in the 1984 fall planting at Haskell.

Combined Analysis and G X E Interactions

Estimations of G X E interactions were made on the genotypes which were present in the seven environments where cold damage was evaluated. Data were averaged over 'nitrogen supply' levels since there were no significant differences among their effects. Homogeneity of variances revealed the existence of two environmental sets. The first set of environments was constituted with 1983 fall plantings while the second

set was made of 1984 fall plantings.

<u>1983 Fall Plantings</u>. The analysis of variance is presented in Table 19. Effects associated with environments, genotypes, and G X E interactions were significant (P < 0.001). The presence of G X E interactions indicated that sensitivity of different genotypes (or at least of some of them) to cold was not consistent among environments.

Differences were not significant (P > 0.05) among M. polymorpha

Circle Valley, T. incarnatum, T. subterraneum, and T. vesiculosum at

Stillwater (Table 20). These four genotypes were ranked best for cold tolerance. Their respective grades to cold damage were less than 4.6.

M. arabica and M. sativa constituted an intermediate group when evaluated for cold damage. All other genotypes were graded between 7.2 and 8.8 on the cold damage scale, and had a lower tolerance level than M. sativa OK-83-Graze. Species which recovered after cold months were M. sativa, Trifolium spp., M. arabica, M. polymorpha, and M. minima.

All genotypes, except those of \underline{M} . \underline{sativa} and \underline{T} . $\underline{vesiculosum}$, were similarly damaged at Haskell with or without liming (Table 20). \underline{M} . \underline{sativa} and \underline{T} . $\underline{vesiculosum}$ were more tolerant than the other species. They survived after the winter months while all others died. \underline{M} . $\underline{arabica}$, \underline{T} . $\underline{incarnatum}$, and \underline{T} . $\underline{subterraneum}$ were significantly (P < 0.05) less sensitive to cold than the other annual medics at Woodward (Table 20). \underline{M} . \underline{sativa} and \underline{T} . $\underline{vesiculosum}$ constituted the highest tolerance group to cold. Their grades were between 5.9 and 6.8. Species which survived after the winter were \underline{M} . \underline{sativa} , \underline{M} . $\underline{arabica}$, and $\underline{Trifolium}$ spp. Other species of annual $\underline{Medicago}$ and \underline{T} . $\underline{pratense}$ were similar in their sensitivity to cold. Cold damage was graded in these

species between 8.1 and 9.

Differences among genotypes for cold damage were significant in the experiment with fewer accessions at Stillwater. \underline{T} . $\underline{vesiculosum}$ (grade = 3.3) was not significantly (P > 0.05) different from \underline{M} . \underline{sativa} . \underline{M} . $\underline{aculeata}$, \underline{M} . \underline{murex} , and \underline{M} . $\underline{polymorpha}$ strain 1 were all more sensitive to cold than \underline{M} . $\underline{blancheana}$ and the standard species of \underline{M} . \underline{sativa} and \underline{T} . $\underline{vesiculosum}$. They were respectively graded 8.5, 8.0, and 7.8 with LSD (P = 0.05) = 1.3. Species which died after frost were \underline{M} . $\underline{aculeata}$ and \underline{M} . \underline{murex} .

1984 Fall Plantings. Environments and genotypes had highly significant effects on cold damage (Table 19). However, G X E interactions were not significant in 1984 (P > 0.05). Mean comparisons among genotypes over environments (Table 20) showed that grades of \underline{M} . sativa, \underline{T} . incarnatum, \underline{T} . subterraneum, and \underline{T} . vesiculosum were not different (P > 0.05). This group of genotypes had the highest cold tolerance and survival in all three environments during the second year. All annual Medicago and \underline{T} . pratense were different from \underline{M} . sativa. They were less tolerant to cold than \underline{M} . sativa and none of them survived the low winter temperatures.

Temperatures recorded in each location and year showed that Stillwater in 1983 was one of the coldest environments (-24°C at Stillwater vs. -19°C at Perkins in Jan. 1985. However, it was the only environment where annual Medicago spp. survived after the winter. Snow covered the experimental area and medic genotypes having some cold tolerance survived. Rating for cold damage obtained in January at Haskell were lower than those at Perkins and Woodward during the second

year (2.8 vs. 6.1 and 8.0 with LSD (P = 0.05) = 0.5). However, all annual medics died at the three locations. Haskell was warmer than Perkins and Woodward in 1984/85. The lowest temperatures recorded at Haskell and Perkins were -15 and -19 $^{\circ}$ C, respectively. Nevertheless, due to frequent frost compounded with waterlogging all annual Medicago died after January.

Genotype X environment interactions observed in the first year of test may have originated from protection by snow cover. Sensitive genotypes (those which died in all environments such as \underline{M} . \underline{rugosa}) and highly tolerant ones (those which survived in all environments such as \underline{M} . \underline{sativa}) performed similarly across all environments. Genotypes with moderate tolerance to cold, such as \underline{M} . $\underline{polymorpha}$ Circle Valley, were not consistent as they survived under snow cover at Stillwater but died at Haskell and Woodward.

Cultivars of M. scutellata, M. rugosa, and M. tornata were the first to be killed with frost in all environments. M. aculeata, M. littoralis, M. murex, and M. truncatula were the second group of species to die after repeated occurrences of frost. M. arabica, M. blancheana, M. minima, and M. polymorpha were the least sensitive medics and had at least some cold tolerance.

No annual <u>Medicago</u> spp., except <u>M</u>. <u>rugosa</u> and <u>M</u>. <u>tornata</u>, changed growth habit over environments. <u>M</u>. <u>blancheana</u> was an upright species. <u>M</u>. <u>scutellata</u> was semi-prostrate. The other medic species were all prostrate. <u>M</u>. <u>rugosa</u> was semi-prostrate and <u>M</u>. <u>tornata</u> was prostrate in fall plantings, however, they were upright in spring plantings. <u>M</u>. <u>sativa</u> was decumbent in all environments as opposed to <u>T</u>. <u>subterraneum</u>, a prostrate species. <u>T</u>. pratense and <u>T</u>. incarnatum were upright species, but \underline{T} . $\underline{vesiculosum}$ was semi-prostrate in fall plantings and prostrate in spring plantings.

Tolerance to cold of annual medic species might be related to their growth habit. M. rugosa and M. scutellata had a semi-prostrate to upright growth habit and were the most sensitive to cold, while M. arabica, M. minima, and M. polymorpha were prostrate and the least sensitive to cold. Decumbent cultivars in many crops were reported by Dexter (37) to be less tolerant to cold than prostrate ones. Such tolerance might also be associated with the ploidy level of annual Medicago as Dexter (37) indicated on other crops. M. rugosa and M. scutellata are tetraploid with 32 chromosomes as opposed to M. minima and M. polymorpha which are diploid (62). However, future studies are necessary to understand the relationship between cold tolerance and growth habit or ploidy level since growth habit depended on growing period in species and since M. tornata is diploid but was highly sensitive to cold.

These findings may explain results obtained by Denman et al. (34) and Kneebone (59) who reported that annual Medicago spp. were poorer than clovers and perennial alfalfa in Oklahoma. They may also explain the limited number of annual medic species to those having some cold tolerance (even though small) in the flora of Oklahoma (89). The present study concurs with data gathered by Cocks (26) in Syria and Francis (44) in Iraq with Australian cultivars which demonstrated more sensitivity to cold than local ecotypes. However, it does not support the lack of winterhardiness of medic species reported by Hanson and Barnes (51) since certain species survived under snow cover and temperature thresholds were not defined in previous reports.

Medic species studied originate in warm areas of the Mediterranean basin (53, 80) and appear to be much less tolerant to cold than M. sativa and T. vesiculosum. Intensive breeding and natural selection have improved cold tolerance in these two forage species to a high level, especially in dormant cultivars (51). Winter kill is not a major problem for medic species in their native habitat, except in high elevations, when they are used as pasture species. Production of these species might be reduced but not to the level of total stand loss in areas where temperatures do not fall below 0°C. Fall planting in a continental climate having very cold winter caused stand failure. Future investigations should include native ecotypes in continental climates with severe cold like in Oklahoma as well as species like M. <u>noeana</u>, M. <u>rotata</u>, and M. <u>rigidula</u> from the colder regions of the Mediterranean basin. Until some intensive breeding is performed for cold tolerance, spring planting will be more successful with present Australian cultivars than fall planting in Oklahoma. When spring-seeded plants were allowed to set seed for regeneration (see Chapter III), offsprings germinated during early fall and they were exposed to winter kill. Such a cycle will not permit a sustained level of seed in soil for persistence of stands from year to year. Hence, intensive breeding is also necessary to develop cultivars which have a high proportion of mechanically and/or physiologically dormant seed that germinate in the spring after frost occurrence.

Forage Yield

Dry matter yield is generally considered the most important parameter in forage crop evaluations. In the present study forage yield was estimated in eight environments; five under continental climatic conditions in Oklahoma and three under Mediterranean conditions in Morocco. Those of Oklahoma were 1983 fall and 1984 spring plantings at Stillwater, 1984 and 1985 spring plantings at Haskell (limed site), and 1985 spring planting at Perkins. Fall plantings of 1985 at Sidi el Aydi, Jemaa Shaim, and Tessaout were the environments where forage yields were obtained in Morocco. Winter kill prohibited harvests from fall plantings in Oklahoma except at Stillwater in 1983. Poor emergence, along with drought, at Woodward and in 1986/87 Moroccan sites resulted in negligible forage yield.

Individual Experiment Analyses

The analysis of variance for dry matter yield production is presented in Table 21. The 'nitrogen supply' factor did not have significant (P > 0.05) effects on dry matter produced when it was included as a main plot treatment. Genotype X 'nitrogen supply' interactions were not significant. Effects associated with genotypes were significantly different in all environments except at Haskell (1984 spring planting).

<u>Combined Analysis and G X E Interactions</u>

A combined analysis was conducted using the same procedure as described for emergence. Data from fall planting at Stillwater in 1983

was not included because many cultivars died during winter. Homogeneity of variances, conducted after averaging data over 'nitrogen supply' levels, showed that Haskell (1984 spring planting) had a small error term compared to those calculated at other experiments. Therefore, these data for forage yield were also not included in the combined analyis. The remaining sites formed two sets of data when testing for homogeneity of variances. One set consisted of Stillwater 1984, Haskell 1985, and Perkins 1985 spring plantings. The second set was composed of Sidi el Aydi, Jemaa Shaim, and Tessaout 1985 fall plantings.

Combined analyses conducted on spring plantings at Stillwater 1984 and Perkins and Haskell 1985 showed that effects associated with environment, genotype, and genotype X environment interactions were significant (Table 22). The presence of G X E interactions indicated that response of different genotypes for forage production was not consistent over environments. Comparisons of genotypic performances in each Oklahoma environment are presented in Table 23. Environments, genotypes, and G X E interactions had significant effects on dry forage yield produced in Morocco (Table 22). The order in ranking of genotypes changes from one environment to the other. Relative yields of cultivars in the 1985 Moroccan fall plantings are in Table 24.

<u>1983 Fall Plantings</u>. Fall planting resulted in stand loss of most medics in all locations, except at Stillwater in 1983 where snow cover prevented winter kill of certain genotypes (Table 23). Genotypes within the top significance group were \underline{M} . <u>sativa</u> OK-83-Graze (98 g/m) and Spredor II and \underline{T} . <u>vesiculosum</u> Yuchi (81 g/m). The other three Trifoliums did not exhibit significant (P > 0.05) differences in their

forage yield. \underline{M} . \underline{minima} , \underline{M} . $\underline{polymorpha}$, and \underline{M} . $\underline{arabica}$ were the only medic species which survived. Their corresponding yields were 2, 21, and 13 g/m (LSD (P = 0.05) = 17 g/m). \underline{M} . \underline{minima} and \underline{M} . $\underline{arabica}$ were not significantly different in forage yield from species which died. However, this agronomic parameter reflected the level of medic tolerance to cold and the three species may be considered as having some potential, even though small, for use in the Great Plains with fall sowing.

1984 and 1985 Spring Plantings. The best yields were obtained from M. polymorpha Circle Valley (109 g/m) and M. scutellata Robinson in the 1984 spring planting at Stillwater (Table 23). Differences were not significant (P > 0.05) among M. littoralis Harbinger and M. truncatula Cyprus, Jemalong 2, and Paraggio which produced 58-71 g/m. M. minima produced the least forage (2 g/m) and was not significantly different from T. pratense. All Trifolium spp. did not differ in dry matter yield and had low production (10-22 g/m). Dry matter production of M. sativa was not different from that of M. tornata, M. truncatula Borung and Jemalong 1, and M. rugosa Paragosa. However, M. rugosa Paraponto 1 had statistically equal dry forage yield (24 g/m) to T. vesiculosum.

Forage yield in 1984 at Haskell was extremely low and differences among genotypes could not be detected with the protected LSD. Dry matter varied between 2 and 8 g/m (Table 23). Poor emergence and waterlogging were the main factors which impaired yield in this environment.

The best yielding genotypes at Perkins in 1985 were M. scutellata

Sava and Robinson (67-72 g/m). However, they were not significantly (P > 0.05) different from \underline{M} . $\underline{polymorpha}$ Circle Valley (40 g/m) and \underline{M} . $\underline{truncatula}$ Paraggio (57 g/m) at Perkins in 1985 (Table 23). Cultivars that produced the lowest yields were \underline{T} . $\underline{pratense}$ and \underline{T} . $\underline{vesiculosum}$ (1 g/m), but they did not statistically differ from

M. tornata Tornafield 1, and M. truncatula Borung and Jemalong 1.

T. subterraneum, T. incarnatum, cultivars of M. rugosa, M. sativa, and

Cultivars of \underline{M} . scutellata were also the highest yielding at Haskell in 1985 (112-125 g/m). Differences were not significant (P > 0.05) within the group made up of \underline{M} . polymorpha, \underline{M} . sativa, and \underline{M} . truncatula Borung and Paraggio (Table 23). All cultivars of \underline{M} . rugosa and \underline{T} . pratense produced less than 1 g/m of dry matter and did not differ in yield from \underline{M} . littoralis, \underline{M} . tornata, \underline{M} . truncatula Jemalong 1, and the other Trifolium spp.

M. truncatula Borung (285 g/m) and M. tornata at Sidi el Aydi (Table 24). Differences were not significant (P > 0.05) among M. littoralis, M. polymorpha Serena, M. scutellata, and the other cultivars of M. truncatula. Genotypes which produced the least amount of forage were T. subterraneum and T. vesiculosum (1 g/m.); however, they did not differ from T. incarnatum, M. truncatula Jemalong 1, M. sativa, M. rugosa, and M. polymorpha Circle Valley. M. truncatula Paraggio yielded the highest dry matter with 329 g/m. at Jemaa Shaim (Table 24); however, it did not differ (P > 0.05) from M. polymorpha Circle Valley, M. scutellata Robinson and Sava and M. truncatula Jemalong 2. M. polymorpha Serena, M. rugosa Paragosa, M. littoralis 1, and M.

<u>truncatula</u> Borung and Jemalong 1 constituted the second highest yielding group with 161-233 g/m . <u>T. subterraneum</u> produced the least dry matter (50 g/m) but it was not significantly different from <u>M. rugosa</u> Paraponto 1 and Sapo, M. sativa, and T. vesiculosum.

The highest forage producing cultivar was \underline{M} . scutellata Robinson (606 g/m) and then Sava at Tessaout (Table 24). Relatively high yields were found (189-260 g/m) using \underline{M} . polymorpha Circle Valley and Serena, and \underline{M} . truncatula Borung, Jemalong 2, and Paraggio. \underline{T} . vesiculosum yielded the least amount of forage (53 g/m) and was not significantly (P > 0.05) different from the other $\underline{Trifolium}$ spp., \underline{M} . truncatula Jemalong 1, \underline{M} . tornata, \underline{M} . sativa, \underline{M} . rugosa Sapo and Paraponto 1, and \underline{M} . littoralis 1.

Genotype X environment interactions were reported to express the lack of response of certain genotypes to environmental variations (28, 68). Certain genotypes were ranked either high or low in almost all environments. Cultivars of M. scutellata had high yields in each environment except at Sidi el Aydi. In contrast, M. rugosa Sapo and Paraponto 1 and Trifolium spp. had poor production. Some of the G X E interactions were caused by varying performance of some genotypes among environments. For instance, M. scutellata Sava and M. truncatula

Jemalong 1 ranked second and third respectively, both at Jemaa Shaim and Tessaout; however, dry matter of the first cultivar increased by 186 g/m , while that of the second decreased by 26 g/m from Jemaa Shaim to Tessaout. A third potential source of G X E interactions may have arisen from the use of wide spectra of genotypes and environments as it was demonstrated by Comstock and Mol1 (28) and Mol1 et al. (68).

 $\underline{\mathsf{M}}.\ \underline{\mathsf{sativa}}\ \mathsf{and}\ \underline{\mathsf{Trifolium}}\ \mathsf{spp}.\ \mathsf{were}\ \mathsf{generally}\ \mathsf{less}\ \mathsf{productive}\ \mathsf{than}$

most of the annual <u>Medicago</u> spp. Spring planting was reported by El Tomi (39) to be less favorable to alfalfa establishment in Oklahoma than fall planting. In Morocco, soils were alkaline with pH = 8.1-8.3 and appeared to be less suitable for these forage species because of some unknown factors. Data obtained from fall plantings in Oklahoma support those of Denman et al. (34) and Kneebone (59) who indicated that annual medics performed poorly compared to alfalfa and clovers. However, when these forage species were planted in the spring, they produced as much or even more than alfalfa and clovers, which supports results found by Rumbaugh and Johnson (77) who demonstrated the superiority of annual <u>Medicago</u> spp. to <u>M. falcata</u> with regard to establishment and rapid growth.

Variation between cultivars of the same species was observed and confirmed the presence of genetic variation in annual medics reported by Crawford (29) and Radwan et al. (74). However, most medic species differed from \underline{T} . $\underline{subterraneum}$ in forage yield and this did not support the conclusions stated by Aitken (3) that annual medics were similar to subclover in growth habit, rate of development, and forage production.

Generally, dry forage yield of annual <u>Medicago</u> spp. were higher in Morocco than from medics (planted in spring) from Oklahoma. This difference was due to duration of the vegetative phase of growth.

Growth occurred during a warmer season with longer photoperiod (April and May) in Oklahoma than in Morocco (Dec.-Feb.). It took over four months for plants to reach flowering in Morocco compared to two in Oklahoma. Flowering stage was hastened with long photoperiods and high temperatures according to Clarkson and Russell (21, 23). Consequently, plants did not have enough time to accumulate biomass prior to the

reproductive growth phase in Oklahoma. Low yields obtained at Haskell in 1984 were related to poor emergence and waterlogging which was reported by Francis and Poole (45) to reduce forage production in annual medics.

Relationships between Emergence Percent and Forage Yield

Linear regression of forage yield on percent emergence was significant (P < 0.001) when plants were spring sown at Stillwater in 1984 and Perkins in 1985 (Table 25). It was also significant (P < 0.05) at Haskell from the 1985 spring planting. However, the linear model was not significant in any Moroccan environment for 1985 fall plantings, which indicated that forage yield was not related to emergence percentage. Significance level decreased when snail medics (\underline{M} . scutellata) and clovers were dropped from the analyses because they were the odd species in most environments; i.e., they performed the best and the poorest for forage production. By dropping these species, there was also a reduction in b values obtained from Oklahoma sites and at Tessaout, but an increase of b at Sidi el Aydi and Jemaa Shaim.

Differences in b observed among environments show that an increase of 1% in emergence percent resulted in an increase of forage yield in Oklahoma, but with a varying magnitude depending on environmental conditions. Deviations in significance level among environments demonstrated that differences in forage yield among genotypes or environments may have originated partly from differences in emergence percent, and this appeared to be likely in Oklahoma environments where linear regressions were significant. Therefore, emergence percent might

be used as a predictor for forage production. Cocks (25) demonstrated that dry matter production increased with plant density and that a 50 kg/ha seeding rate produced the highest forage yield in M. rigidula, M. noeana, and M. truncatula in Syria. In Moroccan environments emergence percent was not a good linear predictor for forage yield. Fall sown plants had enough time prior to harvesting to express their potential of production and to exert competition for light and nutrients. Derkaoui (36) reported a decline in plant density from emergence to flowering stage in medics. He also indicated that when seedlings were between 132 and 228/m, variation in forage yield was minor due to compensations occurring throughout the growth cycle. In Oklahoma, duration of the vegetative growth phase was shorter and thus, interplant competition was smaller.

Simple correlation coefficients between emergence percent and forage yield were generally low (Table 25) and paralleled significance levels of regression analyses. These low correlations showed the weak association between establishment success and dry matter production harvested at flowering stage, particularly in Moroccan environments. The presence of G X E interactions in both emergence percent and forage yield supported further the concept that establishment success, within the limits, may not always be a good predictor for forage yield.

When linear regressions were calculated for each medic species, emergence percent significantly (P < 0.001) affected forage yield in \underline{M} . $\underline{truncatula}$, and to a lesser degree in \underline{M} . $\underline{tornata}$ and \underline{M} . $\underline{littoralis}$ (Table 26). Very low association between the two agronomic parameters existed in \underline{M} . $\underline{polymorpha}$, \underline{M} . \underline{rugosa} , and \underline{M} . $\underline{scutellata}$. Cultivars of these species were earlier maturing and with fewer stems than those of

 $\underline{\mathbf{M}}$. $\underline{\mathbf{truncatula}}$. This may explain the differences observed among species. Establishment success may be used as a predictor for forage in certain species, e.g., $\underline{\mathbf{M}}$. $\underline{\mathbf{truncatula}}$; however, this relationship should be used with caution until further investigations bring more information to this subject.

Forage Quality

Forage produced by annual <u>Medicago</u> spp. was generally of good quality as was estimated in this study by crude protein (CP), neutral detergent fiber (NDF), and acid detergent fiber (ADF) concentrations, and leaf:stem ratio. Chemical components, expressed on an organic matter basis, were determined on samples gathered from six environments: Stillwater 1984; Haskell and Perkins 1985 spring plantings; and Sidi el Aydi, Jemaa Shaim, and Tessaout 1985 fall plantings. Leaf:stem ratio was measured from the Moroccan locations sown in the fall of 1985.

