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# High-yielding anthracnose-resistant *Stylosanthes* for agricultural systems



Editor: S. Chakraborty

# High-yielding anthracnose-resistant *Stylosanthes* for agricultural systems

Edited by Sukumar Chakraborty



Australian Centre for International Agricultural Research  
Canberra 2004

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Chakraborty, S. (ed.) High-yielding anthracnose resistant *Stylosanthes* for agricultural systems. ACIAR Monograph No. 111, 268 p.

ISBN: 1 86320 442 3

1 86320 443 1

Technical editing: Joanne Mason, Mason Edit, Adelaide

Design and layout: Fivefold Creative

Printing: Union Offset

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## Foreword

*Stylosanthes* is a fast-growing tropical legume, with a number of uses. In India it is the most important tropical legume for semi-arid and arid regions, where it is used for livestock production and to restore soil fertility. In China, freshly cut and processed leaf meal is used to feed animals. It is also used as a green manure for soil enhancement.

In both countries *Stylosanthes* species contribute to the supply of forage for cattle, buffalo, goats, sheep and pigs. As a nitrogen-fixing legume, the plant helps replenish soil nutrients when used in ley farming, mixed cropping and intercropping systems. It helps restore marginal lands with infertile acid soils and is a pioneer coloniser in the revegetation of wastelands.

*Stylosanthes* is of great commercial importance to Central and South America, Asia and Africa. The beef industry in northern Australia is increasingly reliant on *Stylosanthes* as a protein-rich pasture plant for cattle. The major constraint to its use, wherever it is grown, is anthracnose, a disease caused by a fungus that is diverse and quick to adapt. With growing international travel and trade, the risk of even more damaging fungal strains entering Australia, India or China is increasing.

Australia has internationally recognised experience in tropical pasture technology in general, and *Stylosanthes* in particular, which was used in the project supported by ACIAR to help combat the anthracnose disease problem.

The aim of the project was to select germplasm of *Stylosanthes* resistant to anthracnose, with the ultimate goal to provide high-yielding, disease-resistant varieties of *Stylosanthes* that will improve on the current varieties in use in India, China and northern Australia.

Some of the achievements of the project were the broad testing of *Stylosanthes* germplasm at 20 sites across the world, the release of several Brazilian and Chinese cultivars, knowledge about the nature of pathogen populations in India, Brazil and Australia and new information about anthracnose disease threats to Australia from China and India.

The project has expanded the scientific base considerably for continued and targeted improvement in anthracnose resistance in *Stylosanthes* in China, India and Brazil, which are major developing countries for livestock production. In India, *Stylosanthes* adoption is being encouraged by opportunities for commercial marketing of leaf meal and seed. Leaf meal production is largely carried out by smallholder farmers who interact with large poultry producers through farmer co-operatives.

The gradual replacement of existing cultivars with high-yielding and resistant varieties will mainly be demand-driven as benefits of growing the new high yielding varieties become apparent through ongoing demonstrations by government and NGOs.

Other practical benefits to the smallholder farmers will flow as the improved technology for stylo cultivation and utilisation becomes widely known through the strategic alliances already in operation.

As well as highlighting the key achievements of the ACIAR project, this volume contains contributions from internationally recognised authorities in *Stylosanthes* research and examines some of the existing and potential commercial utilisation schemes for *Stylosanthes*. It brings together 20 years of research on this important plant and is the latest in ACIAR's monograph series. It is also available on ACIAR's website [www.aciar.gov.au](http://www.aciar.gov.au)



Peter Core  
Director  
Australian Centre for International Agricultural Research





## Preface

Species of *Stylosanthes* (stylo) are among the most versatile, widely adapted and productive tropical pasture legumes commercially used in a range of agricultural systems in many countries with a tropical or subtropical climate. They form the basis of a substantial beef industry in tropical Queensland in Australia, where stylo covers over one million hectares. Stylo is used in many Asian countries including China, India, Thailand, Malaysia, Indonesia, Vietnam, Laos and the Philippines, among others, as a cut-and-carry fodder, a cover crop to suppress weeds and provide forage under plantations of horticulture and forestry species, a green manure to enhance soil fertility and nitrogen, a pioneering coloniser of problem soils that are heavily degraded and, more recently, a cash crop to produce dried leaf meal as an ingredient of animal feed formulations for poultry and other industries. *Stylosanthes* is a promising crop for many countries in Africa where it is principally used as cut-and-carry fodder, protein banks, fallow crops and hay. At its principal centre of diversity in the Americas commercial use of *Stylosanthes* has not expanded as rapidly as in some other countries, but recent developments arising from the release of new cultivars have generated a renewed enthusiasm to offer opportunities for large-scale commercial development.

Helen Stace and Les Edey published the first monograph on stylo, *The biology and agronomy of Stylosanthes*, in 1984. This publication offered a comprehensive treatise on all aspects of *Stylosanthes* biology and production through invited chapters from internationally recognised authors from a number of countries. Since then two other publications have dealt with specific or regional aspects of *Stylosanthes* research and development. De Leeuw et al in 1994 published the proceedings of a regional workshop on the use of *Stylosanthes* in West Africa, held at Kaduna, Nigeria, in October 1992; and Winks and Chakraborty in 1997 published in *Tropical Grasslands* volume 31(5) the proceedings of a workshop on *Stylosanthes* held in Australia. Since Stace and Edey's publication, a large amount of knowledge has accumulated in the past two decades. During this time 15 new cultivars have been released from six species including the previously unknown *S. seabrana*: nine *S. guianensis* cultivars, one each in Brazil, Peru and the USA and six in China; two *S. scabra* cultivars in Australia; two *S. seabrana* cultivars in Australia; one *S. hamata* cultivar in Australia and one *S. capitata* – *S. macrocephala* multiline cultivar in Brazil.

The fungal disease anthracnose continues to be the most significant impediment to the commercial utilisation of *Stylosanthes* worldwide. The disease has devastated productive cultivars in Australia and every other country where *Stylosanthes* is grown and, consequently, many susceptible cultivars have been withdrawn from commercial production. Due to its economic significance, internationally coordinated research

programs in the last two decades have concentrated on the biology, epidemiology and management of anthracnose. The Australian Centre for International Agricultural Research (ACIAR) has been instrumental in funding some of this international collaborative research. Other agencies including the Australian Cooperative Research Centre for Tropical Plant Protection, AusAID, Department for International Development (DFID), the Asian Development Bank and the national agricultural research organisations in many countries have also contributed to this research and development.

This publication summarises new knowledge from *Stylosanthes* research and development in recent years. Given the rapidly declining number of researchers engaged in pasture and forage crops, this timely publication attempts to capture published and unpublished information on the anthracnose disease and the development of new germplasm with anthracnose resistance and high yield potential for existing and new production systems. The book is aimed at active researchers, practitioners, professionals, producers and growers interested in *Stylosanthes*. It is organised in three separate sections: Section A, '*Stylosanthes*: the versatile tropical legume', contains review and overview chapters from internationally recognised authorities in relevant areas in an easy-to-read format in plain English, illustrated with appropriate colour images. Section B, 'Anthracnose-resistant *Stylosanthes*', highlights key achievements of ACIAR projects involving Australia, Brazil, China, Colombia and India in a refereed journal article format. Chapters are organised in two subsections, 'Germplasm evaluation & cultivar development' and 'Pathogen biology and epidemiology'. Contributors are collaborating researchers from partner countries associated with the ACIAR projects on *Stylosanthes*. Section C, 'Practical and commercial utilisation of *Stylosanthes*', offers an analysis of some of the existing and potential commercial utilisation schemes for *Stylosanthes* and practical advice.

Drs Bob Clements, Colin Piggin and Tony Fischer of ACIAR; Dr Peter Kerridge and Carlos Lascano of CIAT; and Dr Don Cameron of CSIRO were instrumental in establishing the ACIAR project. I am indebted to Drs Segenet Kelemu and John Miles from CIAT; Drs Celso Fernandes, Maria Jose Charchar and Ronaldo Andrade from EMBRAPA; Drs C.R. Ramesh, P.S. Pathak, N.P. Melkania, Amaresh Chandra and C.R. Hazra from IGFRI; Mr Liu Guodao, Yi Kexian and Bai Changjun from CATAS; Mr Bob Davis and Ian Staples from QDPI; Drs Raymond Jones, Bruce Pengelly, John Manners, Kemal Kazan, Chunji Liu, Pauline Weeds and Neal White and other colleagues from CSIRO for input into ideas and discussions.

Dr Sukumar Chakraborty  
CSIRO Plant Industry







## Section A

# ***Stylosanthes*: a versatile tropical legume**





# Chapter 1

## History, relationships and diversity among *Stylosanthes* species of commercial significance

Brigitte L. Maass<sup>1</sup> and Mark Sawkins<sup>2</sup>

### Summary

After briefly reviewing the history of discovery of the agricultural value of the genus *Stylosanthes*, this paper elaborates on new insights into phylogeny and taxonomy as well as advances in the understanding of the distribution of species and genetic diversity. With the help of molecular marker techniques, major advances have been achieved in clarifying phylogenetic relationships of species and their genome structures, leading to postulation of putative ancestors of most polyploid species of *Stylosanthes*. The general model for polyploidy of combining diploid species from the two sections *Stylosanthes* and *Styposanthes* in allotetraploid species found in sect. *Styposanthes* has been confirmed, except for *S. capitata*. A number of taxonomic issues are yet to be resolved. As more diverse genetic resources from new areas are studied, inconsistencies and controversies show up. Issues relating to the *S. guianensis* and *S. scabra* species complexes are discussed in greater detail. The recognition and validation of *S. seabrana* has been a major advance; however, a similar situation is still unresolved for *S. hamata* (4n). Despite its present status of *nomen nudum*, *S. hemihamata* is suggested for these tetraploid plants, including cv. Verano. Validation of this taxon is urgently needed. The integration of new techniques, such as molecular markers and geographic information systems, has improved understanding of patterns of diversity, for example in *S. humilis*. The multiple roles of *Stylosanthes* in production systems and their impacts, including potential negative environmental impacts such as soil acidification and invasiveness, are briefly outlined. Finally, prospects for future achievements in plant improvement are presented.

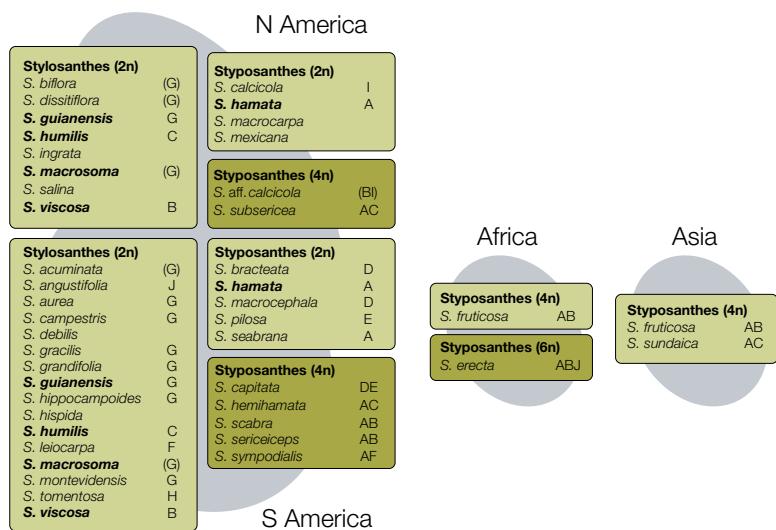
<sup>1</sup> Institute for Crop and Animal Production in the Tropics, Georg-August-University Göttingen, Grisebachstr. 6, D-37077 Göttingen, Germany bmaass@gwdg.de

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### History

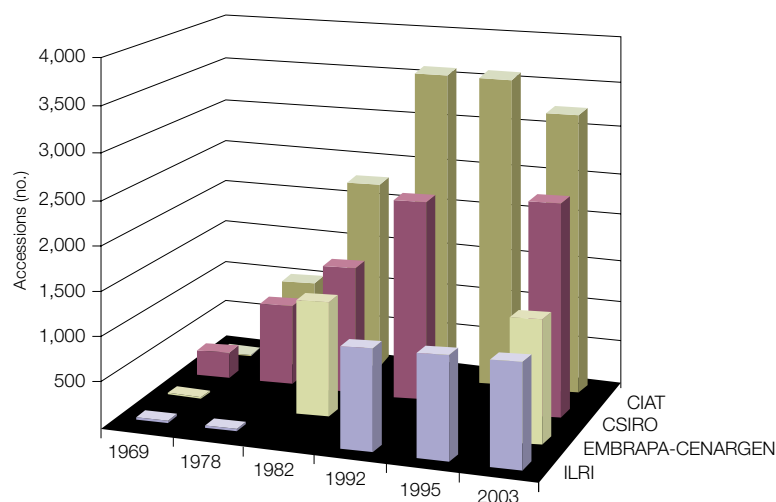
Anyone who has ever collected forage germplasm in tropical Brazil would agree that it is virtually impossible not to step on a *Stylosanthes* plant when the car stops at a new collecting site. Different *Stylosanthes* species (stylo) are very widely distributed throughout Brazil, one of the main centres of diversity (Costa & Ferreira 1984). Except for *S. fruticosa*, *S. erecta* and *S. sundaica*, the genus is native to tropical America (Figure 1.1). Species are part of the natural flora in many regions of South and Central America, Mexico and the Caribbean. It was only in 1914 that the economic value of *S. humilis*, commonly referred to as 'Townsville stylo', was recognised for Australian agriculture. This was followed by *S. guianensis* in Brazil in 1933 (Stace & Edye 1984). In their '*Stylosanthes* story', Burt and Williams (1975) initially described the rising interest in these two species. Graziers and extension officers spread sufficient seed along roads, railways and stock routes that, by 1925, a large-scale invasion was under way. Consequently, by the 1970s *Stylosanthes* had naturalised in at least 0.5 million ha in northern Australia (Miller et al 1997).

However, the annual *S. humilis* suffered in competition with native grasses and, in 1973, was harmed heavily from a serious outburst of anthracnose disease (caused by the fungus *Colletotrichum gloeosporioides*); consequently, interest turned to other perennial legumes for the dry tropics (Burt et al 1979; Edye 1997). This provided the impetus for the exploration of species other than *S. humilis* and *S. guianensis* that led to the gathering of large and comprehensive germplasm collections both by CSIRO and CIAT in collaboration with national organisations (Schultze-Kraft & Keller-Grein 1994; Schultze-Kraft et al 1984). Genetic resources of *Stylosanthes* have been reviewed by Schultze-Kraft et al (1984) and more recently by Hanson and Heering



**Figure 1.1** Species membership to sections *Styposanthes* or *Stylosanthes*, genome and diversity in relation to their natural distribution based on taxonomic, genetic and geographic data (Mannetje 1984; Williams et al 1984; Liu et al 1999; Gama López et al 2001). Bold indicates species with Panamerican distribution; the genome structure is given in capital letters after Liu et al (1999) or Ma et al (unpublished), or derived (in parentheses) from Vander Stappen, de Laet et al (2002) or Vander Stappen, Gama López et al (2002). Note that only species of known ploidy level are included, mostly named as published in the reference, without resolving nomenclatural problems.

(1994). However, the growth of these collections peaked in the late 1980s and early 1990s, and sizes have remained almost stable since (Figure 1.2). The last collecting mission specifically targeting *S. seabrana* in the Brazilian Cerrados was in 1994 by L.A. Edye and R.A. Date (Edye & Maass 1997). Germplasm collections are maintained in compliance



**Figure 1.2** Development of major *ex situ* germplasm collections of *Stylosanthes* (Burt et al 1983; Hanson & Heering 1994; Maass & Mannetje 2002; Maass et al 1997; Schultze-Kraft et al 1984; J. Hanson, pers. comm.; B.C. Pengelly, pers. comm.).

with international standards; however, overall forage genetic resources activities have strongly declined in the past decade (Maass & Pengelly 2001). CSIRO has even discontinued its active collection, but a duplicate of it is held at CIAT to service international requests (B.C. Pengelly, pers. comm. 2004).

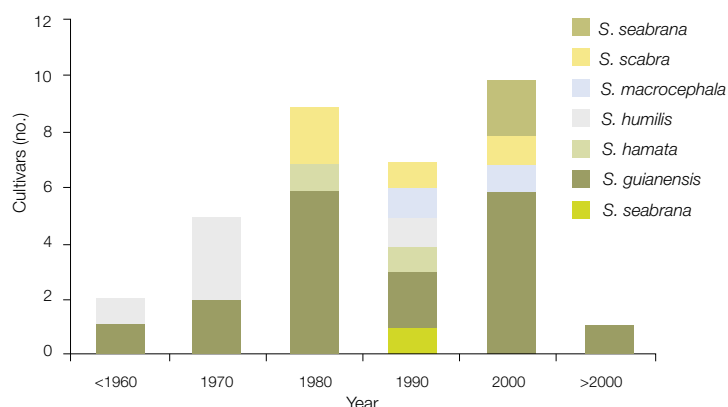
The single most important challenge for *Stylosanthes* as a new genus to agriculture has been the susceptibility to anthracnose disease by most of its species. Identification that several released cultivars were not productive anymore (*S. guianensis* cvs. Schofield, Endeavour, Cook; *S. humilis* Townsville stylo; *S. scabra* cv. Fitzroy) stimulated an intensification of anthracnose research by CSIRO, CIAT and collaborating institutions (Cameron et al 1996; Chakraborty, this volume; Kelemu et al, this volume). It also led to more effective genetic resources activities with systematic germplasm collecting and subsequent agronomic evaluation, resulting in novel species to agriculture such as *S. capitata*, *S. hamata* (4n), *S. scabra* and *S. seabrana*. Breeding programs were initiated by CSIRO, CIAT and EMBRAPA in addition to their germplasm activities (Cameron et al 1997; Miles & Grof 1997). Nevertheless, only two released cultivars have been bred to date, cv. Siran (Cameron et al 1996) and cv. Estilosantes Campo Grande (Grof et al 2001) (Table 1.1). Besides several cultivars registered in Australia, systematic screening and selection (eg Edye 1997; Schultze-Kraft & Keller-Grein 1994) from available natural variability has also resulted in the release of commercial materials in South America and Asia (Table 1.1). Half of the approximately 30 cultivars released are from *S. guianensis* and related species. The early cultivars belonged to different varieties of *S. guianensis* and *S. humilis*, which were widespread and particularly successful legumes in pastures on infertile soils. Novel species, such as tetraploid *S. hamata* and *S. scabra*, have been included in the lists of commercial materials since the mid 1970s. A peak of cultivar development seems to have passed since the 1990s (Figure 1.3), although new, regionally adapted materials have been released in India and China (Guodao et al 2002; Loch & Ferguson 1999; Ramesh et al 1997).

The rapid adoption of pasture improvement technology in Australia has resulted in the continual oversowing of tens of thousands of hectares of native pastures annually, primarily with *S. scabra* cv. Seca and tetraploid *S. hamata* cv. Verano, which today amounts to approximately 1 million ha (Noble et al 2000). Despite similar research efforts and investments, no such success story can be told for the Americas, and spontaneous adoption of *Stylosanthes* cultivars to date has been very limited (Kalmbacher et al 2001; Miles & Lascano 1997). In contrast, in the subhumid region of West Africa, 19,000 ha of *Stylosanthes* were cultivated in fodder banks by about 27,000 smallholder adopters by the mid 1990s (Elbasha et al 1999). Developments in southern China have exceeded this, where a further 0.1 million ha of predominantly *S. guianensis* (CIAT 184) in monoculture, often associated with perennial tree crops, have been established in the past decade (Guodao et al 1997).

**Table 1.1 Released cultivars of *Stylosanthes* species.**

Species	Cultivar	Country	Year	Reference
<i>S. capitata</i>	Capica	Colombia	1982	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>guianensis</i>	Schofield	Australia	(1930s)	Oram 1990
<i>S. guianensis</i> var. <i>guianensis</i>	Cook	Australia	1971	Oram 1990
<i>S. guianensis</i> var. <i>guianensis</i>	Deodoro	Brazil	(?)	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>guianensis</i>	Deodoro II	Brazil	(?)	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>guianensis</i>	Endeavour	Australia	1971	Oram 1990
<i>S. guianensis</i> var. <i>guianensis</i>	Graham	Australia	1979	Oram 1990
<i>S. guianensis</i> var. <i>guianensis</i>	IRI-1022	Brazil	(early 1970s)	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>guianensis</i>	Pucallpa (= Reyán II Zhuhuacao)	Peru, China	1985, 1991	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>guianensis</i>	Savanna	USA	1992	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>guianensis</i>	Mineirão	Brazil	1993	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>intermedia</i>	Common	Australia, Zimbabwe	1965, (early 1970s)	Loch & Ferguson 1999
<i>S. guianensis</i> var. <i>intermedia</i>	Oxley	Australia	1969	Oram 1990
<i>S. guianensis</i> var. <i>pauciflora</i>	Bandeirante	Brazil	1983	Loch & Ferguson 1999
<i>S. guianensis</i>	Reyan 5	China	1998	Guodao et al 2002; Guodao et al, this volume
<i>S. guianensis</i>	Reyan 7	China		Guodao et al, this volume
<i>S. guianensis</i>	Reyan 10	China	2001	Guodao et al 2002
<i>S. guianensis</i>	Reyan 13	China	2000?	Guodao et al, this volume
<i>S. guianensis</i>	907	China	2000?	Guodao et al, this volume
<i>S. hemihamata</i> <sup>a</sup>	Verano	Australia, Thailand	1973, 1975	Oram 1990; Loch & Ferguson 1999
<i>S. hemihamata</i> <sup>a</sup>	Amiga	Australia	1988	Oram 1990
<i>S. humilis</i>	Common	Australia	(pre-1914)	Loch & Ferguson 1999
<i>S. humilis</i>	Gordon	Australia	1968	Oram 1990
<i>S. humilis</i>	Lawson	Australia	1968	Oram 1990
<i>S. humilis</i>	Paterson	Australia	1968	Oram 1990
<i>S. humilis</i>	Khon Kaen	Thailand	1984	Loch & Ferguson 1999
<i>S. macrocephala</i>	Pioneiro	Brazil	1983	Loch & Ferguson 1999
<i>S. macrocephala</i> + <i>S. capitata</i> (mixture)	Estilosantes Campo Grande	Brazil	2000	Verzignassi & Fernandes 2002
<i>S. scabra</i>	Seca	Australia	1976	Oram 1990
<i>S. scabra</i>	Fitzroy	Australia	1979	Oram 1990

<sup>a</sup> tetraploid (4n) species, commonly named *S. hamata* (see text)



**Figure 1.3 Released Stylosanthes cultivars.**

Typical for a new genus to agriculture, research in all aspects was required, ranging from basic biology to agricultural utilisation. From the considerable literature produced, three larger volumes (Table 1.2) comprehensively dealt with the multiple facets of research into *Stylosanthes*. Closing the first international symposium exclusively dedicated to this genus, Humphreys (1984) listed five major areas which he saw as priorities for future research and development, including:

- adoption of correct nomenclature for plant material and accessibility of related biological and agronomic information
- knowledge generation regarding environmental adaptability and target production systems
- understanding of plant strategies of persistence (eg growth, survival, plant replacement, N fixation, tolerance to environmental stresses)
- appreciation of all aspects of biology to underpin agronomic advance, particularly nutritive value
- international collaboration on creation of a newsletter dedicated to the genus.

**Table 1.2 Compilation and exchange of knowledge on Stylosanthes.**

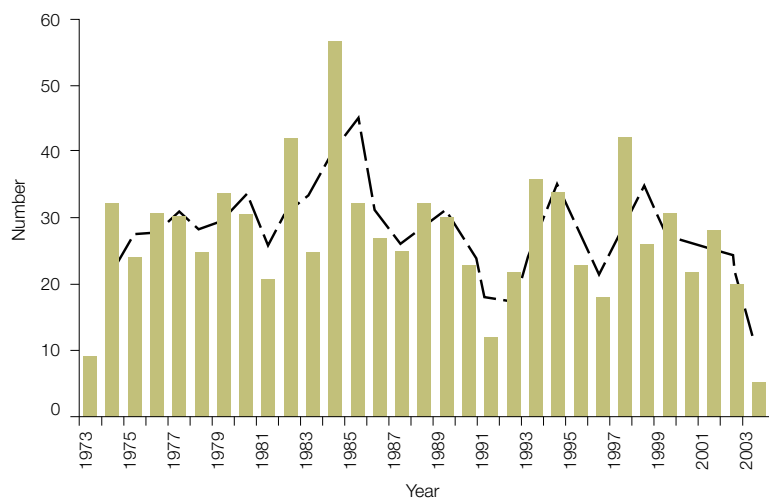
Event	Publication (title)	Authors, year
International study, invited contributions	The role of <i>Centrosema</i> , <i>Desmodium</i> and <i>Stylosanthes</i> in improving tropical pastures	Burt et al 1983
International symposium at Townsville, Australia in 1982	The biology and agronomy of <i>Stylosanthes</i>	Stace & Edye 1984
Regional workshop at Kaduna, Nigeria in 1992	<i>Stylosanthes</i> as a forage and fallow crop	de Leeuw et al 1994
International workshop at Townsville, Australia in 1996	International research and development on <i>Stylosanthes</i>	Winks & Chakraborty 1997
ACIAR international project in Australia, Brazil, Colombia, India and China	<i>Stylosanthes</i> International Newsletter (since 1999)	<a href="http://www.csiro.au/stylointernational">http://www.csiro.au/stylointernational</a>

These issues have since received further attention, largely in collaborative projects, with the annual number of publications on *Stylosanthes* over the past decades remaining remarkably stable (Figure 1.4). Following the last workshop on *Stylosanthes* research and development in 1996 (Winks & Chakraborty 1997), a wide range of research articles has been published on such diverse themes as grazing management and animal nutrition (Paciullo et al 2003), ecophysiological adaptation (Lovato et al 1999), allelopathy (Hu & Jones 1997), seeds (McDonald 2002) and silvipastures (Srinivasan 1999), to only give a few examples. However, our opinion is that over the last 20 years the most important success stories have been concerned with:

- large-scale adoption of *S. guianensis* var. *vulgaris* (CIAT 184) in China
- processing of stylo herbage into leaf meal, pelleting and use for monogastric feed in Asia
- adoption by smallholders of *S. guianensis* cv. Cook together with tetraploid *S. hamata* cv. Verano for use in fodder banks in West Africa
- discovery of the agricultural value of *S. seabrana* and its taxonomic recognition, cultivars releases and adoption in Australia
- new knowledge of the anthracnose pathogen and the release of anthracnose-resistant cultivars in Australia, Brazil, China and India
- advances in the understanding of species relationships and genetic structures.

Advanced understanding of the taxonomic and phylogenetic relationships between *Stylosanthes* species, their distribution and genetic diversity has greatly aided germplasm collection, evaluation and improvement to underpin practical usage of the species described in other chapters. This chapter deals with the relationships and diversity among *Stylosanthes* species of commercial significance without presuming to perform a taxonomic review.