Individual Experiment Analyses

Analyses of variance presented in Table 27 showed that differences among replications in each environment were not significant (P > 0.05) for CP concentration. Effects on CP associated with genotypes were significant at Stillwater, Haskell, Sidi el Aydi, and Jemaa Shaim but not at Perkins and Tessaout. Replications differed (P < 0.05) for NDF at Stillwater, Perkins, and Jemaa Shaim but not in the other environments. Mean squares for the genotypic factor were significant for NDF in Moroccan environments but not in Oklahoma. Acid detergent fiber significantly differed (P < 0.01) from one replication to the other at Stillwater, Perkins, and Sidi el Aydi but not in the other locations. Differences among cultivars for ADF were significant only at Sidi el Aydi and Tessaout. Leaf:stem ratio mean squares associated with genotypes were significant (P < 0.01) at Sidi el Aydi and Jemaa Shaim.

Combined Analysis and G X E Interactions

<u>Crude Protein</u>. Homogeneity of variances revealed that all environments, except Perkins, can be combined for CP. The combined analysis of variance presented in Table 28 indicates that environments had significant (P < 0.001) effects on CP. Differences among genotypes were not significant (P > 0.05); however, G X E interactions were present (P < 0.01). Comparisons among genotypes (Table 29) were made in each environment due to significance of G X E interactions.

The highest CP was obtained with \underline{M} . \underline{minima} (24.7%) at Stillwater; however, this species was not significantly different (P > 0.05) from 13 other genotypes. In contrast, the lowest CP obtained was in \underline{T} . $\underline{pratense}$ (19.4%) but it was not different (P < 0.05) from CP in \underline{M} . $\underline{polymorpha}$ Circle Valley, \underline{M} . $\underline{truncatula}$ Cyprus and Jemalongs, and \underline{T} . $\underline{incarnatum}$. Differences among genotypes were not significant (P > 0.05) at Perkins and mean CP was 21.7% at this location. At Haskell \underline{M} . $\underline{polymorpha}$ Circle Valley had the lowest CP (17.6%); however, it was not different from 10 other entries.

M. sativa OK-83-Graze produced the highest CP (27.4%) at Sidi el Aydi, which was not significantly (P > 0.05) different in CP from M. truncatula Borung and M. tornata Tornafield 1 and 2. The lowest CP concentration was obtained in M. polymorpha Serena (19.7%) but this cultivar did not differ (P > 0.05) from seven others. At Jemaa Shaim the top group for CP was composed of M. truncatula Paraggio and the two Jemalongs, M. sativa OK-83-Graze, M. rugosa Paraponto 1 and Sapo and M. polymorpha Circle Valley. The lowest CP was in M. rugosa Paragosa with 21.0%; however, this cultivar was within a significance group composed

of nine genotypes. Differences among genotypes were not significant at Tessaout where the mean CP was 21.2%.

Genotype X environment interaction effects on CP were not of large magnitude because all genotypes presented a high CP concentration and differences among cultivars were small. Late-maturing species, e.g., M. tornata, were generally higher in CP than early-maturing ones, e.g. M. polymorpha. Harvesting date could be a source of differences observed among entries. Harvest occurred in each experiment when half of the accessions flowered, thus early-maturing entries were penalized with regard to CP concentration and data were slightly biased. Nevertheless, CP concentrations remained high in annual Medicago spp., M. sativa, and Trifolium spp.

The present study demonstrated that annual medic species, alfalfa, and clovers were rich in CP. Jones and McLeod (58) reported that \underline{M} . scutellata contained 22% CP at flowering while \underline{M} . truncatula had 17% at the same stage. Similar results were obtained here in \underline{M} . scutellata; however, higher CP was found in \underline{M} . truncatula. Differences observed by Mohamed and Kovacs (67) between annual and perennial species of Medicago were absent in the present study.

Neutral Detergent Fiber. Two sets of environments were used for combined analysis of NDF concentrations; Oklahoma locations and Moroccan sites. Genotype, environment, and G X E interaction effects were not significant (P > 0.05) when Stillwater, Perkins, and Haskell were combined (Table 28). Corresponding NDF concentrations to these three environments were 34.5%, 32.0%, and 33.8% (LSD (P = 0.05) = 6.6%). Mean NDF over environments and genotypes was 33.4%.

Combined analysis of Moroccan sites showed that NDF was related to environments and genotypes; however, interactions between these factors were not significant (P > 0.05) (Table 28). Neutral detergent fiber was slightly lower at Jemaa Shaim (30.7%) than at Tessaout (32.7%), and 32.5% at Sidi el Aydi (LSD (P = 0.05) = 1.8%). M. truncatula Cyprus contained the highest NDF with 36.7% (Table 30). A intermediate group for NDF was composed of M. littoralis, M. truncatula Jemalong 1 and 2, and M. polymorpha Serena when data were averaged over Moroccan locations. M. sativa had the lowest NDF with 27.3% and was different (P > 0.05) from all annual Medicago spp. The other genotypes contained 29.6% to 32.5% NDF.

The absence of G X E interactions in the two sets showed that NDF concentration is less variable even though wide ranges of environmental conditions existed in each set. Differences among genotypes may have been biased with harvesting time as was pointed out for CP. Neutral detergent fiber was lower in most species than that reported by Belyea and Ricketts (14) (40% NDF in alfalfa at 1/10 bloom stage). Neutral detergent fiber, which reflects the proportion of cell walls in forage (75, 86), is considered negatively correlated to in vitro dry matter digestibility and intake (7). This indicates that annual Medicago spp. would have high digestibility and moderate intake. In grazing situations low intake from low forage availability may often be compensated for by increases in grazing time.

Acid Detergent Fiber. Error mean squares were homogeneous among
Oklahoma and Tessaout environments. Sidi el Aydi and Jemaa Shaim formed
another pair in homogeneity of variance testing. Combined analysis of

each set is presented in Table 28. Effects associated with environments, genotypes, and G X E interactions were not significant (P > 0.05) in the former set (Stillwater, Perkins, Haskell, and Tessaout). Only genotypic effects were significant (P < 0.01) in the other set. Mean ADF value was 23.9% in forage from Oklahoma and Tessaout. Since differences among genotypes were significant (P < 0.01) at Tessaout only, they were attenuated in the combined analysis. At that environment, just four cultivars had significantly (P < 0.05) higher ADF than the lowest value of 17.8%. They were \underline{M} . $\underline{truncatula}$ Cyprus (24.5%), \underline{M} . $\underline{scutellata}$ Robinson (26.0%), \underline{M} . $\underline{polymorpha}$ Serena (23.0%), and \underline{M} . $\underline{littoralis}$ Harbinger (25.5%) (Table 31). Forage from Sidi el Aydi and Jemaa Shaim had 20.5% ADF. Cultivars of \underline{M} . \underline{rugosa} and \underline{M} . $\underline{tornata}$ were significantly (P < 0.05) lower in ADF concentration than \underline{M} . $\underline{scutellata}$ Robinson (22.1%) when ADF was averaged over locations.

Acid detergent fiber was not influenced by G X E interactions in this study. The present ADF percentages were low in most genotypes compared to 30% ADF in alfalfa at 1/10 bloom stage (Beleya and Ricketts (14)). Generally annual medic, alfalfa, and clovers species contained similar proportions of ADF. Since ADF was reported to consist mainly of cellulose and lignin (14, 75, 86), these forage species have moderate amounts of these chemical components at flowering stage. Acid detergent fiber was negatively correlated with digestibility (7), suggesting that the species under evaluation would have a high digestibility and rapid rate of passage in the rumen.

<u>Leaf:Stem Ratio</u>. Genotype X environment interactions were not present (P > 0.05) for leaf:stem ratio; however, both environments and

genotypes had significant (P < 0.001) effects on this parameter (Table 27). Leaf:stem ratio was 1.8, 2.6, and 2.8 at Tessaout, Jemaa Shaim, and Sidi el Aydi, respectively (LSD (P = 0.05) = 0.3). The lowest leaf:stem ratio was in \underline{M} . scutellata Sava (Table 32); however, this cultivar was not significantly (P > 0.05) different from eight other cultivars. \underline{M} . tornata presented the highest ratio with 3.1 but it did not differ (P > 0.05) from \underline{M} . truncatula Paraggio and \underline{M} . sativa.

Differences in leaf:stem ratio among genotypes may be related to harvest date and maturation cycle as was the case for CP. Field data obtained on leaf:stem ratio concur with the value of 2:1 reported by Barnes and Gordon (13) on alfalfa at the vegetative stage. Similar results on this ratio were obtained under greenhouse conditions on annual medic species (see Chap. IV). A high leaf:stem ratio suggests that the species studied have a high digestibility at flowering.

Relationships between Forage Yield and Quality Parameters

Simple correlations (r) between pairs of forage yield, CP, NDF, and ADF varied from -0.58 to 0.93 with no trend both in the magnitude of r and in the sign of the associations across environments (Table 33) or species (Table 34), except that NDF and ADF were positively correlated in both cases. Neutral detergent fiber was also negatively correlated to yield and to CP in medic species except in strand medic where CP and ADF were positively associated.

Barnes and Gordon (13) reported in a review on alfalfa that chemical components of forage may or may not be correlated depending on environmental conditions. Crude protein was reported to be negatively

correlated to NDF and ADF in annual medics in Syria (7). Medics in this study were grown under diverse environments and it appears difficult to draw firm conclusions. Future investigations focusing on these subjects should be conducted under a narrower spectrum of environments and with closely related genetic materials to satisfy specific needs.

On a practical basis, the quality of annual <u>Medicago</u> spp. as estimated by CP, NDF, and ADF concentrations, and leaf:stem ratio appeared to be high at the flowering stage and were similar to that of alfalfa and clovers. These forage species are a good source of CP. Supplementation with cereal grains or low quality forage, as found in pastures and cereal residues, would maximize the production/ha of medic based pastures. Environmental conditions and G X E interactions were important for CP concentrations but did not impact NDF, ADF, and leaf:stem ratio to the same degree.

Conclusions

Emergence was related to environment, genotype, and G X E interactions. Fall planting resulted in higher emergence than spring planting in many genotypes studied, therefore better stand establishment was achieved during fall in Oklahoma. Differences were not apparent for emergence percent between fall sown Oklahoma and Moroccan environments. Since emergence percent depends also on seed quality and seedbed conditions, seeding rate should be adjusted according to the medic cultivar characteristics and to environmental conditions in order to achieve optimum stand densities. Establishment success in spring planting positively influenced forage yield under Oklahoma conditions. Annual Medicago species included in the study may generally be considered to lack cold tolerance at an acceptable level for practical use under continental conditions as in Oklahoma. However, M. polymorpha, M. blancheana, M. minima, and M. arabica, in addition to local ecotypes and species originating from cold regions, will constitute a good germplasm source for selection of cold tolerant cultivars to be used in the future.

Forage yield was another agronomic parameter associated with genotype, environment, and G X E interactions. Several annual medics were higher producing than alfalfa and clovers when planted during spring in Oklahoma or fall in Morocco. \underline{M} . scutellata performed the best in most environments as opposed to \underline{M} . rugosa which performed poorly. The presence of G X E interactions showed that some genotypes were better adapted in certain environments than in others.

Forage quality was as high in annual <u>Medicago</u> species as in perennial alfalfa and clovers. All genotypes contained high CP and leaf:stem ratio, and relatively low NDF and ADF concentrations.

Genotype X environment interactions were important for CP. From these results, it does not appear necessary to place a high priority on evaluation of forage quality during early stages of research to improve annual medics.

Table 1. Monthly mean temperatures and rainfall at Stillwater, OK. and Settat, ${\sf Morocco}^{\left(1\right)}$

Month	Stillwater		Settat		
	T.°C	Pmm (2)	T °C	Pmm	
January	1.1	29.0	10.6	61.0	
February	8.1	34.5	12.2	54.0	
March	8.1	46.5	14.2	47.0	
April	13.1	71.5	16.2	37.0	
May	19.2	115.5	18.8	17.0	
June	25.8	106.0	22.2	3.0	
July	27.5	88.3	25.4	0.0	,
August	28.1	80.3	25.5	0.0	
September	21.9	84.5	23.1	7.0	
October	15.8	69.5	19.5	43.0	
November	9.4	46.3	15.2	60.0	
December	5.8	33.5	11.6	77.0	
TOTAL		767.5		406.00	

^{(1):} Values averaged over 65 and 33 years for Stillwater and Settat, respectively.

^{(2):} Precipitation not including snow.

Table 2. Ecological characteristics of the environments included in the study

	Soil	Annual precipi-	Temp.°	С
Location	type(1)	tation(mm)(2)	Min.	Max.
Stillwater, OK.	Fine-silty, mixed, Thermic Cumalic Haplustolls	860.0	-8.9	36.7
Haskell OK.	Fine, mixed, Thermic Mollic Albaqualfs	1025.0	-6.7	33.9
Perkins, OK.	Fine-loamy, mixed, Thermic Udic Argiustolls	839.2	-10.0	35.6
Woodward, OK.	Sandy, mixed, Thermic Psammentic Haplustalfs	575.0	-11.0	38.0
Sidi el Aydi, Morocco	Calcixerollic chromoxerert	408.0	4.4	33.6
Jemaa Shaim, Morocco	Typic chromoxerert	297.0	7.0	33.5
Tessaout, Morocco	Vertic xero- fluent	257.0	4.3	39.8

^{(1):} Soil names in Morocco are translated according to USDA-Soil taxonomy, Washington D.C., 1975.

^{(2):} Average over 65 and 33 years for Oklahoma sites and Moroccan sites, respectively; precipitations include snow. Temperatures min. and max. are in Jan. and Aug., respectively.

Table 3. List of environments included in the study

0-4-	1	Planting	V
Code	Location	season	Year
StF83	Stillwater	fall	1983
StS84	Stillwater	spring	1984
SsF83	Stillwater (small)	fall	1983
Ss\$84	Stillwater (small)	spring	1984
H1F83	Haskell (limed)	fall	1983
HaF83	Haskell (no lime)	fall	1983
H1S84	Haskell (limed)	spring	1984
HaS84	Haskell (no lime)	spring	1984
H1F84	Haskell (limed)	fall	1984
H1S85	Haskell (limed)	spring	1985
WoF83	Woodward	fall	1983
WoS84	Woodward	spring	1984
WoF84	Woodward	fall.	1984
WoS85	Woodward	spring	1985
PeF84	Perkins	fall	1984
PeS85	Perkins	spring	1985
SiF85	Sidi el Aydi	fall	1985
SiF86	Sidi el Aydi	fall	1986
JmF85	Jemaa Shaim	fall	1985
TeF85	Tessaout	fall	1985
TeF86	Tessaout	fall	1986

Table 4. List of genetic material included in the study(1)

Genus	Species	Common name	Cultivar Sou	rce(2)
Medicago Medicago Medicago Medicago Medicago	aculeata L. arabica (L.) Huds. blancheana L. littoralis Rhode littoralis Rhode	Spotted medic Strand medic Strand medic	Harbinger 1 Harbinger 2	(b) (a) (b) (a) (b)
Medicago Medicago Medicago Medicago Medicago	minima (L.) Mill. murex L. polymorpha L. polymorpha L. polymorpha L.	Little medic Burr medic Burr medic Burr medic Burr medic	Strain 1 Circle Valley Serena	(a) (b) (b) (a) (a)
Medicago Medicago Medicago Medicago Medicago	rugosa Desr. rugosa Desr. rugosa Desr. rugosa Desr. sativa L.	Gama medic Gama medic Gama medic Gama medic Alfalfa	Paragosa Paraponto 1 Paraponto 2 Sapo OK-83-Graze	(b) (b) (a) (b) (c)
Medicago Medicago Medicago Medicago Medicago	sativa scutellata (L) Mill scuttelata tornata (L.) Mill tornata (L.) Mill	Alfalfa Snail medic Snail medic Disc medic Disc medic	Spredor II Robinson Sava Tornafield 1 Tornafield 2	(4) (b) (a) (a) (b)
Medicago Medicago Medicago Medicago Medicago	truncatula Gaertn. truncatula Gaertn. truncatula Gaertn. truncatula Gaertn. truncatula Gaertn.	Barrel medic Barrel medic Barrel medic Barrel medic Barrel medic	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	(b) (b) (b) (a) (a)
Trifolium Trifolium Trifolium Trifolium	incarnatum L. pratense L. subterraneum L. vesiculosum Savi	Crimson clover Redclover Subclover Arrowleaf clover	Redland Mount Barker Yuchi	(d) (d) (d)

^{(1):} Medic seed was provided by Dr. E. J. Crawford and Mr. M. Bounejmate from Australia and Morocco, respectively.

Genotypes used in each experiment are listed in individual tables of comparisons.

Table 5. Mean squares (MS X 10^{-2}) for emergence at each environment

·		Environm	ent	
Source	StF83	H1F83	HaF83	WoF83
Replication Nitrogen Supply (Nit.) Error (a) Genotype (Gen.) Nit. x Gen. Error (b)	4659 5713 2121 20886*** 467 761	3356 5396* 899 15296*** 691* 457	998 276 2195 4925*** 1638 1693	1141 1790** 288 30542*** 1053 988

Table 5. (continued)

-		Environm	ent	
Source	StS84	H1S84	HaS84	WoS84
Replication Nitrogen Supply (Nit.) Error (a) Genotype (Gen.) Nit. x Gen. Error (b)	8178 1184 2856 25603*** 505 738	538 1089 1815 986* 500	309 19 1038 462** 137 192	3976 1017 841 3696* 4899** 2266

Table 5. (continued)

			Environm	ent		
Source	SsF83	SsS84	PeF84	H1F84	WoF84	PeS85
Replication Genotype Error	295 5431*** 432	1683 19157*** 701	840 8609** 1362	1688 4102** 1602	1119 826 1205	658 1755** 740

Table 5. (continued)

				Environmen	t		
Source	H1S85	WoS85	SiF85	JmF85	TeF85	SiF86	TeF86
Replication Genotype Error	953 1410* 780	17 23 16	5403** 3132*** 1222	201 9779*** 520	13384*** 820 945	19 61** 22	5 29** 8

*** : Significant (P < 0.001).

** : Significant (P < 0.01).

* : Significant (P < 0.05).

Table 6. Mean squares for emergence percent in the combined analysis

Source	df	Mean Squares
Environment (Env.)	11	7727.32***
Replication in Env.	24	345.38
Genotype (Gen.)	12	4103.45***
Gen.x Env.	132	368.06**
Error	288	197.34

*** : Significant (P < 0.001)

** : Significant (P < 0.01)

Table 7. Eemergence Percent for genotypes in 1983 fall planting, Oklahoma

Genotype			Environmen	t
Species	Cultivar	StF83	HaF83	WoF83
M. arabica M. littoralis M. littoralis M. minima	Harbinger 1 Harbinger 2	75 88 52 65	54 59 43 45	77 87 50 59
M. polymorpha M. rugosa M. rugosa M. rugosa M. rugosa	Circle Valley Paragosa Paraponto 1 Paraponto 2 Sapo	87 60 55 nd(a) 18	47 40 26 nd 24	88 57 42 nd 8
M. sativa M. sativa M. tornata M. tornata	OK-83-Graze Spredor II Tornafield 1 Tornafield 2	63 81 28 38	38 58 33 31	68 56 25 32
M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Paraggio	55 51 37 67	49 48 44 52	36 52 32 67
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	37 49 78 91	31 32 38 48	38 33 76 95
LSD (P = 0.05)		11	17	13

⁽a) : nd = not determined

Table 8. Emergence percent for genotypes at various nitrogen supply levels in Haskell (limed site), OK.

Genotype		Nitrog	en Supply	levels(1)
Species	Cultivar	I	II	III
M. arabica M. littoralis M. littoralis M. minima	Harbinger 1 Harbinger 2	82 65 41 87	80 62 37 77	89 95 67 96
M. polymorpha M. rugosa M. rugosa M. rugosa	Circle Valley Paragosa Paraponto 1 Sapo	87 50 17 14	93 72 14 10	92 75 29 11
M. sativa M. sativa M. tornata M. tornata	OK-83-Graze Spredor II Tornafield 1 Tornafield 2	86 86 36 49	92 93 37 48	91 97 46 55
M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Paraggio	61 57 33 76	59 53 32 60	43 67 39 98
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	60 43 89 77	36 38 81 92	53 57 93 96
LSD (P = 0.05)		21	17	22

^{(1):} I = No artificial Rhizobium inoculation and no N fertilizer II = Artificial Rhizobium inoculation only

III = N fertilizer only

Table 9. Emergence percent for genotypes evaluated separately at Stillwater, OK. 1983/84

Species	Cultivar '	SsF83	SsS84
M. aculeata M. blancheana M. murex M. polymorpha M. sativa T. vesiculosum	Strain 1 OK-83-Graze Yuchi	36 57 27 30 64 69	18 27 20 19 44 49
LSD (P = 0.05)		16	8

Table 10. Emergence percent for genotypes in 1984 spring planting, at Oklahoma

Genotype		Environment		
Species	Cultivar	StS84	H1S84	HaS84
M. littoralis	Harbinger 1	41	14	9
M. littoralis M. minima	Harbinger 2	76 8	17 14	13 10
M. polymorpha	Circle Valley	83	37	19
M. rugosa	Paragosa	66	29	11
M. rugosa	Paraponto 1	19	14	2
M. rugosa	Sapo	nd(a)	25	nd
M. sativa	OK-83-Graze	67	17	12
M. sativa	Spredor II	66	16	10
M. scutellata	Robinson	48	31	nd
M. scutellata	Sava	nd	35	17
M. tornata	Tornafield 1	22	13	12
M. tornata	Tornafield 2	20	16	3
M. truncatula	Borung	32	20	7
M. truncatula	Cyprus	42	15	11
M. truncatula	Jemalong 1	23	15	6
M. truncatula	Jemalong 2	77	nd	14
M. truncatula	Paraggio	56	28	11
T. incarnatum		18	9	7
T. pratense	Redland	15	nd	13
[. subterraneum	Mount Barker	51	45	31
T. vesiculosum	Yuchi	42	24	8
LSD (P = 0.05)		11	10	6

⁽a) : nd = not determined

Table 11. Emergence percent for genotypes at various nitrogen supply levels in Woodward, OK., in spring planting

Genotype		Nitrog	en Supply 1	evels(1)
Species	Cultivar	I	II	III
M. littoralis M. littoralis M. minima M. polymorpha	Harbinger 1 Harbinger 2 Circle Valley	35 20 59 28	31 52 43 42	33 59 21 41
M. rugosa M. rugosa M. rugosa M. sativa	Paragosa Paraponto 1 Sapo OK-83-Graze	44 31 15 28	28 35 29 58	32 17 5 45
M. sativa M. scutellata M. tornata M. tornata	Spredor II Robinson Tornafield 1 Tornafield 2	60 55 12 10	21 26 30 18	54 17 36 39
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	22 63 55 43 41	58 34 48 32 18	55 26 16 38 21
T. incarnatum T. subterraneum T. vesiculosum	Mount Barker Yuchi	21 55 35	35 54 22	33 56 59
LSD (P = 0.05)		18	21	20

^{(1):} I = No artificial Rhizobium inoculation and no N fertilizer II = Artificial Rhizobium inoculation only III = N fertilizer only

Table 12. Emergence percent for genotypes in 1984 fall planting, at Oklahoma

Genoty	/pe		Environmen	t
Species	Cultivar	PeF84	H1F84	WoF84
1. littoralis	Harbinger 1	62	45	63
1. littoralis	Harbinger 2	nd(a)	54	45
1. polymorpha	Circle Valley	65	49	68
1. rugosa	Paragosa	41	35	63
1. rugosa	Paraponto 1	24	26	56
l. rugosa	Sapo	16	47	nd
l. sativa	OK-83-Graze	72	85	69
l. sativa	Spredor II	69	80	84
1. scutellata	Robinson	53	40	61
1. scutellata	Sava	51	33	81
1. tornata	Tornafield 1	37	35	40
1. tornata	Tornafield 2	43	nd	44
1. truncatula	Borung	61	55	47
1. truncatula	Cyprus	66	67	49
1. truncatula	Jemalong 1	56	29	55
1. truncatula 1. truncatula 1. truncatula	Jemalong 2 Paraggio	63 56	57 62	60 71
[. incarnatum		34	51	50
. pratense	Redland	36	34	37
. subterraneum	Mount Barker	64	46	65
. vesiculosum	Yuchi	57	63	72
.SD (P = 0.05)		17	18	ns(b

⁽a) : nd = not determined
(b) : ns = not significant (P > 0.05)

Table 13. Emergence percent for genotypes in 1985 spring planting, at Oklahoma

Genoty	pe		Environmen	t
Species	Cultivar	PeS85	H1S85	WoS85
M. littoralis M. littoralis	Harbinger 1 Harbinger 2	17 nd(a)	34 17	1 2 9
M. polymorpha M. rugosa	Circle Valley Paragosa	22 12	46 23	9 1
M. rugosa M. rugosa M. sativa M. sativa	Paraponto 1 Sapo OK-83-Graze Spredor II	10 9 14 31	18 24 19 24	1 2 6 1
M. scutellata M. scutellata M. tornata M. tornata	Robinson Sava Tornafield 1 Tornafield 2	33 35 27 21	42 29 20 16	5 6 nd 1
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	12 18 6 33 25	15 32 19 nd 43	6 3 1 2 1
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	14 4 11 12	16 26 26 27	1 1 4 2
LSD (P = 0.05)		8	22	ns(b)

⁽a) : nd = not determined
(b) : ns = not significant (P > 0.05)

Table 14. Emergence percent for genotypes in 1985 fall planting, in Morocco

Genoty	pe		Environme	nt
Species	Cultivar	SiF85	JmF85	TeF85
M. littoralis	Harbinger 1	37	96	33
M. littoralis	Harbinger 2	50	54	48
M. polymorpha	Circle Valley	53	69	47
M. polymorpha	Serena	52	87	36
M. rugosa	Paragosa	44	43	40
M. rugosa	Paraponto 1	40	16	41
M. rugosa	Sapo	22	25	32
M. sativa	OK-83-Graze	87	87	42
M. sativa	Spredor II	55	73	30
M. scutellata M. scutellata M. tornata M. tornata	Robinson	29	22	28
	Sava	89	63	47
	Tornafield 1	42	50	47
	Tornafield 2	40	48	37
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung	82	63	38
	Cyprus	37	41	41
	Jemalong 1	36	52	30
	Jemalong 2	56	73	49
	Paraggio	58	76	45
T. incarnatum T. subterraneum T. vesiculosum	Mount Barker Yuchi	24 45 34	29 56 66	39 28 33
LSD (P = 0.05)		25	16	ns(b)

⁽b) : ns = not significant (P > 0.05)

Table 15. Emergence percent for genotypes in 1986 fall planting, in Morocco

Genot	уре	Enviro	nment
Species	Cultivar	SiF86	TeF86
M. littoralis	Harbinger 1	6	10
M. littoralis	Harbinger 2	8	2
M. polymorpha	Circle Valley	15	13
M. polymorpha	Serena	10	5
M. rugosa	Paragosa	3	1
M. rugosa	Paraponto 1	5	1
M. rugosa	Sapo	2	3
M. scutellata	Robinson	6	4
M. scutellata	Sava	17	7
M. tornata	Tornafield 1	3	2
M. tornata	Tornafield 2	1	3
M. truncatula	Borung	4	9
M. truncatula	Cyprus	4	9
M. truncatula	Jemalong 1	1	7
M. truncatula	Jemalong 2	3	8
M. truncatula	Paraggio	4	10
LSD (P = 0.05)		8	5

Table 16. Mean squares in the regression analysis for emergence

Source	df	MS
Environment (Env.) Env. (linear) Replication in Env. Genotype (Gen.) Gen.X Env.	11 1 24 12 132	2574.01*** 28066.88*** 115.11 1362.28*** 121.68**
Gen. X Env. (linear)	12	212.85***
Pooled deviations	120	114.63
Harbinger 1	10	129.44
Circle Valley	10	92.35
Paragosa	10	104.76
Paraponto 1	10	182.46
OK-83-Graze	10	312.26
Tornafield 1	10	60.13
Borung	10	60.58
Cyprus	10	105.81
Jemalong 1	10	48.30
Paraggio	10	64.21
Crimson clover	10	57.82
Mount Barker	10	67.32
Yuchi	10	90.10
Pooled error	288	65.78

*** : Significant (P < 0.001). ** : Significant (P < 0.01). * : Significant (P < 0.05).

Table 17. Regression coefficients (bi) and residuals (sdi².) about the regression line for emergence

	Species	Cultivar	bi	sdi ²
Μ.	littoralis	Harbinger 1	1.29**	63.66*
Μ.	polymorpha	Circle Valley	0.95	26.57
Μ.	rugosa	Paragosa	0.70***	38.98
Μ.	rugosa	Paraponto 1	0.60***	116.68***
Μ.	sativa	OK-83-Graze	1.59***	246.48***
Μ.	tornata	Tornafield 1	0.60***	0.00
Μ.	truncatula	Borung	1.12*	40.03
Μ.	truncatula	Cyprus	0.84	0.00
М.	truncatula	Jemalong 1	0.80**	0.00
Μ.	truncatula .	Paraggio	0.97	0.00
Т.	incarnatum		0.60***	0.00
Т.	subterraneum	Mount Barker	0.89	1.54
Τ.	vesiculosum	Yuchi	1.32***	24.32

^{*** :} bi and ${\rm sdi}^2$ significantly (P < 0.001) different from 1.00 and 0.00, respectively.

^{** :} bi and ${\rm sdi}^2$ significantly (P < 0.01) different from 1.00 and 0.00, respectively.

^{* :} bi and ${\rm sdi}^2$ significantly (P < 0.05) different from 1.00 and 0.00, respectively.

Table 18. Mean squares (MS \times 10⁻²) for cold damage at each environment of Oklahoma

		Environmen	ts of 1983	· .	
Source	df	StF83	H1F83	HaF83	WoF83
Replication Nitrogen	3	395	11.5	6.0	361
Supply (Nit.)	2	150	5.0	1.7	665
Error (a)	6	3053	9.4	2.2	464
Genotype (Gen.)	19	3173***	87.0***	90.2***	1052***
Gen. X Nit.	38	134	6.7	6.5	70
Error (b)	171	188	6.9	4.9	123

Table 18. (continued)

		Environmen	ts of 1984	
Source	PeF84	H1F84	WoF84	SsF83
Replication Genotype (Gen.) Error	(2) 20a (19)776* (38)413	(3)165 (19)485 (57)378	(3)215 (19)414* (57)183	(3) 370 (5)1950*** (15) 393

a : Values in parantheses are the corresponding degrees of freedom.

*** : Significant (P < 0.001).

** : Significant (P < 0.01).

* : Significant (P < 0.05).

Table 19. Mean squares in the combined analysis for cold damage in Oklahoma in 1983 and 1984

		1983		1984
Source	df	MS	df	· MS
Environment (Env.)	3	75.7***	2	392.5***
Replication in Env.	12	0.7	6	0.9
Genotype (Gen.)	19	8.2***	17	6.6***
Gen. X Env.	57	2.6***	34	2.9
Error	228	0.2	102	2.3

^{*** :} Significant (P < 0.001).

Table 20. Cold damage grades of genotypes in fall plantings at various environments of O(1)

Genotype	-				Environ	nment		
Species	Cultivar	StF83	H1F83	HaF83	WoF83	PeF84	H1F84	WoF84
M. arabica M. littoralis M. littoralis M. minima M. polymorpha	Harbinger 1 Harbinger 2 Circle Valle	6.0 8.0 8.0 7.9 y 4.4	9.0 9.0 9.0 9.0	9.0 9.0 9.0 9.0	7.7 8.7 9.0 8.5 8.3	nd(a) 7.6 nd nd 6.1	nd 5.5 3.0 nd 2.5	nd 7.8 9.0 nd 8.5
M. rugosa M. rugosa M. rugosa M. sativa M. sativa	Paragosa Paraponto 1 Sapo OK-83-Graze Spredor II	8.1 8.9 8.8 5.8 6.3	9.0 9.0 9.0 8.3 8.1	9.0 9.0 9.0 8.3 7.9	8.9 9.0 9.0 6.6 6.8	6.9 7.4 6.9 4.3 4.7	4.0 2.5 4.8 1.8 1.8	6.3
M. scutellata M. scutallata M. tornata M. tornata	Robinson Sava Tornafield 1 Tornafield 2		nd nd 9.0 9.0	nd nd 9.0 9.0	nd nd 9.0 9.0	7.6 7.6 5.7 6.2	3.3 2.5 2.5 nd	8.3 9.0 9.0 8.3
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	7.4 7.8 7.7 nd 7.2	9.0 9.0 9.0 nd 9.0	9.0 9.0 9.0 nd 9.0	8.9 8.8 8.9 nd 8.7	7.0 6.1 7.0 5.9 7.2	3.3 3.3 1.8 2.5 3.3	8.0 9.0 9.0 9.0 8.3
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	4.5 8.1 3.1 4.0	9.0 9.0 9.0 8.4	8.9 9.0 8.9 8.5	7.0 8.7 7.1 5.9	3.8 6.9 3.8 4.9	1.0 1.8 4.0 2.5	7.5 9.0 6.3 6.3
LSD (P = 0.05)		1.1	0.2	0.2	0.9	1.7	ns(b) 1.9

^{(1):} Cold damage was graded visually through a scale of 1 to 9, with 1 = no apparent injury and 9 = leaves and stems were apparently dead.

⁽a) : nd = not determined; (b) : ns = not significant (P > 0.05)

Table 21. Mean squares (MS \times 10^{-2}) for forage yield at each environment

		Environmen	t
Source	StF83	StS84	H1S84
Replication	1267	821	79
litrogen Supply (Nit.)	45	5	148
rror (a)	541	348	66
Genotype (Gen.)	12399***	9679***	28
Nit. X Gen.	651	234	25
Error (b)	447	227	38

Table 21. (continued)

			invironment		
Source	PeS85	H1S85	SiF85	JmF85	TeF85
Replication Genotype (Gen.) Error	378 1267*** 390	3888** 5988*** 681	10320* 23377*** 3183	25390*** 28146*** 2592	8983* 85977*** 2526

*** : Significant (P < 0.001).

** : Significant (P < 0.01).

* : Significant (P < 0.05).

Table 22. Mean squares for forage yield in the combined analyses

	0	klahoma		Morocco			
Source	df	df MS df		MS			
Environment (Env.)	2	6202*	2	165000***			
Replication in Env.	6	1183	9	14857			
Genotype (Gen.)	16	5145***	20	90154***			
Gen. x Env.	32	968***	40	23046***			
Error	96	336	180	5411			

Table 23. Dry matter yield (g/m) of genotypes in Oklahoma

Genotyp	e			Enviro	nment	
Species	Cultivar	StF83	StS84	H1S84	PeS85	H1S85
M. arabica M. littoralis M. littoralis M. minima	Harbinger 1 Harbinger 2	13 0 0 2	nd(a) 58 71 2	nd 4 5 4	nd 34 nd nd	nd 16 13 nd
M. polymorpha M. polymorpha M. rugosa M. rugosa M. sativa M. sativa	Circle Valley Paragosa Paraponto 1 Sapo OK-83-Graze Spredor II	21 0 0 0 98 82	109 53 24 nd 40 39	4 4 3 7 3 8	40 15 18 14 8 21	52 tr(b) tr tr 86 75
M. scutellata M. scutellata M. tornata M. tornata	Robinson Sava Tornafield 1 Tornafield 2	nd nd 0	104 nd 36 33	4 4 5 4	67 72 27 35	112 125 16 14
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	0 0 0 nd 0	41 62 41 70 65	2 6 4 nd 8	23 36 20 36 57	56 42 15 nd 74
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	35 43 48 81	20 10 21 22	5 nd 4 4	10 1 3 1	7 tr 28 20
LSD (P = 0.05)		17	14	ns(c)	33	37

⁽a): nd = not determined
(b): Less than 1 g/m
(c): not significant (P > 0.05)

Table 24. Dry matter yield (g/m) of genotypes from 1985 fall planting in Morocco

			Environment	
Genoty	rpe	Sidi el	Jemaa	Tes-
Species	Cultivar	Aydi	Shaim	saout
M. littoralis	Harbinger 1	118	184	112
M. littoralis	Harbinger 2	124	159	148
M. polymorpha	Circle Valley	50	282	251
M. polymorpha	Serena	101	197	233
M. rugosa M. rugosa	Paragosa Paraponto 1	57 52	213 74	188 116
M. rugosa	Sapo	18	82	69
M. sativa	OK-83-Graze	22	99	40
M. sativa	Spredor II	33	65	41
M. scutellata M. scutellata M. tornata M. tornata	Robinson Sava Tornafield 1 Tornafield 2	124 153 218 214	268 305 157 155	606 491 75 55
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	285 113 73 101 151	233 136 169 286 329	205 128 103 260 223
T. incarnatum T. subterraneum T. vesiculosum	Mount Barker Yuchi	39 1 1	145 50 94	71 66 53
LSD (P = 0.05)		80	72	71

Table 25. linear regression (b) and simple correlation coefficients (r) between forage yield and emergence percentage in each environment

Environment	All genot		No clover no snail	medics
Environment	b	r	b	r
StS84	0.84***	0.69	0.67**	0.72
PeS85	1.67***	0.78	0.84*	0.33
H1S85	1.87*	0.45	0.73	0.28
SiF85	1.29	0.32	2.03	0.51
JmF85	0.88	0.23	1.73	0.52
TeF85	2.88	0.03	0.22	0.10

*** : Linear regression significant (P < 0.001)
** : Linear regression significant (P < 0.01)
* : Linear regression significant (P < 0.05)

Table 26. linear regression (b) and simple correlation coefficients (r) between forage yield and emergence percentage for medics

Species	b	r	
 M. littoralis	1.51*	0.72	
M. polymorpha	2.00	0.39	
M. rugosa	1.85	0.49	
M. scutellata	0.31	0.10	
M. tornata	4.61**	0.75	
M. truncatula	3.01***	0.66	

*** : Linear regression significant (P < 0.001) ** : Linear regression significant (P < 0.01) * : Linear regression significant (P < 0.05)

Table 27. Mean squares (MS) for CP, NDF, ADF, and leaf:stem ratio at each environment

Source(1)	StS84	PeS85	H1S85	SiF85	JmF85	TeF85
CP Replication Genotype Error	2.7 8.9** 2.0	2.8 8.7 7.9	1.5 9.4* 3.7	9.3 16.5*** 4.2	5.9 8.4*** 2.2	2.4 3.8 2.5
NDF Replication Genotype Error	100.2* 32.4 20.2	217.5** 49.5 26.4	14.3 26.5 53.0	8.5 22.0*** 5.0	24.8* 24.2* 11.3	5.0 32.2*** 5.6
ADF Replication Genotype Error	80.9** 23.7 14.9	194.1** 47.4 25.6	46.4 31.2 45.5	20.2** 12.6*** 3.0	3.6 8.5 6.4	4.5 27.6** 9.4
Leaf:Stem Ratio Replication Genotype Error	nd(a)	nd	nd	0.3 1.2** 0.3	0.7 1.0** 0.3	0.5 0.2 0.4

^{(1) :} Three replications and 20 genotypes used in StS84 and HIS85. Three replications and 19 genotypes used in PeS85. Four replications and 16 genotypes used in SiF85, JmF85, and TeF85.

(a) : nd = not determined

^{*** :} Significant (P < 0.001).
** : Significant (P < 0.01).
* : Significant (P < 0.05).

Table 28. Mean squares in the combined analysis for CP, NDF, and ADF

		СР	_	-		NDF		
Source	5 L	ocations MS		<u>0</u> k]	lahoma MS		Mo df	rocco MS
Environment (Env.) 4	65.8***		2	94.8		2	80.6*
Replication in En		3.9		6	102.8		9	12.8
Genotype (Gen.)	11	5.7		16	26.2		15	58.0***
Gen. X Env.	44	7.1**		32	35.4		30	10.2
Error	110	3.2		96	35.9		135	7.3

Table 28. (continued)

	ADF				Leaf:Stem ratio		
Source	Oklahor	ma + TeF85	SiF85	+ JmF85	Morocco	1985/86	
	df	MS	df	MS	df	MS	
Environment (Env.		200.7	1	96.3	2	18.9***	
Replication in Eng		81.1	6	12.0	9	0.5	
Genotype (Gen.)		33.5	15	12.5**	15	1.4***	
Gen. X Env.		31.6	15	8.5	30	0.5	
Error		26.0	90	4.7	135	0.4	

^{** :} Significant (P < 0.001).

** : Significant (P < 0.01).

* : Significant (P < 0.05).

Table 29. Crude protein percentages of genotypes in various environments

		Environment					
Species	Cultivar	StS84	PeS85	H1S85	SiF85	JmF85	TeF85
M. littoralis M. littoralis M. minima M. polymorpha	Harbinger 1 Harbinger 2 Circle Valley	23.2 22.1 24.7 19.8	18.4 nd(a) nd 20.8	20.3 21.6 nd 17.6	23.2 nd nd 23.4	22.4 nd nd 24.3	21.3 nd nd 20.8
M. polymorpha M. rugosa M. rugosa M. rugosa M. sativa	Serena Paragosa Paraponto 1 Sapo OK-83-Graze	nd 22.8 22.6 nd 22.8	nd 21.5 21.0 20.8 20.6	nd 23.4 20.4 18.9 21.0	19.7 21.4 21.7 21.8 27.4	21.8 21.0 24.6 23.5 23.3	21.5 22.4 22.3 21.2 23.8
M. sativa M. scutellata M. scutellata M. tornata M. tornata	Spredor II Robinson Sava Tornafield 1 Tornafield 2	23.9 24.4 nd 23.6 22.7	25.2 25.1 24.1 21.3 21.3	22.5 19.6 19.8 18.8 20.0	nd 19.9 21.9 23.5 24.9	nd 21.8 21.1 21.5 22.5	nd 20.3 19.6 22.0 20.5
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	23.7 21.0 21.7 22.6 23.7	20.5 20.4 22.4 22.3 20.7	21.5 20.0 21.3 nd 21.2	25.9 21.8 21.6 23.1 23.1	22.6 23.0 25.4 25.0 24.3	21.5 19.4 22.5 21.1 21.0
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	20.5 19.4 23.3 22.5	23.8 20.8 23.3 22.2	19.7 20.4 18.8 21.3	nd nd nd nd	nd nd nd nd	nd nd nd nd
LSD (P = 0.05)		2.4	ns(b)	3.2	3.0	2.1	ns

⁽a) : nd = not determined
(b) : ns = not significant (P > 0.05)

Table 30. Neutral detergent fiber percentages of genotypes in various environments

			· ·	Env	vironme	nt	
Species	Cultivar	StS84	PeS85	H1S85	SiF85	JmF85	TeF85
M. littoralis M. littoralis M. minima M. polymorpha	Harbinger 1 Harbinger 2 Circle Valley	37.2 34.6 31.2 30.5	28.7 nd(b) nd 28.2	36.2 40.3 nd 34.2	33.5 nd nd 32.2	29.8 nd nd 31.0	35.2 nd nd 31.5
M. polymorpha M. rugosa M. rugosa M. rugosa M. sativa	Serena Paragosa Paraponto 1 Sapo OK-83-Graze	nd 35.8 38.3 nd 40.6	nd 34.1 40.9 38.8 32.4	nd 36.4 36.9 31.7 32.9	34.0 31.0 32.5 32.5 27.5	30.0 30.5 33.8 32.5 26.5	34.3 29.5 30.8 32.3 28.0
M. sativa M. scutellata M. scutellata M. tornata M. tornata	Spredor II Robinson Sava Tornafield 1 Tornafield 2	38.3 37.0 nd 36.8 34.9	38.0 30.4 32.5 35.4 32.2	32.3 30.0 32.3 33.4 29.0	nd 30.0 30.2 32.2 30.0	nd 29.2 28.2 28.0 27.0	nd 31.8 36.3 30.5 31.0
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	38.3 34.2 33.5 34.5 29.9	27.1 31.3 32.5 29.0 27.4	37.6 30.8 35.4 nd 34.3	33.8 36.2 35.0 35.5 34.0	30.0 34.5 33.0 34.5 31.2	33.5 39.3 33.8 34.8 30.8
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	29.9 31.1 30.3 32.6	28.6 37.0 33.4 32.7	39.4 32.7 34.0 34.7	nd nd nd nd	nd nd nd nd	nd nd nd nd
LSD (P = 0.05)		ns(b) ns	ns	3.2	4.8	3.4

⁽a) : nd = not determined
(b) : ns = not significant (P > 0.05)

Acid detergent fiber percentages of genotypes in vaious environments Table 31.