**Figure 1.4** Publications (no. of hits) on *Stylosanthes* retrieved from Centre for Agriculture and Biosciences International (CABI) herbage abstracts 1973–2003.

## New Insights into Taxonomy and Phylogeny

Mannetje (1984) reviewed the history of the genus *Stylosanthes*, which was established by Swartz in 1788 and later critically revised by Vogel in 1838, Taubert in 1891 and Mohlenbrock in 1958 and 1963. An increase in species described from 6 in 1800 to 25 in 1984 was followed by a relative stability of numbers until the 1990s. However, the validity of

several species has been debated since this time and different views about the taxonomic treatment at specific and infraspecific levels have persisted. For example, Mannetje (1984) recognised seven varieties in *S. guianensis*, in contrast to Ferreira and Costa (1979) and Costa and Ferreira (1984), who regarded many of these taxa as distinct species (Table 1.3). These different taxonomic approaches have been adopted in different institutions and may have led to confusion in the interpretation of data. For example, the International Legume Database and Information System (ILDIS 2001) registered a total of 42 valid species (31 accepted and 11 provisional) in addition to one misapplied species name and 43 synonyms. However, without a well-defined taxonomy, information cannot be properly related to the species of significance.

Within the framework of tropical forage research, it is not unusual that new species are described within a genus in the course of assessing genetic diversity. This also reflects the use of live plants by the genetic resources specialist as opposed to the botanist, who largely studies dried specimens. Since the last review (Mannetje 1984), new species have been described from Brazil – *S. nunoi* (Brandão 1991) and *S. longicarpa* (Brandão & Costa 1992) – and more recently from Guerrero, Mexico – *S. salina* (Costa & van den Berg 2001) – all belonging to sect. *Stylosanthes*. From the so-called ‘Caatinga stylo’ (*S. seabrana*), cultivars ‘Primar’ and ‘Unica’ were registered in Australia in 1996 (Anon. 1996), even before this particular species of *Stylosanthes* sect. *Stylosanthes* had actually been validated by Maass and Mannetje (2002).

**Table 1.3** Comparison of recent findings based on molecular evidence to different taxonomic approaches towards the *S. guianensis* species complex (from Mannetje 1984). Taxa with common symbol in front are closely related; species names as published in the reference.

Ferreira & Costa (1979)	Mannetje (1984)	Kazan et al (1993a)	Vander Stappen et al (1998); Vander Stappen, de Laet et al (2002); Vander Stappen, Gama López et al (2002)
	<i>S. guianensis</i>	<i>S. guianensis</i> ssp. <i>guianensis</i>	<i>S. guianensis</i>
<i>S. guianensis</i> { <ul style="list-style-type: none"> <li>Var. <i>pauciflora</i></li> <li>Var. <i>vulgaris</i></li> <li>Var. <i>microcephala</i></li> <li>Var. <i>canescens</i></li> </ul>	n.d. <sup>b</sup>	n.d.	n.d.
		n.d.	n.d.
	Var.	n.d.	n.d.
		α Var. <i>canescens</i>	n.d.
<i>S. acuminata</i>	Var. <i>marginata</i>	α <i>S. acuminata</i>	n.d.
<i>S. gracilis</i>	Var. <i>gracilis</i>	<i>S. gracilis</i>	<i>S. gracilis</i>
<i>S. campestris</i>	Var. <i>intermedia</i>	–	γ <i>S. campestris</i>
<i>S. hippocampoides</i>		β <i>S. hippocampoides</i>	γ <i>S. hippocampoides</i>
<i>S. grandifolia</i>	Var. <i>robusta</i>	β <i>S. grandifolia</i>	λ <i>S. grandifolia</i>
<i>S. aurea</i>		–	λ <i>S. aurea</i>
<i>S. longiseta</i>	Var. <i>longiseta</i>	–	–
– <sup>a</sup>	Var. <i>dissitiflora</i>	–	ε <i>S. dissitiflora</i>
–	<i>S. biflora</i>	–	ε <i>S. biflora</i>
–	<i>S. montevidensis</i>	–	<i>S. montevidensis</i>
–	( <i>S. macrosoma</i> ) <sup>c</sup>	–	<i>S. macrosoma</i>

<sup>a</sup> not included/studied <sup>b</sup> not differentiated <sup>c</sup> doubtful species in brackets

**Table 1.4 Biochemical and molecular techniques applied in phylogenetic, taxonomic and diversity research of *Stylosanthes***

Technique	Abbreviation	Reference
Seed protein electrophoresis		Robinson & Megarrity 1975; Robinson et al 1976
Isozyme electrophoresis		Gillies & Abbott 1999; Maass et al, unpublished; Marulanda 1996; Robinson et al 1980; Stace 1982; Stace, unpublished; Stace & Cameron 1984; Vithanage & Chakraborty 1992
Randomly amplified polymorphic DNA	RAPDs	Gillies & Abbott 1998; Glover et al 1994; Kazan et al 1993a, 1993b; Liu 1997; Vieira et al 1997
Restriction fragment length polymorphism	RFLPs	Curtis et al 1995; Gillies & Abbott 1996; Liu & Musial 1995, 1996; Liu et al 1999
Amplified fragment length polymorphism	AFLPs	Sawkins 1999; Sawkins et al 2001; Vander Stappen et al 2000
Sequencing of the nrDNA internal transcribed spacer region	ITS-1	Sawkins 1999; Vander Stappen et al 1998; Vander Stappen, de Laet et al 2002
Sequence-tagged microsatellites	STS	Liu & Musial 1996; Liu et al 1996, 1999; Vander Stappen, Van Campenhout et al 1999
Sequencing chloroplast DNA	cpDNA	Liu & Musial 2001; Vander Stappen, Weltjens & Munaut et al 1999

Classical taxonomic treatments of *Stylosanthes* have been mainly based on some aspects of the floral and fruit morphology (Mannetje 1984). In 1838 Vogel established the main division of the genus into the sections *Stylosanthes* and *Stylosanthes* (formerly *Eustylosanthes*), based on the presence of a rudimentary secondary floral axis and two inner bracteoles in the former and no such axis and only one inner bracteole in the latter (Kirkbride & de Kirkbride 1985; Mannetje 1984).

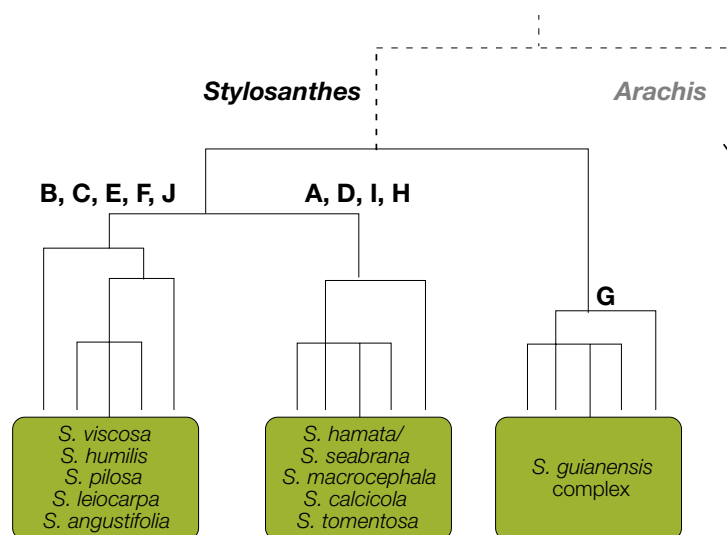
Over the past years, a wealth of taxonomic/phylogenetic studies and investigation of inter- and intraspecific genetic diversity in *Stylosanthes* have been performed by C.J. Liu and collaborators (various publications), Gillies (1994), Vander Stappen (1999) and Sawkins (1999) utilising techniques other than classical taxonomy (Table 1.4). Initially, these techniques relied on variation at the protein level. However, with the rapid advances in molecular genetics, a number of techniques were developed to take advantage of variation at the DNA sequence level. Much of the published work in this area has addressed questions related to taxonomy rather than focusing on detailed analyses of variation within species.

### Phylogeny

The genus *Stylosanthes* has a monophyletic origin (Gillies & Abbott 1994; Vander Stappen, de Laet et al 2002) and is closely related to *Arachis* (Lavin et al 2001), with the *S. guianensis* species complex as the most ancient group, clearly distinct from the rest of the genus (Figure 1.5).

Liu et al (1999) and Ma et al (submitted) identified ten basal genomes, named A to J (Figures 1.1, 1.5). Genome A1 appears to be the maternal donor to all tetraploid species with a known AABB genome (*S. scabra*, *S. sericeiceps*, *S. tuberculata*), while genome C is the maternal donor for all those with a known AACC genome (*S. hamata* (4n), *S. subsericea*, *S. sundaica*). The limited variation between the taxa within each group raises the question as to whether they should be treated as different species

(Liu & Musial 2001). On the other hand, as more than one species have the same ancestral genomes, it seems most likely that more than one hybridisation events has taken place (Vander Stappen, de Laet et al 2002). This independent evolution of similar tetraploid combinations has been suggested by Stace and Cameron (1984) because of the disjunctions of the natural distribution of several *Stylosanthes* species. Only a few species are widely distributed, while many occur in restricted or isolated areas which appear to be local refuges, creating barriers to gene flow and thus allowing the evolution of regionally adapted genotypes (Stace & Cameron 1984). A similar conclusion was reached by Liu (1997) using DNA markers to study intraspecific variation in *S. scabra*.



**Figure 1.5 Schematic phenogram of phylogenetic relationships and genomes (capital letters) of *Stylosanthes* species, derived from Liu et al (1999); Liu & Musial (2001); Vander Stappen, de Laet et al (2002); and Lavin et al (2001); only representative diploid species are given.**

## Putative ancestors of polyploid species

Most species are diploid ( $2n = 20$ ) but polyploid species ( $2n = 40, 60$ ) also exist in *Stylosanthes*. The latter are exclusively allopolyploid. Sect. *Stylosanthes* contains both diploid and polyploid species, while species in sect. *Stylosanthes* are exclusively diploid. Stace and Cameron (1984) postulated that a tetraploid ( $4n$ ) is a combination of a diploid ( $2n$ ) species from sect. *Stylosanthes* and a diploid ( $2n$ ) species from sect. *Stylosanthes*. This model has been fully confirmed by recent molecular work (Liu et al 1999; Vander Stappen, de Laet et al 2002) (Table 1.5), except in *S. capitata*, whose unique genome structure DDEE was donated by its putative ancestors *S. macrocephala* or *S. bracteata* and *S. pilosa*, which all belong to sect. *Stylosanthes* (Liu et al 1999). The ancestors of the hexaploid *S. erecta* are postulated to be *S. scabra* (studied as a representative of the AABB species) and *S. angustifolia* (C.J. Liu, pers. comm. 2004; Ma et al, submitted).

## New species and inconsistencies

The tetraploid nature of *S. hamata* cv. Verano was reported almost three decades ago (Mackay 1975, cited by Stace & Cameron 1987). It is now well established using molecular evidence that *S. hamata* s. str. ( $2n$ ) and *S. humilis* are the putative ancestors of the tetraploid taxon (Curtis et al 1995; Kazan et al 1993a; Liu et al 1999). This new, unnamed species widespread in the tropics through cultivation of its most popular representative, cv. Verano, requires urgent validation. Unfortunately, a wealth of literature referring to this taxon has accumulated since the release of cv. Verano in 1973. The frequent practice in the literature to name this taxon as *S. hamata* without indicating its tetraploid nature will lead to further confusion as this is the correct epithet for diploid plants. In the absence of a valid name for the tetraploid taxon, 'S. *hamata* s.l.'

(eg in Gillies & Abbott 1996) should presently only be used if both the diploid and tetraploid plants are included in this broad sense (*sensu lato*). Stace, in 1987, suggested this species be renamed *S. hemihamata*, which reflects the genetic and prior nomenclatural link with *S. hamata* s. str. (Stace, unpublished a). Though this name was never published according to the rules of the International Botanical Code, this *nomen nudum* will be used where appropriate in this chapter.

The use of the name *Stylosanthes* sp. aff. *hamata* for some tetraploid accessions, identifying other related taxa (eg Burt & Williams 1979a, 1979b; Mannetje 1984), is misleading in that it implies a close relationship of these materials with *S. hamata* or *S. hemihamata*. However, Liu et al (1999) and Liu and Musial (2001) identified the AABB genomic structure in the respective accessions and, hence, demonstrated these plants to be close to *S. scabra*.

It is imperative that accessions that form the basis of recent taxonomic, genetic and phylogenetic conclusions be morphologically studied in a comprehensive revision of this genus. An example is the listing of *S. mexicana* as a tetraploid species with an AACC genome by Liu and Musial (2001); diploid by Stace and Cameron (1984); and both diploid and tetraploid by Vander Stappen, Van Campenhout et al (1999). Similarly, Liu and Musial (2001), using accession CPI 76259, and Vander Stappen, Weltjens & Munaut et al (1999), using accession CIAT 1608, found *S. ingrata* to be tetraploid, a species listed under sect. *Stylosanthes* that exclusively contains diploid species (Mannetje 1984). In both cases clarification is needed about the identity of taxa studied by the different authors. Fortunately, these recent studies either clearly referred to herbarium specimens (eg Vander Stappen, de Laet et al 2002) or germplasm accessions mainly from CSIRO or CIAT.

**Table 1.5 Identification of putative progenitors for tetraploid *Stylosanthes* taxa by the use of molecular markers (derived from Vander Stappen, de Laet et al 2002; Vander Stappen, Gama López et al 2002; Liu et al 1999; Liu & Musial 2001).**

Tetraploid species	Genome a	Putative maternal parent	Putative paternal parent	Remarks
<i>S. fruticosa</i>	AB	<i>S. viscosa</i>	<i>S. sp. (A)</i>	
<i>S. scabra</i> s. lat.	AB	<i>S. seabrana</i>	<i>S. viscosa</i>	
<i>S. sericeiceps</i>	AB	<i>S. seabrana</i> / <i>S. hamata</i> s. str.	<i>S. viscosa</i>	
<i>S. hemihamata</i>	AC	<i>S. humilis</i>	<i>S. hamata</i> s. str.	
<i>S. mexicana</i> <sup>b</sup>	}	AC	<i>S. sp. (C)</i>	Liu et al
			<i>S. sp.</i>	Vander Stappen et al
<i>S. subsericea</i> <sup>b</sup>	}	AC	<i>S. sp. (C)</i>	Liu et al
			<i>S. sp.</i>	Vander Stappen et al
<i>S. sundaica</i>	AC	<i>S. humilis</i>	<i>S. hamata</i> s. str.	
<i>S. sympodialis</i>	AF	<i>S. leiocarpa</i>	<i>S. hamata</i> s. str.	
<i>S. capitata</i>	DE	<i>S. macrocephala</i> / <i>S. bracteata</i>	<i>S. pilosa</i>	
<i>S. sp. nov. (aff. calcicola)</i>	BI? <sup>c</sup>	<i>S. calcicola</i>	<i>S. viscosa</i>	

<sup>a</sup> according to Liu et al (1999)

<sup>b</sup> plant material requires morphological verification of species identity

<sup>c</sup> from Vander Stappen, Gama López et al (2002)

Revising the taxonomy of *Stylosanthes*, Marnette (1984) doubted the validity of some species, among them *S. pilosa*, which has been shown to have a distinct genome (E) (Liu et al 1999; Liu & Musial 2001; Vander Stappen, de Laet et al 2002; Vander Stappen, Weltjens & Munaut et al 1999). As the putative ancestor of *S. capitata*, this taxon merits species status. However, its relationships to *S. ruellioides* (Marnette 1984) and *S. bahiensis* (Costa & Ferreira 1984) need critical revision, and nomenclatural rules must be followed.

The recent discovery of a new Mexican allotetraploid species, initially named *S. aff. calcicola* (Vander Stappen, Gama López et al 2002) and whose one putative ancestor was identified as the diploid *S. calcicola*, points to a common pattern of diploid and related, morphologically similar, tetraploid species such as *S. macrocephala* (2n) and *S. capitata* (4n) or *S. hamata* (2n) and *S. hemihamata* (Verano-type, 4n). The improved understanding of polyploidy may help to better spot new species in geographic regions that have received less attention.

New insights into phylogeny and taxonomic status of taxa from molecular studies have revealed an unevenness of specific or subspecific treatment within different species complexes. For example, several species related to *S. scabra* have been recognised that may rather be reduced to intraspecific variants (Liu & Musial 2001; Marnette 1984), while recent publications (eg Vander Stappen, de Laet et al 2002) accepted a higher level of interspecific variation within the *S. guianensis* species complex, thereby partly opposing Marnette's view (1984).

### **The *S. guianensis* species complex**

Molecular studies have confirmed that the complex of species related to *S. guianensis* is phylogenetically the most ancient (Gillies & Abbott, 1994, 1996; Liu et al 1999; Vander Stappen, Van Campenhout et al 1999; Vander Stappen, Weltjens & Munaut et al 1999). Accessions from this species group usually cluster into a well-defined clade (Vander Stappen, Weltjens & Munaut et al 1999). Consequently, Liu et al (1999) proposed the same genome (G) for a number of species in that group (*S. campestris*, *S. grandifolia*, *S. guianensis*, *S. hippocampoides*, *S. montevidensis*). Accessions belonging to *S. gracilis* were genetically differentiated from *S. guianensis* by the magnitude of a species level in other clades (Vander Stappen, Weltjens & Munaut et al 1999), which was also observed by Vieira et al (1997). In contrast, *S. montevidensis* was more similar to *S. guianensis*, which may call into question its species status (Vander Stappen, Weltjens & Munaut et al 1999). From the degree of genetic differentiation between different taxa within the *S. guianensis* complex (also based on pollen stainability), Kazan et al (1993a) strongly supported the taxonomic treatment by Ferreira and Costa (1979). The findings by Vieira et al (1997) corroborate this conclusion.

A recent morphological study of newly collected *S. guianensis* from Mexico failed to corroborate any previous taxonomic treatment of *S. guianensis* in Mexico (Vander Stappen et al 1998). Consequently, *S. dissitiflora* should be recognised as a valid species (Gama López

et al 2001). The newly described species from Brazil, *S. nunoi* and *S. longicarpa*, seem to be related to the *S. guianensis* species complex as well, while *S. hispida* (syn. *S. cayennensis*) is not related to it (Vander Stappen, de Laet et al 2002). Table 1.3 compares recent findings to previous taxonomic treatments by Ferreira and Costa (1979) and Marnette (1984).

In conclusion, there is good evidence to retain several of the species identified by Ferreira and Costa (1979) as the genetic differentiation determined among them is of similar magnitude to that observed among other *Stylosanthes* species. L. 't Marnette (pers. comm. 2004) also agreed that most of the taxa treated as varieties before any molecular data was available are separate species. However, he argued that the correct nomenclatural rules according to the International Botanical Code need to be applied for naming them. On the other hand, the taxa of the *S. guianensis* species complex are all genetically closely related (genome G), and some gene flow and subsequent hybridisation can be assumed as long as they grow conspecifically. Thus, the ultimate taxonomic solution will depend on the overall 'splitting' or 'lumping' approach taken for intrageneric organisation.

### **The *S. scabra* species complex**

*Stylosanthes scabra* and related species have been considered difficult to delineate due to large morphological variability within *S. scabra* (Edge & Maass 1997). Costa and Ferreira (1984) recognised two distinct morphotypes, while Maass (1989) determined four morpho-agronomic (MA) types, one of which was later validated as the diploid species *S. seabrana* (Maass & Marnette 2002). These MA-types partly agreed with groupings based on molecular data (Liu 1997). Marnette (1984) placed *S. scabra* together with *S. tuberculata* and *S. nervosa* as taxonomically problematic species (Table 1.6), difficult to distinguish as they have virtually identical pods. It would be sensible to reduce these three taxa to *S. scabra*, while recognising the morphological similarity between *S. scabra* and *S. fruticosa*, and the difference between *S. sympodialis* and the other two species. The phylogenetic analysis by Gillies and Abbott (1996) supports the view that *S. sympodialis* is genetically associated with *S. scabra*, *S. hemihamata*, *S. leiocarpa* and others, but also sufficiently distinct to merit species status. Recognising the genome structure (AAFF) for *S. sympodialis*, with *S. hamata* and *S. leiocarpa* as its diploid ancestors, Liu et al (1999) further corroborated this concept.

Liu and Musial (2001) raised the question of the validity of a separate species status, at least for *S. scabra*, *S. sericeiceps* and *S. tuberculata*, as they all share the AABB genomic structure and may not be sufficiently divergent to be treated as different species, but rather as subspecies or varieties. They may have developed into morphologically distinguishable taxa because they have evolved in isolation, disconnected from the main savanna regions (Stace & Cameron 1984). In addition, the different genetic groups may have evolved from several tetraploid plants formed

locally by independent hybridisation events between the two diploid progenitor species *S. viscosa* and *S. seabrana* (Liu 1997; Liu & Musial 1997).

Another issue concerns the South American *S. scabra* and the African and Asian *S. fruticosa*. Glover et al (1994) demonstrated that there was little differentiation between *S. fruticosa* and *S. scabra*, but Gillies and Abbott (1994) found the two species sufficiently distinct to merit different species status. Interestingly, in the study by Glover et al (1994) two *S. scabra* accessions from Espirito Santo and Rio de Janeiro in Brazil, located on the Atlantic coast, appear closest to the African *S. fruticosa* accessions. However, Vander Stappen and Volckaert (1999) found the two species identical, except for one repeat length variation in the *trnLF* intergenic spacer, and the fact that the two species most likely have the same maternal progenitor. Recent results from C.J. Liu's team suggested that *S. scabra* and *S. fruticosa* were reciprocal products involving the same two donor species. In other words, they share the same nuclear genome (AABB) but have different cytoplasms (C.J. Liu, pers. comm. 2004) (Table 1.5).

Another relationship within this species complex has been documented with the hexaploid *S. erecta* from Africa. Liu et al (1999) postulated *S. scabra* (as a representative of the AABB species) to be one of the ancestors of this species. Vander Stappen van Campenhout et al (1999), however, showed *S. fruticosa* to be closely related to *S. erecta*, which seems reasonable as *S. fruticosa* is also an African species and overlaps with *S. erecta* in distribution.

In conclusion, we support the taxonomic view of Mannerje (1984), who suggested the use of a wider species concept for *S. scabra*, sensu lato, that includes *S. tuberculata* and *S. nervosa*. Whether other less-studied species, such as *S. sericeiceps*, should be integrated under this species concept remains to be investigated.

## Distribution of Species and Genetic Variation

### Ecogeographic distribution

Before the advent of computer-based technologies such as geographic information systems (GIS), plant collectors had to manually identify the ecological conditions where previous collections had been made, and then use this information to identify regions with a similar ecological profile to target new collecting localities. This approach was used to great effect by many *Stylosanthes* researchers to identify regions with a high potential for containing new useful genetic resources (Burt et al 1976, 1979; Burt & Reid 1976; Burt et al 1980). These ecogeographic surveys form a vital component in the development of strategies for effective conservation and use of plant genetic resources (Maxted & Guarino 1997; Maxted et al 1995). At its core, an ecogeographic survey is the gathering and synthesis of data relating to the ecology, geography and taxonomy of a target taxon, which can be obtained from passport data associated with herbarium specimens and germplasm accessions, literature and other sources.

Two recent ecogeographic surveys have been undertaken on *Stylosanthes*. Tse-ring (1996) surveyed herbarium specimens to identify and prioritise areas for further collection and conservation, emphasising nine species. Sawkins (1999) studied four species using GIS and showed that *S. guianensis* has the widest geographic distribution, as compared to *S. viscosa*, *S. humilis* and *S. capitata*. In GIS several levels of different information can be easily superimposed and mapped. *Stylosanthes guianensis* occurred in the widest range of vegetation, classified using FAO (1974) or the Holdridge system, from savanna to thorn scrub, tropical forest, dry forest and montane tropical forests adapted to cool temperatures. Overlaying maps of conservation reserves on maps of the species' geographical distribution by point collection provided useful datasets for the identification and design of potential in situ reserves.

**Table 1.6 Comparing recent findings based on molecular evidence to different taxonomic approaches towards *S. scabra* and related species.**

Williams et al (1984)	Mannerje (1984)	Liu et al (1999)	Genome
<i>S. fruticosa</i>	<i>S. fruticosa</i>	<i>S. fruticosa</i>	AB
<i>S. nervosa</i>	} <i>S. scabra</i> s. lat. }	} <i>S. scabra</i> s. lat. }	– <sup>a</sup>
<i>S. scabra</i>			AB
<i>S. tuberculata</i>			AB
<i>S. suffruticosa</i>			–
	<i>S. aff. hamata</i>		AB
<i>S. sericeiceps</i>	<i>S. sericeiceps</i>		AB
<i>S. sympodialis</i>	<i>S. sympodialis</i>	<i>S. sympodialis</i>	AF

<sup>a</sup> not studied



A number of GIS-based programs for the analysis of collection data are predictive in nature, for example Floramap (Jones 2002; Jones, Galwey et al 1997) or DIVA-GIS (Hijmans et al 2001). The information on climatic parameters at collection points can be used to identify potential areas of species distribution based on a similar climate. However, these maps are only as good as the input data and depend heavily on correct species identification. For instance, the map showing the potential distribution of *S. guianensis* ssp. *guianensis* (Figure 1.6) may have included other subspecies or varieties, despite best efforts.

The diversity of different *Stylosanthes* species has been characterised through GIS mapping approaches (Jones et al 1996; Jones, Sawkins et al 1997; Sawkins 1999; Sawkins et al 1999) using germplasm accessions that are considered to adequately represent the known distribution of major species (Schultze-Kraft & Keller-Grein 1994). Each map (Figures 1.6, 1.7) identifies a range of probabilities from no climate similarity to high climate similarity.

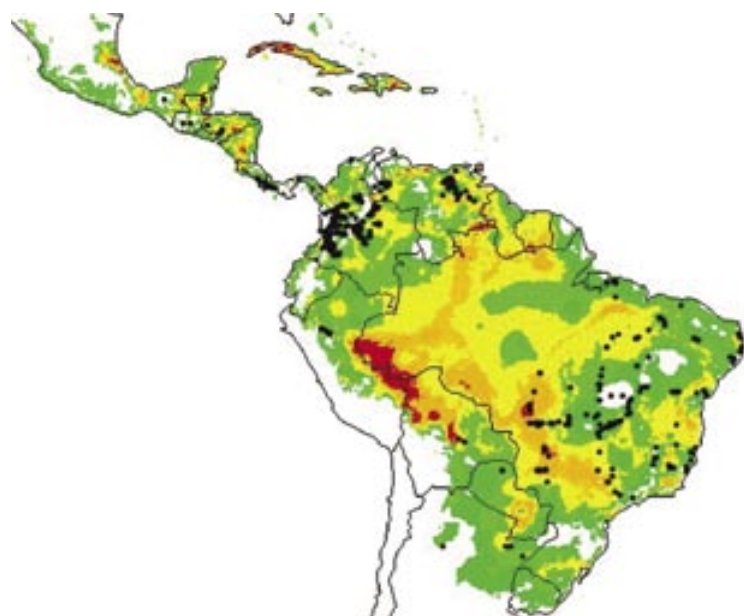
The plotted collection points fell well inside the known distribution areas of *S. humilis* (Williams et al 1984), while some high probability areas indicated that there may be important as yet uncollected areas of *S. humilis* in western Brazil, adjacent to Bolivia, and in Venezuela, Nicaragua and Mexico (Figure 1.7). Interestingly, recent studies by Gama López (2002) and collaborators have confirmed new *S. humilis* diversity from Mexico (Vander Stappen et al 2000), which they regard as a major centre of *Stylosanthes* diversity. In addition, other new species have recently been identified from Mexico (eg Costa & van den Berg 2001; Gama López et al 2001a; Vander Stappen, Gama López et al 2002),

and some previously described doubtful taxa (Mannetje 1984), such as *S. dissitiflora* and *S. subsericea*, appear to have been validated by this research (Gama López et al 2001; Vander Stappen et al unpublished).