		Environment					
Species	Cultivar S	tS84	PeS85	H1S85	SiF85i	JmF85	TeF85
M. littoralis M. littoralis M. minima M. polymorpha M. polymorpha	Harbinger 1 Harbinger 2 Circle Valley Serena	26.2 21.9 20.0 20.1 nd	19.3 nd(a) nd 23.6 nd	23.3 30.7 nd 27.2 nd	21.5 nd nd 19.5 22.2	20.0 nd nd 21.5 21.2	25.5 nd nd 19.3 23.0
M. rugosa M. rugosa M. rugosa M. sativa M. sativa	Paragosa Paraponto 1 Sapo OK-83-Graze Spredor II	24.0 26.8 nd 28.1 27.8	27.6 30.6 31.6 31.2 28.6	32.5 23.5 29.4 26.3 24.6	18.2 20.0 18.8 23.0 nd	18.0 18.5 19.2 20.2 nd	17.8 20.8 18.5 18.2 nd
M. scutellata M. scutellata M. tornata M. tornata	Robinson Sava Tornafield 1 Tornafield 2	26.9 nd 23.2 24.9	24.0 26.0 28.5 23.6	24.4 26.4 24.0 20.1	21.8 22.0 21.0 19.8	22.5 19.0 18.5 17.5	22.0 26.0 22.0 17.8
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung Cyprus Jemalong 1 Jemalong 2 Paraggio	26.4 24.0 23.0 21.9 19.0	20.5 25.1 27.1 22.8 21.0	28.5 24.1 29.6 nd 24.9	21.8 25.2 22.8 22.5 21.5	19.5 18.0 20.5 21.0 18.5	21.0 24.5 20.6 22.2 21.0
T. incarnatum T. pratense T. subterraneum T. vesiculosum	Redland Mount Barker Yuchi	19.6 22.3 22.5 22.0	23.3 30.7 30.1 30.0	30.2 23.3 23.4 23.8	nd nd nd nd	nd nd nd nd	nd nd nd nd
LSD (P = 0.05)		ns(b)) ns	ns	2.5	ns	4.4

⁽a) : nd = not determined
(b) : ns = not significant (P > 0.05)

Table 32. Leaf:stem ratio of genotypes at each environment in Morocco from 1985 fall planting

Genotype		Environment				
Species	Cultivar	SiF85	JmF85	TeF85	Mean	
M. littoralis	Harbinger 1	2.8	2.5	2.0	2.4	
M. polymorpha	Circle Valley	3.0	2.5	1.8	2.4	
M. polymorpha	Serena	2.3	2.0	1.8	2.0	
M. rugosa	Paragosa	2.5	2.3	1.8	2.2	
M. rugosa	Paraponto 1	2.5	2.5	1.8	2.3	
M. rugosa	Sapo	2.3	2.5	2.3	2.3	
M. sativa	OK-83-Graze	2.8	3.5	2.3	2.8	
M. scutellata	Robinson	2.3	2.5	1.8	2.2	
M. scutellata	Sava	2.3	1.8	1.3	1.8	
M. tornata	Tornafield 1	3.8	3.3	1.8	2.9	
M. tornata	Tornafield 2	4.0	3.5	1.8	3.1	
M. truncatula	Borung	2.3	2.5	1.5	2.1	
M. truncatula	Cyprus	2.8	2.5	1.8	2.3	
M. truncatula	Jemalong 1	3.3	2.8	1.5	2.5	
M. truncatula	Jemalong 2	3.0	2.0	1.8	2.3	
M. truncatula	Paraggio	3.3	2.8	1.8	2.6	
LSD (P = 0.05)		0.8	0.8	0.9	0.5	

Table 33. Simple correlation coefficients between forage yield and quality parameters in each environment

Envir- onment	Yield/ CP	Yield/ NDF	Yield/ ADF	CP/ NDF	CP/ ADF	NDF/ ADF
StS84	-0.05	0.11	0.04	0.41	0.34	0.91
PeS85	0.19	-0.41	-0.58	0.12	-0.17	0.84
H185	-0.04	-0.38	-0.13	0.31	0.31	0.59
SiF85	0.28	0.19	0.16	-0.27	0.06	0.32
JmF85	-0.08	-0.06	0.35	0.62	-0.17	0.40
TeF85	-0.52	0.22	0.16	-0.68	0.64	0.56

Table 34. Simple correlation coefficients between forage and quality parameters for medics and alfalfa

Species	Yield/ CP	Yield/ NDF	Yield/ ADF	CP/ NDF	CP/ ADF	NDF/ ADF
M. littoralis	0.49	-0.51	-0.44	0.35	0.25	0.93
M. polymorpha	0.45	-0.02	-0.43	-0.30	-0.61	0.30
M. rugosa	0.09	-0.68	-0.61	-0.14	-0.35	0.89
M. scutellata	-0.48	0.14	-0.29	0.15	0.23	0.78
M. tornata	0.62	-0.38	-0.62	0.33	-0.30	0.67
M. truncatula	0.51	0.06	-0.48	0.07	-0.36	0.55
M. sativa	0.25	-0.34	-0.90	0.69	-0.10	0.18

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CHAPTER III

SEED PRODUCTION AND RESEEDING OF ANNUAL MEDICAGO SPP.

Abstract

Productivity and persistence of legume-based pastures are strongly dependant on seed yield and seed quality. This study was designed to evaluate seed production and seed characteristics of annual medics when grown under non-irrigated, clean-tilled conditions of continental and Mediterranean climates, and to assess regeneration of these species in the U.S. Southern Great Plains. Two planting dates, fall and spring, were evaluated at Stillwater, OK. in 1983/84 and at Perkins, OK. in 1984/85. Fall planting was repeated in 1985 and 1986 at Sidi el Aydi, Morocco. Treatments were plant species represented by 23 annual Medicago spp., two grazing strains of perennial alfalfa (M. sativa L.), and four Trifolium spp. Seeds were inoculated with Rhizobium spp. and planted in a randomized complete design with three replications. Accessions were grown in single rows 2 m long and 1 m apart.

Fall plantings in Oklahoma resulted in winter kill of all annual Medicago species in Oklahoma except M. blancheana L. and M. polymorpha L. which produced 1,540 and 2,240 seeds/ m^2 , respectively. Spring planted annual medics produced more seed than fall planted in Oklahoma; however, genotypes and genotype X environment interactions had

significant (P < 0.001) effects on seed yield. Snail medic (M. scutellata L. (Mill.)) produced about 2,300 seeds/m² compared to alfalfa (M. sativa) with 3,560 seeds/m². All other annual Medicago species yielded less than 1,000 seeds/m², except M. truncatula Gaertn. Cyprus, which had 1,677 seeds/m² during the first year at Stillwater, OK. Spring-sown Trifolium spp., M. rugosa Desr., M. arabica (L.) Huds., and M. minima (L.) Mill. did not produce seed in Oklahoma. Total seed numbers ranged between 6,580 and 30,582 seeds/m² in annual Medicago grown at Sidi el Aydi from the 1985 fall planting. Negligible seed quantities were produced from the 1986 fall planting in Morocco with no seed from M. sativa and Trifolium spp. in either year. The highest producing species were M. truncatula, M. littoralis Rhode, and M. tornata (L.) Mill. having greater than 19,300 seeds/m² at Sidi el Aydi, Morocco, in 1985/86.

Genotype X environment interactions were significant (P < 0.001) for number of seeds/pod and percentage of hardseed. M. scutellata had the highest number of seeds/pod with 3.7 in Oklahoma, but M. truncatula had 6.0 seeds/pod at Sidi el Aydi in Morocco. All annual Medicago possessed a high proportion of hardseed except M. truncatula Paraggio (60%). Highly significant (P < 0.01) simple linear correlations existed among all seed properties measured: seed weight, total number of seeds, number of seeds/pod, 1000-seed weight, percent germination, and percent hardseed. Offspring plants regenerated in the subsequent fall of 1984 at Stillwater were 238, 165, and 180 plants/m² in M. scutellata Robinson and Sava, and M. sativa, respectively. Other accessions regenerated less than 20 plants/m² despite the high amount of seed produced indicating the possibility to soil seed reserve constitution in some

annual $\underline{\text{Medicago}}$; e.g., $\underline{\text{M}}$. $\underline{\text{truncatula}}$ Cyprus. All seedlings were exposed to frost and died. At Perkins there was no emergence in fall from pods produced in spring of 1985. Cold tolerance will be necessary for medics studied for successful use in cold environments.

Key words : <u>Trifolium</u>, annual medic, alfalfa, clover, G X E interaction, Oklahama, Morocco

Introduction

Characteristics of Annual Medic Seed

Quality and quantity of dry matter production are the most important concerns for forage crops. However, reliable production of viable seed in adequate quantities is imperative for annual pasture legume persistence. Natural selection tends to evolve certain traits which are not always conducive to commercial seed production and ease of establishment nor to palability by animals. Thick, tough spiny pods are difficult to thresh; however, they sometimes protect seeds from predators.

Annual medics (Medicago spp.), along with subclover (Trifolium subterraneum L.), are a major component in the ley-farming system in Australia (7), and are being reintroduced to North Africa and the Middle East (31). According to Heyn (23), annual medics originated in the Mediterranean basin which is characterized by a hot, dry summer season (5 - 8 months), occasional summer rainstorms, and high inter- and intraannual variability. To avoid seedling death during periods of erratic precipitations, these species have developed features such as thickwalled spined pods and hard (impermeable) seed coats which assists in seed dissemination and survival (15).

Seeds will not germinate immediately after maturation, either because of physiological embryo dormancy (1) and/or mechanical dormancy associated with an impermeable seed coat (26). Nearly 100% hard seed during the summer and autumn followed by a rapid change to at least 30% permeable seed by mid-October is the suggested optimum for natural pasture regeneration in Australia (15). However, Crawford (17)

indicated that some cultivars do not have a high level of hardseededness, e.g., \underline{M} . $\underline{truncatula}$ Gaertn. cv. Paraggio, and therefore are subjected to seedling loss during brief summer rains. On the other hand, extremely hardseeded cultivars might have limited plant density in pastures immediately after the establishment year in Australia (15).

High hardseed proportions are common among medics. From 3,583 genotypes representing 28 species, Crawford (15) indicated that only 6.8% of them exhibited more than 30% permeable seed after field exposure from maturation in November to mid-April. He also found that gama medic (M. rugosa Desr.) was the most permeable species (48%), followed by strand medic (M. tornata (L.) Mill.) (20%), while the others had less than 10%. Similar results were reported by Quinlivan (26). Association between seed coat permeability and number of seeds/burr were reported by Andrew (3) and Salisbury et al. (29) in medics and subclover.

Subjecting medic seed to low temperatures, to fluctuating high temperatures, and to scarification were shown by Quinlivan (25, 26) to be effective in breakdown of hard seed coats, thus promoting seed inbibition and germination. Aitken (1) reported that increased osmotic pressure also helped to break seed coats. Lipp and Ballard (24) demonstrated removal of physiological dormancy with carbon dioxide at 2.5%. Thiourea and 2-chloroethylphosphoric acid were reported to have similar results as carbon dioxide (20). The response to such treatments varied among medic species and was related to application conditions.

Seed Production in Medics

Economically optimum seeding rate for seed production in Australia is about 10 kg/ha as suggested by Ragless (27). Cocks (12) in Syria

found that seed yield increased with increased \underline{M} . $\underline{rigidula}$ L. seeding rate, but not in \underline{M} . \underline{noeana} L. and \underline{M} . $\underline{truncatula}$. He indicated that seed weight and number of seeds/pod were not related to plant density in these species. However, increased plant density resulted in low levels of hardseededness. When medics were interseeded with wheat ($\underline{Triticum}$ $\underline{aestivum}$ L.), 25-30 plants/m were required to ensure good seed production and regeneration (Brownlee and Scott (5)). On the other hand, Gintzburger (19) demonstrated better crop persistence when sowing pods than naked seed, particularly under rangeland conditions because of large quantities of hard seeds in pods.

Flower initiation in medics may be accelerated by a long photoperiod (>18 hours) or low temperature $(-1 \, ^{\circ}\text{C})$ in most species (except M. scutellata (L.) Mill.), and these two factors substitute for each other (9). Consequently, flower initiation occurs sooner for winter than for summer plantings as observed by Aitken (1) in Australia. In contrast, when vernalization was extended more than seven weeks, flower initiation was delayed in M. littoralis Rhode and M. truncatula (1, 9). Ruiter and Taylor (28) indicated that lack of vernalization is a minor factor in delaying flowering. They also showed that short photoperiod, e.g., 8 hrs., delayed flowering by 35 days. However, there was considerable variation among medic species in their level of response. After vernalization, increase in temperatures accelerated the rate of development of all growth stages in medics, except for flowering in M. scutellata, if temperatures did not exceed 30/23 °C day/night (11). Duration of daylight to which the mother plants are exposed during the last 8-12 days of seed maturation may affect seed coat color, structure, and permeability to water as well as germination (22). The duration of

the vegetative growth varies greatly between cultivars. In South Australia for 171 genotypes among 28 species, Crawford (14, 16) estimated the range from 65 to 164 days.

Cocks (12) has shown that potential seed yield is not achieved in medics due to massive flower abortion in Syria. This was also reported in clover species (29). Flower abortion usually reduces the number of seeds/burr, but differences between cultivars in the level of abortion and number of seeds/burr have been recorded (12, 29).

Water stress (10) and nitrogen deficiency (2) during the vegetative period were reported to delay the onset of flowering and reduce percentage of viable hard seed, especially when they occured at or after flowering. Gintzburger (19) indicated, however, that early-flowering medics were capable of producing limited amounts of viable seeds in severe conditions with as little as 100 mm of rain during the growing season and up to 16 days of frost at ground level. Some insects, e.g., blue green aphid (Acyrthosiphon kondoi Shinji) were reported to impair seed production in susceptible cultivars (27).

Seed yield and forage yield appear to behave independently, to a large extent, in \underline{M} . aculeata L. and \underline{M} . rigidula, according to Ceccarelli and Somaroo (8), who stated that selection of medic varieties combining high seed yield and high dry matter could be made. Crawford (13) reported the presence of significant associations between seedling vigor and seed weight, pod weight, and degree of spininess of pods, and between productivity and pod yield. Seed number/burr is reported to be a heritable trait in subclover (29). Many environmental variables affect seed production during either flowering stage or seed maturation. Seed yield ranging from 0 to 1,300 kg/ha was reported in Australia (14).

Medic Pasture Persistence

Pasture persistence depends on seed yield and hard seed in the seed bank at the top 5 cm of soil (7). Crop management, particularly grazing, may affect persistence. In summer, when forage is reduced by grazing, pods constitute a feed source to sheep. Seed and pod intake may be as high as 470-600 g of organic matter/ewe/day (18, 30). Carter and Lake (7) mentioned that grazing of medic pods by sheep could cause loss in seed reserve and pasture persistence because less than 2% of ingested seed escaped digestion (6, 30). Moreover, with intensive late spring and summer grazing, there was a highly significant linear decline in total pasture, medic pods, pod weight, seeds/pod, seed weight, and viable seeds (7). On the other hand, when light grazing is practiced in medic pastures during the growing season, Carter and Lake (7) noted a reduction in dry matter production. They concluded that excessive weed competition for light and nutrients caused these reductions.

The potential for seed production and reseeding of medics under Oklahoma continental climate and the Mediterranean climate in Morocco has not been investigated. The objectives of this study were to evaluate seed production and seed characteristics of annual medics when grown under dry clean-tilled conditions of continental and Mediterranean climates, and to assess regeneration of these species in Oklahoma. A secondary objective was to estimate the relationships existing among seed characteristics.

Materials and Methods

Site Description

Three locations were utilized in the study: Stillwater and Perkins in Oklahoma in the U.S. Southern Great Plains, which have a continental climate with very cold winters, and Sidi el Aydi in Morocco, which has the Mediterranean climate with temperatures rarely as low as 0°C. Physical and climatic descriptions of each site have been discussed previously (see Chapter II). Both fall and spring plantings were used in Oklahoma. The combination of sites and planting dates resulted in six environments under which the study was conducted. These environments are: Stillwater 1983 fall planting; Stillwater 1984 spring planting; Perkins 1984 fall planting; Perkins 1985 spring planting; Sidi el Aydi 1985 fall planting; and Sidi el Aydi 1986 fall planting.

Field Plot Organization

The field study in Oklahoma consisted of three types of management to respond to three distinct questions:

- I- To evaluate seed production potential, seed was harvested two months after plant senescence. This management type was the only one conducted at Sidi el Aydi, Morocco.
- II- To study the reseeding potential, pods and seeds were allowed to form and fall to the soil as the plants matured and senesced.
- III- To monitor the importance of late germination and emergence of original sown-seed, plants were harvested prior to seed set.
 In this part of the study no late emerging seedlings were

found; thus, there was no need to adjust data in the other management types to compensate for late emergence.

Each management type was arranged in a randomized complete block design with three replications. Treatments were plant accessions (genotypes) representing eight annual Medicago species, perennial alfalfa (M. sativa L.), and four clovers (Trifolium spp.). There were 25 plant accessions as identified in Table 1. Seed availability for certain entries was limited and it was necessary to interchange some accessions among environments. Those evaluated in a given environment are listed in Table 1. Medic accessions represented wide ranges in maturation and seed size (4, 23).

Four medic species, M. aculeata, M. blancheana L., M. murex Willd., and M. polymorpha L. (strain 1) were studied in a separate experiment to compare their seed production to that of alfalfa and arrowleaf clover (T. vesiculosum Savi). Plants were grown at Stillwater during two growing seasons; fall of 1983 and spring of 1984. The design was a randomized complete block with four replications and treatments were plant species.

Conduct of Experiments

All entries were tested for germination and those possessing a high proportion of hard seed were scarified with sandpaper. Seeds were preinoculated with proper <u>Rhizobium</u> spp. and hand-sown at the rate of 100 viable seed per linear meter. Plot sizes (experimental units) were single rows 2 m long and 1 m apart. Phosphorus and potassium in the soil were adjusted with fertilizer application according to Oklahoma State University soil test recommendations with reference to alfalfa,

and 40 kg N/ha were applied at sowing. Planting dates were in the third week of October or March in Oklahoma and in the third week of November in Morocco. All experiments were under non-irrigated clean-tilled conditions and weeded by hand.

Observations and Measurements

From plots on seed production (management type I), pods produced were harvested after full maturation. They were collected with a vacuum cleaner after slight soil disruption. They were separated from soil by sieving, then rapidly rinsed with water to remove dust and dried at 40 °C for storage and quality testing. Total clean pods from each plot were weighed and 20 randomly-chosen pods were weighed and hand-decorticated to count number of seeds/pod and for germination tests. The other pods were mechanically threshed and seeds were cleaned. Seeds were weighed and the weight of 1,000 seeds was measured. Total number of seeds was calculated from total seed weight and the weight of 1,000 seeds. Immature seeds were not included in measurements and counts. Germination tests were made on four 25-seed samples with and without scarification about two months after harvest. It was conducted in Petri dishes under dark conditions at 22 \pm 3 $^{\circ}$ C. Final evaluations were made after 11 days. Hard seeds were those which did not inbibe water during germination tests without scarification. They were included in germinating seeds in addition to those which germinated normally (sprouts with root and shoot).

In plots conducted for regeneration (management type II), where pods were allowed to fall on soil to constitute soil seed bank, emerged seedlings at Stillwater 1984 in the subsequent growing season were

counted. No emergence resulted from the pods left on the soil at Perkins for 15 months.

Statistical Analysis

Data were subjected to analysis of variance and comparisons among genotypes were made with protected least significant difference (LSD) at P=0.05 (21) in each environment. Combined analyses of variance were performed on environments with homogeneous variances using the 'fixed' model; environments and genotypes were considered not random. These analyses were calculated to examine genotypic, environmental, and genotype X environment (G X E) interaction effects. Where significant G X E effects were present, data are exhibited for all genotypes in each environment. Simple linear correlations between all combinations of seed characteristics were calculated and tested against zero with t tests to detect the type of associations existing between pairs of seed characters.

Results and Discussion

Data were collected and analyzed from management type I from spring planting at Stillwater (1984) and Perkins (1985) and from 1985 fall planting at Sidi el Aydi. Management type II was analyzed only at Stillwater. The observations that no late emergence occurred in the management type III plots clearly indicate that this precaution was unnecessary because sown seeds either germinated within a few days or not at all. All fall plantings in Oklahoma resulted in the clear conclusion that cold tolerance in these medics was insufficient for winter survival in a continental climate; thus, any potential seed production must come from spring plantings.

The drought caused seed crop failure in the 1986/87 planting at Sidi el Aydi (< 150 mm rainfall from Oct. 1986 to May 1987). This demonstrated that, even in the center of origin for medics, seed production is not assured on an annual basis. It also clearly indicated the necessity to work with medics having reliable seed dormancy so that the soil seed bank can provide seed for reestablishment more than one growing season later.

Seed Production

Seed yield is an important parameter which, when high in terms of quantity, maximizes the returns. It does have a special meaning for selfseeded legume pastures in terms of number of seeds which will constitute a soil seed bank and regenerate the pasture after grazing and/or herbicide control. Therefore, it appeared judicious to present seed yield with these two aspects. Homogeneity of variances revealed

that Oklahoma environments presented much lower error mean squares for seed production and seed characteristics when compared to the Moroccan one (Table 2). Therefore, comparisons among environments were limited to those of Oklahoma.