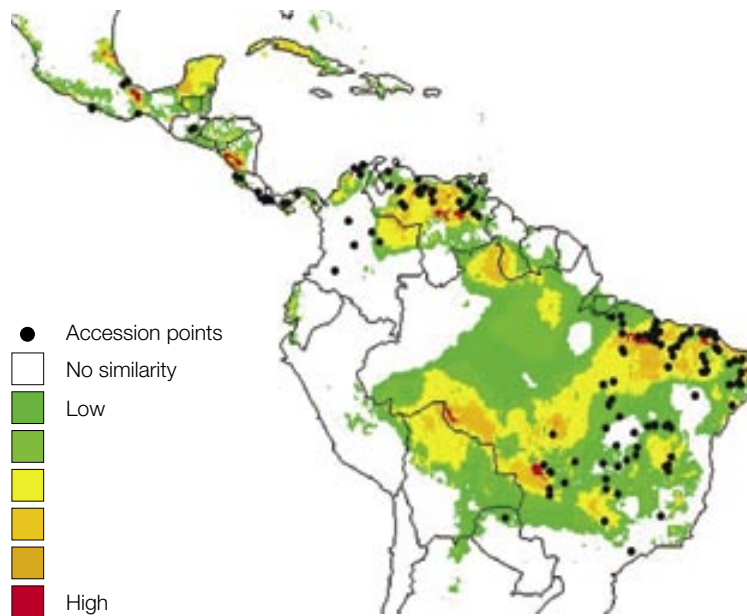
### Distribution of genetic diversity

In early attempts to understand the geographic patterns of diversity, the distribution of specific and intraspecific diversity of MA-types has been related to regions of origin (Edye et al 1974). For example, Maass (1989) showed that MA-types of *S. scabra* were unevenly distributed ecogeographically, which has been partly confirmed through the genetic study by Liu (1997).

Early information on genetic diversity in *S. humilis* based on isozyme data (Stace, unpublished b) revealed the occurrence of a common genotype throughout the species range, with distinct regional variants at one or more loci. Recent insights into *S. humilis* genetic diversity were provided by two studies using AFLPs that complement each other in the geographic range of the materials investigated. Sawkins et al (2001) examined the genetic variation in a collection of accessions of *S. humilis* and *S. viscosa* and found that *S. humilis* contained less variation than *S. viscosa* but that geographical patterns of variation were generally similar between the two species. Without exception, they also confirmed the regional types of *S. humilis* postulated by Stace (unpublished b). Vander Stappen et al (2000) investigated the diversity of Mexican accessions of *S. humilis* and contrasted this to the diversity observed in few accessions collected from South America. The authors concluded that Mexico may contain unique diversity that could be used for the



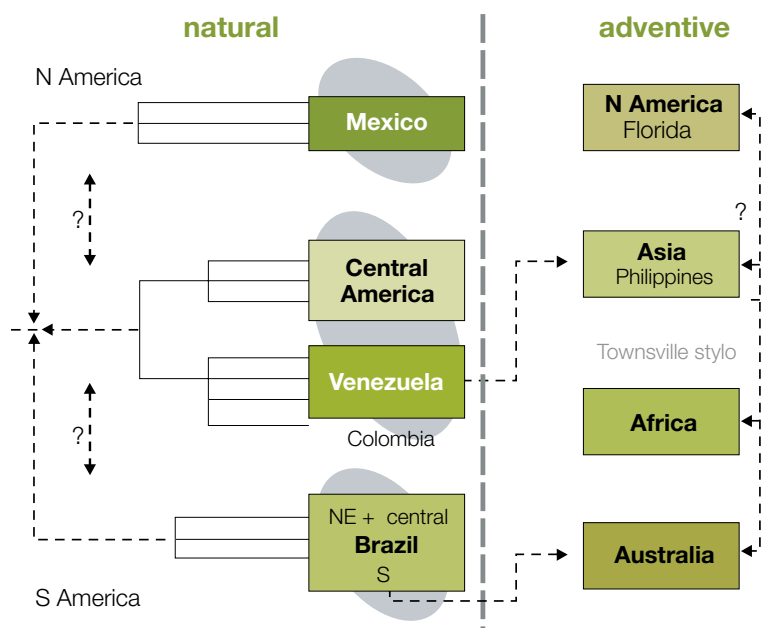
**Figure 1.6** *Stylosanthes guianensis* probability density distribution produced by applying FloraMap based on 449 germplasm accessions (Sawkins 1999).



**Figure 1.7** *Stylosanthes humilis* probability density distribution produced by applying FloraMap based on 210 germplasm accessions (Sawkins 1999).

conservation and utilisation of *S. humilis* germplasm. Both studies support the conclusion by Stace (unpublished b) that there are genetic races in *S. humilis*, a species that most likely has at least two independent gene pools, one in Mexico and the other in South America (Figure 1.8). Due to the different materials studied by Vander Stappen et al (2000) and Sawkins et al (2001), it could not be established with certainty whether the apparent probable third gene pool from Central America and Venezuela was closer to the Mexican or the Brazilian gene pool. ‘Townsville stylo’, widely distributed in Australia and transferred to African countries and Florida, was closely related to the southern Brazilian genotype, while Philippino *S. humilis* showed isozyme patterns of the Venezuelan genotype (Stace, unpublished b). Finally, none of these studies detected relations of commercial plants with the Mexican gene pool (Figure 1.8).

In addition, and as seen in other studies, novel variation was likely to be found in Mexico and Central America for this species. Areas in Baja California, at the Mexican Atlantic coast and in Guatemala were singled out by an ecological evaluation at the point of collection (Burt et al 1979). However, previous Australian collecting missions have not prioritised these areas because it was impractical to visit all regions that most likely would provide forage legumes for dry tropical and subtropical regions of Australia. In addition, *S. hemihamata* cv. Verano came from Venezuela, and the then promising *S. scabra* introductions from northeastern Brazil (Burt et al 1979), which led to emphasis on collections from these regions.



**Figure 1.8** Schematic phenogram of *Stylosanthes humilis* gene pools and their genetic relationships derived from studying isozymes (Stace, unpublished b) and AFLPs (Sawkins et al 2001; Vander Stappen et al 2000); thick broken arrows indicate known or probable (?) gene flow.

## Usage and significance

Detailed treatise on *Stylosanthes* utilisation is given in other chapters (Cameron & Chakraborty; Pengelly et al; Phaikaew et al; Hall & Glatzle, among others, this volume). Here we outline only some of the outstanding successes achieved by developing and using *Stylosanthes* species in agriculture, and discuss concerns regarding possible negative environmental impact of this forage legume.

## Grazed pastures and fodder banks

*Stylosanthes* species have improved extensive pastures, particularly in Australia, where the large extent of native pastures oversown with *Stylosanthes* has been amply documented (eg Miller et al 1997). It appears that the impact of *Stylosanthes* on commercial livestock production has not been great in the Americas, and the selection focus for a pasture legume has narrowed the choice of species and genotypes that might have otherwise been successful in different production systems (Miles & Lascano 1997). Roadside grazing has been promoted in Thailand (Guodao et al 1997), and seed of *Stylosanthes* species has been sown along roads, railways and stock routes, for example in Australia, Thailand and Ethiopia (A. Robertson, pers. comm. 1998).

Developing *Stylosanthes* species for ley farming or improved fallows has been the focus more recently in the Americas, for example with *S. guianensis* (CIAT 184) in the Amazonian region of Peru, but to date with little recognisable adoption outside projects run by research and/or development organisations (Miles & Lascano 1997). Development, since the 1990s, of *S. seabrana* (cvs. Primar and Unica) for heavy textured soils and its subsequent rapid adoption in Australian cropping systems shows there is a role for this species in this production niche (Pengelly & Conway 2000; Pengelly et al, this volume). Research and development in Africa strongly emphasised the use of *Stylosanthes* not only as a forage but also as a cover crop and green manure (de Leeuw et al 1994). The wide adoption of the fodder bank technology in West Africa now demonstrates a significant impact from such R & D (Elbasha et al 1999; Tarawali et al 2002). Although a wide range of species has been tested, *S. guianensis* cv. Cook and *S. hemihamata* cv. Verano have been the main species recommended for this technology and strongly promoted by ILCA (now ILRI), particularly in West and Central African countries (de Leeuw et al 1994; Tarawali et al 1998, 2002). By the mid 1990s about 19,000 ha were cultivated by about 27,000 adopters, especially in the subhumid zones of 15 West African countries surveyed (Elbasha et al 1999; Tarawali et al 2002). However, the most serious drawback to adoption is anthracnose susceptibility of the *S. guianensis* component, which has driven scientists to experiment with more diverse legume–legume mixtures and not only *Stylosanthes* species (Muhr et al 1999a, 1999b; Peters et al 1994).



## Cut-and-carry and leaf meal production

Phaikaew et al (this volume) summarise the use of *Stylosanthes* in Asia including as a cut-and-carry fodder. Commercial leaf meal production from *S. guianensis* in southern China is an important success story of the past two decades and leaf meal is an emerging technology in India (Guodao et al 1997; Guodao et al, this volume). Leaf meal is used as feed concentrate to supply protein and other nutrients for poultry and pigs (eg Chanphone Keoboulapheth & Choke Mikled 2003).

## Environmental impact

Positive effects of *Stylosanthes* species on the environment have been documented many times. This particularly refers to biological nitrogen fixation in both grazed pastures and mixed crop–forage systems. For example, Muhr et al (1999c) measured a significant nitrogen input of more than 90 kg/ha from *S. guianensis* (CIAT 184) in a maize crop in Nigeria. Such increase in residual soil fertility is a major input to an otherwise low-input mixed cropping system. Similarly, to introduce a ley phase with *S. seabrana* is seen as a solution to restore soil fertility after continuous cropping (Pengelly & Conway 2000). Obi (1999) showed that a cover crop of *S. gracilis* was the most efficient among the tropical grasses and legumes in improving soil conditions of a degraded Ultisol in Nigeria by regularly adding organic matter.

An environmentally negative impact, on the other hand, has been reported, where *Stylosanthes*-dominated stands can bring about soil acidification, particularly on light-textured soils of low fertility, by leaching of unused nitrates down the soil profile (Noble et al 1997, 2000). This development has not only been observed in extensive Australian pastures but also in *Stylosanthes* stands in China and Thailand (Noble et al 2000). In Australia the introduction of competitive, productive grasses together with the development of management options that would avoid legume dominance are regarded as measures against this long-term soil degradation.

Invasiveness has been recognised as an important feature of a successful pasture legume, which should be able to survive and spread unassisted (Miller et al 1997). This property, however, may also cause problems if the species is too aggressive and invades areas not intended for pastures. Conservationists have blamed *Stylosanthes* species for this, in particular *S. guianensis*. This and other species are listed among invasive naturalised plants in Queensland (Batianoff & Butler 2002), Taiwan (Shan-Hua Wu et al 2003), Pacific islands (PIER 2003) and Hawaii (Daehler & Denslow 2003).

## Prospects

A considerable change in research themes has taken place since the deliberate use of *Stylosanthes* in agriculture. Initially, research concentrated on understanding the basic biology, genetic resources and adaptation of *Stylosanthes*, and exploring its regional potential in a global venture (Burt et al 1983; Stace & Edye 1984). Ten years later (de Leeuw et al 1994) the main emphasis was placed on the utilisation of different species for grazing, fodder banks and as cover crops in mixed cropping systems. This reflects an emphasis on downstream research. Australian research on *Stylosanthes* was initially focused on *S. humilis*, with significant research teams operating in the Northern Territory, and North and Central Queensland. Exploitation of a wider range of *Stylosanthes* species was initiated in the 1960s and has become the exclusive focus for *Stylosanthes* research following anthracnose devastation of *S. humilis* from 1973 onwards. In the 1980s and 1990s there was a major integrated research effort in Australia, Brazil, Colombia, China and India on *Stylosanthes*, with projects on plant introduction and evaluation, anthracnose epidemiology, plant breeding and cultivar development, genetic engineering of disease resistance and molecular plant nutrition. This research has delivered a suite of moderate to highly resistant Australian cultivars, namely Seca, Verano, Amiga, Siran, Unica and Primar; and has contributed to new cultivars in Brazil and China. In Australia and South America basic research has maintained an importance, in particular with regard to understanding and overcoming the major constraints placed on the utilisation of *Stylosanthes* worldwide by the anthracnose disease (Winks & Chakraborty 1997; several papers, this volume).

The basic and applied research of these past decades has produced a substantial body of scientific knowledge of a genus that was practically unknown as a cultivated plant half a century ago. Concerning tropical forages, research and development in *Stylosanthes* has reached a remarkable pinnacle, catching up with the R & D level of temperate genera such as *Trifolium* and *Medicago*. However, despite various shortfalls in different *Stylosanthes* species, no one would argue that research in this genus should be given up and efforts redirected towards other legumes (Miles & Lascano 1997). In the early days of tropical forage research this might have happened, as there was a large selection of available but largely unexplored genera and species (Williams 1983). However, if it is accepted that *Stylosanthes* has reached such an advanced stage in research and development, then we should also accept that for tropical forage research, as in every widely used crop in agriculture, new challenges for crop improvement will continue to arise for breeders, pathologists and scientists of other disciplines.

In the future, further releases of bred cultivars from known *Stylosanthes* species can be expected after an initial stage of exploring natural diversity. This was suggested by Cameron (1983), who considered that the development of improved cultivars through plant introduction may be difficult once well-developed cultivars of the same species were already in use for well-defined agroecological zones. It is less likely that additional novel species will be discovered for agricultural use, as recently occurred with *S. seabrana*. Great advances have been made in the understanding of anthracnose resistance and the selection and development of new resistant cultivars (see other chapters, this volume), and perhaps less so for other breeding goals, such as forage and seed yield, or drought tolerance and persistence in mixed swards under grazing, as summarised by Miles and Grof (1997). With the stock of existing molecular information, the identification of specific genes or quantitative trait loci should advance rapidly. The first steps in this direction have already been taken (McIntyre et al 1995; Sawkins 1999; Stines et al 1996; Thumma et al 2001). Comparative mapping, for example between *Stylosanthes* and *Arachis*, should be possible and could speed up advances in many areas of interest (M. E. Ferguson, pers. comm. 2004). Methods such as marker-assisted breeding will most likely make breeding efforts quicker and more efficient, as long as the objectives are clearly defined.

With regard to utilisation and adoption, important progress has taken place especially in China and West Africa, but also in India, where the development of production systems, processing and cultivars are now pursued independently from plant materials released elsewhere (see other chapters, this volume). Nevertheless, the comprehensive germplasm assembled in large collections maintained ex situ at CIAT, ILRI, EMBRAPA and CSIRO still provides the basis for the research and development work in these regions. It is likely that new *Stylosanthes* species will be discovered by additional collection and/or more detailed characterisation of existing germplasm accessions (Vander Stappen & Volckaert 1999). As a consequence, the taxonomy may remain in flux for a considerable time to come, which is only to be expected for a genus still relatively new to agriculture.

## Acknowledgments

We are very grateful to C.J. Liu for his rapid and constructive reading through the article. Due acknowledgment is also given to L. 't Mannetje who, under much time constraint, commented on some critical issues and, certainly, significantly contributed to the paper. We also thank H.M. Stace for having unconditionally shared, many years ago, unpublished results whose validity has been proven by recent research.

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## Chapter 2

# Forage potential of *Stylosanthes* in different production systems

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### Summary

*Stylosanthes* is by far the most economically significant pasture and forage legume in the tropical regions of the globe. It is used in a variety of feeding systems ranging from freshly cut fodder to dry leaf meal supplements. Its contribution to rural communities comes from the added nutritional value to domesticated animals in both broadacre and cut-and-carry systems as well as in adding nitrogen to the soil. Several species are of commercial significance in Asia, Africa, South and Central America and Australia. This paper provides an overview of the different production systems in which *Stylosanthes* species are successfully used around the world.

### Introduction

Over the last 15 years commercial development of *Stylosanthes* (stylo) in the tropics has made substantial progress. This success has been built on multidisciplinary research efforts which have been extended to many different countries. In Australia, India, Thailand and other subhumid to semi-arid regions there has generally been steady increase in the seed production and use of *S. scabra* and *S. hamata*. Under extensive grazing systems in northern Australia the longevity of commercial pastures is now well established as the Seca (*S. scabra*) and Verano (*S. hamata*) cultivars have demonstrated durable field resistance to anthracnose caused by *Colletotrichum gloeosporioides*. Both Thailand and India have produced high tonnages of these cultivars (chiefly Verano; Guodao et al 1997). In Thailand, there have been extensive sowings of common grazing land through government sponsorship; similarly, the large increases in seed production in India have been underpinned by government programs for wasteland revegetation. More recently, private demand for seed has risen significantly in India with the use of *Stylosanthes* in a wide range of production systems, both at the village and individual farmer level.

In other tropical countries good progress has been made towards the development of commercial production systems but various limitations remain to be overcome. Widespread research success in contrasting production systems in semi-arid regions of Africa has generally failed to be translated into commercial practice. Socioeconomic problems and seed production have been suggested as serious obstacles to adoption by smallholder farmers (Dzowela 1993). The large research effort with *Stylosanthes* in the humid forests and savannas of South America has not met with significant commercial success to date. Comprehensive disease and pest screening programs have been successful in

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identifying resistant accessions. Cultivars have been released in several countries but lack of persistence under grazing and difficulties with seed production appear to have mitigated against commercial adoption. In China and South-East Asia, selections from the Australian and South American programs have enjoyed greater success (Guodao et al 1997). *Stylosanthes guianensis* cultivars, in particular CIAT 184, have been commercialised in southern China for intensive smallholder management systems, including use of leaf meal. Promising research results and early seed production success with CIAT 184 have also been obtained in several countries in South-East Asia.

Successful development of *Stylosanthes*-based production systems has been underpinned by significant progress in the development of adapted cultivars. Research on ecology, management and animal production has been important in the development of viable production systems. In this chapter we will briefly review recent advances in cultivar development, management and use. A notable feature of *Stylosanthes* usage patterns is the wide diversity of production systems that have been developed. The main focus of the chapter will be to review the contribution and potential of *Stylosanthes* species for this diversity of production systems.

## Cultivar Development and Use

Early success with the annual *S. humilis* ('Townsville stylo') and perennial *S. guianensis* (common and fine-stem stylos) laid the foundation for current development of *Stylosanthes* technology. In Queensland, Australia, the advent of cultivars of *S. hamata* ('Caribbean stylo'), *S. scabra* (shrubby stylo) and *S. seabrana* ('Caatinga stylo') has increased the estimated potential area for *Stylosanthes* from 9.7 million ha (based on *S. humilis*, Townsville stylo) to approximately 33 million ha (Edye 1997). Genetic resource collection and evaluation have continued as the major contributors of new cultivars. The difficulties posed by *C. gloeosporioides*, the highly variable anthracnose pathogen, have prompted intensive work on the epidemiology and genetics of this fungus. Significant progress has been made in implementing breeding solutions to develop cultivars and populations with broadly based resistance.

### ***S. scabra* and *S. hamata***

Research and commercial experience with Seca shrubby stylo has shown the broad climatic and edaphic adaptation of this cultivar. Use in northern Australia extends between 14 and 25 degrees latitude; useful yields in years of very low rainfall suggest the arid boundary for use may extend to the 500 mm isohyet (Hall et al 1995). Mixtures of Seca with Verano are favoured in the Australian tropics as insurance against anthracnose and for complementarity of growth patterns. Seca shows advantage when perennial plants are the main mechanism for persistence; where persistence is principally through seedling recruitment, higher yields result from the more vigorous Verano seedlings (Edye & Hall 1993). Screening of Caribbean stylo collections has so far failed to detect accessions, which are highly productive in subtropical regions.

Amiga Caribbean stylo has been released on the basis of improved perenniality under semi-arid conditions and better production than Verano at elevated tropical sites (Edye et al 1991). Siran shrubby stylo, a composite of three bred and selected lines incorporating four sources of resistance, has been developed to provide insurance against future race specialisation of *C. gloeosporioides* on Seca. New multivariate techniques of analysis have been developed to aid the identification of new fungal races and to discriminate between host accessions used for race surveys (Chakraborty et al 1996; Chakraborty et al 2002). Discovery of an asexual mechanism for chromosome transfer between different biotypes of *C. gloeosporioides* may explain how new races of the fungus can be generated (Manners & He 1997).

Molecular markers have been used effectively to characterise genetic diversity of the pathogen on different continents (Chakraborty et al 1997; Kelemu et al 1997). These techniques also identify prospective collection sites for plant germplasm on the basis of species relationships and geographical patterns of diversity (Liu 1997). Genetic maps currently under development, linking molecular markers to useful agronomic traits such as anthracnose resistance, would allow marker-assisted breeding to pyramid resistance genes for production of cultivars with broadly based resistance. Good progress has been reported on the use of recurrent selection to build anthracnose resistance by recombining genes from accessions with partial resistance (Cameron & Trevorrow 1998).

### ***S. seabrana***

Caatinga stylo *S. seabrana* is a new species from Bahia in Brazil (Maass & Mannetje 2002). Due to its similarity with *S. scabra*, Caatinga stylo had been previously referred to as *S. sp. aff. scabra* (Jansen & Edye 1996). This diploid species and *S. viscosa* appear to be the diploid progenitors of the allotetraploid *S. scabra* (Liu & Musial 1997). It is well adapted to heavy clay soils where its persistence and productivity are superior to shrubby stylo. Effective strains of root nodule bacteria have been collected from Brazil, as this species does not nodulate effectively with Australian native strains or existing commercial inoculant strains for *Stylosanthes* (Date et al 1996). Current research is aimed at new methods for establishment of the new strains in clay soils in semi-arid environments. Caatinga stylo is expected to make significant contributions to livestock production in other semi-arid regions of the tropics and subtropics. Two cultivars, Primar and Unica, have been recently commercialised in Australia. Primar is earlier flowering and may be better suited to regions where the length of growing season is reduced by low rainfall or low temperature. Both cultivars have recovered well from frosty winters where top growth was killed.

### ***S. guianensis*, *S. capitata* and *S. macrocephala***

The Oxley cultivar of *S. guianensis* has been an effective and persistent legume in the subtropics of Australia and Zimbabwe. Substantial commercial success has been achieved in tropical regions of China with

*S. guianensis* CIAT 184, designated as cultivar 'Reyen II - Zhuhuacao'. The superior anthracnose resistance of CIAT 184 has seen it replacing cv. Graham, which it outyields by 20% in Guangdong (Devendra & Sere 1992). Use in China can be extremely intensive. Production of leaf meal is an important innovation that may be extended to other countries in South-East Asia (Guodao et al 1997). The broad adaptation of CIAT 184 has seen it released in several South American countries; it has also been widely and successfully tested in South-East Asian countries where initial seed production has been developed. Nevertheless, the anthracnose susceptibility of CIAT 184 under severe disease pressure in the Colombian llanos gives cause for concern. In recent years four new cultivars, Reyen 2, 5, 7 and 10 with higher anthracnose resistance than CIAT 184, have been released (Huaxuan & Kexian 2002).

Plant collection in Brazil has given rise to *S. guianensis* cv. Mineirão, a vigorous, anthracnose-resistant line that was released in Brazil in 1993. Yields are very high but commercial success has been slow to develop because of low seed production from this very late flowering cultivar. Intensive selection for anthracnose resistance, flowering time and seed production in breeding populations of *S. guianensis* has developed several promising lines which are undergoing regional evaluation in various tropical countries (Miles & Grof 1997). Problems of persistence unrelated to anthracnose attack have been identified in *S. guianensis*. Selection for earliness of flowering, seed production and basal leaf area remaining after defoliation may improve persistence under grazing (Grof unpublished, in Miles & Grof 1997).

The early promise of *S. capitata* for acid soils of the humid tropics has not translated into commercial success despite good animal production from farm development work with cultivar Capica. Released by the Colombian national agricultural research institute in 1983, Capica is a mixture of five *S. capitata* accessions. Poor seedling recruitment in grazed pastures and severe anthracnose challenges in the Brazilian savannas appear responsible for the low level of commercial adoption. Recent research with mixtures of *S. capitata* and *S. macrocephala* derived from persistent plants selected from old grazed plots has seen the release in 2001 of a new multiline cultivar 'Estilosantes Campo Grande' (Grof et al 2001).

## Ecology and Management

### Soil type and fertility

The greatest success with *Stylosanthes* pastures has been on light textured, infertile soils where adaptation to low fertility confers a competitive advantage on *Stylosanthes*. Even with phosphorus levels as low as 3–4 ppm available P (bicarbonate method), *Stylosanthes* cultivars can establish with minimal soil disturbance. They have the ability to dominate the pasture gradually under a wide range of stocking rates (Jones et al 1997). The potential distribution range of *Stylosanthes* in northern Australia has been considerably expanded by successful research on adaptation to heavier textured soils. *Stylosanthes scabra* has

proven to be persistent on medium clays and clay duplexes (Partridge et al 1994). Even the heavy clays appear suitable for development with *S. seabrana*, which has been highly productive and a strong coloniser on these soils during initial evaluation trials in northern Australia (Edye et al, unpublished).

### Associated species

In semi-arid regions the strong drought resistance of *S. scabra* and *S. hamata* helps to give these species a competitive advantage over grasses and weeds. This advantage can be further strengthened by diet preference for grasses in the early part of the growing season (Jones et al 1997). In northern Australian woodlands the native grasses are usually intolerant of the increased grazing pressure that follows introduction of *Stylosanthes* cultivars. Improved grasses, such as *Urochloa mosambicensis*, *Cenchrus ciliaris* and *Bothriochloa pertusa*, are better able to persist with *Stylosanthes* under this increased grazing pressure. The improved grasses are more competitive where phosphorus availability is moderate to high; in the case of *C. ciliaris* the legume may disappear from the pasture at moderate to high levels of soil P.

In the humid tropics improved grasses such as *Brachiaria* spp., *Andropogon gayanus* and *Panicum maximum* are highly competitive. Although *S. capitata* can be competitive in the early years of grazing, perennial plants appear to be short lived, and seedlings have not been able to provide adequate recruitment for maintenance of the *S. capitata* component (Miles & Lascano 1997). Similar problems have been experienced with the early cultivar releases of *S. guianensis* and *S. macrocephala* in South America.