Total Seed Weight

Spring planting of medics was much more successful than fall planting in Oklahoma. Total seed weight produced differed significantly (P < 0.001) among genotypes grown in spring at Stillwater in 1984 and at Perkins in 1985 (Table 2). The combination of the two environments showed no effect (P > 0.05) due to environment but differences due to genotypes and genotype X environment interactions were detected (P < 0.001) (Table 3).

The highest total seed weight was achieved with snail medics (\underline{M} . scutellata (L.) Mill) 'Robinson' and 'Sava' which produced 21.0 and 17.7 g/m², respectively, at Stillwater (Table 4). Alfalfa (\underline{M} . sativa) produced 12.7 g/m². Other annual medic genotypes and arrowleaf clover did not differ statistically (P > 0.05) in total seed weight and did not differ from zero although the plants grew vegetatively. Species evaluated separately at Stillwater in the spring of 1984 also had very low total seed weight. \underline{M} . blancheana yielded 3.2 g/m² and was significantly (P < 0.05) different from \underline{M} . polymorpha, \underline{M} . aculeata, and \underline{M} . murex. Respective total seed weights of these accessions were 1.5, 0.4, and 0 g/m² (LSD (P = 0.05) = 1.1 g/m²). Alfalfa produced 10.3 g/m² and was higher than medics.

At Perkins, snail medics Robinson and Sava produced the highest total seed weight, as they did at Stillwater, with 33.9 and 32.8 g/m^2 ,

respectively, in 1985 spring planting (Table 4). Strains of alfalfa did not differ (P > 0.05) and yielded 10.0 and 10.1 g/m^2 of seed. However, they were lower than snail medics. Barrel medic (M. truncatula) 'Cyprus' was the only other medic accession to produce (2.7 g/m^2) significantly (P < 0.05) more than zero.

In Morocco, genotypes planted at Sidi el Aydi in the fall of 1985 differed significantly (P < 0.001) in the total seed weight produced (Table 2). Snail medics (M. scutellata) Robinson and Sava yielded the highest total seed weight as they did in both Oklahoma environments, but were not significantly (P > 0.05) greater than barrel medics (M. truncatula) 'Paraggio' and 'Borung' or disc medic (M. tornata)

Tornafield 1. Total seed weight in these cultivars ranged between 112.7 and 132.2 g/m² (Table 4). The lowest yielding cultivar was gama medic (M. rugosa) 'Sapo' with 53.2 g/m²; however, it did not differ (P > 0.05) from M. littoralis, M. polymorpha, and M. truncatula 'Jemalong'.

Differences were not significant (P > 0.05) among cultivars of the same species except in M. truncatula when Jemalong was lower than Borung and Paraggio but similar to Cyprus which produced 86.7 g/m². Genotypes M. sativa and Trifolium spp. did not produce any seed and differed (P < 0.05) from the annual Medicago spp.

Very few medic plants survived the cold winter temperatures; however, a few plants from \underline{M} . $\underline{blancheana}$ and \underline{M} . $\underline{polymorpha}$ 'Circle Valley' and 'strain 1' were present to produce seed during the spring from 1983 fall planting at Stillwater, Oklahoma. They produced 13.1, 5.0, and 6.2 g/m^2 of seed, respectively. Subclover (\underline{T} . $\underline{subterraneum}$) also survived in that environment and produced 7.3 g/m^2 of seed. Alfalfa (\underline{M} . \underline{sativa}) and arrowleaf clover (\underline{T} . $\underline{vesiculosum}$), the reference

genotypes, yielded 26.0 and 10.6 g/m^2 of seed, respectively.

Differences among annual medic genotypes for cold survival have already been discussed (see Chapter II). However, at Stillwater, snow covered the experimental area in the winter of 1983/84. The survived medic species, M. blancheana and M. polymorpha, reproduced with an adequate amount of seeds which indicated that they were more cold tolerant than those which died when temperatures did not descend below those under snow cover. Therefore, it may be possible to find a source for cold tolerance from these two species if large screening to develop cultivars with an acceptable level of tolerance to cold for regions with snow was undertaken, but future investigations are necessary at first to strengthen these findings which were reported from one environment only. At the present stage of knowledge annual medic genotypes studied should not be fall planted in the U.S. Southern Great Plains.

Total Seed Number

Seed yield expressed on the basis of number of seeds/m provided similar information as that obtained when yield was on a weight basis regarding the significant (P < 0.001) presence of genotypic effects at each environment (Table 2). These effects were also highly significant (P < 0.001) as were those associated to genotype X environment interactions when Oklahoma environments were combined (Table 3). However, total number of seed was different from seed weights in that snail medics (\underline{M} . scutellata) Robinson and Sava produced fewer seeds than alfalfa at Stillwater from 1984 spring planting (Table 4). These two medic cultivars did not differ significantly (P > 0.05) in their total number of seeds from barrel medic Cyprus, but produced much more seed

weight. Strand medic (\underline{M} . <u>littoralis</u>) Harbinger 1, burr medic (\underline{M} . <u>polymorpha</u>) Circle Valley, and barrel medic Jemalong 2 were the other medic cultivars yielding significantly (P < 0.05) more seeds than zero.

Total number of seeds from accessions evaluated apart at Stillwater was 780 and 830 seeds/ m^2 in \underline{M} . $\underline{blancheana}$ and \underline{M} . $\underline{polymorpha}$, respectively, and the difference shown in total seed weight between the two species became not significant in the total number of seeds produced. At Perkins, snail medics produced the highest number of seeds, like at Stillwater, within the medic group in the spring of 1985 (Table 4). All the other medic genotypes, except \underline{M} . $\underline{truncatula}$ Cyprus, which yielded 805 seeds/ m^2 , produced seeds not significantly (P > 0.05) greater in number than zero. Alfalfa had as much or slightly higher seed number than snail medic. Spring planted clovers did not produce any seed prior to senescence.

While medic seed production was low, the seed numbers appear to be adequate for reseeding purposes in Oklahoma when plants were spring sown, at least for M. scutellata, M. truncatula, M. littoralis, and M. polymorpha, assuming reasonable quality characteristics. Differences in reproduction observed among genotypes could be based upon several explanations. Flowering did not occur in M. arabica, M. rugosa, and Trifolium spp., which could be due to absence of vernalization as was reported by Aitken (1) and Clarkson and Russell (9) in annual medics. There was high flower abortion in M. tornata and M. murex which resulted in few pods formed. Cocks (12) indicated massive flower abortion in M. rigidula, M. rotata, and M. truncatula in Syria. Flower abortion and incomplete seed development as indicated by high proportions of immature brown, light seeds could be due to insect infestation. Lygus sp.

nymphs, blue green aphids (\underline{A} . \underline{kondoi}), spotted alfalfa aphids ($\underline{Therioaphis}$ maculata Buckton), pea aphids (\underline{A} . \underline{pisum} Harris), and other insects (Appendix) were present on all $\underline{Medicago}$ spp., but at different levels. These insects were reported by Ragless (27) to impair medic seed production.

Flowering stage was not reached at the same time in genotypes which bloomed. Snail medic (M. scutellata), M. polymorpha, and M. blancheana were the first to set flowers two months after planting in the spring in Oklahoma. They were followed one week later by M. littoralis, M. truncatula Cyprus, M. aculeata, and M. minima. The other M. truncatula and M. tornata were late flowering, three to four weeks after M. scutellata. Length of the vegetative period was much shorter for all medic species than that reported by Crawford (16) from fall planting in Australia. Aitken (1) indicated that flowering stage was more rapid with winter than with summer plantings. Clarkson and Russell (11) showed that high temperatures accelerated the rate of development stages in medics and this was the case under which this study was investigated.

Generally, the number of seeds produced at the Moroccan environment of Sidi el Aydi when plants were sown in the fall of 1985, was much higher in all medic genotypes than that obtained under Oklahoma conditions. Genotypic effects were also significantly (P < 0.001) present at that environment. The highest number of seeds was achieved in barrel medic (\underline{M} . $\underline{truncatula}$) Borung with 30,582 seeds/ \underline{m}^2 ; however, it did not significantly (P > 0.05) differ from that obtained with strand medic (\underline{M} . $\underline{littoralis}$) Harbinger, disc medic (\underline{M} . $\underline{tornata}$) Tornafield, \underline{M} . $\underline{truncatula}$ Cyprus, Jemalong, and Paraggio. The lowest seed producing medic genotype was snail medic \underline{M} . $\underline{scutellata}$ Sava with 6,580 seeds/ \underline{m}^2 ,

but it did not differ significantly (P > 0.05) from Robinson, gama medics (\underline{M} . \underline{rugosa}) Sapo, Paraponto 1, and Paragosa, and burr medics (\underline{M} . $\underline{polymorpha}$) Circle Valley and Serena. Alfalfa and snail medics produced the highest seed number under continental environments of Oklahoma, but performed as the poorest in the Mediterranean environment of Morocco.

These seed yields of medics from 1985 fall planting in Morocco were unquestionably higher than necessary for reseeding. They were higher than those reported from Australia by Crawford (14) which seems to show a potential for commercial seed production in Morocco. Additional research is required to identify the best ways to exploit this capacity.

Differences observed among medic cultivars might have originated from the duration of the vegetative period as well as the reproductive period. Such periods were both longer in late maturing genotypes than in early maturing ones, and generally the former produced slightly more seeds than the latter; e.g., Borung vs. Cyprus. Also, small-seeded cultivars performed better than large-seeded ones.

Seed production was higher with 1985 fall planting in the Mediterranean environment of Morocco than with spring plantings in the continental environments of Oklahoma. Duration of the vegetative period was longer at Sidi el Aydi than at Stillwater or Perkins. First flowers appeared three months after planting in \underline{M} . scutellata at Sidi el Aydi in comparison to two months at Stillwater. Low temperatures allowed vernalization and milder temperatures during seed formation prevailed at Sidi el Aydi. Insect infestation was not important with absence of Lygus spp. and only few aphids which did not appear until maturation.

Seed Characteristics

Medic pastures rely heavily on seed quality, in addition to total yield, for both establishment, regeneration (selfseeding), and persistence. Seed characteristics (number of seeds/pod and the weight of 1,000 seeds, proportion of hard seed, and germination percentage) are considered as important attributes of a medic cultivar. Genotypes differed in number of seeds/pod, weight of 1,000 seeds, germination percentage, and the proportion of hard seed for spring grown plants in Oklahoma and fall sown plants in Morocco; however, the significance level was not the same from one environment to the other, particularly in percent germination (Table 2). When Stillwater and Perkins data were combined, environment, genotype, and genotype X environment interaction effects were found, except for 1,000-seed weight where G X E interactions were not significant (P > 0.05) and in germination percent where genotypic effects were not significant (P > 0.05) (Table 3).

Number of Seeds/pod

Seed number/pod varied from 1.1 to 3.7 at Stillwater (Table 5). Pods of \underline{M} . scutellata had 3.7 seeds/pod and differed (P < 0.05) from those of \underline{M} . sativa and \underline{M} . truncatula Cyprus which had 3.3 seeds/pod. The fewest number of seed was present in \underline{M} . polymorpha Circle Valley with 1.1 seeds/pod. Differences among cultivars of the same species were present in \underline{M} . truncatula. Numbers of seeds/pod were 2.1, 2.3, and 0.5 in \underline{M} . blancheana, \underline{M} . polymorpha (strain 1) and \underline{M} . murex, respectively. Burr medic (\underline{M} . polymorpha) Circle Valley and \underline{M} . littoralis Harbinger contained fewer seeds/pod than the other genotypes

at Perkins (Table 5). \underline{M} . scutellata Robinson developed the highest number of seeds/pod with 3.5. It was not significantly (P > 0.05) different from \underline{M} . scutellata Sava and \underline{M} . truncatula Cyprus. Differences among genotypes of \underline{M} . truncatula were significant at Perkins.

Numbers of seeds/pod from plants grown in spring under Oklahoma conditions were lower than those reported by Heyn (23) in annual medics under their native habitat conditions. Considerable proportions of brown immature seeds were thrown out during cleaning. They were not fully developed, possibly because of insect infestation and premature senescence during their formation. Andrew (3) demonstrated sequential softening of seeds contained in a pod. He also found a positive correlation between the number of seeds/pod and percent dormant seed. Therefore, the higher the number of seeds/pod, the better is pasture persistence. Conditions such as insect control to improve the number of seeds/pod should be investigated for spring sown medics in Oklahoma.

Number of seeds/pod ranged from 1.3 to 6.4 at Sidi el Aydi from 1985 fall sown medics in Morocco (Table 6). Cultivars of \underline{M} . $\underline{truncatula}$ contained more seeds/pod than those of the other medic species. Pods of \underline{M} . $\underline{scutellata}$, \underline{M} . $\underline{polymorpha}$, \underline{M} . $\underline{tornata}$, and \underline{M} . $\underline{littoralis}$ did not differ (P > 0.05) in the number of seeds/pod. Genotypes of \underline{M} . \underline{rugosa} developed fewer numbers of seeds/pod than any other medic cultivar. Differences among accessions within a species were not significant. Number of seeds/pod at Sidi el Aydi was good in annual medics, except in gama medic (\underline{M} . \underline{rugosa}), and concurred with Heyn's description (23) of annual medic species. This indicated that seed formation was under favorable conditions in Morocco during the spring of 1986.

1,000-Seed Weight

Snail medic (\underline{M} . scutellata) developed the heaviest seeds at all environments (Tables 5 and 6). All other Medicago spp. had seeds only 25 to 30% as heavy as \underline{M} . scutellata; however, \underline{M} . blancheana with 8.67 g/1,000 seeds at Stillwater in the spring of 1984 was heavier than small seeded species, and such was the case for \underline{M} . rugosa at Sidi el Aydi.

The seed size for all medics, expressed by the weight of 1,000 seeds under Moroccan conditions in comparison to that obtained under Oklahoma conditions, was a good indicator that seed development was under favorable conditions in Morocco and that the potential for seed production in Morocco is high. Differences among medic species and among environments demonstrated that with the same seed quantity two medic cultivars may contain different seed numbers and also a cultivar may have different seed numbers produced per unit of weight from one environment to another. Seed is now marketed in Morocco without any distinction among cultivars. Hence, medic seed prices would be more truthfully determined when based on number of viable seeds present rather than on weight.

Hard Seed Percent

Barrel medic (M. truncatula) Paraggio, along with alfalfa, was the only medic cultivar which presented a fairly low proportion of hard seed when plants were grown in spring of Oklahoma or from the fall in Morocco (Tables 5 and 6). Hard seed in this cultivar was between 56 and 63%. Snail medic (M. scutellata) and gama medic (M. rugosa) demonstrated moderate levels of hardseededness in Oklahoma and Morocco, respectively.

A slight increase in hardseededness occurred when plants were grown at Sidi el Aydi in comparison to Stillwater and Perkins. Environmental factors were demonstrated by Andrew (2) and Clarkson and Russell (10) to affect the proportion of viable hard seed in medic species. Ragless (27) reported that insect infestation weakened seed viability in Australia.

Generally the level of hardseededness, which is a good attribute regarding pasture persistence according to Carter and Lake (7) and Crawford (15), was high except in M. truncatula Paraggio and to a lesser degree in M. rugosa. Crawford (15, 17) indicated that Paraggio and M. rugosa possessed low proportions of hard seed in comparison to other medics. Paraggio may in some cases, such as at Sidi el Aydi 1985-86, compensate hardseededness with high seed yield. Results obtained in this study demonstrated the possibility to create a soil seed bank ensuring pasture persistence under Oklahoma and Morocco conditions for medics which produced reasonable amounts of seeds. Additional investigations are needed to follow changes of hard seeds in time under field conditions.

Germination Percent

Harvested seeds, when scarified and tested under controlled conditions, presented germination percentages between 76% and 99% in all genotypes which reproduced (Tables 5 and 6). Snail medic (\underline{M} . scutellata) and barrel medic (\underline{M} . truncatula) Paraggio were the only medics which germinated with less than (P < 0.05) 85% in the spring of 1985 at Stillwater. A slight increase in germination percentages was observed when plants were grown at Perkins from the 1985 spring planting

in comparison to those from the 1984 spring planting at Stillwater. The increase was more noticeable from plants sown in the fall of 1985 at Sidi el Aydi.

Assuming adequate conditions in the field for germination and emergence, seed produced when scarified will generate good establishment. Natural regeneration will be possible as a high proportion of viable medic seeds was found under both continental and Mediterranean climates. Additional research on germination as time advances for both stored seeds and those destined to constitute soil seed reserve will certainly progress knowledge about medics under continental conditions and in Morocco.

Relationships Between Seed Characters

Total seed weight/ m^2 , seed number/ m^2 , number of seeds/pod, 1,000-seed weight, hardseed percentage, and germination percentage were all positively correlated. Simple correlation coefficients between these parameters were all significantly (P < 0.01) higher than zero (Table 7). Association between seed weight/ m^2 and seed number/ m^2 was less than 1.0 due to differences among genotypes in seed size which was shown by the lower correlation between seed number/ m^2 and 1,000-seed weight.

Andrew (3) and Salisbury et al. (29) also found positive correlations between seed coat impermeability (hardseededness) and number of seeds/pod in annual medics and subclover. It appears from all these results that favorable conditions, either natural or through management, which tend to increase seed yield on a weight basis, will

increase the total number of seeds, number of seeds/pod, and hard seed percentage. Consequently, regeneration in terms of number of offspring plants per unit area and pasture persistence will be ameliorated. Also, an increase in seed size, within limits so that number of seeds is not reduced, will promote better seedling vigor as was demonstrated by Crawford (13) in barrel medic (\underline{M} . $\underline{truncatula}$).

From a breeding standpoint, selection of medic lines having a high number of seeds/pod, which can easily be done according to Salisbury et al. (29) who indicated that this trait had high heritability in subclover, will improve seed yield in weight and number of seeds, and seed quality (hard seed and germination percentages) in these lines. Further studies are needed to estimate heritability of seed yield and quality in medics so that realized improvement of these traits through selection of number of seeds/pod can be predicted.

Reseeding in Oklahoma

Medic regeneration was monitored in fall of 1984 from pods produced by 1984 spring grown plants at Stillwater. The number of offspring seedlings was significantly (P < 0.001) dependent on genotype at Stillwater (Table 8). Snail (M. scutellata) Robinson produced the highest number of offspring seedlings with 238 plants/m² (Table 9), which represented about three times the number of parent plants; however, it did not differ (P > 0.05) from alfalfa (M. sativa). Its related cultivar Sava regenerated well with 165 plants/m². All other entries in the test produced less than 19 plants/m² and were statistically (P > 0.05) similar. At Perkins in 1985 there was no emergence from pods which remained on the soil. These pods contained

more than 70% of viable seeds (Table 10).

Hardseededness was the primary factor which kept annual medic seeds in pods from not emerging in the subsequent fall. High proportions of hard seed in all medic genotypes were found (Table 5). Gintzberger (19) demonstrated that pod sowing resulted in better pasture than naked seed sowing probably because pods contained more hard seeds. Future investigations are needed to assess seed destiny remaining in the soil.

No single factor related to sites or weather appears to explain why germination occurred in one situation in Oklahoma but not in the other. One could speculate that differences in rainfall amounts and distribution in relation to temperatures during and after seed formation played a role. Gutterman (22) demonstrated that duration of daylight to which the mother plants were exposed during the last 8-12 days of seed maturation may affect seed coat permeability and seed germination. This clearly shows that reestablishment or selfseeding of medics in the U.S. Southern Great Plains is not predictable.

Emerged seedlings at Stillwater in the fall of 1984 had good vigor but did not survive winter. Cold sensitivity of annual Medicago spp. studied will be the most limiting factor for medic use under a continental climate like in Oklahoma. Intensive breeding or identification of more winter hardy material will be necessary to obtain acceptable cold tolerance.

Conclusions

At present, several of the annual medics evaluated in these experiments may have potential to produce seed for developing a soil seed bank for regeneration (reseeding) when spring planted in Snail medic (M. scutellata), M. blancheana, M. polymorpha, and M. truncatula are the most promising species with regard to the number of offspring seeds if attempts are made to select for cold tolerance or to investigate the fate of hard seed of these species in the soil over time. Hardseededness limited the number of regenerated plants, while winter cold did not allow either fall sown and fall regenerated plants to survive. Clovers (Trifolium spp.) did not produce seed when spring planted in Oklahoma or fall sown at Sidi el Aydi, Morocco, nor did alfalfa (M. sativa) from fall planting at Sidi el Aydi. Rainfall in Morocco is an important factor influencing medic seed production, and when this is at an adequate level, all medics, particularly small-seeded species, possess excellent prolificity and seed quality which encourages the development of a commercial seed production. The positive correlations observed between seed production traits suggests that selection for highly heritable traits such as seed number/pod would lead to seed yield and quality improvement.