### Grazing and fertiliser management

Under stocking rates used by Australian graziers (2–>10 ha/beef animal), the *S. scabra* and *S. hamata* cultivars are highly persistent. In the early years of pasture development, light grazing pressure allows better seeding and a more rapid increase in density and yield of the legume component (Jones et al 1997). Spread from initially sparse stands is aided by passage of seed through the digestive tract of cattle (Gardener 1984). High levels of hardseededness provide the mechanism for maintenance of substantial seedbanks, a key factor in the excellent persistence of both *Stylosanthes* species (Jones & Mannelje 1997). At the very high grazing pressures encountered with uncontrolled grazing of village common land in other tropical countries, neither species is able to persist. Cultivars with a more prostrate morphology such as *S. humilis* cv. Khon Kaen are better adapted to this high grazing pressure.

At very low levels of soil P (<3–4 ppm, bicarbonate extraction), fertiliser application would normally be required to produce adequate growth of *Stylosanthes* (Jones et al 1997). Kerridge et al (1990) have used results from a series of long-term grazing experiments to compare the merits of fertiliser and mineral supplementation for animal production. Recommendations for the use of fertiliser and/or supplement for the

Australian semi-arid tropics have been derived for a wide range of soil P levels. The high costs of P fertiliser dictate that P supplementation gives better economic returns in northern Australia where soil available-P lies in the 4–8 ppm range. In practice, it is not economical to use fertiliser except in areas of high rainfall where high stocking rates can be maintained (Coates et al 1997).

Under Australian conditions, where *Stylosanthes* are usually established by broadcasting seed into native pasture communities, the legume will commonly dominate the pasture unless management options are implemented to favour the native grass component. Dominance of *Stylosanthes* in long-term permanent pastures is undesirable because of acidification and the poor protection of surface soil provided by *Stylosanthes* plants. Light grazing in the early years will help the legume to spread from increased seed production, and avoid damage to the grass from overgrazing. Resting the pasture from grazing in the early wet season may help to maintain the desired botanical composition by avoiding grazing damage to grass plants in early stages of regrowth. Judicious burning of pasture dominated by *Stylosanthes* can kill many legume plants and restore a higher proportion of native grasses (Miller et al 1997).

## Use in Production Systems

Through its contribution to the beef industry, by far the most significant use of *Stylosanthes* has been in broadacre pastures and rangelands of northern Australia (Coates et al 1997). Similarly, in tropical America the widest experience with *Stylosanthes* has been as a component of grass–legume pastures (Miles & Lascano 1997). Besides its major role as a pasture legume, *Stylosanthes* is currently used or has the potential for use in a number of other production systems including ley farming, intercropping, agroforestry, silvipasture, soil conservation, land reclamation, as a protein bank, and as processed or unprocessed cut-and-carry feed. In recent years work by international forage networks using participatory research methodology has vastly expanded options for forage production and use, including those for *Stylosanthes* (Stür et al 2002).

In many countries successful integration of *Stylosanthes* into various production systems has been demonstrated through ‘on-station’ and ‘on-farm’ research and development efforts of federal and state governments and agricultural and other universities. In many developing countries research sponsored by international aid agencies and,

**Table 2.1 Major commercial cultivars currently in use in different countries and total seed production in the last two decades as an indication of the commercial utilisation of *Stylosanthes*.**

Country	Total production (t)		Major cultivars currently in commercial use				
	Previous	Recent	<i>S. guianensis</i>	<i>S. scabra</i>	<i>S. seabrana</i>	<i>S. capitata</i>	<i>S. hamata</i>
Australia (Hopkinson & Walker 1984)	>150 ('76)	100 ('96)	Oxley	Seca, Siran	Primar, Unica		Verano, Amiga
Brazil (Miles & Lascano 1997; Grof et al 2001)	n.a.	20 ('96)	Mineirão			<i>S. capitata</i> - <i>S. macrocephala</i> <i>multiline</i> <i>Estilosantes</i> <i>Campo Grande</i>	
China (Guodao et al 1997; Huaxuan & Kexian 2002)	n.a.	43/year ('88-'96)	Reyen II — Zhuhuacao, Reyán 5, 7, 10	Seca			Verano
Colombia (Ferguson et al 1989)	14 ('86-'88)	Negligible ('97)				Capica	
India (Ramesh et al 1997)	61 ('88)	1200 (94-95)		Seca, Fitzroy			Verano
Indonesia (Guodao et al 1997)	n.a.	Small quantity	CIAT 184				
Malaysia (Guodao et al 1997)	None before '94	0.5 ('94)	CIAT 184				
Nigeria (Agishi 1994)	>20/year ('75-'80)	20/year ('81-'90)					Verano
Philippines (Guodao et al 1997)	5/year (late '70s)	>0.5/year (since '84)	Cook, CIAT 184				
Thailand (Humphrey 1984; Phaikaew 1997)	43 ('76-'79)	>150 ('95)	Graham, CIAT 184				Verano
Zimbabwe (Clatworthy 1984)	>9	n.a.	Oxley				

more recently, non-government organisations (NGOs) has provided a stimulus for development. However, the paucity of information makes it impossible to gauge the true extent of its commercial utilisation. Available seed production trends for the major commercial cultivars suggest an overall increase in commercial utilisation in various countries between the 1970s and 1990s (Table 2.1). More up to date seed production figures are given for some countries in this volume. In many countries, however, commercial seed production has ceased for some cultivars and there has been a decline in the quantity of seed produced as a direct result of devastation caused by the anthracnose diseases. Also, acceptance by producer communities in most countries has not met the expected rapid adoption rates often predicted for *Stylosanthes* technology. Socioeconomic issues and susceptibility to anthracnose disease and other biotic stresses are the greatest limitations to its use and persistence.

### Extensive grazing systems

In northern Australia long-term funding support for grazing experiments has provided good information on the management requirements and production outcomes for extensive grazing of *Stylosanthes* pastures. Recent reviews have summarised the long-term botanical and production trends and the plant physiological and dietary preference aspects which explain the performance data (Jones et al 1997; Coates et al 1997).

### Establishment and Management

Complete or strip cultivation was initially employed for establishment of *Stylosanthes* cultivars. With the continuing cost-price squeeze on Australian beef producers and expansion of commercial development to more remote and less productive environments, lower cost methods of establishment have been preferred. Establishment is now commonly undertaken by broadcasting seed into native vegetation without tree clearing or cultivation. Burning or heavy grazing may be employed to reduce competition from the existing herbaceous community. Where a two–four-year time interval for legume establishment is economically unacceptable, cultivation and clearing may still be undertaken to shorten the time interval to full establishment.

Desirable management practices have already been briefly discussed. Under the more extensive management systems there is usually little departure from continuous grazing. High stocking rates in the establishment phase will inhibit seed production and spread, and therefore delay full development of the pasture. In the longer term high stocking pressure, especially early in the growing season, can damage the native grass component, leading to *Stylosanthes* dominance of the pasture. Although *Stylosanthes* dominance appears to have little immediate effect on animal production level, long-term effects of major rises in soil acidity, a decline in biodiversity and increased risk of soil erosion are undesirable (Jones et al 1997).

### Nature and Level of Production

In the northern Australian beef industry, stocking rates of native pasture oversown with *Stylosanthes* are commonly two to three times higher than those in untreated native pasture. Nevertheless, increases in stocking rate through oversowing with *Stylosanthes* vary greatly with location, from up to ten-fold down to no increase. Annual live weight gains of 140–160 kg/head are achieved in most years on good pastures. Under experimental conditions this represents an advantage of 30–60 kg/head annual live weight gain over grazing native pastures (Coates et al 1997). Somewhat surprisingly, this difference in favour of the *Stylosanthes* pastures is maintained over highly variable seasons, with growing season length in the range 20–43 weeks. The pattern of live weight gain on these pastures is highly seasonal, with the biggest advantage to the *Stylosanthes* pastures being recorded during the late wet and dry seasons.

Diet selection studies provide an explanation for these strong seasonal patterns. Typically, cattle show a marked preference for young green grass early in the wet season; during the late wet and early dry seasons this pattern is reversed in favour of the *Stylosanthes*. The proportion of *Stylosanthes* in the diet can vary from 10 to 60% and is influenced by the species, with the perennial *S. scabra* showing a more even pattern of consumption than annual or short-lived perennial *Stylosanthes*.

In northern Australian production systems *Stylosanthes* pastures have traditionally been used for the last 6–12 months before turnoff of finished steers and bullocks (Miller et al 1997). With current demand by customers for higher quality product, producers are employing a range of options to deliver finished animals of younger age. Mineral, protein and energy supplements allied with greater use of *Stylosanthes* pasture can even out much of the seasonal effect and ensure better eating quality. Other parts of the overall production system also need improvement to lift reproductive performance of heifers and improve the nutrition of weaners, especially under early weaning systems designed to relieve the lactation stress on breeders. *Stylosanthes* pastures are being successfully used by leading producers in these roles (Barrett 1997).

### Intercropping and ley farming

Successful intercropping of *S. hamata* with cereals such as sorghum, maize and bajra (pearl millet) under rainfed conditions has been shown to increase grain yield of cereals by 6–26% at various sites in India (Ramesh et al 1997). In many countries crop residues from cereal and other crops form a major feed source for livestock. Compared with cereal monoculture, residue from legume–cereal intercropping improves animal nutrition due to high nitrogen content. In the subhumid zone of Nigeria, maize yield in an *S. hamata* intercrop can increase three-fold compared with that under monoculture without fertiliser (Mohamed-Saleem & Otsyina 1986). In large parts of Africa millet intercropping with cowpea is already one of the most common farming practices, and integrating *Stylosanthes* into this farming system seems a logical choice. In very dry environments, however, legume–cereal intercropping can reduce



grain yield in the cereal crop. In the drier Sahelian environment in Nigeria (annual rainfall 560 mm), integration of *Stylosanthes* into the low-input cereal-based farming system over a three-year period caused a grain yield depression in millet. Yields averaged 300 kg/ha compared with 700 kg/ha in pure stands (Kouame et al 1994). Management options can improve yield (Muhr et al 2002); measures such as delayed planting and reduced density of the legume have been successfully used in millet, maize and rice to introduce *Stylosanthes* to cereal production systems without yield penalties. Crop canopy management using early and late maturing varieties is another option (de Leeuw et al 1994). *Stylosanthes* is productive in paddy bunds under irrigated rice, as a companion crop in upland rice in South-East Asia (Humphreys 1984; Roder & Maniphone 1995) and in upland rice production systems in South America (Miles & Lascano 1997).

Use of legume leys helps maintain soil cover, provides additional forage and improves soil nutritional and physical properties. In Africa *Stylosanthes* is promoted to maintain soil fertility and supply feed for work oxen in the wetter and semi-arid regions of Mali and Ivory Coast (de Leeuw et al 1994). Farmers in this region grow cotton as a cash crop and sorghum and maize as food crops. Use of *Stylosanthes* as a ley legume can improve maize yield by up to the equivalent of adding 100–120 kg N/ha. Depending on soil and weather conditions, the benefit from improved soil nutrition and structure can last for more than one season (de Leeuw et al 1994). *Stylosanthes* and other legumes are best sown at the time of final weeding or shortly before harvest on more moist sites within annual cropping systems in India. In addition to providing additional quality fodder, this practice maintains soil cover and adds nitrogen for the next cropping cycle. The underlying aim must be to retain enough seed of *Stylosanthes* in the soil seedbank so that once it is established, it can be used for one or more years without reseeding. In Australia *S. seabrana* could prove useful as a ley legume under some farming systems. However, given the availability of other high-yielding legumes such as *Lablab* and *Macroptilium*, success of *S. seabrana* as a ley legume will be dependent on both improving its establishment and introduction of effective strains of root nodule bacteria (R.M. Jones, pers. comm.).

### Silvopasture and plantation systems

Use of forage grass is commonplace in forest and other plantations worldwide. The forage resource is used as a cut-and-carry fodder for stall-feeding or grazed in situ. Due to relatively low shade tolerance, use of *Stylosanthes* and other legumes has not become very widespread in tree plantations until recently. Increasing use of *Stylosanthes* and other forage species has resulted from an emphasis on agroforestry in countries like India and China. Factors which influence forage production, for example tree density, can now be better managed to obtain reasonable forage production for up to six to seven years after establishment. However, forage production still peaks in the second or third year. Benefits of *Stylosanthes* in silvopasture go beyond providing

forage for grazing animals. The legume can increase soil fertility, suppress weed (Ramaprasad & Prasad 1991) and accelerate fuel wood production and dry matter accumulation in trees including *Leucaena leucocephala*, *Eucalyptus* spp., *Acacia auriculiformis* and bamboo (Gill 1992).

*Stylosanthes* is being increasingly used in the restoration of degraded forests in India. In the state of Tamil Nadu, around 1500 ha of degraded forest was oversown with 3 t of *Seca* and *Verano* seeds in 1996 (Robertson 1996). State and federal government authorities which manage forest resources in India are by far the largest consumer of the 1000–1500 t of commercial *Stylosanthes* seed produced each year since the early 1990s (Ramesh et al 1997). Many forests are community resources, with open access for grazing animals from adjoining villages and large migratory herds of mainly sheep and goat. Although forests can be protected from grazing for two years to promote *Stylosanthes* establishment, subsequent uncontrolled grazing and poor management leave very little understorey of *Stylosanthes*. Some of the biggest successes in tree-based farming systems in India can be seen on privately owned land. Here producers have collaborated with NGOs such as BAIF Development and Research Foundation (Anon. 1997), Watershed Organization Trust (Lobo & Gudrun 1995) and others, and use a participatory approach based on sound economic management.

In southern China *S. guianensis* and *S. hamata* are generally grown on marginal soils, frequently in combination with perennial tree crops such as mango, orange, rubber, coconuts, coffee, watermelon and cassava and, more recently, in reforestation areas (Guodao et al 1997). In Guangdong and Hainan, smallholder farmers and state and commercial farms all follow this practice, although the extent is not



Use of *Stylosanthes guianensis* in a mango orchard in southern China (photo: Bai Changjun).

known. Both government and private sectors use *Stylosanthes* as forage and/or cover crops in plantations in India. *Stylosanthes* has the potential to be used in horticultural orchards in Tamil Nadu (Dasthagir & Suresh 1990); with *Anona nepalensis* and pineapple on the hilly terrain of Meghalaya (Chauhan et al 1993); and with grapes, mango and tamarind, among others (Ramesh et al 1997).

There are more than 20 million ha of coconut, rubber and oil palm plantations in South-East Asia (Horne et al 1997). High forage productivity is limited to the initial three–five-year period in rubber and oil palm plantations, but good long-term production systems are possible in coconut plantations. Research has demonstrated the usefulness of *Stylosanthes* in integrating sheep farming in rubber plantations in Malaysia (Ng et al 1997). *Stylosanthes scabra* (Seca) and *S. guianensis* (CIAT 184) were both productive and persistent in these plantations, but only the latter seeded. Both *S. guianensis* and *S. hamata* have been well adapted as effective pioneer plants in coconut plantations in the Philippines, Bali and eastern Malaysia (Humphreys 1984). Forage grasses and legumes, including *S. guianensis*, are grown in coconut plantations in parts of Indonesia in the so-called ‘cocobeef’ system (Soedarmadi 1995). *Stylosanthes guianensis* covers large areas of the 0.7 million ha of coconut plantations in Kerala. In coconut plantations productivity of *Stylosanthes* can progressively improve as the quantity of light penetrating the canopy increases from 10% in a 10-year-old crop to about 70% in a 70-year-old stand (Pillai et al 1987).

### Soil conservation and wasteland development

Globally, some 2 billion ha of once productive land is estimated to have suffered from degradation (Lal & Stewart 1990). Nowhere is this more relevant than in India, where over 50% (175 million ha) of the land is subject to land degradation of one form or another (FAO 1986). An estimated 31% of China’s 1973 million ha, large tracts of the African Sahel, southeastern Nigeria, East African highlands and South and Central America also suffer from serious degradation. Over-grazing and deforestation has resulted in by far the biggest area of eroded land. As a pioneering coloniser, *Stylosanthes* establishes well on poor soils under dryland conditions, even when the topsoil is severely eroded. Additionally, it can fix 20 to >200 kg N/ha/year depending on agronomic, edaphic and environmental conditions (Vallis & Gardener 1984), making it a useful green manure crop in the Amazonian basin of Peru (Miles & Lascano 1997). Its ability to improve bulk density, infiltration rate and water holding capacity makes it a useful species in the conservation, stabilisation and sustainable development of land and water resources (de Leeuw et al 1994), especially in fragile environments.

In Mindanao, Philippines, *S. guianensis* CIAT 184 is used to control soil erosion and suppress growth of *Imperata cylindrica* in plantation strips when establishing young forestry plantations (Horne et al 1997), and CIAT 184 is used to rehabilitate degraded *I. cylindrica* uplands in the east Kalimantan region of Indonesia (Guodao et al 1997).



Use of *Stylosanthes* in the restoration of heavily eroded soils (top) and as a pioneer colonising species for degraded soils (above) in China (photo: Bai Changjun).

In the last seven to eight years a number of agroforestry-based programs have been sponsored by Indian and overseas funding agencies and managed by federal and state governments or non-government agencies to address this important issue, with varying degree of success. Wasteland and watershed developments are two such schemes which have been the backbone of integrated holistic rural development. Vital to the physical development is the socioeconomic development of both individual producers and the community as a whole through appropriate training under a participatory research approach. Generally speaking, the success of these schemes has depended on the level of active community participation and ownership, and on the delivery of direct and indirect economic benefits to the community. Consequently, long-term success of development schemes on community/government owned land has generally been poorer compared with schemes undertaken on privately owned land. There is considerable information on agroforestry and wasteland development in India (Hegde 1987; Hazra et al 1996; Singh et al 1994) and comments in this paper will largely be restricted to the role of *Stylosanthes*.

Although there are many approaches and philosophies at work, most developments have components of soil conservation; water conservation, harvesting and utilisation; transfer of improved technology on crop and livestock production; regeneration and development of land through agroforestry; and processing and marketing of the produce. In more heavily degraded watersheds, following basic soil and land conservation practices through use of contour trenches and bunds, afforestation may start with small hills and ridges in the catchment area. Pioneering grass



and legume species, including *Stylosanthes*, are established to retain and enrich the soil and the area is closed off to prevent uncontrolled grazing. Trees are selected for fuel and timber (*Albizia lebbek*, *Azadirachta indica*, *Eucalyptus camaldulensis*, *E. tereticornis*, *Leucaena leucocephala*, *Prosopis cineraria* etc), food (*Anacardium occidentale*, *Annona squamosa*, *Emblica officinalis*, *Tamarindus indica*, *Ziziphus mauritiana*), medicine and other useful purposes (*Acacia catechu*, *Bambusa arundinacea*, *Dendrocalamus strictus*, *Madhuca indica*, *Sapindus trifoliatus*, *Tectona grandis*, *Terminalia arjuna* etc). In the agricultural lands more intensive cropping and horticultural plantations result from improved soil and land conservation. That part of the area unsuitable for cropping is used for forestry and forage production. *Leucaena*, *Bambusa* and native trees line the paddocks while *Desmanthus*, *Stylosanthes*, *Heteropogon* and other improved pasture species are used to stabilise boundaries between properties. Dry matter production of *Stylosanthes* can vary with the degree of soil degradation, associated trees, extent of shading and competition from other forage species, among others.

### Other utilisation schemes

**Processed feed: leaf meal and hay** Large numbers of commercial, state and smallholder farms grow *Stylosanthes* for leaf meal production in China. A small quantity (2–5%) is mixed in feed rations for poultry, pigs, cattle, ducks and fish. Freshly cut material is either dried outdoors on racks in the field or machine dried at low temperatures, and then ground and sieved to give particles in the range 1–1.5 mm with 9–12% water and 10–40% crude protein (Guodao et al 1997). CIAT 184 and Seca are the main cultivars, which yield about 5 t/ha/year and sell for US\$140–170/t. In Guangdong province more than 100,000 farm families have planted

over 0.1 million ha of *Stylosanthes*, primarily as fresh feed or for leaf meal production. Recent interests in Thailand to develop feed pellets using leaf meal could further expand this utilisation scheme. The key to leaf meal production may be access to markets (Horne et al 1997). As a hay crop, *S. humilis*, *S. guianensis* and *S. hamata* have been used in parts of West Africa and, to a limited extent, in Australia. Use of leaf meal as a component of broiler ration has been a significant recent development for India that can lead to commercial partnership between smallholder farmers and poultry integrators (Guodao et al, this volume).

**Fodder or protein bank** Fodder banks are stands of forage legumes established to provide high protein supplemental feed to livestock during the dry season. Under West African conditions a 1–4 ha area is fenced off and a thick stand of *Stylosanthes* is established in a seedbed fertilised with phosphorus. Controlled and strategic grazing is employed to manage grass competition, reduce risk of fire and provide forage to livestock without depleting soil seed reserves (Ajileye et al 1994). Fodder banks can be sustained for many years with a stocking rate of up to five animals per ha. From an initial 5 banks in 1981, numbers peaked at over 400 in 1990. However, the adoption rate has gradually declined due to several economic and ecological reasons and the technology may be more suitable for the subhumid zone than the semi-arid region. In South America fodder banks have served to improve persistence of palatable species such as *S. capitata*, but banks in Colombia have not improved animal performance during the dry season despite higher protein intake. In Brazil banks of *S. guianensis* have improved daily weight gain in heifers; better performance was attributed to green leaf retention and tolerance to drought and acid soils. Lower palatability of *S. guianensis* compared with *S. capitata* and more intensive management of the banks in Brazil are suggested as reasons for this difference (Miles et al 1994).

**Roadside sowing** In Thailand, India, Nigeria and Ethiopia *Stylosanthes* has been introduced through roadside sowings. This cost-effective strategy has caused rapid spread and increased awareness among farmers due to greater visibility. During 1995–96, the Tamil Nadu Department of Animal Husbandry sowed over 25,000 km using an estimated seeding rate of 1 kg/km (Robertson 1996). Some of the more spectacular successes with roadside plantings of *S. scabra* in India with and *S. guianensis* in China are shown as examples.

**Seed production** Commercial seed production of *Stylosanthes* occurs on a small to medium scale in Australia, India, Thailand, China and Nigeria, among others. Smallholder and commercial farmers, privately owned seed companies and government departments produce and market *Stylosanthes* seed. In Australia the bulk of the commercial seed production is handled by a very small number of privately owned seed companies. In contrast, over 90% of Verano seeds in Thailand are produced by village farmers on contract to the Thai Department of Livestock Development (Phaikaew 1997), involving more than 1100 smallholder farmers. In India a network of 25 villages in the Anantpur district of Andhra Pradesh (Ramesh et al 1997) and government



Lush growth of *Stylosanthes guianensis* CIAT 184 is ideal for hay making. In China, bundles are often made in the field before transportation. (Photo: Bai Changjun).



Spectacular planting of *Stylosanthes scabra* on the median strip dividing a road in southern India (left, photo: C.R. Ramesh) and roadside planting of *S. guianensis* CIAT 184 in Hainan, southern China (right, photo: Liu Guodao).

organisations such as the Indian Grassland and Forage Research Institute produce most *Stylosanthes* seed. In more recent years Indian producers have formed growers' cooperatives to manage production and marketing of *Stylosanthes* seeds. The technology for seed production and processing is mechanised in Australia, and header or vacuum harvesting is mainly used; average seed yield for Seca is 550 kg/ha and that of Verano is 800 kg/ha (Hopkinson & Walker 1984). In contrast, production, harvesting and processing are largely manual in China, India and Thailand, and seed yields are higher. For example, in Thailand and India yields greater than 900 kg/ha are common for Verano. With good seed prices, seed production has become profitable and growers in these countries often choose *Stylosanthes* in preference to other more drought-prone crops.

### Limitations to Commercial Utilisation

Anthrachnose disease is by far the biggest limitation to the commercial utilisation of *Stylosanthes*. The disease affects the establishment, growth, seed production and persistence of most, if not all, species. It has been the single most important factor in the demise of a large number of cultivars around the world. Of the 15 cultivars released in Australia since 1969, commercial seed production has ceased for all but 7 cultivars due to overcoming of resistance as a result of adaptation by the anthrachnose pathogen (Chakraborty 1997; Chakraborty, this volume). The situation is very similar in South America where as many as eight cultivars from three species have been released since 1977 (Miles & Lascano 1997). Susceptibility to anthrachnose in all except Mineirão, among other factors, has meant that commercial seed production has never been seriously attempted. In Africa and Asia the pattern of changes in cultivar use

has closely followed those in Australia and South America because cultivars used have largely been those developed in these countries, with the exception of Khon Kaen stylo (*S. humilis*), released by Khon Kaen University in Thailand. The wide range of genetic and pathogenic diversity in *C. gloeosporioides* at its centre of origin in South America (Kelemu et al 1997) suggests that use of genetic diversity in the plant, with genotype mixtures or pyramids to contain more than one source of anthrachnose resistance genes, may provide protection. For countries such as Australia, where genetic and pathogenic diversity is more limited, risk of serious damage comes from adaptation of the endemic pathogen population and from incursions of exotic, more virulent and complex races from overseas. Other pest and disease problems include two stem borers (*Caloptilia* sp., *Platyomopsis pedicornis*) which have caused serious damage to stands in Brazil and Colombia; and legume little leaf (phytoplasma) and *Botrytis* head rot, which can significantly affect seed crops (Kelemu et al, this volume).

Poor seed yield, often due to late flowering, has affected the use of some cultivars. In particular, *S. guianensis* cultivars Bandeirante and Mineirão have been poor seed producers. In some countries lack of a well-established protocol for cultivar release, promotion and commercialisation and an absence of a national seed industry has inhibited transfer of an effective *Stylosanthes* technology package to the end-users. Poor establishment has been a common problem with cultivars of most species. Long-term persistence of the tropical *S. guianensis* cultivars is generally poor both under grazing and cut-and-carry systems. Recent research in Australia has shown that, as with temperate legumes, soil pH can decline over time when a high content of *Stylosanthes* is maintained (Jones et al 1997). The potential problem of soil acidification will be more

common where the crop has a high nitrogen fixing ability, is irrigated such as in seed crops, and where plant material is removed either as hay or through vacuum harvesting. Although it may take 25–50 years for pH to increase by one unit, soil acidification can lead to fertility decline. The extent of acidification will depend on the buffering capacity of the soil, soil type and pasture composition. Use of vigorous, deep-rooting pasture grasses will prevent legume domination, stop nitrate leaching down the soil profile and help to slow acidification.