Table 1. Genotypes evaluated in various environments

	Genotyp	e	Environment						
C		01+4	Stillwater		Pe	rkins S	Sidi el	Aydi	
Spe	ecies	Cultivar	Fall 1983	Spring 1984	Fall 1984	Spring 1985	g Fall 1985	Fall 1986	
М.	arabica		X(a)	Χ	nd	nd(b)	nd	nd	
Μ.	littoralis	Harbinger 1	Χ	X	X	X	Х	Х	
Μ.	littoralis	Harbinger 2	X	Х	Χ	nd	Х	X	
		01 1 1/11	X	X	nd	nd	nd	nd	
Μ.	polymorpha	Circle Valley	y X	X	Χ	Χ	Х	X	
Μ.	polymorpha	Serena	nd	nd	nd	nd	X	Х	
Μ.	rugosa	Paragosa	X	X	X	X	X	X	
Μ.	rugosa	Paraponto 1	Χ	X	Χ	Χ	Χ	Х	
Μ.	rugosa	Paraponto 2	nd	Χ	nd	nd	nd	nd	
Μ.	rugosa	Sapo	Х	Χ	Х	Χ	Х	Х	
М.	sativa	OK-83-Graze	Χ	X	Χ	Х	Х	nd	
М.	sativa	Spredor II	Х	nd	X	X	X	nd	
Μ.	scutellata	Robinson	Χ	X	Χ	X	Χ	Х	
Μ.	scutellata	Sava	Χ	Χ	Χ	Χ	X	Х	
Μ.	tornata	Tornafield 1	Χ	X	Χ	Χ	Х	Х	
Μ.	tornata	Tornafield 2	nd	Х	Х	Х	Х	nd	
Μ.	truncatula	Borung	X	Χ	Χ	Χ	Χ	Х	
Μ.	truncatula	Cyprus	Χ	X	Χ	Χ	Х	Χ	
Μ.	truncatula	Jemalong 1	X	Χ	X	X	Χ	Х	
Μ.	truncatula	Jemalong 2	Х	X	X	Χ	X	nd	
М.	truncatula	Paraggio	X	X	Χ	Х	Х	Х	
Τ.	incarnatum		nd	nd	X	Χ	Х	nd	
Τ.	pratense	Redland Programme Redland	nd	nd	nd	Χ	nd	nd	
Τ.	subterraneum	Mount Barker	Χ	nd	Χ	X	Х	nd	
Τ.	vesiculosum	Yuchi	Χ	X	Χ	Χ	Х	nd	

(a) : X = evaluated
(b) : nd = not evaluated

Table 2. Mean squares for seed yield and seed characteristics at each environment (1)

Source	df	Stillwater MS	<u>Perkins</u> MS	Sidi df	el Aydi MS
Seed wt.					
Rep.	2	13.7	0.4	2	565.7
Genotype	19	117.0***	367.0***	20	5914.8***
Error	38	7.0	1.9	40	393.8
Seed number					
Rep.	2	1.4	1.1	2	157.8
Genotype	19	25.0***	51.4***	20	3013.0***
Error	38	0.7	0.6	40	465.3
Seeds/pod					
Rep.	2	4.1	0.3	2	220.7
Genotype	9	1209.0***	1286.0***	15	10168.0***
Error	18	59.4	58.0	30	653.0
Wt./1000 seeds					
Rep.	2 9	0.6	0.2	2	3.7
Genotype		493.5***	462.8***	15	774.4***
Error	18	0.3	1.0	30	2.7
Hardseed %					
Rep.	2	12.4	4.8	2	185.2
Genotype	9	205.7***	135.2***	15	483.6**
Error	18	5.3	13.4	30	134.7
Germination %					
Rep.	2	0.7	5.7	2	42.8
Genotype	9	66.6***	42.0*	15	93.4**
Error	18	7.4	12.1	30	28.4

^{(1):} Mean squares for seed number need be multiplied by (10^5) Mean squares for seeds/pod need be multiplied by (10^{-3}) Mean squares for Wt./1000 seeds need be multiplied by (10^{-1})

^{***:} Significant (P < 0.001)

**: Significant (P < 0.01)

*: Significant (P < 0.05)

Table 3. Mean squares for seed production and seed characteristics in Oklahoma

		Mean squa	res
Source	df	Seed weight	Seed number
Environment (Env.)	1	56.8	0.04
Replication in Env.	4	8.5	1.21
Genotype (Gen.)	15	527.0***	65.72***
Gen. X Env.	15	56.8***	6.23***
Error	60	5.5	0.72

Table 3. (continued)

	-	Mean squares						
Source	df	Seeds/ pod	1000- seed wt.	Hard- seed %	Germi- nation %			
Environment (Env.) 1	445.5***	34.7*	240.0**	400.4***			
Replication in En	ıv. 4	2.2	3.6	8.6	3.2			
Genotype (Gen.)	9	2229.0***	9555.1***	279.5*	74.2			
Gen. X Env.	9	265.8***	8.3	61.3***	34.4**			
Error	36	58.8	6.1	9.3	9.8			

^{***:} Significant (P < 0.001). ** : Significant (P < 0.01). * : Significant (P < 0.05).

Table 4. Seed production of genotypes at Stillwater 1984, Prekens 1985, and Sidi el Aydi 1985/86

Genotype		Seed	weight	(g/m²)	See	Seed number/m²		
Species	Cultivar	Still- water 1984	Perkins 1985	5101 el	Still		Sidi el ns Aydi	
M. arabica M. littoralis M. littoralis M. minima M. polymorpha	Harbinger 1 Harbinger 2 Circle Valle	0 1.2 0.9 0 ey 1.9	nd(a) 0.8 nd nd 0.7	nd 76.8 54.7 nd 68.2	0 461 309 0 660	nd 254 nd nd 273	nd 24870 23700 nd 17600	
M. polymorpha M. rugosa M. rugosa M. rugosa M. rugosa	Serena Paragosa Paraponto 1 Paraponto 2 Sapo	nd 0 0 0	nd 0 0 nd 0	54.2 74.8 77.2 nd 53.2	nd 0 0 0	nd 0 0 nd 0	14850 11420 7780 nd 8430	
M. sativa M. sativa M. scutellata M. scutellata M. tornata	OK-83-Graze Spredor II Robinson Sava Tornafield 1	12.7 nd 21.0 17.7 0.2	10.0 10.1 32.8 33.9 0.1	0 0 123.7 132.2 112.7	3628 nd 1698 1456 64	3221 3829 2815 3219 41	0 0 6980 6580 21760	
M. tornata M. truncatula M. truncatula M. truncatula M. truncatula	Tornafield 2 Borung Cyprus Jemalong 1 Jemalong 2	0 0.7 4.2 0.9 1.3	0.1 0.5 2.7 0.7 0.6	94.4 113.0 86.7 75.9 74.5	0 279 1677 384 542	21 383 805 308 266	21790 30580 20180 19350 22850	
M. truncatula T. incarnatum T. pratense T. subterraneum T. vesiculosum	Paraggio Redland Mount Barker Yuchi	0.4 nd nd nd	0.3 0 0 0	117.4 0 nd 0	167 nd nd nd	124 0 0 0	28480 0 nd 0	
LSD (P = 0.05)		4.4	2.3	32.8	451	396	11260	

⁽a) : nd = not determined

Table 5. Seed characteristics of genotypes at Stillwater and Perkins, Oklahoma

	See	ds/pod	Wt. of seed	1000 s (g)	_% Ha	rdseed	% Germ nati	
Cultivar	StS84	PeS85	StS84	PeS85	StS84	PeS85	StS84	PeS85
M. littoralis Harbinger 1	1.9	1.6	2.68	2.60	85	83	88	92
M. polymorpha Circle Valley	1.1	1.2	2.37	2.59	85	84	90	92
M. sativa OK-83-Graze	3.3	2.9	2.50	2.65	62	63	85	83
M. scutellata Robinson	3.7	3.5	12.30	12.07	70	78	77	85
Sava	3.7	3.4	12.20	11.67	71	75	77	87
M. truncatula Borung	2.2	2.1	2.49	2.37	81	80	83	84
Cyprus	3.3	3.2	2.63	2.52	84	87	89	93
Jemalong 1	2.0	2.1	2.37	2.39	79	70	86	89
Jemalong 2	2.7	2.0	2.48	2.37	78	79	82	86
Paraggio	2.4	2.2	2.45	2.10	63	64	79	84
LSD (P = 0.05)	0.2	0.4	0.28	0.53	4	6	5	6

Table 6. Seed characteristics of genotypes at Sidi el Aydi, Morocco

Species	Cultivar	Seeds/ pod	1000- seed wt. (g)	Hard- seed %	Germi- nation %
M. littoralis M. littoralis M. polymorpha M. polymorpha M. rugosa	Harbinger 1	3.1	3.09	82	87
	Harbinger 2	3.0	3.10	83	92
	Circle Valley	4.3	3.88	96	99
	Serena	3.9	3.69	96	96
	Paragosa	1.4	6.51	79	89
M. rugosa M. rugosa M. scutellata M. scutellata M. tornata M. tornata	Paraponto	1.4	6.40	77	81
	Sapo	1.3	6.22	72	83
	Robinson	4.1	20.74	95	99
	Sava	4.4	20.21	91	98
	Tornafield 1	3.2	4.54	81	96
	Tornafield 2	3.1	4.34	93	99
M. truncatula M. truncatula M. truncatula M. truncatula M. truncatula	Borung	6.1	3.71	84	92
	Cyprus	6.1	4.34	88	94
	Jemalong 1	5.7	3.96	84	99
	Jemalong 2	6.4	4.33	79	91
	Paraggio	6.4	4.12	56	91
LSD (P = 0.05)		1.3	0.88	19	9

Table 7. Simple correlation coefficients between seed characteristics

	Seed weight	Number of seeds/pod	Wt./ 1000 seeds	Germina- tion %	Hard- seed %
Number of seeds/pod	0.622***				
Wt./1000 seed	0.613***	0.527***	en de la companya de		
Germination %	0.530***	0.815***	0.590***		
Hardseed %	0.499***	0.772***	0.597***	0.980***	
Seed number	0.806***	0.640***	0.230**	0.473***	0.422**

***: Significantly different from 0.0 (P < 0.001) **: Significantly different from 0.0 (P < 0.01)

Table 8. Mean squares for reseeding at Stillwater, Oklahoma

Source	df	MS	
Replication	2	5000*	•
Genotype	19	14000***	
Error	38	1500	

***: Significant (P < 0.001) *: Significant (P < 0.05)

Table 9. Number of regenerated plants in fall 1984 at Stillwater, OK.

Species		Cultivar	Number of regenerated plants/m ²
M. arab	ica		-(a)
M. litte	oralis	Harbinger 1	6
M. litto	oralis	Harbinger 2	15
M. minir			-
M. polyr	norpha	Circle Valley	10
M. rugos	sa	Paragosa	-
M. rugos		Paraponto 1	-
M. rugos		Paraponto 2	-
M. rugos	sa	Sapo	-
M. sativ	/a	OK-83-Graze	180
M. scute	ellata	Robinson	238
M. scute		Sava	165
M. torna	ata	Tornafield 1	0
M. torna	ata	Tornafield 2	-
M. trun	catula	Borung	1
M. trun	catula	Cyprus	19
M. trun		Jemalong 1	
M. trun	catula	Jemalong 2	3 0
M. trun		Paraggio	1
T. vesi	culosum	Yuchi	-
LSD (P	= 0.05)		64

⁽a) : Number of offspring plants is zero because no seed was produced.

Table 10. Germination and hardseed percentages of genotypes with pods remaining on soil for 15 months at Perkins, Oklahoma

Species	Cultivar	Germination %	Hardseed %
M. littoralis	Harbinger 1	80	70
M. polymorpha	Circle Valley	80	60
M. sativa	OK-83-Graze	-(a)	-
M. scutellata	Robinson	72	56
M. scutellata	Sava	74	50
M. truncatula	Borung	82	80
M. truncatula	Cyprus	86	76
M. truncatula	Jemalong 1	84	70
M. truncatula	Jemalong 2	-	-
M. truncatula	Paraggio	72	54

⁽a) : - = not determined

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CHAPTER IV

BIOMASS PARTITIONING AND ROOT DEVELOPMENT IN ANNUAL MEDICAGO SPP.

Abstract

The relative contribution of various plant parts to the total biomass in forage legume species at a given growth stage is a subject of biological and economical concern, particularly when species are cropped in mixture. This study was conducted under greenhouse conditions to estimate changes in biomass partitioning over time and to evaluate the root system and stem development in annual medics (Medicago spp.). The experiment was a split-plot in time, with main plots arranged in randomized complete block design with three replications. Main plots were plant ages 13, 24, 34, 44, 57, 66, and 76 days from emergence. Subplots contained plant species including four annual medics; M.

littoralis Rhode, M. polymorpha L., M. scutellata (L.) Mill., and M.

truncatula Gaertn., one alfalfa (M. sativa L.), and one subclover (Trifolium subterraneum L.).

Dry weights of the whole plant and of roots, stems, leaves, and pods were significantly influenced by age, species, and age X species interactions. Differences observed were related to seed size and maturation cycle. Source:sink ratio was associated with plant age and age X species interactions. However, there was a decrease in this ratio during seed formation. Leaf:stem ratio depended on plant age and age X

species interactions. These interactions were absent in the root:total weight ratio. Flowering was the stage when most development pattern changes occurred within and among species. Medics and subclover developed a fibrous root system as opposed to alfalfa which had a taproot system. Maximum root and stem lengths were 71.8 and 83.1 cm in M. truncatula, respectively. However, root length:stem length ratio was associated with age, species, and their interactions.

M. littoralis, M. polymorpha, and M. scutellata constituted one group of species when all six measured parameters were considered in a multivariate analysis. M. sativa was different from this group.

M. truncatula, despite its similarity to the other medics in root and pod weights, did not differ from M. sativa with respect to the remaining parameters. T. subterraneum formed a separate group. Mixture of the four medics studied may have some advantage in crop compensation but would be difficult to manage.

Key words: Medic, alfalfa, subclover, Trifolium, mixture

Introduction

Importance of legumes in maintaining fertility and structure of soil under pasture conditions has been well demonstrated by Amor (2) and Jones (15). In addition to their forage value, their residues make significant contributions to soil nitrogen status (3). Medics (Medicago spp.) and clovers (Trifolium spp.) are major components of annual pastures where the Mediterranean climate prevails in Australia (8). They are being reintroduced for extensive use into North Africa, their native habitat from where they have been collected (4, 5).

Alston (1) reported that seasonal changes in nitrogen fixation in barrel medic (M. truncatula Gaertn.), estimated by the acetylene reduction technique, were closely correlated with dry weight and photosynthetic area. He indicated dry weight and nitrogen concentration decreased at the end of the growth cycle in medics. There was also movement of photosynthates from vegetative plant parts to reproductive organs. Cocks (10) noted massive flower abortion in medics, which reduced seed yield in these species. Abortion could be a way of adjusting high demand of sink compartments (pods and seeds) for photosynthates when the source is a limiting factor.

Predictions of plant development in the field from controlled conditions were investigated by Clarkson and Russell (9) in medics. They indicated that predictions were satisfactory for all development stages from planting to formation of the first mature pod for snail medic (\underline{M} . scutellata (L.) Mill)), and for all stages except flowering for \underline{M} . truncatula. Daily increments of development were a function of daily mean temperatures. Increased temperature accelerated rate of

development of all growth stages until temperatures reached $30/23\,^{\circ}\text{C}$ for day and night, respectively (9, 25).

Understanding the root system and its development is essential to managing any crop, particularly forage species. Root development has marked effects on plant survival and its ability to compete for water and nutrients (12, 23). Jung and Larsen (16) reported that alfalfa (M. <u>sativa</u> L.), under irrigation, took 46% of absorbed water from the first 30 cm of soil, 22% from the second 30 cm, and 10% from each additional 30 cm of depth. However, these figures can vary depending on climatic conditions, depth and amount of roots within the soil profile, and soil texture. Humphries and Bailey (13) observed large genetic variation for root weight, root length, and root:shoot ratio among clover species and within cultivars of subterranean clover (T. subterraneum L.). Similar results were obtained by Sinskaya (21) for root development rate in annual medics and wild perennial alfalfas. The latter indicated that at 10 days of age, roots of wild alfalfa were ten times longer than stems, and were more developed than those of annual medics. However, Weise (24) found that root lengths of annual weedy species were not significantly different from those of perennials at early growth stages. When the genetic variation is expressed, as in the case of sorghum (Sorghum bicolor (L.) Moench), differences in root growth were shown to be related to cultivar tolerance to drought (17). More tolerant lines had higher root:shoot ratios. Simpson et al. (20) demonstrated that extensive root systems were associated with higher yields. Root development can also be related to maturation cycle. Humphries and Bailey (13) reported that late-maturing subclover cultivars produced deeper and heavier roots than early-maturing ones.

Data on biomass partitioning in medic species used as mixtures are limited. A better understanding of root growth of annual medics should result in better management of these forage species. The objectives of this study were to describe changes in biomass partition over time in six forage species and to evaluate root and stem development.

Materials and Methods

The experiment was a split-plot in time, arranged in a randomized complete block design with three replications. Replications were separated in time during Nov. 1984-Jan. 1985, Jan.-Mar. 1985, and Apr.-June 1985, respectively, for the first, second, and third replications. Main plots were plant ages 13, 24, 34, 44, 57, 66, and 76 days after emergence chosen to monitor growth over time. There were six sub-plot levels (plant species): four annual medics, one alfalfa, and one clover (Table 1). Alfalfa and subclover were used as controls since they behave as a true perennial and a true annual, respectively.

Twenty scarified seeds of each treatment combination of entry and age were inoculated with the proper $\underline{Rhizobium}$ spp. and sown in greenhouse as single rows in a wooden flat (1.2 m long, 1.0 m wide, and 1.0 m deep), with one side as a door to be removed for intact plant excavation. The flat was divided into small units (0.15 X 0.15 X 1.00 m) with fiberglass sheets to avoid root mixtures among subplots. At emergence seedlings were thinned to leave 11 plants per subplot. Plants were grown on an inert medium (Ditomite) which does not stick on roots like natural soil or sand. Ditomite was rinsed several times to remove dust and gas prior to use. Nutrients were supplied weekly with a solution made of a complete fertilizer. The fertilizer contained 15, 30, and 15% of N, P_2O_5 , and K_2O , respectively, in addition to micronutrients. Plants were watered as necessary to maintain growth.

On each sampling date plants were uprooted and rinsed with tapwater to remove Ditomite particles. Ten randomly chosen plants from each subplot (which constituted the experimental unit) were measured. Stem

number was counted at the crown level. Stem length was measured from soil level to the apical growing point. Root length within the profile was recorded. Root diameter was measured at the thickest point of the largest root produced by each plant. Plants were fractioned into roots, stems, leaves, and pods. All parts were dried at $70 \pm 5^{\circ}\text{C}$ for 48 hr. Dry weights represent the total of plant material from the experimental unit. Lengths correspond to the average length of the 10 plants measureed. The experiment was terminated after 76 days from emergence when annual medics and clover began senescence.

Root:total and leaf:stem ratios were calculated on a dry weight basis. Source:sink ratio was approximated by the ratio of leaf dry weight divided by the sum of dry weights of other plant parts; roots, stems, and pods, according to Brown (7). Root:stem ratio was determined on a length basis.

Statistical analyses included analyses of variances with all factors considered fixed and protected least significant difference (LSD) at P = 0.05 for mean separations (22). Both orthogonal contrasts and multivariate analyses were conducted on linear regression coefficients of different parameters measured. Each response measured was regressed linearly on age, and values of linear regression coefficients obtained were tested for their significance and computed in the analyses of variance according to Potthoff and Roy (18) and Sanders (19), who demonstrated the superiority of this method in fitting better growth curve comparisons and species grouping. Orthogonal contrasts for mean squares of regresssion coefficients were made to assess for homogeneous grouping among species for each parameter measured.

Multivariate analysis was used to obtain more information on species

grouping. In this procedure an analysis of variance was performed for two or more variables at a time. Determination of Hotelling-Lawly trace in the variance-covariance matrix was the criterion used in hypothesis tests (11). Data are presented as the total weight of ten plants for dry weights and as the average of ten plants for stem number, root diameter, and lengths

Results and Discussion

Analyses of variance (Table 2) showed that significant differences existed (P < 0.001) among species and ages for all characters measured and that age X species interactions were found. Replication effects were significant, as expected, since replications differed in time and certain growing conditions, particularly light and temperature.

Clarkson and Russell (9) demonstrated that light and temperature had marked effects on medic growth and development. Plant age effects were manifested as tissue accumulated in roots, stems, leaves, and pods over time. Hunt (14) characterized the increased dry matter production of plant parts as affected by growing conditions and genetic material used. Therefore, changes in dry weight of plant parts over time were expected.

Amount of dry matter accumulated in each plant organ depended on species. Interactions between species and ages were significant, indicating that these species accumulated dry matter within each plant part differently over time. Due to such interactions it was necessary to evaluate the differences among species at each age (Table 3).

Root dry weight was not different (P > 0.05) among species at 13 days of age (Table 3, Fig. 1). However, \underline{M} . scutellata with 0.18 g had heavier roots than the other species at 24 days of age. Differences were not apparent at 34, 44, and 57 days and mean root weight was 0.47, 1.28, and 1.83 g, respectively. Alfalfa had heavier roots at 66 and 76 days of age than annual medics and subclover, indicating the root had begun serving as a storage organ in the perennial. Barrel medic (\underline{M} . truncatula) produced heavier roots at 76 days than other medics and subclover but less than alfalfa.

Stem weights did not differ (P > 0.05) among entries from the beginning of growth to 57 days of age (Table 3, Fig. 2). M. sativa and M. truncatula had more dry matter in stems than the other medics and subclover at 66 and 76 days. At 76 days M. sativa and M. truncatula had 11.27 and 10.10 g, respectively, while \underline{T} . subterraneum had only 3.23 g as stems which were similar to the other medics.

Leaf dry matter did not differ significantly (P > 0.05) among species up to 24 days of age (Table 3, Fig. 3); however, \underline{M} . scutellata produced significantly (P < 0.05) heavier leaf weights (0.35 g) than \underline{M} . littoralis and \underline{M} . polymorpha, which were superior to \underline{M} . truncatula, \underline{M} . sativa, and \underline{T} . subterraneum at 24 days. These differences were statistically negligible at 34, 44, and 57 days of age. Toward the end of the growth cycle at 66 and 76 days, \underline{M} . truncatula and \underline{M} . sativa produced much more leaf material than the other species.

Pod weight differed among cultivars from 44 days after emergence (Table 3, Fig. 4). At 44 days, \underline{M} . scutallata and \underline{M} . polymorpha were the only species to have green pods. \underline{M} . scutellata had more pod material than \underline{M} . littoralis and \underline{M} . truncatula at 57 days. At this age subclover had not set burrs. Strand medic pod weight (2.34 g) was less than that of snail medic (13.27 g) at 66 days. At the end of the growth cycle, 76 days of age, \underline{M} . littoralis produced the least amount of pods among medics and was not different (P > 0.05) from \underline{T} . subterraneum. Alfalfa did not produce pods in this study.

Total dry weight of the whole plant did not differ significantly (P> 0.05) among species at early growth (Table 3, Fig. 5). However, by 24 days, M. scutellata had accumulated more dry weight (0.61 g) than other species. Such differences were attenuated at 34, 44, and 57 days

of age; however, \underline{M} . $\underline{truncatula}$ had produced three to four times more dry weight than \underline{M} . $\underline{littoralis}$ and \underline{T} . $\underline{subterraneum}$ by 66 days. Total dry weight did not increase significantly (P > 0.05) after 66 days as plants reached maximum growth.