There are specific limitations to *Stylosanthes* use under certain farming systems. Tolerance to shade is a limitation in agroforestry and silvipasture, although *Stylosanthes* can be used as a pioneer coloniser and in subsequent years as gaps appear from tree harvesting. Socioeconomic factors strongly influence the use of forages in a mixed-farming enterprise. Williams et al (1992) illustrate this for a millet–*Stylosanthes* production scheme in West Africa. In poor rainfall years, as prices of millet go up and at the same time livestock prices fall, emphasis will be on producing grain; in good rainfall years millet prices drop. Producers may feel secure due to grain self-sufficiency, and may sell surplus millet to buy livestock and grow *Stylosanthes* to feed livestock. Thus forage production would only occur in good years when producers can afford to grow feed together with grain. There is also competition for time and labour from more important food and cash crops and lack of control over nomadic livestock grazing.

## Future Prospects

*Stylosanthes* is by far the most successful tropical forage legume worldwide. Many of its significant limitations (eg susceptibility to anthracnose, adaptation to a range of soil and environmental conditions) have been adequately addressed through research and development in several countries. Multidisciplinary teams with expertise in selection and breeding, agronomy, pathology, nutrition, ecology, animal production and economics have helped lay a foundation for the current *Stylosanthes* technology. Formal and informal arrangements for cooperation between researchers in the various countries have been important (Cameron & Lenné 1994). Free and open exchange of germplasm between countries has been a major strength of this collaboration. Collections held at CIAT, CSIRO and ILCA have been widely evaluated in Africa, Asia, Australia and South America. In the past, collaboration in epidemiology, genetics and management of anthracnose have mainly involved researchers from Australia, Brazil and Colombia. With expansion in the number of areas under various *Stylosanthes* production systems, this collaboration is now extending to many Asian countries, notably China and India.

Diseases and pests remain as threats to the persistence and productivity of *Stylosanthes*, particularly in more humid regions. Expansion of the genetic resource base is of critical importance to meet this challenge. With the liberalisation of trade, international cooperation will be more important in the future in protecting primary industries from incursions of exotic pests and diseases. The new molecular tools for plant

improvement have provided new insights into the population structures of both plant and pathogen populations. Where breeding is warranted, molecular markers can allow breeders to manipulate genes for complex characters through indirect selection for linked markers.

As a versatile legume, *Stylosanthes* is already making significant contributions to agricultural and environmental production systems. The emergence of a commercially viable seed industry in several countries is an indication of the maturity of *Stylosanthes*-based technology. Nevertheless, the major clients of *Stylosanthes* seed in countries other than Australia are still government departments, the private sector using less than 10% of seeds. Further research and transfer of technology must be aimed at private sector utilisation schemes; there is need to improve the contribution of *Stylosanthes* to smallholder production systems. The holistic mixed farming approach being developed in India has been extremely effective in raising the standard of living among holders of small and marginal lands. Economic benefits are based on improvements in soil and water resources, which have directly contributed to the restoration of severely degraded lands. This holistic approach with its strong emphasis on participatory research offers a new paradigm for successful adoption of *Stylosanthes* in diverse production systems.

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## Chapter 3

# *Stylosanthes* as a forage legume at its centre of diversity

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### Summary

This chapter reviews the literature on *Stylosanthes* research and development from Latin America and USA published during the last 12 years or so. In reviewing past and current research, it offers a critical appraisal of significant agronomic, economic and social drivers that may have been responsible for the poor adoption of *Stylosanthes* cultivars in Latin America. It identifies critical gaps in knowledge that can lead to the development and utilisation of new cultivars based on sound scientific principles. A collation of literature and experiences from past releases show that high seed yields, anthracnose resistance, persistence under grazing and alternative usage of a pasture species are important traits of successful cultivars. Although these traits have direct effects on seed cost, demand and availability in the market, they have not been an explicit objective of many previous *Stylosanthes* improvement programs. The paper looks beyond the unsuccessful past experiences with anthracnose-susceptible Australian and other early cultivars; it focuses on the new opportunities offered by the recently released *S. capitata*-*S. macrocephala* cv. Campo Grande and the renewed interest in *S. guianensis* var. *vulgaris* cv. Mineirão.

### Introduction

Historically, in Latin America, the main attempts to grow *Stylosanthes*-based pastures have happened in the well-drained savannas. This ecosystem covers an area of about 226 million ha, encompassing the Cerrados of Brazil (204 million ha) and the Llanos region of Colombia and Venezuela (22 million ha). These savanna regions present characteristics such as a dry season of variable length and highly weathered low fertility soils, which have significant impacts on livestock productions systems and pasture development. Climate and soils of this savanna-like region have been described by Cochrane et al (1985) and Adamoli et al (1985).

Low forage availability and quality of native or cultivated grass pastures during the dry season are the main constraints on livestock production in the savanna region. The use of tropical forage legumes to enrich native pastures or to establish grass-legume pastures was recognised as an economical and feasible alternative solution for this shortcoming. Among the tropical forage legume genera, *Stylosanthes* is considered the most suitable for savanna conditions. The ability of native *Stylosanthes* species to grow and produce good quality forage in low fertility soils and the successful history of species of this genus in Australia under similar climatic conditions corroborate the choice.

There are records of *Stylosanthes* as a key legume for pasture improvement in Brazil since the middle of the last century (Otero 1952). The establishment of a huge pasture development program, the beginning of CIAT activities and the strengthening of some National Research Institutions all happened during the end of the 1960s and beginning of the 1970s, and brought a further stimulus for research in the genus in Latin America.

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However, as pointed out by Miles and Lascano (1997), ‘the impact of *Stylosanthes* spp. on tropical American livestock production is not proportional to the research literature generated over the past 30 years or so’. Compared to the successful role of *Stylosanthes* cultivars in Australia or China, it is very pertinent to question why *Stylosanthes* cultivars did not succeed in Latin America. Are biotic factors, which impose a much stronger pressure on an endemic genus, such an overwhelming constraint that the search for new cultivars becomes a fruitless endeavour? How did inadequate utilisation strategies or cultural and economic barriers contribute to the few effective attempts to introduce *Stylosanthes* spp. into the existing production systems?

Understanding the reasons for the lack of adoption is a key issue in assessing the feasibility of using *Stylosanthes* spp. as a major species for pasture development in Latin America.

## A Review of *Stylosanthes* spp. Research and Development

The bulk of literature on research and development of *Stylosanthes* species (stylo) in Latin America and the USA, up to the middle of the 1990s, were fully reviewed by Thomas (1984), Kretschmer and Brozman (1984), Thomas and Grof (1986), Miles et al (1994), Miles and Lascano (1997) and Miles and Grof (1997).

In the CAB abstracts there are 998 references on *Stylosanthes* between January 1990 and September 2003, from which 305 are from Latin America and the USA. These articles, classified according to subjects

and topics, are presented in Table 3.1. Almost 50% of these are from Brazil (144), with the remaining from Colombia (48), Cuba (29), USA (15), Peru (11) and Venezuela (11). A small group of 17 articles involves authors from different countries and, from this group, 10 are related to studies on anthracnose. Brazil is the country with the largest area of tropical savannas in the world, it is the centre of origin for a number of *Stylosanthes* species (Costa & Ferreira 1984) and it has a strong history of agricultural research; all these explain the predominance of papers on *Stylosanthes* from Brazil.

Among the articles from South America, 52% (160 references) are exclusively about *S. guianensis* and 9% (27 references) refer to *S. capitata*. This proportion reflects the importance of *S. guianensis* to tropical America due to its adaptation and superior agronomic characteristics (Andrade & Karia 2000; Karia & Andrade 1996). Thomas (1984) and Thomas and Grof (1986) indicated that *S. scabra* was a species deserving more research because it is well adapted to well-drained savannas of South America, a region with environmental conditions similar to those in Australia where *S. scabra* is widely used. This species, however, suffers a virus infection in research station plots and shows typical little leaf symptoms.

Among the 24 articles on genetic resources (Table 3.1), 10 deal with morphological characterisation, 10 relate to geographical distribution and collection trips and a further 4 deal with taxonomy, with 3 of these describing new species: *S. seabrana* Maass & ‘t Mannetje, *S. salina* Sousa Costa & Van den Berg and *S. longicarpa* Brandão & Sousa Costa.

**Table 3.1 Main subject and topics covered in the literature referring to *Stylosanthes* spp. from Latin America and USA between 1990 and 2003.**

Subject	Number of articles	Specific topics covered
Genetic resources	24	Collecting/introduction; taxonomic studies; morphological, ecological and molecular characterisation
Genetics and breeding	27	Cytogenetics; reproductive biology; ecological genetics; evolution; breeding and biotechnology
Agronomy	61	Establishing methods; forage production; regional trials
Grazing	37	Utilisation; pasture recuperation; pasture management; persistence
Abiotic stress	37	Water stress; inundation; salinity; mineral nutrition
Biotic stress	33	Disease; pests; competition
Seeds	21	Storage; germination; production technology
Alternative uses	19	Green manure; cover crops; biological control
Microbiology and nutrient cycling	31	Biological fixation; micorrhiza association
Forage nutritive value and quality	10	Nutritive value; chemical composition; animal nutrition
Review	5	Review papers



## Germplasm collection, diversity and evaluation

It is likely that one of the first research activities associated with *Stylosanthes* spp. in Brazil was germplasm collection, in particular the collection and evaluation of *S. guianensis* (Otero 1952). Other more recent efforts in Latin America were summarised by Miles and Lascano (1997). Seeds from these collections are in long-term storage facilities at CIAT and EMBRAPA. Cataloguing these collections and developing databases are essential to facilitate the use of this germplasm by national research institutions. There is a lack of stable and reliable morphological descriptors for *Stylosanthes* species and the use of molecular markers has significantly improved the description and identification of taxa (Maass & Sawkins, this volume). This information, along with morpho-agronomical data and ecological characteristics of the collection site, are invaluable contributions to the organisation of genetic resources, germplasm evaluation and plant breeding (Karia et al 2001). Instead of actively maintaining an entire collection, the establishment of a core collection facilitates the use and conservation of large germplasm collections (Brown & Spillane 1999). A core collection is a subset containing 10–15% of the accessions which represents the variability of the main collection (Brown 1989a, 1989b).

Following a worldwide trend, molecular studies have become an important aspect of *Stylosanthes* research in the last ten years. From 27 articles on genetics, 14 deal with biotechnology using molecular tools (Table 3.1), including tissue culture (6 on somaclonal variation and tissue culture media), plant transformation (5 on electroporation and *Agrobacterium*), molecular markers (2 articles) and the analysis of genetic distance between species (1 article). In Brazil a direct transfer of a methionine-rich 2S albumin gene from Brazil nuts (*Bertholletia excelsa*) was successfully attempted using microparticle bombardment and protoplast electroporation of *S. guianensis* cv. Mineirão by the Genetics and Plant Breeding Department of ESALQ/USP (Piracicaba-Brazil) in 1999 (Quecini 1999).

Of the 5 articles on ecological genetics, 3 were about genetic variability of adaptive characteristics of *S. scabra* and *S. humilis* under distinct environments in northeastern Brazil. There were 3 papers on the use of gamma radiation to induce mutation.

Germplasm collection, evaluation and selection were the main activities in *Stylosanthes* research in Latin America and USA, mostly following the International Tropical Pastures Evaluation Network (RIEPT) model, which uses a sequence of regional trials in four phases. Using a participative approach, researchers from all partner national research institutes have defined the methodology outlined in 'Regional Trial Manuals' (Lascano & Pizarro 1986; Paladines & Lascano 1983; Toledo 1982). Initial evaluation of large *Stylosanthes* germplasm collections, Type A trials, were carried out at sites representing the four major target ecosystems: Carimagua in the Colombian Llanos for the isohyperthermic savannas; Planaltina in the Brazilian Cerrados for isothermic savannas; Pucallpa in Peru and San

Isidro and Guapiles, both in Costa Rica, for the humid tropics; and Atenas in Costa Rica representing the subhumid tropics. Selected accessions from Type A trials were evaluated in small plot cutting trials (Type B trials) in each of the major target ecosystems. Plant response to grazing was evaluated in Type C trials and animal response in Type D trials. The RIEPT model has been very successful in selecting pasture cultivars with broad adaptation to the target environment. However, it does not consider alternative uses for a pasture species such as cover crop, fallow or companion crop, green manure etc. There is also the concern that Type A trials do not fully exploit the genetic diversity in the collection.

Despite this, it is not practical to test large numbers of accessions at various ecosystems for all potential utilisation schemes, and dealing initially with a core collection (Karia et al 2001) may be adequate. Exposing this core collection to a variety of utilisation options and environmental conditions can help identify accessions or 'morphological types' for specific usage. The entire collection can then be scanned to expand the number of accessions with the desired trait for further evaluation and selection.

## Breeding programs

There were only 2 papers specifically on breeding programs: Miles and Grof (1997) reviewed the *Stylosanthes* breeding programs in Latin America, and Hutton and Grof (1993) provided a more detailed description of one of these programs. Since 1978 there have been four *Stylosanthes* breeding programs in South America (Miles & Grof 1997). Of these, the *S. capitata* breeding program developed by CIAT (Carimagua, Colombia) and Empresa Brasileira de Pesquisa Agropecuária (EMBRAPA) Cerrados (Planaltina) have not succeeded in releasing a cultivar. The selected *S. capitata* hybrid with high dry matter and seed yields (Hutton 1987) was heavily attacked by anthracnose at the seed multiplication phase, and the program was aborted. Further details of this program appear in Hutton and Grof (1993). The second breeding program, on *S. guianensis* at CIAT in Carimagua, has developed anthracnose-resistant materials with good seed yields. Although two bulk populations with high seed yield were selected for a second phase, the program has been discontinued. The third breeding program, developed by CIAT and EMBRAPA Beef Cattle (Campo Grande, Brazil) using *S. guianensis* hybrids from the CIAT-Carimagua breeding program, has succeeded in selecting 20 lines with improved seed production, ten times the seed yield of Mineirão. These lines are undergoing further evaluation with good prospects for the release of a new cultivar (C.D. Fernandes, pers. comm.). The fourth program, developed by EMBRAPA Beef Cattle, has resulted in the release of stylo cv. Campo Grande (*S. capitata* + *S. macrocephala*).

In general, *Stylosanthes* breeding programs in Latin America have not been successful, although the existing germplasm collections present enough variability and *Stylosanthes* reproductive biology and genetics are well known (Miles 2001; Miles & Grof 1997). This is partly due to a lack

of clear focus in some of the programs, where the breeding objectives may not have considered farmer and seed producer perspectives of what characteristics are necessary and how any new cultivar will be used in existing production systems. The choice of selection parameters has been another reason. For example, selection for 'grazing persistence' can be made under long-term grazing trials, but this is costly, slow and inefficient. Easily measurable physiological, morphological or agronomical characteristics with strong relationships to plant persistence can provide an efficient alternative but demand a deeper understanding of pasture dynamics. Anthracnose resistance is a case in point, where advanced knowledge of *Colletotrichum gloeosporioides* diversity has provided strong support to the search for resistance.

### **Agronomy**

Among the 36 references on agronomy, the majority (18 articles) referred to dry matter production from small plots cutting trials (Table 3.1). Establishment of *Stylosanthes* spp. was the object of 9 articles, while 3 articles covered undersowing of rice with *Stylosanthes* spp. and 2 dealt with the oversowing of native savannas with *S. capitata*. There were 25 articles reporting results from the RIEPT trials, mainly from small plots cutting trials. Most of these were from Brazil and Colombia, but there were also some from Argentina, Cuba, Puerto Rico and Venezuela, showing the broadness of environments covered by these regional trials. The common methodology used in these trials has produced valuable datasets, which can be explored for further insights on the interactions between the environment and accession performance. Exploiting this data, Amezcua et al (1991) found that temperature and soil texture were the main characteristics defining the performance of *S. guianensis* cv. Pucalpa in a range of environments. Although adaptation to environment is not the only determining factor for the success of a cultivar, the baseline information from such trial networks, when combined with data from experiments under controlled conditions using geographical information systems (GIS), can be a useful strategy to climatically match sites to target different cultivars.

Although Thomas (1984) stressed the need for more grazing trials to measure animal performance and to gain insights into legume persistence mechanisms, during the last decade the number of articles on *Stylosanthes* spp. grazing trials was only 12% of the total publications on the genus (Table 3.1). Most of these trials had the single objective of comparing beef production in grass pastures with and without *Stylosanthes* spp., and were without any consideration of other variables. The effect of legume-based pastures on dairy production was the object of a few trials, most of them in the humid tropics. A lack of persistence of *Stylosanthes* spp. was the main concern in these intensive dairy production systems. Few studies proposed pasture management guidelines based on grazing intensity or frequency, pasture height or grazing systems. Stylo persistence or disappearance was merely verified and there was no in-depth examination to give the necessary insight into the causes of the disappearance or of stylo collapse.

### **Abiotic and biotic stress**

Only the edaphic component was considered in 37 articles that referred to abiotic stress (Table 3.1). Alternatives and doses of phosphorus fertilisation and aluminum toxicity dominated as research topics, always under a low input system and most dealing with *S. guianensis* in glasshouse trials. Some important topics, such as the maintenance of fertilisation and its effects on productivity and botanical composition of grass–stylo pasture, were not covered.

Of the 33 articles on biotic stress, 20 deal with anthracnose, probably because of its devastating damage to the earlier Australian cultivars and its endemic nature in Latin America (Table 3.1). *Colletotrichum gloeosporioides* diversity and characterisation and plant resistance were the most important disease aspects reported. Although there are reports of pest attack, mainly from stem-borers (*Caloptilia* sp.) and bud worms (*Stegasta boquela*), among others, there were no papers on these in the CAB abstracts.

### **Seed production and biology**

Eleven of the 21 articles on seeds dealt with seed germination and methods to overcome dormancy or hardseededness (Table 3.1). These papers, however, followed an academic approach and the research results have little chance to impact on existing stylo seed production activities. Aspects of seed production technology, such as techniques for mechanical scarification of large seed lots, irrigation, seeding rates and row spacing, and specific seed production practices for commercial cultivars were covered in 6 papers. These papers provided practical and useful agronomic guidelines for seed producers.

A high and reliable seed yield is a key characteristic in a successful cultivar as it affects seed availability in the market. High seed yield and indirect seedling recruitment also ensures persistence in grass–stylo pastures. High seed yield is inherent in some species such as *S. macrocephala* and *S. capitata*, but not in others such as *S. guianensis* var. *pauciflora* and some late flowering accessions of *S. guianensis* var. *vulgaris*. Low commercial seed yields have constrained adoption of late flowering cultivars from these species. Choice of suitable sites for seed production is a definitive question to be addressed by research, before or during the release of any new cultivar. The environmental requirements for flowering, seed formation, maturation and harvesting must be at least roughly known at the start of commercial seed production of a new cultivar. Probably, this is the single most important factor leading to a sustainable availability of inexpensive seeds in the market.

An important objective, sought in all *S. guianensis* breeding programs in Latin America, was to combine high seed production with anthracnose resistance. However, it is difficult to measure seed production due to shattering in many species. Consequently, plant characteristics that have a high correlation with potential seed yield will facilitate evaluation at the initial selection stages.

## Alternative uses

Nineteen articles deal with alternative uses for *Stylosanthes* spp., covering agroforestry systems in the humid tropics; fallow cropping in grain production; and cover cropping for cassava, citrus, coconut and banana plantations. The potential for using *Stylosanthes* spp. as a biological control for ticks and nematodes was also studied. Exploring the possibilities of using *Stylosanthes* spp. as a cover crop, green manure or as a component of agroforestry systems will further enlarge the seed market. A large seed market can offer a better perspective on potential profits from a newly released cultivar and so stimulate a greater number of seed producers to engage in seed production.

## Microbiology and nutrient cycling

Nitrogen fixation, microbiology and nutrient cycling were covered in 31 articles (Table 3.1). Besides measuring the response to inoculation with rhizobia or endomycorrhizal fungi, some quantified biological nitrogen fixation and transfer from *Stylosanthes* spp. to companion grasses in mixed pastures. However, most followed a qualitative and exploratory approach, in which *Stylosanthes* spp. were compared against other legume species, under a single management and/or environmental condition. A fixed N pool in the leaf litter appears to be the most important condition for system sustainability, and *Stylosanthes* persistence and forage productivity are necessary conditions for adequate N supply in the system.

## Forage nutritive value and quality

There were relatively few articles in the last decade on the nutritive value and quality of forage. Most of these studied the nutritive value of *S. guianensis* hay or its effect on grass consumption when combined in different proportions. No comparison has been made between cultivars or species and most articles did not provide insights into how genotypes, environmental conditions or management practices influence the forage nutritive value. In addressing the main challenges on legume persistence and adaptation to biotic and abiotic factors, forage quality has played a secondary role in the evaluation and selection of new *Stylosanthes* cultivars. However, considering the morphological variation, there is room to improve *Stylosanthes* spp. forage utilisation through improving nutritive value. Characteristics like leafiness and reduced lignin will increase consumption and digestibility, although scarce information on protein digestion in the rumen makes it difficult to predict the role of *Stylosanthes* as a low cost protein supplement. Another important characteristic is the rate of decline in nutritive value; cultivars with a slower rate can be particularly suitable for utilisation systems relying on forage accumulation, eg in protein banks. Palatability is another characteristic that influences plant survival and forage accumulation

A review of the literature on *Stylosanthes* spp. has shown a scarcity of information on topics of crucial importance to germplasm evaluation and selection and, consequently, to the release of successful cultivars. For

instance, mechanisms of persistence under grazing, seed production technology, alternative usage of forage legumes, fertiliser effects on legume persistence in grass–stylo pasture, and several others previously identified by Thomas (1984), Miles and Lascano (1997) and Miles (2001) have received inadequate attention.

## Utilisation of *Stylosanthes* Cultivars in Latin America

The objective of a pasture research program is to bring improvements to livestock production systems. This positive and sustainable impact occurs when a new technology, such as a new cultivar or a management strategy, is widely adopted by farmers. Adoption and utilisation of *Stylosanthes* spp. cultivars in Latin America have been limited and, to date, have not had any impact on cattle production (Miles & Lascano 1997). Of the three introduced Australian cultivars and the ten cultivars released in the market, only Mineirão and, more recently, Campo Grande are available in the seed market. Therefore, it is strategically pertinent to question why these cultivars did not succeed.

Until the 1960s *Stylosanthes* spp. were restricted to research stations, although materials with good forage quality were already available and the importance of native *Stylosanthes* as a component of natural pastures had been identified by Otero (1941, 1952). A strategic decision was made at the end of 1960s to set up an international program for beef production in Latin America by using the vast areas not suitable for cropping or areas that were not cropped due to economical constraints. The objective was to produce cheap beef and milk, the two main components of the diet of the poor classes, in order to reduce famine and social inequality. The approach was to combine capital resources with technical know-how, and financial support from international and national banks was directed to provide livestock development assistance in 15 countries in Latin America (Fransen 1975). Technology and resources were transferred to encourage rational use of land for beef and milk production, with emphasis on low-cost production systems through the use of improved pastures, particularly by introducing legumes and fertiliser. The target areas were low fertility soils and pastures with low protein content during the dry season, which imposed limitations on beef and milk production in the then existing extensive livestock production systems. At that time, Australia was the only country in the tropical belt with a successful history of pasture development; *Stylosanthes*-based pastures had been shown to be a viable and economical solution to the dry season protein shortage. A great international seed trade in Australian tropical pasture cultivars was established to support the Latin America pasture development program and the Australian cultivars of *Stylosanthes guianensis* became an important item in this seed trade portfolio.

At the same time, research on *Stylosanthes* accelerated at international research institutions like CIAT, in partnership with the national research institutions. This has resulted in the identification of several promising

**Table 3.2 Released *Stylosanthes* spp. cultivars and main constraints to their adoption/utilisation in South and Central America.**

Species	Cultivar	Availability in the market (years after introduction or release)	Possible causes for failure or low adoption
<i>S. guianensis</i>	Cook	Few	Anthraco-nose susceptibility
<i>S. guianensis</i>	Endeavour	Few	Anthraco-nose susceptibility
<i>S. guianensis</i>	Schofield	Few	Anthraco-nose susceptibility
<i>S. guianensis</i>	cv. IRI 1022	Few	Anthraco-nose susceptibility
<i>S. guianensis</i>	cv. Bandeirante	None	Seed availability
<i>S. macrocephala</i>	cv. Pioneiro	None	Seed availability
<i>S. guianensis</i>	cv. Pucalpa	3–4 years?	Seed availability
<i>S. guianensis</i>	cv. Savana	3–4 years	Anthraco-nose susceptibility
<i>S. guianensis</i>	cv. Mineirão	Currently available	High seed price in the market
<i>S. capitata</i>	cv. Capica	2–3 years	Seed availability
<i>S. capitata</i> and <i>S. macrocephala</i>	cv. Campo Grande	Currently available	Success/failure not yet determined due to its short history
<i>S. capitata</i>	cv. Lavradeiro	In pre-release process	

materials, and at least 12 cultivars of *S. guianensis*, *S. capitata* and *S. macrocephala* have been released or imported from Australia for use in Latin America (Table 3.2). Some, including *S. guianensis* cvv. Deodoro I and II (Costa & Curado 1980) and La Libertad (CIAT 1972), only appear in the literature and there are no records on their release, detailed evaluation data, on-farm trials or seed production. The Australian *S. guianensis* cvv. Schofield, Cook and Endeavour were the first to be used in commercial pasture development in tropical America. Availability of imported seeds and an efficient technology transfer strategy linked to funds from government programs were the main factors behind this initial spike in *Stylosanthes* use in Brazil. To a much lesser extent, these cultivars were also used in other Latin American countries. Selected for Australian conditions without a severe anthracnose disease pressure, these cultivars, when reintroduced to the centre of origin of the anthracnose pathogen, were devastated by the disease within one to two years of their establishment in commercial pastures (Hutton 1978). The widespread failure of these cultivars brought forth great disappointment among farmers, extension workers and consulting technicians involved in the projects (Barcellos et al 2001).

In the mid 1960s the International Basic Economy Corporation's Research Institute (IRI) released *S. guianensis* cv. IRI 1022, an accession collected in Matão, São Paulo State, Brazil. IRI assured ample seed distribution to other Brazilian research institutions (Hymowitz et al 1967) and there was commercial seed production in Brazil (John Landers, former IRI staff, pers. comm.). This cultivar was available at least up to 1977, with SEMEL, a Brazilian seed enterprise, producing seeds from a 15 ha area (Cardoso 1994). Once planted in large commercial pastures in diverse environments, IRI 1022 became susceptible to anthracnose and, like the Australian cultivars, was also devastated by the disease.

*Stylosanthes guianensis* var. *pauciflora* cv. Bandeirante, released in 1983 (Souza et al 1983a), had anthracnose resistance, excellent capacity to retain green foliage during the dry season, good forage yield and adaptation to low fertility soils. Seed yields of only around 50 kg/ha, under research station conditions (Andrade et al 1983), was the main constraint. *Stylosanthes macrocephala* cv. Pioneiro, also released in 1983 (Souza et al 1983b), was the first cultivar from this species. Good persistence under severe grazing, high anthracnose resistance, good seed production and adaptation to low fertility soils were the main attributes of cv. Pioneiro. At that time the perception was that the release of anthracnose-resistant cultivars would favour a rapid and wide adoption by farmers to naturally create a stimulus for commercial seed production. Both cultivars, however, never reached the market because of inappropriate release processes, insufficient promotion involving public and private extension agents, and a lack of interest from seed companies in commercial seed production (Andrade, R.P. unpublished).

*Stylosanthes capitata* cv. Capica, released in 1983 by CIAT and ICA (Instituto Colombiano Agropecuario) for the Colombian Llanos region, was a mixture of five *S. capitata* accessions. High forage productivity, excellent adaptation to soil and climatic conditions of the Llanos, good seed yields and resistance to both anthracnose disease and stem borer were the main agronomical attributes for this cultivar (ICA-CIAT 1983). Unlike cv. Bandeirante and cv. Pioneiro, the release of Capica followed an organised planning process, involving basic seed production, on-farm validation and diffusion to the Colombian farmers (Ferguson et al 1989). The release efforts involved farmer associations and the official extension service, and included arrangements such as funds to support seed production and marketing (Díaz & Sánchez 1994; Giraldo & Sanchez 1994). There was reasonable initial adoption of cv. Capica while official institutions led the process, but this did not lead to a strong demand

for the cultivar and, consequently, commercial seed production did not endure and the cultivar is no longer available in the market.

*Stylosanthes guianensis* cv. Savana was released in 1994 in Florida, USA (Chambliss et al 1994). Commercial seed was available for three to four years after its release and anthracnose attack was the main reason for its disappearance from the market (C.G. Chambliss, pers. comm.)

*Stylosanthes guianensis* cv. Pucalpa was jointly released by IVITA (Instituto Veterinario de Investigaciones Tropicales y de Altura) and INIPA (Instituto Nacional de Investigación y Promoción Agropecuaria del Perú) in 1985, following a five-year evaluation in distinct locations in Perú (Amezquita et al 1991). Besides productivity, there was outstanding anthracnose resistance under humid tropical conditions, which was attributed to the presence of antagonistic bacteria on leaf surfaces and to the small variation between day and night temperatures, both exclusive to tropical humid environments (Lenné & Ordóñez 1997; Lenné et al 1985). From 1988 to 1991, a total of 1500 kg of cv. Pucalpa seed was produced in a CIAT-IVITA-INIAA collaborative seed production project (Vela et al 1991). Initially, on-farm trials and technology transfer activities were the main users of the seed. After 1991 FUNDEAGRO, an NGO that supported the Peruvian agriculture development, took over the seed production and implemented a more private enterprise approach (Hidalgo 1994). Although this has been a pioneering attempt at technology transfer to establish seed production in a frontier region, cv. Pucalpa did not show the expected performance under grazing (Miles & Lascano 1997) and seeds are not available in the market.

*Stylosanthes guianensis* var. *vulgaris* cv. Mineirão, released in 1993 by EMBRAPA, was the first cultivar released by a national research institution to effectively reach the market and be adopted by farmers (CPAC-CNPG 1994a, 1994b). This cultivar was the top performing accession in a cutting trial network covering the Cerrados region and was also tested under grazing in on-station and on-farm trials before release (Ayarza et al 1999; Zoby et al 1993). It shows reliable anthracnose resistance, high forage production, adaptation to low fertility soils and remarkable green forage retention during the dry season (Embrapa Cerrados 1998). The release process was well organised, although the efforts to produce basic seed were not efficient. Following a widespread initial interest and eventual commercial seed production by seed companies, only one firm is currently producing cv. Mineirão seeds in Brazil. Effective commercial seed production started in 1996 in the northwestern region of Minas Gerais state. This seed firm has invested in promotion and marketing and, based on the seed sale since 1997, it is estimated that an area of about 30,000 ha has been planted in Brazil (Mario Martins, Sementes Mineirão owner, pers. comm.). Most of the cv. Mineirão adopters are concentrated in the Cerrados region, but over the years there were buyers from other regions. To date, corroborating the research data on its excellent anthracnose resistance, there has been no report of serious disease outbreaks. However, low seed yields and a lack of long-term persistence under grazing remain the main drawbacks for this cultivar.

The main uses for Mineirão have been in protein banks, pasture renovation and the establishment of grass-legume pastures. Farmers have recognised the benefits of high yields of good quality forage and the retention of green foliage during the dry. Consequently, the use of protein banks to supplement cattle grazing grass pastures has become widespread for this cultivar, mainly in semi-intensive dairy farms, where Mineirão protein banks reduce protein supplementation costs. Protein banks also demand less labour than other cut-and-carry supplementation options using elephant grass (*Pennisetum purpureum*) or sugar cane (*Saccharum officinarum*). Mineirão protein banks are established in a 15–20% area of a grass paddock to allow grazing from June to September, the dry season peak. Cattle access to the protein bank can be free or controlled, for two to four hours per day. Subdividing the protein bank into sections, each grazed for a month during the dry season, can increase efficiency and avoid unnecessary trampling which reduces forage availability (Barcellos et al 2001).

Another main use for Mineirão is in pasture reclamation, in particular of degraded *Brachiaria decumbens* pastures, which involves adding fertilisers, tilling the soil and broadcasting the seed. The purpose of the tillage, which can vary from plowing to light harrowing depending on the soil type and pasture condition, is to incorporate lime and fertilisers and break the compacted soil surface. Soil tillage also eliminates or reduces the regrowth vigor of *B. decumbens*, and ensures favourable conditions for the establishment of Mineirão seedlings (Barcellos & Vilela 2001). Recommendations on using cv. Mineirão in the establishment of mixed grass-legume pastures are restricted to *Andropogon gayanus* cv. Planaltina and *B. decumbens* grasses. The *B. brizantha* and *Panicum maximum* cultivars are too aggressive and, when associated with these grasses, Mineirão persistence is restricted to the first year of establishment. On the other hand, *A. gayanus* and *B. decumbens* allow Mineirão to persist as a component of the mixed pasture for about four years. In these mixed pastures Mineirão constitutes over 60% of the botanical composition of the pastures during the first and second years. After this climax, stylo composition falls to a minimum in the fourth year (EMBRAPA Cerrados 1998). For all the above uses, the recommended seeding rate for Mineirão is 0.5–0.7 kg/ha of Pure Live Seed (PLS), which is relatively low compared to seeding rates of 1–2 kg/ha, normally recommended for this species.

The cultivar Campo Grande, released in 2000, is a mixture of *Stylosanthes capitata* and *Stylosanthes macrocephala* lines. Plants of these two species were collected in 1990 from residual populations in an abandoned experimental area on a low fertility sandy soil that had been subjected to grass invasion and uncontrolled heavy grazing. At the EMBRAPA Beef Cattle Research Centre, plants collected from this old experimental area, along with other pre-selected *S. capitata* and *S. macrocephala* lines were evaluated for high forage yield, anthracnose resistance, flowering and uniform seed maturity. Some degree of outcrossing might have happened, and after six selection cycles for each



Nelore steers grazing a section of the protein bank in Brazil (photo: A Barcellos).

species, seeds of the top lines were mixed (EMBRAPA Gado de Corte 2000; Miles & Grof 1997). The cultivar is a seed mixture containing, on a weight basis, 80% *S. capitata* and 20% *S. macrocephala*, which resembles the species proportion surviving at the old experiment site. Seed production is carried out separately for each species and seed mixing occurs before marketing. Campo Grande has shown good anthracnose resistance and, due to the mixing of different lines from two species, stable anthracnose resistance is expected. It has excellent adaptation to sandy soils and in regional trials has emerged as the best performer in the southern part of the Brazilian Cerrados, where the dry season is less severe. It is a prolific seed producer and seedling recruitment is its main persistence mechanism. On-farm experiences have shown that seedling recruitment is greatest in sandy soil and, consequently, it shows good persistence in association with *B. decumbens* (EMBRAPA Gado de Corte 2000). In areas with a long and intense dry season, cv. Campo Grande does not maintain foliage after the end of the rainy season. Under these conditions, its main contributions are nitrogen fixation and improvement of cattle's diet during the rainy season. There is virtually no way to assure that seed companies will maintain the recommended mixture of the two species in commercial seed lots, and there is the possibility of a shift in balance among the lines within each species once seed production activities occur under a different set of climatic conditions.

Campo Grande stylo is used in mixed grass–legume pastures and for pasture reclamation. Its use in the recuperation of degraded pastures follows a similar route to the one used for Mineirão. In another instances it is sown with fertilisers in rows using zero tillage seeders, following a herbicide application to suppress grass growth (EMBRAPA Gado de Corte 2000).

The new *S. capitata* cv. Lavradeiro was selected from the *S. capitata* cv. Capica at EMBRAPA Roraima for use in the savanna-like regions in the northern part of Brazil. It is in a pre-release state and basic seed production is under way. This cultivar can be used for establishment of

*Andropogon* grass–stylo pasture or as a cover crop to add nitrogen to a corn zero-tillage cropping system (Vicente Gianlupi, pers. comm.).

## Adoption Constraints

*Stylosanthes* spp. can be useful under a number of production systems, such as in mixed grass–legume pastures, for renovation of degraded pasture, as a protein bank to supplement native and cultivated grass pastures, and in ley farming systems (Miles et al 1994). It is surprising that a plant with so many optional uses has not succeeded in Latin America; Miles and Lascano (1997) have discussed the main reasons for its low adoption. Broadly, there are agronomic, economic and cultural reasons for the poor adoption of *Stylosanthes* on this continent. Brazil is the country in Latin America with the greatest experience in both releasing cultivars and in the commercial seed production and use of some cultivars at the farm level. Therefore, it is useful to analyse the constraints from the Brazilian experience with *Stylosanthes* cultivars.

Anthracnose susceptibility has been the main constraint to the adoption of Australian stylo cultivars. The sudden and rapid degradation of stylo-based pastures due to anthracnose damage has discouraged the use of grass–stylo pastures among farmers. It is arguable, however, whether this negative experience with anthracnose-susceptible cultivars is strong enough to be a major impediment to the adoption of cultivars released some years later. In fact, the anthracnose resistance of *S. guianensis* cv. Bandeirante and *S. macrocephala* cv. Pioneiro, released in 1983, has not improved their adoption in Brazil. Similarly, the anthracnose-resistant cv. Pucalpa has not succeeded in Peru.

Probably, seed related problems are the most common cause of unsuccessful release and poor adoption of cultivars. High seed yield is vital to the agronomic performance of a successful cultivar (Ferguson 1985). Low seed yield was the main cause for the failure of *S. guianensis* cv. Bandeirante and for the poor adoption of cv. Mineirão. In general, pasture seed producers do not choose cultivars with low seed yield or cultivars that require complex harvesting techniques. Producers of cv. Mineirão seed consider the presence of great amounts of viscous green foliage at harvest a serious disadvantage for combine harvesting. Low seed yields imply a high production cost per kilogram and, consequently, a high seed price in the market. Use of low seeding rates is a subterfuge to reduce establishment costs. However, experience with cv. Mineirão, for which a seeding rate of 0.5–0.7 kg/ha of PLS is recommended, is showing that this is a risky strategy since low seeding rates increase establishment failures and create a negative perception about the cultivar. One recent initiative from seed companies, apparently successful, is to sell a mix of Mineirão (1 part) and Campo Grande (3 parts) seeds with a recommended seeding rate of 2 kg/ha of the mixture. Campo Grande seeds are about 10 times cheaper than Mineirão seeds, which lowers the market price for the mixture and, consequently, the seed expenditure per hectare. At this seeding rate, stylo seedlings outcompete weeds to rapidly establish a legume stand.





A typical *Stylosanthes guianensis* cv. Mineirão protein bank in Brazil. (photo: A Barcellos)

Unsuccessful releases of *S. macrocephala* cv. Pioneiro and *S. capitata* cv. Capica, respectively in Brazil and Colombia, showed that high seed yield assures neither seed availability nor widespread adoption. Both cultivars had good forage production, tolerance to low fertility soils, anthracnose resistance and good seed yields for easy harvesting using combine harvesters. However, seed firms in both countries did not foresee a big enough market for the cultivars, and avoided commercial seed production. To some degree, this situation has been repeated with other cultivars.

The ability of the legume to persist in a mixed pasture has been taken as an essential characteristic for the success and adoption of a new stylo cultivar. Although highly desirable, it appears that a lack of persistence is not always an important adoption constraint. For instance, farmers who have adopted Mineirão accept that it will not last more than four years, but they are willing to use it if it provides a good profit margin (Mario Martins, Sementes Mineirão owner, personal communication). The need for a persistent pasture legume is not important in integrated crop–livestock production systems, where the concern is whether the legume may become a weed in the cropping phase.

In general, farmers in tropical America are accustomed to grass pastures and do not recognise the economic and ecological benefits from the use of legume pastures (Miles & Lascano 1997). This is perhaps the most overwhelming constraint to adoption of even the most outstanding cultivar. It may require well-structured education campaigns based on real life on-farm results including benefit–cost analysis to overcome this constraint, and the handful of leading producers who have pioneered adoption would be invaluable spokesman in this process.

## Future Prospects

Research priorities will have to be realigned to screen, select and release cultivars that are readily adopted. The perceptions and experiences of seed producers and early adopter farmers will be an important element in determining the mix of attributes for any new cultivar. As pointed out by Miles (2001), defining the key plant characteristics that make cultivars readily adopted has been hindered by inadequate communication between breeders and the private sector. The recent partnership between EMBRAPA and Unipasto, an association of Brazilian pasture seed firms which partially supports EMBRAPA's tropical forage evaluation and breeding programs, will provide a first-time opportunity to fill this gap. Cuts in research funding have affected both CIAT and the national research institutions, and have greatly reduced the momentum of pasture research throughout Latin America. Such partnership, facilitated by the adoption of plant breeding rights legislation in Brazil, may also help overcome budget shortages in addition to providing direct industry inputs to research. Brazilian pasture seed firms undertake significant export to markets in South and Central America. Although currently dominated by grasses, pasture legume seeds of commercially successful cultivars, including stylo, can take a share of this market.

It is clear that inadequate release and follow-up procedures have been a major reason for the lack of adoption of some good cultivars. Efficient release strategies are designed to achieve widespread use of a new cultivar and constitute seed production, diffusion and promotion activities (Ferguson 1985). The establishment of a seed supply system, including basic and commercial seed production, to supply high quality cheap seed to the market is a key ingredient. Although commercial seed production and supply are a function of market demand, concatenated and strategic diffusion to promote the new cultivar can help generate demand (Ferguson et al 1989).

More recently, there has been a growing acceptance of crop–livestock integrated systems in the Cerrados of Brazil. Other countries in South America are willing to adopt integrated crop–livestock production systems to develop their savannas, and there is a clear tendency for the border region between the Cerrados and the Amazonian regions to become the main beef production zone in Brazil (Judson Valentin, pers. comm.). This will generate new scenarios for pasture development in South America to influence *Stylosanthes* research and development.

Many farmers have little knowledge of the potential benefits to be gained from the use of *Stylosanthes*. Ongoing communication and targeted education must accompany the release of cultivars with cheap and readily available seeds. Despite some agronomic limitations, Campo Grande stylo, which is now available in the Brazilian market at relatively low prices, is expected to play this role. An appropriate and transparent technology transfer strategy that does not exaggerate performance will also help create a stimulus for widespread adoption.

## Acknowledgments

The authors are thankful to Dr Gaston Sauma (SEFO Semillas, Bolivia), Dr Carrol Chambliss (University of Florida, USA), Dr Eduardo Cuadros (SAIU S. R. Ltda, Peru) and Dr Luis Ramires Avilés (UADY, Mexico) who provided information about ongoing *Stylosanthes* utilisation/seed production in their respective countries. Thanks are also due to Mr Mario Martins (Sementes Mineirão) for sharing his experiences in *S. guianensis* cv. Mineirão seed production and marketing.

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## Chapter 4

# Cattle production from *Stylosanthes* pastures

Trevor J. Hall<sup>1</sup> and Albrecht Glatzle<sup>2</sup>

### Summary

Introducing legumes to improve soil fertility and animal production from grass-dominant pastures on low fertility soils of the tropics has been a long-proven successful management system. The *Stylosanthes* genus (stylo) is being grown on all continents with varying levels of success and it has provided some of the most successful species for light-textured tropical soils, in both the high rainfall and the seasonally dry tropics. Although stylos will grow well at low fertility, the addition of nutrients, particularly phosphorus, have been required for grazing animals to capitalise on the higher protein and digestible energy intake available from a mixed grass–*Stylosanthes* pasture.

The annual increase in beef production from adding stylos to a pasture, providing phosphorus is not severely limiting, is around 30–60 kg/head, with over 100 kg/head achievable in some environments by also supplementing animals directly with nutrients. An increase of 2 litres of milk per day can be obtained from lactating cows on stylo-based pastures. Increases in stocking rates of over two-fold are normal in well-established *Stylosanthes* pastures.

The increasing market demand for younger and better-conditioned animals has provided the impetus for improving cattle nutrition by sowing *Stylosanthes* species in tropical and coastal subtropical areas around the world. The improved animal production from *Stylosanthes* pasture technology has increased the financial viability of cattle grazing in the tropics and provided greater management options to meet consumer demands for higher quality products. Cattle, sheep, goats, pigs and poultry are being fed from stylo pastures. Future roles for *Stylosanthes* species will expand into mixed grazing and farming systems of tropical countries.

This chapter outlines the adaptation of *Stylosanthes* species in grazing systems and reviews the latest research on cattle grazing performance from stylo pastures.

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### Introduction

The *Stylosanthes* genus is a diverse group of species with a wide distribution throughout tropical Central and South America (Williams et al 1984). The genus has become well known in tropical agriculture over the last four decades. The natural distribution, taxonomy and environmental adaptation of pan American, South and North American, African and Asian species of *Stylosanthes*, the diseases and pests, genetic resources, cultivar development, utilisation, potential and limitations for animal production were reviewed by Edye (1987).

Species of *Stylosanthes* have proved to be widely adapted to low fertility, light textured, acid soils of the tropics. Stylos have been introduced to countries around the world with tropical and subtropical pasture grazing systems since the 1960s, to compliment native grasses and improve sown pasture quality. At least two reviews have appeared on animal production from *Stylosanthes* species (Coates et al 1997; Gillard & Winter 1984), comparing animal production from sown stylo pastures with native grass pastures. The Australian trials were conducted in central and northern Queensland and in the northern half of the Northern Territory from the early 1960s, and some have included the effects of adding fertilisers and direct nutrient supplements to the cattle.

The annual extra liveweight gain by adding a stylo into grass pasture has varied markedly, from no benefit in drought years to over 100 kg/head in long growing seasons. The common range is 30–60 kg/head extra liveweight per year from the legume. Animal performance cannot always be improved simply by introducing a stylo if soil fertility is poor, which in turn grows a nutrient deficient pasture. The addition of fertiliser, usually superphosphate, and direct mineral supplements, such



as phosphorus, during the pasture growing season will increase cattle growth rates on stylo pastures on these low fertility soils.

New cultivars have been developed from a range of *Stylosanthes* species suited to both high and low rainfall environments. These cultivars include a range from proven species, such as *S. guianensis* in China (Changjun et al this volume), *S. capitata* and *S. macrocephala* (Grof et al 2001), and a more recently identified species, *S. seabrana* (Edye et al 1998), which has been evaluated for animal production (Hall 2000a). The release of the *S. capitata*-*S. macrocephala* cultivar Campo Grande in Brazil has reignited interest in the commercial development of stylos (Valle et al 2001). In northern Australia the current area of over 600,000 ha of commercial *Stylosanthes* pastures, predominantly *S. scabra* and *S. hamata*, is expanding annually (Miller et al 1997). This chapter serves as a timely review of recent research on cattle production from stylo pastures.

### **Stylosanthes Species in Production Systems**

*Stylosanthes* species are among the most important forage legumes across northern Australia (Gillard & Winter 1984) and South America (Miles & Lascano 1997; Thomas 1984), and they are playing an increasingly important role in Asia and India (Ramesh et al 1997), China (Guodao et al 1997) and more recently in Africa (Peters et al 1994). Stylos are now being used to feed many forms of livestock, including cattle, goats, sheep, pigs and poultry.

Across the tropics in Australia there are extensive areas of low fertility soils with a light textured surface, which are suitable for introducing *Stylosanthes* species. *Stylosanthes guianensis* is the most suited species to the wet tropics; *S. hippocampoides* (cv. Oxley) does well in the subcoastal subtropics; and *S. scabra* and *S. hamata* have been successful species across the seasonally dry tropics (Miller et al 1997). Recently, *S. seabrana* has proved adapted to heavy clay soils of the inland subtropics (Edye et al 1998).

*Stylosanthes capitata* and *S. guianensis* were considered by Mannetje and Jones (1992) to be the best species for tropical and wetter areas of South-East Asia, and *S. hamata* and *S. scabra* were adapted to semi-arid areas. These species are all suitable for the cut-and-carry forage systems in this region (Phaikaew et al, this volume). Peters and colleagues (2001) reviewed the role of improved forages in enhancing smallholder productivity and maintaining ecosystem health in the tropics and presented studies of forage adoption of *S. guianensis* in China. The high-yielding, cut-and-carry forage systems using *Stylosanthes* enhanced year-round animal productivity, as well as improving land-use efficiency and reducing labour requirements.

In West Africa Muhr and colleagues (2001) reported that cropping and dry season feeding strategies were increasingly being limited by land availability, so the agronomic performance of legume species, in particular *S. guianensis*, promised substantial productivity gains once they could be integrated into the traditional fallow systems. Little and Agyemang (1992)

have reviewed the role of *Stylosanthes* species in the African context and reported on its evaluation, population dynamics, animal production, agronomy, seed production and integration into cropping systems. Pengelly et al (this volume) have summarised the success of stylo in cropping systems of Africa and Australia.

### **Stylosanthes Species Research in Animal Production**

The morphological and environmental adaptation variation within the *Stylosanthes* genus has provided commercial cultivars from species suited to a wide range of animal production systems in wet and dry tropical and subtropical environments around the world. Some of the successful species are:

#### **S. capitata**

*Stylosanthes capitata* has shown promise for low pH soils, with high aluminium and manganese saturation in South America (Grof et al 1979), and it has produced a positive effect on animal production (Thomas et al 1987). Cv. Capica, a blend of five similar ecotypes, was released in Colombia but has not been widely adopted (Miles & Lascano 1997). There is excellent performance and animal production from cv. Campo Grande, the multiline composite developed from selected lines of *S. capitata* and *S. macrocephala* (Valle et al 2001). At Fazenda Ribeirão in Mato Grosso do Sul, 11,000 ha are under cv. Campo Grande pastures in association with *Brachiaria decumbens* or *Andropogon gayanus* (B. Grof, pers. comm. 2003). There have been no trials on animal production from *S. capitata* in Australia, although high yields have been produced in small plot evaluation trials (T. Hall, unpublished data). Rhizobium specificity was seen as a limitation to its commercial application in the Australian tropics.



**Nelore cattle on cv. Campo Grande stylo (*Stylosanthes capitata* and *S. macrocephala*) pasture in Brazil (photo: B. Grof).**



## ***S. guianensis***

The first cultivars released in Australia were varieties of *S. guianensis* for the wet tropics and coastal Queensland, eg cv. Schofield, Cook and Endeavour. These cultivars were exported to Asia and South America in the 1970s, where they succumbed to anthracnose disease or did not tolerate grass competition from well-adapted grasses such as *Brachiaria* species or *Pangola*. Endeavour has produced 138 kg/head/year on infertile sands when it was sown with improved grasses and well fertilised (Winter et al 1977).

*Stylosanthes guianensis* cultivars adapted to South America have been developed, eg cv. Mineirão (var. *vulgaris*), which was released in 1993 (Anon. 1993) and is well adapted to the Cerrados region of Brazil. It has thick stems, may grow to a height of 2.5 m with good soil fertility in the second season, is highly anthracnose resistant, and retains green leaf during the dry season. However, it has not been well accepted by farmers or seed producers because of its poor seed yields.



**New *Stylosanthes guianensis* hybrid pasture in northern Queensland (photo: B. Grof).**

Cv. Pucallpa (CIAT 184) was released in Peru for the humid tropics and low pH soils. This accession has been used in southern China as cv. Reyan II – Zhuhuacao, and is the second most used stylo after cv. Graham, originally an Australian cultivar (Devendra & Sere 1992). Reyan 5 is another cultivar to have originated from CIAT 184 in China, and two others, Reyan 7 from CIAT 136 and Reyan 10 from CIAT 1283, have been recently released in China (Guodao et al, this volume). Leaf meal concentrate is produced for poultry and pigs from these stylos.

In the Chaco in Paraguay, *S. guianensis* cv. Cook grows vigorously in the first year, but does not set seed before frosts in an average year, and even light frosts kill well-established plants.

Although *S. guianensis* cultivars have not been widely sown in Australia in recent years, there are new cultivars being promoted. A new generation of improved, hybrid-derived lines of *S. guianensis* has been developed. This material may have application in coastal and subcoastal regions of the tropics in areas with higher than 1500 mm annual rainfall. Two new lines, Nina and Temprano, were initially selected from South American material (B. Grof, pers. comm. 2003).

In trials of relative palatability of tropical legumes, Peters and colleagues (2000) reported only *S. guianensis* accessions were positively selected by cattle throughout the year.

## ***S. hamata***

A grazing trial comparing the established annual *S. humilis* with the new species *S. hamata* (Caribbean stylo) in northern Queensland demonstrated the benefit of the higher yielding and often biennial species. Selection within *S. hamata* accessions led to the release of cv. Verano in 1973. Subsequent field adaptation trials under grazing led to the release of *S. hamata* cv. Amiga in 1988, because of its superior adaptation to a range of more harsh environments than where Verano was suited (Edye et al 1991).

Native perennial grasses in tropical Australia are notoriously poor competitors at high grazing pressure. This has been observed extensively in commercial pastures, eg at 'Wrotham Park', and it has been widely reported. The poor competitive ability of native perennial grasses was measured in a *S. hamata* grazing trial in northern Queensland, where the native grasses decreased with increasing grazing pressure (by increasing the stocking rate) and they were virtually eliminated at high stocking rates (Jones 2003).

In coastal northern Queensland, Verano is a more successful grazing legume than the trailing *Macroptilium atropurpureum* (Siratro). It was becoming the dominant species at high and medium grazing pressure until *Bothriochloa pertusa* (Indian couch), an introduced grass, invaded and reduced the legume's contribution (Jones 2003). Indian couch was less invasive in the Verano pasture at low grazing pressure.

Over a four-year period, liveweight gains of steers grazing Verano-dominant pastures during the wet season were similar in northern Queensland (Lansdown) and in the Northern Territory (Katherine). However, losses in the dry season at Katherine were far greater than at Lansdown (Jones 2004). Mean annual liveweight gain at Katherine was 99 kg/head compared with 155 kg/head at Lansdown. The soils in the Northern Territory were of lower fertility than the Queensland site. Jones (2004) speculated that on infertile soils in the seasonally dry tropics, where stylo leaf can have low plant nitrogen (eg <1% N in Verano leaf) in the dry winter season, this may be insufficient to sustain steer liveweight, even on stylo-dominant pastures. Under these conditions, Verano pastures may require protein supplementation to prevent animal weight loss in the dry season.