Dissimilarities occurring among species at early growth in dry weights of roots, leaves, and total may have been related to seed size and earliness of cultivars used. Snail medic (\underline{M} . scutellata) seeds are four to six times bigger than those of the other medics and it produced the most vigorous seedlings with large and thick cotyledons. All other medics are small-seeded species and their seedlings were relatively less vigorous. Barrel medic (\underline{M} . truncatula) cv. 'Jemalong' is a latematuring cultivar and grew more slowly from emergence until 34 days than strand medic (\underline{M} . littoralis), burr medic (\underline{M} . polymorpha), and snail medic (\underline{M} . scutellata) cultivars which are early-maturing. Alfalfa behaved as a late-maturing species, because of its perenniality, with slow growth at the beginning of the growth cycle similar to that of \underline{M} . truncatula.

Differences at the end of the growth cycle (66 and 76 days) were associated with maturation. First flowers appeared at the age of 36 days for M. scutellata, M. polymorpha, and M. littoralis; at 46 days for M. truncatula and T. subterraneum; and at 56 days for M. sativa. Flowering stage is a characteristic of the cultivars used in the study and not to species since large variation for this trait exists within each species (10). In this study there was much variation in time to reach maturity after 57 days of age with concommitent effects on biomass partitioning. Barrel medic (M. truncatula) and subclover reached

maturation later than did \underline{M} . polymorpha, \underline{M} . littoralis, and \underline{M} . scutellata. However, strand medic (\underline{M} . littoralis) did not reach its potential because it was heavily infested with mites ($\underline{Tetranychus}$ sp.) and it was hurt with insecticides used to control mites during the third replication.

Dry weight of roots, stems, and leaves increased with time until the age of 66 days (57 days for snail medic and subclover), then decreased slightly at the end of the growth cycle. Such a decrease was not demonstrated in M. truncatula and M. sativa which reached a plateau. At this period there was appearance of crown buds in alfalfa. Reduction in dry weight of nonreproductive organs could be due to breakdown of some components (proteins and carbohydrates) and their movement to the reproductive organs (pods and seeds) and to leaf drop (1). Differences in pod dry weight among species may partly be due to pod and seed size which differ from one medic species to another. M. scutellata has large pods of about 1 cm diameter, as opposed to M. littoralis which pods have less than 0.5 cm diameter.

The source:sink ratio was calculated by dividing leaf dry weight (including cotyledons) by the sum of dry weights of the other plant parts. Roots, stems, pods, and seeds were considered as parts of the sink in this study. Source:sink ratios were affected by plant age and age X species interactions (Table 4). Differences among species for this ratio were present (P < 0.05) at early growth, 13 and 24 days of age (Table 5). M. scutellata had a larger source:sink ratio (1.56) than M. littoralis, M. polymorpha, and T. subterraneum at 13 days primarily because of the large cotyledon size of this species. In contrast, M.

<u>sativa</u> had a lower ratio (0.63) than the medics at 24 days of age because it formed an upright stem with few leaves, while the medics, except \underline{M} . <u>scutellata</u>, and subclover formed a rosette with more leaves and shorter stems.

There were two distinct periods in the growth cycle of medics and subclover for the trend of source:sink ratio despite the presence of age x species interactions at early growth (Table 5). The first period was characterized as having higher source:sink ratio than was the second, and the passage from the first to the second occurred between 44 and 57 days of age. The decline in the source:sink ratio at this time could be related to the high demand of pod and seed formation for photosynthates. Evetts (12) demonstrated that leaf area was highly and positively correlated with leaf dry weight (r2 = 0.84); therefore, it would be possible to think that sink demand exceeded source photosynthate supply at flowering, limiting forage and seed yield in medics; which may also explain flower abortion observed by Cocks (10) in annual medics.

Leaf:stem ratios, which reflect forage quality (6), did not differ significantly (P > 0.05) among plant species but did differ among ages (Table 4). Proportion of photosynthates stored in stems compared to that in leaves appears to be similar among the species included in the study. Leaf:stem ratio increased slightly up to the age of 24 days, except in alfalfa (Table 5). This may have been due to the formation of the rosette stage in annuals, which induced a high number of leaves with little stem elongation. The ratio decreased between 24 and 44 days (except for alfalfa) which could be associated with changes occurring within the plant during flowering prior to pod formation. The rate of stem growth was higher than that of leaf growth during the reproductive

phase since the species studied possess an indeterminant growth. This decrease observed in the ratio at flowering might indicate that nutritive value of medics would be reduced after about 44 days. No important changes appeared in the leaf:stem ratio as plants approached maturation. Values obtained for leaf:stem ratio were higher than those reported by Barnes and Gordon (6) in M. sativa. They were 4:1 and 1:1 (mean values over species) at 24 and 57 days of age, respectively, while Barnes and Gordon (6) reported 2:1 and 1:2 at the vegetative and the early seed stages of M. sativa.

The root:total weight ratio was influenced by both plant age and species (Table 4). Age x species interactions were also present (P< 0.05) for this ratio. Differences among species were apparent at 13, 66, and 76 days of age (Table 5). M. scutellata had a lower root:total ratio (0.21) than other annual Medicago species and Trifolium, which may reflect the presence of sizeable cotyledons in this species at early growth. Toward the end of growth at 66 and 76 days, T. subterraneum and M. sativa had higher root:total ratio (1:5) than the annual medics. The development of taproot kept the ratio high in alfalfa, whereas limited growth of stems in subclover resulted in low dry weight of shoots and consequently a high root:total ratio. These results on $\underline{\mathsf{T}}$. subterraneum were similar to those reported by Humphries and Bailey (13). The root:total ratio generally decreased with time (Table 5), indicating that as time advanced there were less photosynthates transported to roots than those used by the above-ground plant parts.

Root diameter varied significantly (P < 0.01) according to plant species (Table 6). The mean root diameter of \underline{M} . sativa was 2.5 mm

compared to 1.4 mm in M. truncatula and T. subterraneum (Table 7).

Annual Medicago and T. subterraneum developed a fibrous root system as opposed to M. sativa which developed a large taproot by the end of the study. Williams (25) observed that clovers developed a taproot under field conditions. Environmental conditions during growth have been reported to play a determinant role in root system development and characteristics (20, 23). Therefore, this deviation may have been related to the type of medium used in the present study as large ditomite particles allowed space for root penetration. M. sativa started to develop a taproot after the age of 24 days, while medics and clovers maintained fibrous roots. These findings support the similarity in the root system between annual and perennial species at early growth stages (12, 24) and their differences at advanced growth stages (20).

Root length was related to plant age and species. However, age x species interactions were absent (P > 0.05) (Table 6). Differences in root length among species did not become statistically significant (P < 0.05) until the age of 57 days. When maximum lengths reached by the different species, not necessarily at the same age, were compared (Table 8), roots of \underline{M} . truncatula (71.8 cm) and \underline{M} . sativa (70.3 cm) were longer than those of \underline{M} . littoralis (61.4 cm), \underline{M} . scutellata (60.2 cm), and \underline{T} . subterraneum (63.2 cm). Sinskaya (21) indicated that wild perennial alfalfa roots are longer than those of annual medics at the first 10 days of age. Evetts (12) and Weise (24) found no differences between perennial and annual grasses and weed species. Root length increased with age throughout the growth cycle, 8.4, 33.3, and 60.0 cm, respectively, at 13, 34, and 57 days. However, after 57 days of age,

there was no significant (P > 0.05) increase in root length (Table 8)

Stem number depended (P < 0.001) on plant age and species, but not on their interactions (P > 0.05) (Table 6). \underline{T} . $\underline{subterraneum}$ produced more stems than \underline{M} . $\underline{scutellata}$ and \underline{M} . \underline{sativa} (Table 9). \underline{M} . $\underline{scutellata}$ had fewer stems than the other medics. This feature may be associated with growth habit. \underline{M} . $\underline{scutellata}$ and \underline{M} . \underline{sativa} exhibited an upright habit as opposed to the other annual $\underline{Medicago}$ and \underline{T} . $\underline{subterraneum}$ which were prostrate. Upright main stems might inhibit continued stem emergence from the crown at early growth while there was no dominance among stems in the prostrate situation.

Stem length was associated with significant (P < 0.001) age, species, and age X species interaction mean squares (Table 6, Fig 6). Length increased with age, however, increments were not uniform across species due to the presence of age X species interactions. Differences among species were not apparent at 13 and 24 days of age, but they became significant (P < 0.05) at subsequent sampling dates (Table 10). T. subterraneum had the shortest stems throughout the growth cycle. M. sativa had longer stems (17.4 cm) than the medics, except for M. scutellata, at 34 days of age. M. littoralis was the only medic with shorter stems (19.8 cm) than M. sativa (36.5 cm) at 44 days. M. truncatula demonstrated longer stems than the other medic species toward the end of the growth cycle, 80.0 and 83.1 cm, respectively, at 66 and 76 days. From these data on stem length it appeared that M. scutellata elongated its stems at an early stage as opposed to M. truncatula, which produced longer stems by the end of the growth cycle.

Root length:stem length ratios were associated with plant age, species, and age X species interactions (Table 6). Before 34 days such a

ratio was about 7:1 with no difference among species. This value was not much lower than the 10:1 ratio reported by Sinskaya (21). At 34 days of age \underline{T} . Subterraneum had the highest ratio (25.0) (Table 10), while \underline{M} . Subterraneum demonstrated a higher ratio (14.9) than the other species of the same genus. The ratio remained higher (11.8) in \underline{T} . Subterraneum than in the other species at 44 days of age. Changes occurring in the root length:stem length ratio were related to maturation cycle since late-maturing cultivars of \underline{M} . Subterraneum did not elongate their stems proportionally to roots as early maturing ones. After 44 days of age the ratio was statistically similar (P > 0.05) for all cultivars with a value of 1:1 at the end of the growth cycle. Generally, such a ratio tended to decrease with age in all species indicating that the plants first develop their root system, then stems.

Species grouping was determined with variance analyses for linear regression coefficients (18, 19) of roots, stems, leaves, and whole plant dry weights, and for root and stem lengths on age. All linear regressions were significantly (P < 0.001) different from zero. The trend for these regression coefficients indicated that root weight in alfalfa increased with a rate twice or more as high as that in annual medics and subclover (Table 11). Linear growth rates of stem and leaf materials were higher for alfalfa and barrel medic than for other species, as was the case for root and stem elongations. Strand medic and subclover demonstrated lower rates of dry matter accumulation than the other species. Simple correlation coefficients between each response measured and age were generally more than 0.50, particularly

for root and stem lengths (Table 11).

Test of contrasts using regression coefficients mean squares against residuals showed that \underline{M} . $\underline{truncatula}$ did not differ statistically (P > 0.05) from \underline{M} . \underline{sativa} for the responses measured, except for root dry weight (Table 12). The group constituted by \underline{M} . $\underline{truncatula}$ and \underline{M} . \underline{sativa} differed (P < 0.01) from that constituted by \underline{M} . $\underline{littoralis}$, \underline{M} . $\underline{polymorpha}$, and \underline{M} . $\underline{scutellata}$ with respect to the six measured parameters taken separately. The group containing only annual medics was not different (P > 0.05) from subclover in root, stem, and leaf dry weights and root length regression coefficients.

Combination of the six responses in the multivariate analysis with Hotelling-Lawley trace (Table 12) showed that barrel medic (\underline{M} . $\underline{truncatula}$) did not differ significantly (P > 0.05) from alfalfa. Thus, the deviation for root weights between the two species was diluted when other parameters were included in the analysis of variance. The group composed of \underline{M} . $\underline{littoralis}$, \underline{M} . $\underline{polymorpha}$, and \underline{M} . $\underline{scutellata}$ differed (P < 0.01) from that composed of \underline{M} . $\underline{truncatula}$ and \underline{M} . \underline{sativa} . However, when $\underline{truncatula}$ was considered with the other three medics in the same group, annual medics reacted differently (P < 0.01) from alfalfa. This may suggest that \underline{M} . $\underline{truncatula}$ was an intermediate species between true annuals; e.g., \underline{M} . $\underline{scutellata}$, and true perennial alfalfa. Subclover differed (P < 0.05) from true annual medics and formed a separate group.

Multivariate analysis, with Hotelling-Lawley trace, was a good tool to group species with common traits. However, it sometimes hid some differences existing among individuals of the same group for a particular variate, as was the case between \underline{M} . $\underline{truncatula}$ and \underline{M} . \underline{sativa} for root dry weight. It might also have hidden some similarities among

groups as was demonstrated between the annual $\underline{\mathsf{Medicago}}$ group and $\underline{\mathsf{T}}$. $\underline{\mathsf{subterraneum}}$. Both orthogonal contrasts and multivariate analysis were conducted on linear regression coefficients of dry weights and lengths of plant parts. This approach of condensing information should be considered as a complementary tool to regular individual analysis of variance and LSD comparisons.

The tendency of \underline{M} . $\underline{truncatula}$ Jemalong to present many growth characteristics like those of \underline{M} . \underline{sativa} may be related to the maturation cycle. This cultivar is late-maturing. It grew slowly at the beginning of the cycle, but since its growth cycle is longer than that of the other medics it accumulated more dry matter and partitioned photosynthates as alfalfa did except for root dry weight that demonstrated its annual growth characteristic.

Conclusions

The study showed that annual medics do not react similarly throughout their growth cycle for biomass partitioning and accumulation or for root and stem development. Strand medic (M. littoralis), burr medic (M. polymorpha), and snail medic (M. scutellata), represented by Harbinger, Circle Valley, and Robinson, respectively, constituted one group of species. However, the large-seeded snail medic differed from small-seeded medics at early growth for most parameters. Alfalfa (M. sativa) had to be considered individually because of its distinct growth characteristics. Barrel medic (M. truncatula) represented by Jemalong was intermediate. It behaved like the other medics for root dry weight and pod formation, but it was not different from alfalfa in total dry weight, root and stem elongation, and the derived ratios such as leaf:stem, source:sink, and root length:stem length. Barrel medic formed a separate group with regard to flowering date. Subclover (T. subterraneum) differed from Medicago spp as it produced less dry weights and shorter stems. The presence of age x species interactions for some parameters was a good indication of changes occurring in the plant at different levels among species. Flowering stage was the most determinant time for these changes and reorganization inside the plant. Seed size and maturation cycle were also important factors in species descrimination in biomass partitioning and root and stem development. Because of these factors, it appears that management of a mixture made of the four medics studied will be difficult, even when compensation for better nutrient use is present, when maximum output is desired. Any grazing period or cutting date, except after full maturation of all spcies, might favor at least one species over another in a mixture.

Table 1. Species and cultivars used in the study

Code	Genus	Species	Common name	Cult- ivar		.(g)/ seeds
1	Medicago	littoralis	strand medic	Harbinger	1	3.1
2	Medicago	polymorpha	burr clover	Circle Val	ley	4.2
3	Medicago	scutellata	snail medic	Robinson		19.6
4	Medicago	truncatula	barrel medic	Jemalong 1		3.9
5	Medicago	sativa	alfalfa	0K-83-Graz	e	3.1
6	Trifolium	subterraneum	subclover	Mount Bark	er	5.2

Table 2. Mean squares for dry weight og plant parts and total

			М	ean squares		
Source	df	Root Wt.	Stem Wt.	Leaf Wt.	Pod Wt.	Total Wt.
Repli- cation	2	9.82*	48.67**	22.50*	73.50*	546.12*
Age	6	19.57**	132.91***	95.83***	123.83**	1315.66***
Species	5	7.09***	36.25***	26.60***	88.56***	272.20***
Age X species	30	2.12***	10.42*	8.97***	19.86**	70.12**

*** : Significant (P < 0.001) ** : Significant (P < 0.01) * : Significant (P < 0.05)

Table 3. Dry weight of plant parts for species at different ages

			y weight (
Species	Root	Stem	Leaf	Pod	Total
		Age	e = 13 days	<u>.</u>	
M. littoralis	0.02	0.01	0.02	-(a)	0.04
M. polymorpha	0.02	0.01	0.02	-	0.05
1. scutellata	0.03	0.02	0.09	-	0.14
1. truncatula	0.02	0.01	0.03	-	0.06
1. sativa	0.02	0.01	0.03	-	0.06
Γ. subterraneum	0.02	0.01	0.03	. -	0.06
SD (P = 0.05)	ns(b)	ns	ns		ns
		<u>A</u>	ge = 24 day	<u>/s</u>	
1. littoralis	0.10	0.05	0.18	-	0.34
1. polymorpha	0.10	0.03	0.17	- .	0.34
1. scutellata	0.18	0.08	0.35	-	0.61
1. truncatula	0.07	0.02	0.09	-	0.18
1. sativa	0.07	0.05	0.08	-	0.20
Γ. subterraneum	0.06	0.03	0.09	-	0.17
SD (P = 0.05)	0.05	ns	0.08	-	0.11
		<u>A</u>	ge = 34 day	<u>/s</u>	
M. littoralis	0.23	0.28	0.41	-	0.93
1. polymorpha	0.57	0.43	1.04	-	2.04
1. scutellata	0.74	0.51	1.20	-	2.46
1. truncatula	0.36	0.53	0.61	-	1.50
1. sativa	0.27	0.29	0.42	-	0.98
. subterraneum	0.63	0.39	0.89		1.92
SD (P = 0.05)	ns	ns	ns	-	ns
		<u>A</u>	ge = 44 day	<u>/s</u>	
1. littoralis	0.69	0.91	1.62	0.00	3.23
1. polymorpha	1.44	2.38	3.80	2.22	9.84
1. scutellata	1.29	3.49	4.08	3.28	12.14
1. truncatula	1.47	3.21	3.94	0.00	8.63
1. sativa	1.69	2.49	2.77	0.00	6.96
. subterraneum	1.14	1.86	2.36	0.00	5.36
SD (P = 0.05)	ns	ns	ns	1.57	ns

Table 3. (Continued)

		ח	ry weight	(a)	
Species	Root	Stem	Leaf	Pod	Total
		<u>A</u>	ge = 57 d	ays	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	1.09 1.46 1.81 2.27 2.43 1.94	1.70 3.81 4.94 4.64 4.94 3.46	1.53 3.54 4.99 3.66 4.39 3.29	0.94 4.73 8.05 1.99 0.00 0.00	5.27 13.54 19.79 12.56 11.56 8.69
LSD $(P = 0.05)$	ns	ns	ns	4.29	ns
		<u>A</u>	ge = 66 d	ays	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	1.26 1.98 1.74 3.41 6.01 1.67	3.23 4.08 4.82 11.79 11.03 3.24	2.58 2.77 5.05 8.87 10.38 2.36	2.34 7.42 13.27 7.97 0.00 0.20	9.40 16.25 24.88 32.04 27.42 7.46
LSD $(P = 0.05)$	2.21	4.64	3.87	9.42	14.41
		<u>A</u>	ge = 76 d	ays	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	0.73 1.39 1.88 2.86 5.49 1.15	2.33 3.50 4.51 10.10 11.27 3.23	2.51 2.72 4.13 9.10 10.36 3.10	3.90 8.41 12.77 11.28 0.00 0.37	9.48 16.02 23.30 33.33 27.12 7.85
LSD (P= 0.05)	0.85	5.70	4.63	7.22	16.37

⁽a) : - = no pod produced.
(b) : ns = not significant (P > 0.05)

Table 4. Mean squares for derived ratios

		Mean squares		
Source	df	Source:sink	Leaf:stem	Root:total
Replication	2	0.195	1.750	0.003
Age	6	1.677*	33.387***	0.163***
Species	5	0.068	1.942	0.020**
Age X species	30	0.123**	1.991	0.005*

*** : Significant (P < 0.001)
** : Significant (P < 0.01)
* : Significant (P < 0.05)

Table 5. Ratios of each species at different ages

Species	Source:sink	Leaf:stem	Root:total
		Age = 13 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	0.85 0.81 1.56 1.07 1.30	3.37 3.18 3.34 3.50 3.39 3.28	0.40 0.41 0.21 0.34 0.28 0.34
LSD $(P = 0.05)$	0.49	ns	0.09
		Age = 24 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	1.32 1.29 1.29 1.00 0.63 0.96	6.00 6.57 4.17 3.59 1.60 3.54	0.32 0.36 0.31 0.37 0.42 0.35
LSD $(P = 0.05)$	0.49	ns	ns
		Age = 34 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	0.82 1.09 1.02 0.81 0.82 1.12	1.53 3.22 2.70 3.95 1.62 4.27	0.25 0.30 0.30 0.28 0.27 0.33
LSD $(P = 0.05)$	ns	ns	ns
		Age = 44 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	1.03 0.89 0.82 0.89 0.76 1.02	1.84 1.72 1.38 1.38 1.29 1.81	0.22 0.16 0.16 0.18 0.23 0.21
LSD $(P = 0.05)$	ns	ns	ns

Table 5. (Continued)

Species	Source:sink	Leaf:stem	Root:total
		Age = 57 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	0.41 0.45 0.42 0.58 0.61 0.64	0.91 0.95 1.03 0.93 0.88 1.03	0.22 0.12 0.11 0.18 0.19 0.22
LSD $(P = 0.05)$	ns	ns	ns
		Age = 66 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	0.40 0.26 0.31 0.42 0.63 0.49	0.82 0.76 1.05 0.75 0.95 0.74	0.14 0.12 0.08 0.12 0.21 0.21
LSD $(P = 0.05)$	ns	ns	0.07
		Age = 76 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	0.34 0.22 0.19 0.40 0.62 0.77	1.10 0.77 0.83 0.90 0.98 1.24	0.08 0.09 0.08 0.09 0.21 0.16
LSD ($P = 0.05$)	ns	ns	0.05

ns : not significant (P > 0.05)

Table 6. Mean squares for root and stem development

				Mean square	S	
		Diam-	Root	+2		t length by tem length
Source	df	eter	Length	Number	Length	ratio
Repli- cation	2	4.54	1354.55*	6.37*	183.41	27.87
Age	6	3.57	9436.90***	23.76***	11913.43***	185.33*
Species	5	5.77**	149.02**	9.42***	2128.72***	162.72***
Age X species	30	0.50	64.21	0.92	306.50***	31.02**