Verano Caribbean stylo (*Stylosanthes hamata*) pasture in coastal northern Queensland (photo: D. Coates).

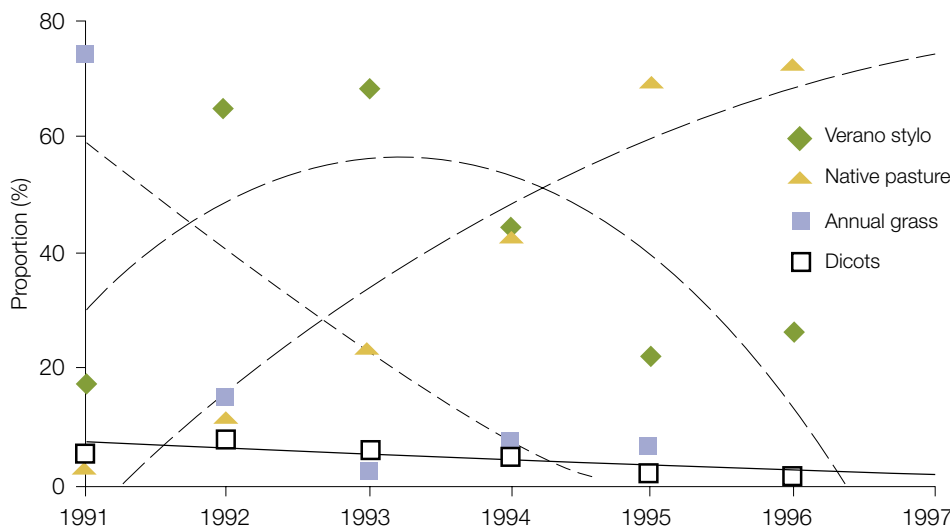
The first experiences with Verano and Amiga in the Chaco of South America in the early 1990s looked very promising. When sown at high seeding rates in low fertility regosol soils, where grasses requiring high fertility would not readily grow, these Caribbean stylos became the dominant pasture component by the second or third year. Thereafter, however, their populations declined, as shown in Figure 4.1. Being a biennial species, the stylos need to regenerate every second year from soil seed reserves, and the seedlings could not compete with the

established perennial grasses. Re-establishment of Caribbean stylo seedlings was slow and inconsistent with the increasing sward density of invading perennial grasses. However, Caribbean stylos, along with *Alysicapus vaginalis*, have been successful in a ley farming system in the Chaco, where they regenerate from soil seed reserves following an intermediate crop of silage sorghum (Glatzle & Ramirez 1993).

### **S. hippocampoides**

In Central Queensland the mean annual liveweight gain on *S. hippocampoides* (formerly *S. guianensis*) finestem stylo (cv. Oxley) was 167 kg/head at a stocking rate of 1.47 head/ha. Over the same period unsown native grass pasture, cleared of timber, gave a gain of 62 kg/head at 0.62 head/ha (Bowen & Rickert 1979). These relative differences in sown stylo pastures compared with native grass pastures have been reported with other stylo species across northern Australia, although total liveweight gains are usually less on the poorer tropical soils than on subtropical soils of Central Queensland.

In the Paraguayan Chaco, Oxley is the only species that persists and increases its proportion in permanent pastures over the years, given appropriate soil conditions (Figure 4.1). Finestem stylo prefers a coarse-textured soil with a sand fraction above 50% and a clay fraction below 5%. As it is frost tolerant, finestem stylo remains green the entire winter, being far less conspicuous during summer and autumn. It has shown high grazing tolerance, persisting even after being grazed close to the ground (1 cm) for months. Best regeneration from seed occurs when the grass component is kept short and the soil is disturbed by animal traffic. Anthracnose incidence, which was observed during prolonged wet periods, does not appear to be a threat to persistence.



**Figure 4.1** Development of pasture composition on a sandy soil (72% sand and 7% clay) in the Chaco of Paraguay. *Stylosanthes hamata* cv. Verano was sown as a sole species in October 1990. Annual grasses were *Digitaria sanguinalis* and *Cenchrus echinatus*, and perennial grasses *Panicum maximum* cv. Gatton and *Cenchrus ciliaris* invaded the pasture.



In South Africa, Kelly and Tiffin (1983) found that Oxley was difficult to establish on veld, and steer liveweight gains in the first year were not improved compared with grass-only pasture. In the second year Oxley reduced steer weight losses during the dry season and increased wet season weight gains. At peak weight, steers grazing Oxley were 13% heavier than those on unimproved veld.

In Zimbabwe the cattle liveweight gain on veld oversown with Oxley was curvilinearly related to stocking rate and was higher under continuous than rotational grazing systems (Lungu et al 1995).

### ***S. humilis***

In the late 1950s *S. humilis* was the first species evaluated for animal production from the *Stylosanthes* genus in Australia. It was called Townsville lucerne or Townsville stylo and was naturalised across extensive patches of the dry tropics and subcoastal Queensland. Examples of animal production include the following: Shaw (1961) reported an extra 48 kg/head from superphosphate-fertilised stylo compared with unfertilised native grass pasture at about one-third the stocking rate of the stylo in Queensland; and Norman and Stewart (1964) reported gains of 36 kg/head- from *S. humilis* compared with native grass pastures in the Northern Territory. Shaw (1978) suggested this increased animal production from the stylo was mainly due to increased legume production from adding fertiliser, and that there was an increase in stylo yield in fertilised pastures at high stocking rates. Grass competition declined at high grazing pressure and these stylo pastures often became legume dominant, while still maintaining good ground cover for most of the year.

*Bothriochloa pertusa* has invaded extensive areas of native pasture across the dry tropics of Queensland, replacing *S. humilis* and *Heteropogon contortus* (black spear grass) pastures on texture contrast (duplex) soils, as the *S. humilis* disappeared with the rapid spread of anthracnose disease (*Colletotrichum gloeosporioides*) in the early 1970s (McCaskill 1992).

This disease eliminated *S. humilis* from commercial pastures across northern Australia within several years. *Stylosanthes hamata* cv. Verano became its replacement. Liveweight gains were greater on *S. hamata* than on *S. humilis* pastures at low legume yields, although there were no differences in liveweight gain when the legume yield of both stylos exceeded 600 kg/ha (Gillard et al 1980).

### ***S. macrocephala***

The commercial material in Brazil, eg cv. Pioneiro, has not received widespread adoption by farmers, even though it is well adapted to western and central Brazil but not to higher rainfall areas (Miles & Lascano 1997). It is a component of the newly released cultivar Campo Grande in Brazil which has provided encouraging results in animal production from stylo-grass pastures (Valle et al 2001). However, it is too early to realistically assess its commercial success. An analysis of the

nutritive characteristics of *S. macrocephala* suggest it has a higher fibre content (>68.8%), which produces a lower dry matter digestibility (37.4% in vitro), than that of *S. guianensis* (Villaquiran & Lascano 1986).

Adapted genotypes of *S. macrocephala* with its specific rhizobium have not been introduced successfully to Australia. In field evaluation of limited genetic material, there was good establishment on a sandy acid (pH 5.5) soil but it was not as productive as *S. scabra* and *S. hamata* accessions, so research never progressed to the animal production stage (T. Hall, unpublished data).

### ***S. scabra***

*S. scabra* cv. Seca (shrubby stylo) has been the most commercially successful tropical legume for the dry tropics of Australia. Cattle producers are convinced of the improved liveweight, increased stocking rates and better husbandry and management options provided by pastures of this species.

On an infertile, sandy-surfaced, duplex soil with P of <5 ppm (acid extractable) in northwestern Queensland, cattle performance on *S. scabra* pastures has continued to improve as the legume content in the pasture has increased over time. On a mixed pasture of Seca and Verano, with native *Sorghum plumosum* (perennial sorghum), *Chrysopogon fallax* (golden beard grass) and *Aristida* spp. (wire grasses), steer liveweight gains of 0.76 kg/head/day were produced on a nine-year-old stylo pasture, compared with 0.43 kg/head/day on native grass pasture, during the dry season from May to September.

**Brahman steer grazing Seca stylo-dominant (*Stylosanthes scabra*) pasture in summer in northern Queensland (photo: K.A. Shaw).**



The steers on the stylo pasture were in finished condition for market, while the native pasture fed steers required an additional year to achieve the same liveweight and they were still not in a finished condition for marketing. There were no consistent animal production benefits in the early establishment years on this pasture (Hall et al 1996). Much higher production, averaging 213 kg/head/year over five years (0.58 kg/day), has been produced from fertilised Seca pastures in more favourable coastal environments of central Queensland (Middleton et al 1993).

In southern South America, as in the Chaco, *S. scabra* cvv. Seca and Siran set seed before frost in an average year. Foliage death due to mild frosts is common, although in the following spring regrowth occurs from the crown. Severe frosts (close to  $-5^{\circ}\text{C}$  at ground level, once every five to ten years) kill established *S. scabra* plants. Regeneration from seed is both poor and slow. Ten years after sowing, shrubby stylo populations decline to below 1%, even in grass pastures where stylo proportions in the second or third year were above 10%.

### ***S. seabrana***

Edye et al (1998) identified and commercialised two accessions of *S. seabrana*, cvv. Primar and Unica (Caatinga stylo), which performed well on fertile, heavy clay soils in the frost-prone inland subtropics, extending the contribution that *Stylosanthes* species can make to beef production in northern Australia. Of the *S. seabrana* accessions evaluated, the two cultivars were the best adapted and amongst the most anthracnose resistant (Trevorrow et al 1998), but they have since suffered anthracnose damage in the high rainfall, commercial seed production areas of northern Queensland (Chakraborty, this volume).

Increased steer growth rates on Caatinga stylo pastures have been measured in a frost-prone environment of southern inland Queensland on a grey-brown cracking clay soil previously supporting brigalow (*Acacia*

*harpophylla*) and wilga (*Geijera parviflora*) forest. Good quality native pastures or sown buffel grass (*Cenchrus ciliaris*) on these soils is used for cattle breeding and fattening (Hall 2000b). Here Primar and Unica with sown Bambatsi (*Panicum coloratum*) and native Queensland bluegrass (*Dichanthium sericeum*) produced over 100 kg/ha liveweight gain each year for three consecutive years. Steer daily growth rates were 0.62 kg/head for ten months of the year, from August to June, at continuous stocking. The native pastures produced 0.35–0.5 kg/head at near half the stocking rate for the same periods (Figure 4.2). The steers on the Caatinga stylo were fat and ready for sale after ten months. Under this heavy and continuous grazing management, the stylo population increased annually and reached dominance in patches, while two other legumes adapted to clay soils, *Clitoria ternatea* (Milgarra butterfly pea) (Hall 1985) and *Desmanthus* spp. (Jaribu desmanthus, a mix of three cultivars) (Cook, Graham et al 1993), declined in population and could not seed (Hall 2000a; Hall & Douglas 2000).

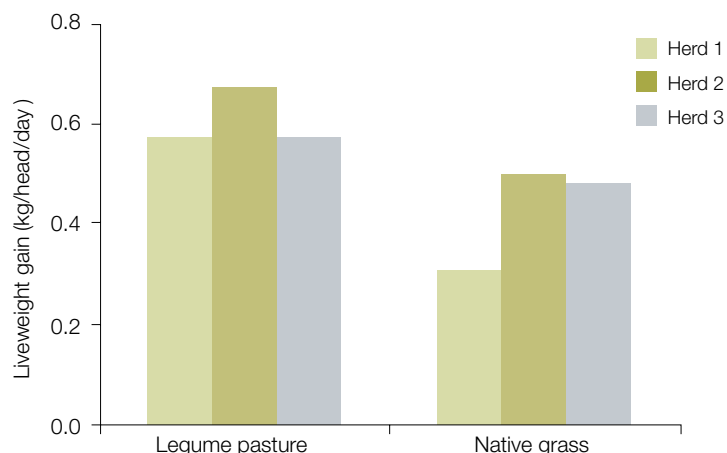
On fertile, medium to heavy basaltic clay soils, pH neutral to alkaline, in subcoastal Queensland, Caatinga stylo produced liveweight gains of 140–200 kg/head/year, equivalent to 160 kg/ha/year. This production was 18% higher than from sown grass pastures. The density and yield of Caatinga stylo increased over time at these production levels (R. Clem, pers. comm. 2003).

Caatinga stylo has the potential, as a ley pasture on clay soils sown for cropping, to increase soil fertility, break disease cycles and be used for animal production to improve the economics of crop and pasture rotation systems.

In the Paraguayan Chaco, Unica is showing promise by being more frost tolerant than *S. scabra* and better adapted to fine-textured soils. Conclusive results on persistence or animal production are not yet available.



Steers grazing Caatinga stylo (*Stylosanthes seabrana*) pastures in winter in the subtropics of southern inland Queensland (photo: T.J. Hall).



**Figure 4.2** Annual liveweight gain (kg/head/day) of three herds of steers grazing *Stylosanthes seabrana*-based pasture (Caatinga stylo cvv. Unica and Primar) with Bambatsi panic (*Panicum coloratum*) compared with native grass pasture (*Dichanthium* and *Chloris* species) in the subtropics of Queensland.

## Animal Production – Australian Experience

### Production on native pastures

Native grass pastures on soils suited to stylos in the Australian tropics are most deficient for animal production due to low phosphorus concentration during the wet season, when plants are green and growing, and low nitrogen (protein) when plants are maturing during the dry season. Energy levels can be limiting, especially during the drier months (Miller & Webb 1990). They are also intolerant of heavy grazing pressure (Jones 2003).

The length of the growing season affects pasture yield, the green leaf period and nutrient dilution, particularly the limiting of soil nitrogen. A range of annual animal production has been reported from tropical grass pastures, varying from low levels of 53 kg/head in the Northern Territory (Norman & Stewart 1964) to 145 kg/head in central Queensland (Middleton et al 1993).

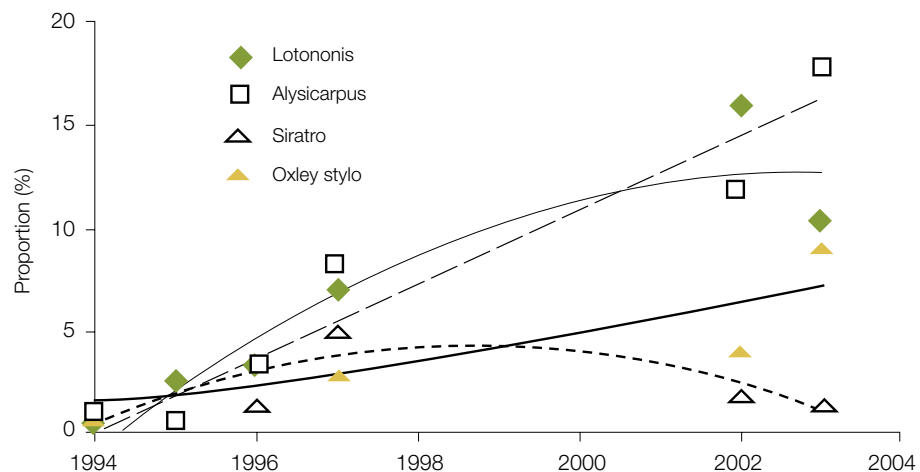
### Production on stylos

In northern Australia the annual cattle liveweight produced from stylo pastures is normally in the range 89 kg/head (Norman & Stewart 1964) to 176 kg/head (Coates et al 1997). Up to 200 kg/head has been recorded on stylo–grass pastures on higher quality soils in central Queensland (R. Clem, pers. comm. 2003).

The annual liveweight gain advantage to cattle grazing stylo–grass pastures, compared with grass-only pastures, is usually 30–60 kg/head (Coates et al 1997), with an additional benefit of increased stocking rates possible. Adding a stylo legume to grass pastures increases cattle intake and reduces nutritional limitations of low nitrogen, which leads to poor digestibility. Coates et al (1993) reported an increase of 18% in dry matter intake during the dry season, and an increase of 27% in the intake of digestible dry matter, on stylo–grass pasture compared with a grass only pasture.

**Figure 4.3** Establishment and persistence of legumes in a continuously grazed *Pangola* grass pasture in the Chaco of Paraguay. Finestem stylo cv. Oxley was the only *Stylosanthes* that increased in proportion during the 10-year period. Other legumes, sown<sup>1</sup> but not shown, either did not persist or remained at proportions below 1%.

<sup>1</sup> Legumes sown in December 1992, in rows 8 m apart with four rows of each species or cultivar, across a 3 ha plot of established *Pangola* pasture on sandy soil (58% sand and 4% clay): *Stylosanthes hippocampoides* cv. Oxley, *Stylosanthes hamata* cv. Verano and Amiga, *Stylosanthes scabra* cv. Seca and Siran, *Stylosanthes guianensis* cv. Cook, *Lotononis bainesii* cv. Miles, *Alysicarpus vaginalis* (CIAT 17360), *Macroptilium atropurpureum* cv. Siratro, *Centrosema pubescens* cv. Belalto, *Chamaecrista rotundifolia* cv. Wynn and *Desmanthus virgatus* (locally collected accession). No fertiliser was applied. Proportions of pasture components were determined by a point intercept method in autumn of every year.



In the late wet and dry seasons of northern Australia the advantage due to incorporating stylo into a pasture can average 0.25 and 0.15 kg/head/day respectively. These increases are associated with increased stylo selection at these times as well as higher nitrogen and digestible energy intake (Coates et al 1997).

It is the green component of a pasture which has the higher nutritional value and the most effect on animal production. In a grass–stylo pasture in northern Queensland there was a close linear relationship between cattle annual liveweight gain and a pasture growth index of number of weeks of green pasture available (Jones et al 1990).

By 1996 *Stylosanthes* pastures contributed some \$20 million annually to beef production in Australia, through higher turn-off weights, improved weaner and heifer nutrition, reduced death rates and reduced drought risk (Miller et al 1997).

## Animal Production – Chaco Experience, Paraguay

There is little information available on animal production from legumes in the Paraguayan Chaco that has been documented with scientific rigour. However, it is commonly accepted that pastures improved with persistent legumes produce more dry matter and tolerate a higher stocking rate than grass-only pastures. In the early 1990s, Oxley, Verano, Amiga, Seca, Siran and Cook stylos were introduced to the Chaco from Australia. These cultivars were widely sown into the regosol soils, partly by seed broadcasting or drilling following thorough soil tillage, and partly by sod seeding into established *Pangola* pastures using a band seeder of Australian origin (Cook, Clem et al 1993).

In the seventh to ninth years after legume establishment, pasture with a legume composition as shown in Figure 4.3 was cut twice per year for hay. It consistently yielded 10–11 large bales/ha (400 kg each) compared to 5–7 bales/ha harvested from an adjacent plot of *Pangola* alone. In addition, when leguminous pastures were rotated to a cropping phase,



**Table 4.1 Pasture composition, stocking rate and annual steer performance (kg/ha) on a *Pangola* pasture, with and without legumes in Paraguay (see footnote of Figure 4.1).**

Years after legume establishment	1	2	4	6	10
Legume proportion (%)	4	8	29	49	36
Stocking rate (steers/ha) with legumes	1.3	1.0	1.0	2.4	2.5
Stocking rate (steers/ha) without legumes	1.3	1.0	1.0	1.5	-
Live weight gain (kg/ha/year)					
In <i>Pangola</i> grass with legumes	292	270	284	464	401 <sup>1</sup>
In <i>Pangola</i> grass without legumes	284	231	227	283	-
Liveweight gain in favour of legumes	8	39	57	181	-

<sup>1</sup> Liveweight gain in 7 months (January to August, 2003)

yields of silage and grain sorghum increased up to 150% and 30% respectively, compared to preceding weedy fallow or non-leguminous crops (Glatzle 1999).

Dairy farmers report that lactating cows produce about 2 litres more milk per day when shifted from a grass-only pasture to a pasture with legumes, particularly Oxley. Grazing typically accounts for 25–50% of the dry-matter ration, with the rest consisting of sorghum silage and concentrates.

On *Pangola* pasture with an annual legume development as shown in Figure 4.3, growing steers were used to compare stocking rates and liveweight gains. Finestem stylo was one of three dominant and persistent legumes. Between the first and sixth years, and again starting with the tenth year, the pasture was continuously grazed at the stocking rates shown in Table 4.1. *Pangola* with legumes produced consistently higher liveweight gains than *Pangola* without legumes. For example, in the sixth year the legume advantage was 181 kg/ha. Furthermore, live weight gain per ha increased over the years in the pasture with legumes, as the legume proportion increased and contributed more nitrogen to the system. Legume proportion in the sward stabilised at about 35%. Despite below-average rainfall in the tenth year, steers gained 400 kg/ha of live weight in only seven months at a stocking rate of 2.5 head/ha. This performance, on a previously rundown sandy soil, compares favourably with production from grass pastures on recently cleared virgin soil.

One conclusion is that adapted and persistent legumes, eg from finestem stylo, can contribute considerably to soil rehabilitation and reconstitution of animal production on rundown arable lands in the Chaco.

*Stylosanthes recta* is a small native species on the Chaco (Hacker et al 1996) but it never attains a sufficiently high proportion within pastures to significantly improve animal production.

## Animal Production – Cerrados Experience, Brazil

A cattle grazing trial at Fazenda Ribeirão, Chapadão do Sul-MS, on a typical oxisol soil in the Brazilian Cerrados compared cv. Campo Grande stylo and *Brachiaria decumbens* (signal grass) with *B. decumbens* alone. Liveweight gains on the legume-based pasture at stocking rates of 0.6 AU/ha, 1 AU/ha and 1.4 AU/ha were 7%, 18% and 20% respectively, above those of the grass-only pasture (B. Grof, pers. comm. 2003).

Mineirão is recommended for use as a protein bank to supplement native grass pastures in Brazil and it can be grazed at 0.3 ha/head. The crude protein content during the season ranges 12–18% and in vitro dry matter digestibility (IVDMD) ranges 52–60%. It has better persistence under rotational grazing with rest periods of 21 and 28 days than under continuous grazing. Mineirão in association with *Andropogon gayanus* has produced liveweight gains of 0.8 kg/head/day in the wet season and 0.15 kg/head/day during the dry season, at stocking rates of 1.8 AU/ha and 1.3 AU/ha respectively. In the Campo Grande region a liveweight gain of 0.5 kg/head/day has been recorded on Mineirão (B. Grof, pers. comm. 2003).

## Management of *Stylosanthes* Pastures for Animal Production

The use of stylo legumes have been relatively low cost and easy to manage in combination with native and sown grasses in Australia. In the early years of stylo introduction, adaptation to low fertility soils and response to low rates of superphosphate increased the rate of commercial adoption. The ability of stylos to increase in population under continuous grazing was seen initially as a benefit; however, as pastures became more widespread and in use for many years, the steady increase of stylos at the expense of the grass component has become a management problem. With extensive property sizes, livestock management on native pastures is minimal, and continuous grazing or periodic spelling systems are practised.



## Stylosanthes dominance

Stylo pastures can withstand constant stocking because there is greater grazing pressure on young green grass leaf in the early wet season, allowing the legume to become better established, grow and seed. A grass–stylo pasture in northern areas of Australia, in particular, can also support two to four times higher stocking rates than native pastures alone, and this causes excessive pressure on the less grazing-tolerant grass component, eventually creating a stylo-dominant pasture. This change in botanical composition in native pastures after the introduction of stylos has been reviewed by Jones et al (1997). The opposite problem, grass dominance over stylos, has been a limitation to sown pasture performance in South America.

Initially, stylo pastures are easy to manage; however, maintaining a balanced grass–legume pasture under grazing requires strategic spelling in early summer to promote grass to compete with the legume, and management can require occasional burning to reduce the legume (Cooksley et al 2003). There may be long-term detrimental effects on the pasture and soil condition by this grazing-induced stylo dominance if left unchecked. Jones (2003) reported that native perennial grasses declined with increasing stocking rates and that they were virtually eliminated at high stocking rates, while sown sabi grass (*Urochloa mosambicensis*) persisted. It may be important therefore to include a grazing-tolerant grass when introducing these perennial stylos into native pasture to prevent subsequent stylo dominance.

Spelling alone can restore the native grass component when mixed grass–stylo pastures are grazed at low stocking rates, while at medium stocking rates burning is necessary and there is a further significant response to spelling. At high stocking rates spelling is ineffective in restoring desirable native perennial grasses (Cooksley et al 2003). These authors concluded that a 30–50% grass component could only be achieved with a burning program to reduce stylo competition, regardless of stocking rate.

The increase in populations of *S. seabrana* under continuous grazing on clay soils in subtropical Queensland (Hall 2000a) suggests this species may perform similarly to the more tropical stylos under Australian conditions.

Townsville stylo dominance over native grasses occurred naturally across northern Australia in the 1960s until anthracnose disease destroyed the populations. This sudden disappearance of stylo has created landscape degradation problems due to the very slow or negligible reintroduction of perennial grasses on some soil types. The granitic soils of inland Queensland are one example where perennial grasses have not recovered in over 25 years, and the annual grasses only provide limited soil surface erosion protection from early summer storms.

## Soil acidification

The strong adaptation of stylos under higher grazing pressure in Australian conditions has potential impacts on future landscape stability and productivity. Detrimental effects include: loss of soil surface stability, increased erosion, nutrient depletion, profile acidification and vegetation changes including weed invasion. Noble et al (2000) reported on strategies to reduce the negative impacts of stylo dominance and on sustaining productive legume pastures. The extent of acidification in northern Queensland has been assessed (Noble et al 2002), and soil buffering capacity was shown to affect the rate of acidification under a stylo pasture.

## Cattle Producer Experience with Stylo Pastures

Naturalised *S. humilis* across the dry tropics demonstrated the animal production benefits of a legume in tropical grazing systems. Prime condition bullocks could be produced at high stocking rates on this stylo on country that otherwise was suitable only for breeding store animals (Barrett 1997). With the disappearance of this species in the early 1970s, Seca and Verano became the most widely planted pasture legumes in the Australian tropics. In coastal Queensland these commercial stylo pastures have produced steer liveweight gains of 0.73 kg/head for 200 days through the dry season and periods of gain to 1.6 kg/head for short periods during the wet season (Barrett 1997).

The first commercial grazing trials with Verano and added superphosphate demonstrated improved animal production and increased stocking rates at 'Wrotham Park', northern Queensland. Periodic aerial applications of superphosphate, to 100 kg/ha, maintained the stylo pastures and up to a 10-fold increase in animal production, compared with native grass pastures (Arnold 1997). These pastures fluctuated from Verano dominant to annual grass dominant, eg *Digitaria ciliaris* (annual summer grass) and *Eragrostis* spp. (love grasses), as soil nitrogen levels fluctuated. Weed populations such as *Sida* spp. also fluctuated markedly. There were no native perennial grasses capable of tolerating the high grazing pressures applied to these pastures. When superphosphate applications declined, *Melaleuca* species trees re-established and competed with the stylos, allowing a more balanced, although less productive, stylo–grass pasture.