*** : Significant (P < 0.001) ** : Significant (P < 0.01) * : Significant (P < 0.05)

Table 7. Root diameter and maximum root length of species

Species	Root diam- eter (mm)	Root length (cm)
M. littoralis	1.2	61.4
M. polymorpha	1.3	66.1
M. scutellata	1.1	60.2
M. truncatula	1.4	71.8
M. sativa	2.5	70.3
T. subterraneum	1.4	63.2
LSD (P = 0.05)	0.5	6.9

Table 8. Root length at different plant ages

Age (days)	Root length (cm)
13	8.4
24	17.3
34	33.3
44	49.6
57	60.0
66	61.7
76	65.7
LSD (P = 0.05)	10.5

Table 9. Stem number in each species at flowering

	Species	Stem number
	M. littoralis	4.0
	M. polymorpha	4.0
	M. scutellata	2.0
*	M. truncatula	4.0
	M. sativa	3.0
	T. subterraneum	5.0
	LSD (P = 0.05)	1.5

Table 10. Stem length and root length:stem length ratio of species at different ages

Species	Stem length (cm)	Root length by stem length ratio	
		Age = 13 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	1.6 1.5 1.4 1.2 1.2	7.08 6.40 6.56 7.61 5.67 9.22	
LSD $(P = 0.05)$	· ns	ns	
		Age = 24 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	2.1 3.5 3.8 2.0 3.9 2.6	9.69 7.35 5.18 7.78 4.42 7.64	
LSD $(P = 0.05)$	ns	ns	
		Age = 34 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	6.9 9.7 14.4 5.6 17.4 1.5	6.10 6.20 2.50 14.90 1.40 25.00	
LSD $(P = 0.05)$	6.7	7.02	
		Age = 44 days	
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	19.8 32.0 33.5 29.3 36.5 4.2	2.30 1.90 1.56 2.13 1.04 11.80	
LSD $(P = 0.05)$	13.8	6.34	

Table 10. (Continued)

Species	Stem length (cm)	Root length by stem length ratio		
	Age = 57	days		
M. littoralis M. polymorpha	42.0 54.7	1.32 1.13		
M. scutellata M. truncatula M. sativa	47.5 59.9 53.8	1.27 1.08 1.26		
T. subterraneum	23.2	4.11		
LSD $(P = 0.05)$	16.5	ns		
	<u>Age = 66</u>	Age = 66 days		
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	55.8 55.2 55.4 80.0 69.3 26.1	1.10 1.16 1.05 0.84 0.98 7.06		
LSD $(P = 0.05)$	15.1	ns		
	<u>Age = 76</u>	o days		
M. littoralis M. polymorpha M. scutellata M. truncatula M. sativa T. subterraneum	67.4 65.1 57.3 83.1 78.4 29.6	0.83 1.01 1.18 0.88 0.91 4.00		
LSD (P = 0.05)	15.8	ns		

ns : not significant (P > 0.05)

Table 11. Linear regression coefficients of plant part dry weights and lengths no age for each species

Species	Root wt.	Stem wt.	Leaf wt.
M. littoralis	0.018	0.050	0.045
M. polymorpha	0.030	0.074	0.053
M. scutellata	0.030	0.093	0.086
M. truncatula	0.048	0.200	0.160
M. sativa	0.102	0.203	0.187
T. subterraneum	0.027	0.065	0.055

Table 11. (Continued)

Species	Total wt.	Root length	Stem length
M. littoralis	0.071	0.817	1.150
M. polymorpha	0.309	0.947	1.150
M. scutellata	0.460	0.951	1.030
M. truncatula	0.577	1.689	1.544
M. sativa	0.492	1.153	1.346
T. subterraneum	0.152	0.878	0.403

Table 12. Mean squares for linear regression coefficients of growth parameters on age

Source(1)	Mean squares (x10 ⁻³) Weight) Leng		Hotelling/ Lawley
(groups)	Root		Leaf	Ttl.	Root	Stem	trace
4 vs. 5	4***	0.1	1	12	6	59	29.72
Error	0.07	2	2	17	13	63	
1,2,3 vs. 4,5	8***	59**	45***	172*	167**	405*	15.73**
Error	0.07	2	2	17	13	65	
1,2,3,4 vs. 5	12***	24*	24**	28	97*	39	43.14**
Error	0.07	2	1	17	13	63	
1,2,3 vs. 6	0.2	3	2	124*	13	1597**	5.16*
Error	0.07	2	2	17	13	63	

^{(1):} 1 = M. littoralis, 2 = M. polymorpha, 3 = M. scutellata, 4 = M. truncatula, 5 = M. sativa, and 6 = T. subterraneum

*** : Significant (P < 0.001)

** : Significant (P < 0.01)

* : Significant (P < 0.05)

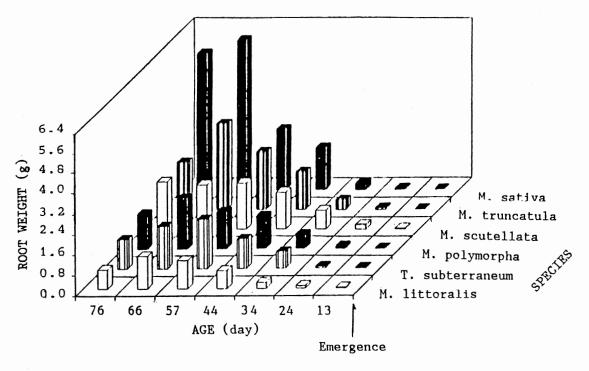


Fig. 1. Changes in Root Dry Weight (g) of the Species Through Time (days)

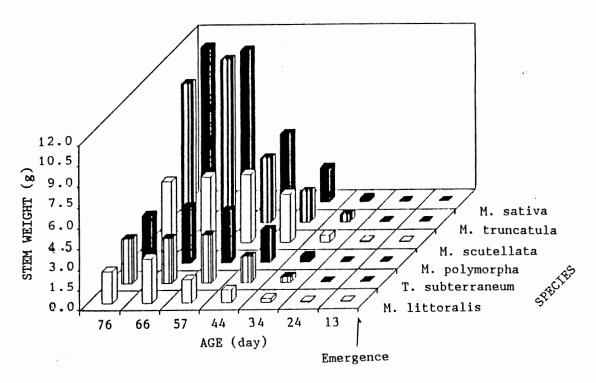


Fig. 2. Changes in Stem Dry Weight (g) of the Species Through Time (days)

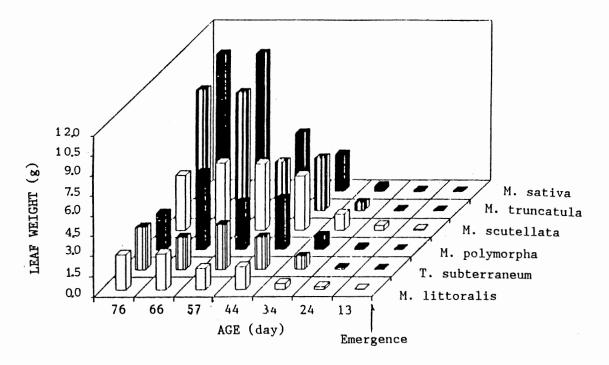


Fig. 3. Changes in Leaf Dry Weight (g) of the Species Through Time (days)

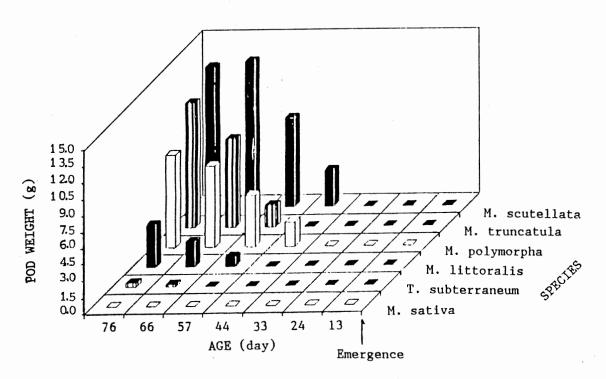


Fig. 4. Changes in Pod Dry Weight (g) of the Species Through Time (days)

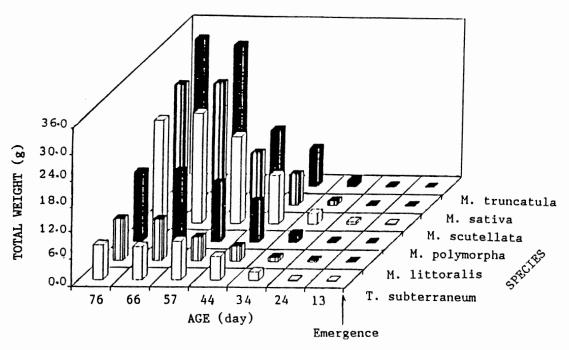


Fig. 5. Changes in Total Dry Weight (g) of the Species Through Time (days)

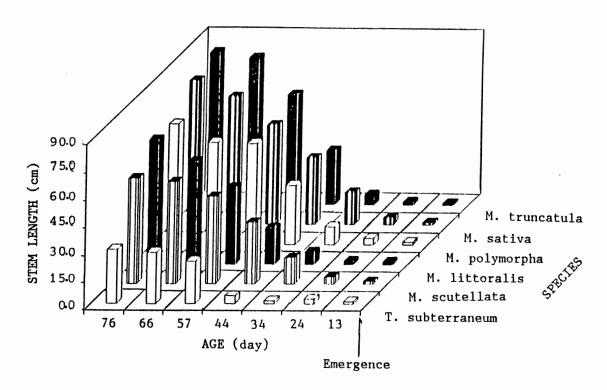


Fig. 6: Changes in Stem Length (cm) of the Species Through Time (days)

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CHAPTER V

GENERAL CONCLUSIONS

Annual <u>Medicago</u> species present a wide range of genetic material.

Depending on environmental conditions, they may be more or less productive than perennial alfalfa (<u>M. sativa</u>) and clovers (<u>Trifolium</u> spp.). Genotype X environment interactions were strongly present for emergence percentage, forage and seed yields, but to a lesser extent for cold survival, growth habit, and forage quality.

Neither artificial inoculation nor nitrogen fertilizer affected the agronomic traits measured; thus, it is strongly suggested that soils of the Southern Great Plains contain effective <u>Rhizobium</u> spp. to inoculate annual <u>Medicago</u> spp., <u>M. sativa</u>, and <u>Trifolium</u> spp. It was difficult to draw conclusions about effects of soil acidity on production of genotypes studied because of waterlogging and frost damage which occurred during these investigations.

Fall sown accessions studied in Oklahoma resulted in higher emergence percentage than in spring planting. However, due to the existence of G X E interactions, desirable stand densities ought to be planned according to emergence percent and seeding rate for each cultivar at a given environment. Application of the same seeding rate over environments and for all cultivars will generate different stand densities, not only because of G X E interactions, but also since

emergence depends on seed quality and seedbed conditions, and because annual Medicago species have different seed sizes. On the other hand, fall planting resulted in total stand loss of annual Medicago species under continental winter conditions in Oklahoma when snow did not cover plants. Nevertheless, M. arabica, M. blancheana, M. minima, and M. polymorpha possessed some cold tolerance when temperatures stayed above OoC, while the other medic species were very cold sensitive.

M. sativa and Trifolium spp. were much more tolerant to cold than medics. Large screening and/or intensive selection programs will be necessary to develop new cultivars with tolerance to cold in the U.S. Southern Great Plains if these species are to be managed as selfseeded pasture species. The four mentioned medics, in addition to local ecotypes and other germplasm from cold regions, may be good sources of germplasm for future investigations.

Certain genotypes of annual <u>Medicago</u>, e.g., <u>M. scutellata</u>, produced as much forage yield as <u>M. sativa</u> or <u>Trifolium</u> spp., or even more when they were spring planted in Oklahoma. All medics were more productive than alfalfa and clovers when they were fall planted in their naturally adapted mediterranean habitat, as shown from the Moroccan plantings in these studies. Some genotypes, e.g., those of <u>M. scutellata</u>, excelled in a wide range of environments, whereas some, e.g., <u>M. rugosa</u> Sapo, performed poorly in all environments; others were not consistent in their response. Establishment success positively influenced forage yield in Oklahoma environments with spring planting, but not in Moroccan ones with fall planting.

Forage of annual <u>Medicago</u> species, as well as of perennial \underline{M} . <u>sativa</u> and <u>Trifolium</u> species, was of good quality at flowering. It contained high crude protein concentrations, low proportion of neutral and acid detergent fibers, and fairly good leaf:stem ratio. Forage quality in these species appeared to be the less variable agronomic parameter over a wide range of environments.

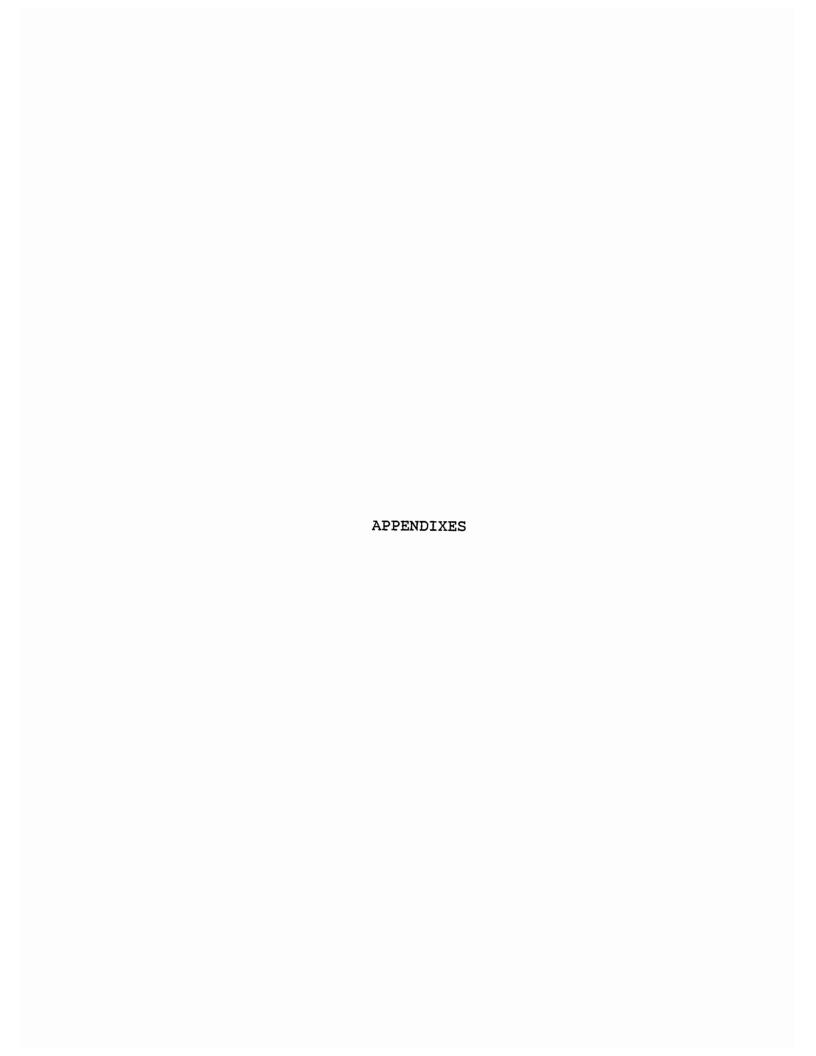
Good seed production was obtained using certain genotypes with spring plantings in Oklahoma and the best seed yield was from M. scutellata, which was similar to alfalfa in number of offspring seeds. However, pest infestation and probably lack of vernalization were major factors contributing to lack of flowering or flower abortion, and incomplete seed development observed in most medic genotypes studied. Further studies on these factors will improve seed yields in annual Medicago species. In Morocco, insect infestation and vernalization did not appear to be important factors. Thus, all annual Medicago species, particularly small-seeded ones, produced a high amount of seed during the first year, but a very small amount in the second year when low rainfall limited stand establishment and growth. Trifolium species did not produce seed in Oklahoma nor in Morocco, and nor did M. sativa in Morocco. Conclusions from Moroccan studies on seed need to be used with caution as medics were evaluated in one location, Sidi el Aydi. Total number of offspring seeds was positively correlated to number of seeds/pod and to the proportion of hardseed. Therefore, selection for high number of seeds/pod will improve seed yield and seed quality.

Regeneration from offspring seeds was excellent with \underline{M} . $\underline{scutellata}$ and \underline{M} . \underline{sativa} in the subsequent fall at Stillwater, OK, but it was very limited with the other species. Regeneration was absent at Perkins, OK. High proportion of hardseed in medics was the main reason for low number

of offspring seedlings in most species. Further investigations are necessary to follow the fate of remaining seeds in soil. Medic offspring plants germinated in fall and died during the winter which emphasized the need to improve cold tolerance in these species if natural reseeding is to be expected in the Southern Great Plains of the U.S. Another possible alternative to escape winter kill will be the development of cultivars with reliable seed dormancy to allow germination only after danger of freezes are past.

Annual Medicago species did not behave in the same way in their dry matter accumulation, biomass partitioning, and root and stem development. Seed size and maturation cycle were the source of differences observed among species. Management of mixtures containing cultivars which differ in their seed size and maturation cycle will be difficult to optimize.

Differences associated with genotypes and environments and the presence of G X E interactions were characterized in this study. They demonstrated that annual Medicago species do not constitute a homogeneous group of forage species. This concept in medic research is strongly defendable since the same seedlots of different genotypes were used over soil types, seasons, and years in the study (in addition to adjustments made each time for seed viability). This concept has already been elaborated in the important work of Heyn (51) and Lesins and Lesins (60) concerning the taxonomy of the genus Medicago. Possible advances in selection for desirable agronomic traits seems promising with further categorization and delineation of variability in the genus Medicago.



List of insects present on each plant genotype at Stillwater, May 1984

Cultivar	Insect
M. arabica	Thrips, Pea aphids, Spotted aphids, Blue aphids, Corn ear worms, Lady beetle larvae, Leaf hopper nymphs
M. aculeata	Thrips, Lygus nymphs, Leaf hopper nymphs and adults, Blue aphids, Damsel nymphs, Spotted aphids, Pea aphids, Long-horned grasshopper nymph, Lady beetle larvae
M. blancheana	Thrips, Spotted aphids, Lygus nymphs, Leaf hopper nymphs, Damsel nymphs, Corn ear worms, Blue aphids, Lady beetle larvae, Pea aphids
M. littoralis Harbinger 1	Thrips, Lady beetle adults and larvae, Spotted aphids, Pea aphids, Leaf hopper nymphs and adults, Damsel nymphs, Blue aphids, Corn ear worm
M. littoralis Harbinger 2	Thrips, Pea aphids, Spotted aphids, Damsel nymphs, Lady beetle larvae and adults, Blue aphids, Leaf hopper nymphs
M. minima	Thrips, Spotted aphids, Lygus nymphs, Damsel nymphs,
M. murex	Thrips, Springtails, Spotted aphids, Leaf hopper nymphs, Damsel nymph, Corn ear worm, Pea aphids, Lygus nymphs
M. polymorpha strain 1	Thrips, Springtails, Damsel nymphs, Spotted aphids, Leaf hopper nymphs and adults, Lace wing larvae, Pea aphids, Lygus nymphs, Blue aphids

APPENDIX (Continued)

Cultivar	Insect
M. polymorpha Cercle Valley	Thrips, Pea aphids, Spotted aphids, Lygus nymphs and adult, Lady beetle adult, Leaf hopper nymph
M. rugosa Paragosa	Spotted aphids, Pea aphids, Lygus nymphs, Thrips
M. rugosa Paraponto 1	Thrips, Leaf hopper nymphs and adults, Spotted aphids, Damsel nymphs, Lady beetle adults
M. rugosa Paraponto 2	Thrips, Spotted aphids, Pea aphids, Leaf hopper nymphs, Blue aphids, Lady beetle larvae
M. rugosa Sapo	Thrips, Lygus nymph, Spotted aphids
M. sativa OK-83-Graze	Thrips, Pea aphids, Spotted aphids, Leaf hopper adult and nymph, Lady beetle larvae, Blue aphid, Damsel adult, Lygus nymph
M. sativa Spredor II	Thrips, Spotted aphids, Leaf hopper nymphs, Pea aphids, Lygus nymphs, Lady beetle larvae
M. scutellata Robinson	Pea aphids, Blue aphids, Spotted aphids, Yellow striped army worm, Lygus nymphs, Damsel nymphs, Lady beetle larvae, Thrips
M. scutelatta Sava	Thrips, Pea aphids, Leaf hopper nymphs
M. scutellata Sava	Lygus nymphs, Corn ear worms, Springtails, Lady beetle larvae
M. tornata Tornafield 1	Thrips, Lady beetle larvae and adults, Pea aphids, Lady nymphs and adults, Spotted aphids, Leaf hopper aphids, Lygus nymphs

APPENDIX (Continued)

Cultivar	Insect
M. tornata Tornafield 2	Thrips, Damsel nymphs and adults, Pea aphids, Leaf hopper nymphs and adults, Spotted aphids, Lady beetle larvae, Lygus nymphs
M. truncatula Borung	Thrips, Lady beetle larvae, Spotted aphids, Pea aphids, Lygus nymphs, Leaf hopper nymphs, Damsel nymphs
M. truncatula Cyprus	Thrips, Pea aphids, Spotted aphids, Blue aphids, Leaf hopper adults, Corn ear worms, Lady beetle adults, Damsel nymphs
M. truncatula Jemalong 1	Thrips, Lady beetle adult, Spotted aphids, Leaf hopper nymph and adults, Lygus nymphs, Pea aphids, Damsel nymphs
M. truncatula Jemalong 2	Thrips, Spotted aphids, Leaf hopper nymphs and adults, Pea aphids
M. truncatula Paraggio	Thrips, Pea aphids, Spotted aphids, Blue aphids, Damsel adults
T. subterraneum Mount Barker	Thrips, Lygus nymphs, Leaf hopper nymph, Pea aphids, Corn ear worms, Blue aphids, Reduviidae
T. vesiculosum Yuchi	Thrips

(1): Insect population was rated though a scale from 1 to 9 with 1 represented less than 50 insects while 9 represented more than 1000 in three sweeps.

Insects collected were determined by Phoebe Courtney a technician at the Entomology Dept. Okla. State Univ.

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IN ANNUAL MEDICAGO SPP.

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