The commercial seeding of extensive areas in Australia involved mixing stylo seed and 30 t of superphosphate on the ground, and then aerially spreading the mixture onto recently cleared woodlands at rates of 1–3 kg/ha seed and 100 kg/ha superphosphate. Seca established in open eucalypt woodlands when spread aerially at low rates of 0.25 kg/ha seed with 25 kg/ha superphosphate. These pastures took several years longer to develop, but the stylo could still become dominant, due to increased stocking rates and reduced burning.

In trials on commercial properties across Queensland, superphosphate-fertilised stylo pastures have consistently produced liveweight advantages of 30–40 kg/head over native pastures, with a higher stocking rate and a higher sale price per kg because of the better condition of the animals (Anon. 1994). In stylo pastures with higher rates of phosphorus fertiliser and wet season phosphorus supplementation, a liveweight advantage over native pastures of 70 kg/head can be achieved on low fertility soils, while in the absence of phosphorus fertiliser the liveweight advantage from stylos was only 9 kg/head.

With the increase in live cattle exports from northern Australia, improving the nutrition of cattle by better quality pastures, which include stylos, is necessary to meet market requirements for well-grown, younger animals. Returns to producers from these markets are greater than from the traditional marketing of older and less well-conditioned cattle for the chilled manufacturing beef trade (Winter et al 1996).

Miller et al (1997) estimated that the 600,000 ha of grazing land in northern Australia sown to stylo pastures was expanding annually by some 50,000 ha, using approximately 100 t of predominantly Seca, Verano and Amiga seed. Prices of cattle, seed and superphosphate and good summer rainfall have a great influence on the rate of commercial sowing.

## Limitations of *Stylosanthes* Pastures for Animal Production

There has not always been a positive result in animal production from introducing stylos. Jones et al (2000) reported that there was no effect from legumes, including stylos, on liveweight gain during a ten-year period. This was largely attributed to poor rainfall, a low quantity of legume produced, and the fact that the site had enhanced soil nitrogen status from a previous productive legume pasture and applied nitrogen fertiliser.

The susceptibility of *Stylosanthes* to anthracnose (*C. gloeosporioides*) has been a major limitation of the genus and cultivar availability (Irwin et al 1984), as well as limiting the environmental adaptation and farmer adoption of stylos in animal production systems.

There are other biotic and abiotic constraints, ie poor seed production and an intolerance of heavy grazing, that limit stylo adoption in South America (Kelemu et al, this volume). When anthracnose-resistant stylo cultivars were developed, replacing Australian cultivars, these other biotic constraints limited their widespread adoption by farmers (Miles & Lascano 1997).

In South-East Asia and China factors limiting *Stylosanthes* adoption include: anthracnose damage, poor persistence of *S. guianensis* and low seed yields in the low latitude humid tropics (Guodao et al 1997). Cutting tolerance, an attribute not usually evaluated in Australia, is a necessary characteristic of a successful legume for the forage systems in this region.

There is increasing pressure on land resources in India and stylos have shown promise as a fodder in different management systems. They enrich soil nutrients and stabilise soil degradation. Anthracnose, head blight, virus diseases and insect pests threaten their potential role (Ramesh et al 1997). However, the availability of *S. seabrana* and promising new commercial utilisation schemes, including the use of stylo leaf meal in poultry rations, have generated a renewed interest in stylos in India (Ramesh et al, this volume) and other parts of Asia (Changjun et al, this volume; Guodao et al, this volume; Phaikaew et al, this volume).

## Soil fertility

The wide variability reported in animal production from stylo pastures is due to soil fertility, companion grass species, mineral deficiencies, seasonal rainfall amount and distribution, stocking rates and animal class. The increase in animal production by incorporating stylo compared with a grass-only pasture is strongly influenced by soil fertility, especially the phosphorus status. Stylos are efficient at extracting phosphorus (Probert 1984). Superphosphate, which provides phosphorus and sulphur, has been the main fertiliser required for optimum legume establishment and growth and cattle performance on stylo pastures in the dry tropics of Australia. For example, Winter et al (1989) suggest that the mediocre growth of cattle could be due to the low levels of some nutrients, particularly phosphorus and sulphur, in native grass–stylo pastures. Superphosphate fertiliser tends to promote higher liveweight gains of cattle during the wet season and reduces the losses during the dry season. Jones (2004) suggests nitrogen limitation on some tropical soils, even with a productive stylo pasture, may still be limiting animal production during the dry season. Nitrogen, available energy (digestibility) and phosphorus are the main nutritional limitations to cattle production from pastures on infertile tropical soils of Australia (Coates et al 1997).

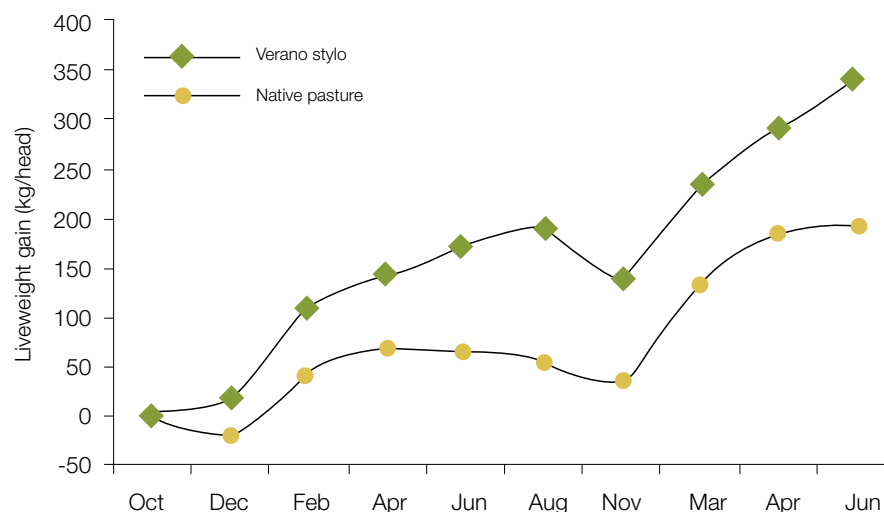
## Supplements on *Stylosanthes* pastures

The value of additional supplements to cattle grazing stylo pastures is dependent on soil fertility status and especially the phosphorus level in Australian soils. Seca and Verano can grow at low phosphorus levels of 3–5 ppm (bicarbonate extractable); however, both plant growth and animal production will be restricted. Supplemental phosphorus fed directly can help alleviate this problem. There is no extra animal production benefit from feeding a phosphorus supplement to cattle grazing stylo pastures on soils with a P level above 8–10 ppm (Coates et al 1997).

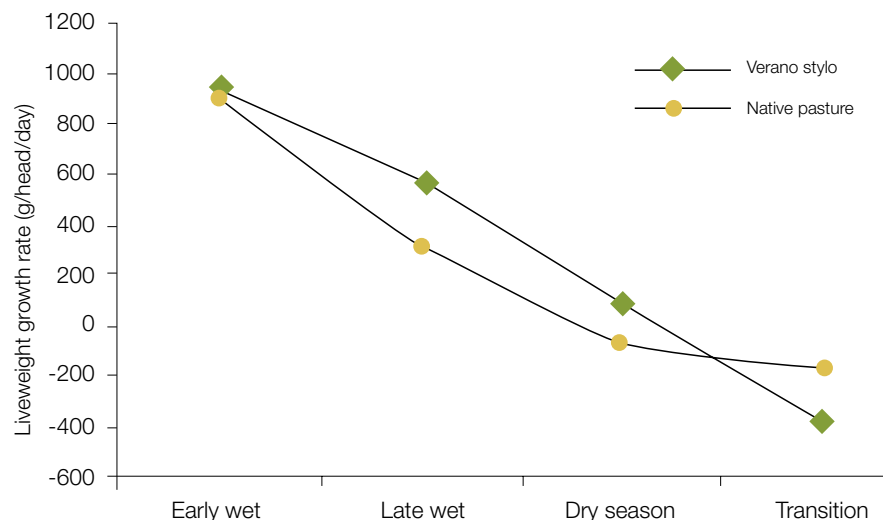
## Seasonal effects

The stylos in a pasture contribute to an extended weight gain period of several months after grasses have matured. These benefits improve the viability of cattle production by allowing access to higher priced markets for younger well-grown cattle. Reproductive performance is also enhanced, providing greater management and economic options to cattle producers.

**Figure 4.4** Seasonal liveweight change in steers grazing *Stylosanthes hamata* cv. Verano and native grass pastures in northern Australia (after Gillard et al 1980).



**Figure 4.5** Steer growth rates (g/head/day) on *Stylosanthes hamata* cv. Verano and native grass in northern Queensland at four annual growth phases (after Gardener et al 1993). Note – the growth phases are of varying duration (weeks to months).



An example of steer liveweight improvement over seasons from adding a stylo to native pastures in northern Australia is shown in Figure 4.4. The extended weight gain period from the stylo into the winter dry season (May–August) has the most significant effect. Late in the dry season (October–November) there can be rapid weight loss for a short transition period if dry matter intake becomes limited after early summer storms produce new green grass leaf.

Gardener et al (1993) quantified four seasonal periods of liveweight change in northern Australia, which show that the main advantage of stylo in a pasture over native pasture alone is in the late wet and dry seasons, or mid-summer through winter (Figure 4.5). These periods are of very different durations (weeks to months) and vary annually with rainfall patterns. There is little advantage in the early wet season when green grass leaf is most plentiful and actively selected.

### Diet selection

Seasonal diet selection studies show that the periods of greatest liveweight gain on stylo pasture relative to grass alone are when the diet has the highest proportion of stylo, such as in the late wet and dry seasons. The selection preference for green grass leaf over stylos in the early wet season has been reported consistently in Australia (eg Coates 1996; Gardener 1980; Hunter et al 1976). The strong perennial Seca is selected over a longer period than the short-lived perennial Verano (Coates 1996), providing an opportunity for a longer period of liveweight gain. The seasonal pattern of stylo selection is affected by rainfall amount and distribution, grass to stylo proportions and the associated grass species (Coates et al 1997).

## Conclusions and Future

Sowing *Stylosanthes* species has provided animal production benefits from low quality tropical grass pastures, and the genus has the potential to provide even greater improvements in tropical and subtropical environments around the world. In recent years the increased cattle production from adding stylos has become well accepted, although not well documented experimentally. The increasing role of animal products in many people's diets and the higher food quality demands of consumers provide pressure to improve animal growth rates by improving their year-round nutrition. Stylo has the capacity to help in these evolving animal management strategies.

There are *Stylosanthes* species and ecotypes within current well adapted cultivars in South America that have not been included in detailed environmental adaptation or grazing production studies in Australia or in other tropical countries around the world. The genetic variation within these species, including *S. capitata* and *S. macrocephala*, require evaluation of their adaptation across the tropics and subtropics. This could lead to additional legume species for the grazing industry and may expand the role of legumes in land management.

Another area requiring research is the commercial development and management of stylo pastures, to provide optimal integration into sustainable and viable grazing and farming production systems. Species such as *S. seabrana*, that are adapted to heavier textured soils, have potential for incorporation into farming systems on these better cropping soils. Well-adapted stylos may help provide quality forage to increase animal growth rates and produce the carcass quality demanded by consumers, improve soil fertility and crop production, and provide management options for maintaining landscape sustainability demanded by the wider community.

## Acknowledgments

We thank B. Grof and R.L. Clem for access to unpublished data, C.E. Lascano for assistance with South American information and R.J. Jones for comments on the review.

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## Chapter 5

# Utilisation of *Stylosanthes* as a forage crop in Asia

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### Summary

*Stylosanthes* species (stylo) are used in many ways in Asian farming systems. By smallholders stylo is often used as a fresh cut-and-carry fodder for ruminant and monogastric animals such as pigs, chicken and rabbits, and for feeding fish. Smallholder farmers have limited areas of land available for planting feed for animals and generally integrate forages with food crops and trees. This means that forages need to be cut rather than grazed to avoid damage to trees and crops by grazing animals. In China stylo is used mainly as a cover crop in fruit tree and other tree plantations, and then used as fresh feed for animals, cut and dried for leaf meal production or left uncut for seed. Stylo is also used as a cut-and-carry fodder. In India, where it is grown in different production systems including for environmental purposes such as wasteland development, soil conservation and mine rehabilitation. The most widely used species in Asia today are *S. hamata* Verano and *S. guianensis* CIAT 184, to a lesser extent Graham in China, and recently *S. seabrana* for leaf meal production in India. *Stylosanthes hamata* is used mainly in northeastern Thailand for cut-and-carry situations and inclusion in heavily grazed pastures. Graham and CIAT 184 are grown on more than 130,000 ha in southern China. In the last few years CIAT 184 has gained popularity in more countries in South-East Asia because of its broad adaptation, potential for multiple uses and high productivity in acid, infertile soils. *Stylosanthes seabrana* has gained popularity in India because of its adaptation to heavy soils and the ease of its seed production. Prospects for increased use of these species, particularly in smallholder farming systems, are excellent.

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### Introduction

*Stylosanthes* species have made a significant contribution to animal nutrition in the tropics and subtropics. It is used in a variety of production systems, eg supporting large commercial operations in Australia, or on small-scale farms in developing countries where resource-poor farmers use stylo as a feed for farm animals. The demand for meat and dairy products in Asia is growing rapidly. For instance, with a production of >1.4 billion broilers each year, the Indian poultry industry is growing at 10–15% per year. It is the fourth largest egg producer and the eighth largest broiler meat producer in the world and contributes 3% to India's GDP. Similarly, the dairy industry is growing at 4–5% each year. As a component of poultry and dairy rations, stylo can contribute to address the significant shortages in green and dry fodder in India and generate income for smallholder farmers.

Much of the research and development on stylo has focused on its use in extensive grazing systems (Stace & Edye 1984). Recent work by international forage networks has tried to redress this balance through the introduction of forages into smallholder production systems in Asia using farmer participatory research methods. This has resulted in an increase in the adoption of forage technologies by smallholder farmers, including the use of stylo (Stür et al 2002). The production of farmer-friendly publications on forage technologies and ways of working effectively with farmers has contributed to the uptake of forages (Horne & Stür 1999, 2003; Stür & Horne 2001). A paper summarising the use of stylo and the extent of seed production in China and South-East Asia has been published recently by Guodao et al (1997). This chapter updates information on the current usage of stylo in Asia with particular emphasis on the use of stylo in cut-and-carry systems.

## History of Introduction and Use of Stylo in South-East Asia

*Stylosanthes* spp. were first introduced to South-East Asia in 1949, generally following the development of commercial cultivars in Australia. *Stylosanthes guianensis* cultivars were introduced to humid and subhumid areas in Malaysia, Indonesia, southern Thailand, the Philippines, India and China; and *S. humilis*, *S. hamata* and *S. scabra* were introduced to drier areas in the region such as northeastern Thailand, eastern Indonesia and southern China. One of the main limitations of stylo has been the lack of resistance of cultivars to anthracnose. This disease severely reduced growth and survival of many cultivars; for example, an outbreak of anthracnose in *S. humilis* almost completely decimated this species in Thailand and prompted a switch to *S. hamata* Verano in 1976. *Stylosanthes guianensis* Schofield was later similarly affected in many countries and was replaced by Cook and Graham, which in turn were damaged by anthracnose some years later and have now largely been replaced by *S. guianensis* CIAT 184.

In India many different *Stylosanthes* spp. have been grown in agricultural systems as (a) pure legume pastures, (b) mixed legume/grass pastures, (c) mixed legume/cereal crops, (d) legumes with rangeland, (e) legumes for seed production and (f) legumes in short fallows, agroforestry and silvipastoral systems, for cut-and-carry fodder, grazing and cover crops. Details on the use of stylo in India have recently been published by Ramesh et al (1997). As in Thailand, *S. humilis* was decimated by anthracnose. Only *S. hamata* Verano and *S. scabra* Fitzroy and Seca continue to be cultivated, and seed of these cultivars is produced mainly by smallholder farmers. Multi-location trials with new *Stylosanthes* spp. showed the potential of *S. seabrana* for a range of agroclimatic zones. Smallholders in Hulkoti and Shurashettykoppa near Dharwad have accepted this species for cut-and-carry fodder and leaf meal production. It has also been used to address environmental issues such as wasteland development, soil conservation, wildlife sanctuaries and reclamations of mining land, especially of iron ore and mud slurry in the aluminium industry.

In China *S. humilis* was first introduced by the Chinese Academy of Tropical Agricultural Sciences (CATAS) in 1962. Subsequently, more species and varieties including *S. guianensis* Cook, Graham, CIAT 184, Mineirão; *S. hamata* Verano; *S. scabra* Seca, and most recently *S. seabrana* have been introduced from Australia and South America. Stylo is well adapted to many areas in southern China including Guangdong and Hainan Island. The main national research institution responsible for tropical forage germplasm collection and evaluation in China is the Tropical Pasture Research Center (TPRC) of CATAS. CATAS has introduced more than 1200 new forage accessions from South America, Africa and Australia, including a range of forage species introduced from CIAT. Five stylo cultivars have been released by CATAS originating from an evaluation of 170 accessions. *Stylosanthes guianensis* CIAT 184

has been outstanding and has yielded approximately 20% more than Graham. A new cultivar Reyan No 2 – Zhuhuacao, originating from CIAT 184, was released in 1987 and has almost completely replaced Graham in southern China (Devendra & Sere 1992). A major advantage of CIAT 184 is its higher tolerance to anthracnose, which severely damaged Schofield in 1982 and Cook in 1987. Since 1990 Graham has started showing signs of anthracnose infection and has been replaced by CIAT 184. Another *S. guianensis* cultivar Reyan No 5, also originating from CIAT 184, was also released. Other cultivars are *S. guianensis* Reyan No 7 (originating from CIAT 136), *S. guianensis* Reyan No 10 (from CIAT 1283) and *S. scabra* Seca (from Australia). Of these varieties Reyan No 2 (CIAT 184) is well adapted to conditions in tropical and subtropical area in China, with high forage yield (15–20 t/ha/year dry matter) and quality, and has been widely planted by smallholder farmers. It continues to be used in many provinces in southern and southwestern China such as Hainan, Guangdong, Guangxi, Fujian, Guizhou, Sichuan and Yunnan. The total area planted is over 130,000 ha, which has contributed significantly to animal production and the economy in these regions. Stylo has become the most important legume in forage production in South China, with a popular slogan: 'Alfalfa in the North and stylo in the South'. Some signs of anthracnose infection have been noted on CIAT 184 for several years but CIAT 184 based cultivars still show a high level of field resistance in China.

In Thailand more than 200 *Stylosanthes* species accessions were introduced and evaluated in the early 1970s. *Stylosanthes humilis* was initially selected and developed. Severe anthracnose damage forced a switch to *S. hamata* Verano (Hare 1993) and over 1400 t of Verano seed was produced from 1990–95, largely by smallholder farmers (Phaikaew 1997). Graham and CIAT 184 are other important varieties, and the superior performance of CIAT 184 has meant that the Department of Livestock Development (DLD) has only produced seed of CIAT 184 since 1996. Khon Kaen University has also been active in stylo research and has released an anthracnose-tolerant Khon Kaen stylo, originating from *S. humilis* CPI 61674, as a replacement for Townsville stylo. Although considered to be more grazing tolerant than Verano, its commercial use has been limited, with relatively minor seed production. In recent years cultivar development has progressively focused on composites rather than single varieties. New accessions have been introduced from CIAT, CSIRO and China during 1999–2002 and evaluated at Pakchong and Ubon Ratchathani University for more durable tolerance to anthracnose and higher seed yield. Two composites selected by Bert Grof, ATF 3308 and ATF 3309, and a black-seeded CATAS selection from CIAT 184 are among potential replacements for CIAT 184 (Phaikaew et al 2002). ATF 3308 was more productive and had a higher seed yield than ATF 3309 and CIAT 184 in experiments in northeastern Thailand. Graham was completely destroyed by anthracnose in the same experiment (Ganda Nakamane, pers. comm.).

In the Philippines the Bureau of Animal Industry (BAI) introduced stylo varieties from Australia and produced about 5 t/year of Schofield seed on government stations in Masbate and Bohol in the late 1970s (Guodao et al 1997). This seed was used for oversowing native pastures and with introduced grasses such as *Brachiaria decumbens*. Following the anthracnose mediated demise of Schofield in the early 1980s, Cook and Seca were introduced in 1983 and, since 1984, 500 kg/year of Cook seed produced by BAI has been used for sowing on government stations. The Southeast Asian Regional Forage Seeds Project and its successor, the Forages for Smallholder Project, introduced CIAT 184 in 1992. It showed excellent growth in many parts of the Philippines (Lanting et al 1995) and in other countries in the region (Stür et al 1995). Although seeds of both Cook and CIAT 184 were produced at Isabela and Quirino in northern Philippines in 1995, Cook was severely damaged by anthracnose at both sites, while CIAT 184 only had minor infection. Consequently, CIAT 184 is used more widely while Cook is mostly used in Masbate and Bohol, where it does not suffer from severe anthracnose.

In Indonesia *S. humilis* Verano, Schofield and Cook were introduced to South Sulawesi in the early 1970s and other introductions were targeted for Java and eastern Indonesia. Before it was severely damaged by anthracnose in the early 1980s, Schofield was used to suppress the serious weed *Imperata cylindrical* in southern Sumatra. Sporadic seed production and evaluation of Verano, Seca, Cook, Graham and CIAT 184 have occurred in the 1990s (Tuhulele et al 1995). Only CIAT 184 is currently grown by smallholders, and small quantities of seed are produced on government stations.

In Malaysia Schofield was first introduced from Australia in 1949 (Vivian 1959), covering some 240 ha in Kelantan, but it was vegetatively propagated using stem cuttings due to poor seed set in this wet tropical environment. Although several studies have demonstrated its usefulness, there has been little commercial use of stylo in Malaysia. *Stylosanthes guianensis* was identified as the most promising legume for Malaysia (Wong 1982; Wong et al 1982); CIAT 184 was found as the most productive and persistent legume in rubber plantations (Ng et al 1997); and nearly 500 kg of CIAT 184 were produced for distribution to farmers (Chen et al 1995).

### Thailand: current utilisation including as a cut-and-carry fodder

The demand for forages in Thailand has been driven to some extent by government programs promoting livestock production. Although emphasis has been generally on grasses, large quantities of Verano stylo and CIAT 184 are produced every year. Verano has been a key cultivar for Thailand. During 1976–90 it was used for sowing communal grazing areas such as natural grassland, roadsides, paddy bunds, forest land and other non-cropping areas. Since that time, Verano has been used mainly for sowing in association with grasses such as *Brachiaria ruziziensis* for cut-and-carry systems and grazing by cattle

Several studies have demonstrated the usefulness of Verano and CIAT 184 as cut-and-carry fodders for animals. Studies on dry matter production under different environments and cutting regimes (Table 5.1)

**Table 5.1 Dry matter yield (DMY) of *Stylosanthes hamata* Verano and *Stylosanthes guianensis* CIAT 184 under different cutting intervals at various locations in Thailand.**

<i>Stylosanthes hamata</i> Verano				
Site	Rainfall (mm/year)	DMY (t/ha/year)	Cutting frequency	References
Chiang Yuen (Maha Sarakham) Upland infertile sand	934	7.6–10.8	30–75 days	Phaikaew et al 1984
		9.7–10.0	45–60 days	Jantharasiri et al 1988
Pak Chong (Nakhon Ratchasima) Upland fertile silty clay	1100	10.9–11.9	45-days at 1–5 inch cutting height	Thinnakorn & Wittayanupapuyenyong 1992
Chai Nat Upland fertile clay	1300	9.8	45 days	Punyavirocha et al 1992
		7.4	30 days	Nakamaneet et al 1994
Narathiwat Upland infertile sand	3200	1.2–4.1	2 cuts	Maneedool et al 1986
Khon Kaen Upland infertile sandy loam	1100	8.0	45 days (3 cuts)	Topark-Ngarm 1980
		5.3	45 days (1 cut)	Wilaipon et al 1982; Wilaipon & Wilaipon 1982
		7.5	1 month after flowering	
		3.9	45 days (2 cuts)	
<i>Stylosanthes guianensis</i> CIAT 184				
Narathiwat Lowland infertile sand	3200	8.6, 9.7 and 10.2	Spacing 25x50 cm, 40, 50 and 60 days (8, 7 and 6 cuts)	Sukkasem et al 2003
		9.1, 9.1 and 10.3	Spacing 25x50 cm; cutting height 10, 15 and 20 cm	

**Table 5.2 Chemical composition of *Stylosanthes hamata* Verano under different cutting intervals.**

Cutting interval (days)	Chemical composition							Reference
	CP	CF	EE	Ash	NFE	NDF	ADF	
30	20.0	28.2	2.2	10.3	39.2	48.5	38.9	Phaikaew et al 1984
45	18.1	31.4	2.5	9.1	38.9	52.8	42.6	Phaikaew et al 1984
60	15.2	35.2	2.3	8.0	39.3	57.9	48.1	Phaikaew et al 1984
75	13.8	40.6	2.0	6.3	37.3	55.1	44.6	Phaikaew et al 1984
45	17.0	28.8	2.6	6.9	44.7	46.2	36.5	Jantarasiri et al 1988
60	16.0	29.0	2.4	6.1	46.6	43.4	35.5	Jantarasiri et al 1988
71	12.9	30.9	1.8	9.1	35.2	51.3	47.3	Snitwong et al 1979
90	15.4	25.3	2.2	10.8	34.8	43.3	39.2	Snitwong et al 1979
120	12.2	28.5	2.1	7.9	38.9	49.1	43.8	Snitwong et al 1979
Blooming	14.2	31.3	1.3	8.5	44.8	57.8	41.5	Wilaipon et al 1982
Post bloom	12.4	35.7	1.4	7.0	43.5	57.5	45.5	Wilaipon et al 1982

(CP, crude protein; CF, crude fibre; EE, ether extract; NFE, nitrogen-free extract; NDF, detergent fibre; ADF, acid detergent fibre)



**Cut-and-carry Verano stylo used in stall-feeding of cattle in Thailand.**

have been used to develop optimum cutting management techniques to maximise fodder yield. Other work has shown the tremendous yield potential with different cutting intervals and the high nutritive value of Verano (Tables 5.1 and 5.2). Verano can be cut many times and forage yield is often higher in the second year after establishment. The annual dry matter yield is about 8–10 t/ha/year, with high yields during the rainy season. The recommended cutting interval is every 45–60 days in the rainy season and 75 days in the dry season, at a cutting height of 15–20 cm above ground level. However, crude protein content is highest when cut young, ie every 30 days (Table 5.2). The decrease in

crude protein content with longer cutting intervals is associated with a corresponding decrease in leaf:stem ratio. In Verano leaf:stem ratio gradually increases from 1:1.7 to 1: 2.7 when cutting interval is increased from 30 to 75 days, respectively (Phaikaew et al 1984).

With 16% protein in the dry matter, stylo supplies a gross energy of more than 100,000 MJ per ha, which is two to three times higher than rice and 65–69% higher than rice–corn (Kexian 2002). In addition to its use as a fresh feed, stylo is successfully used in China and India as hay, hay cake and leaf meal powder for concentrate, often as a substitute for grains in animal feed. In the future DLD envisages the production of stylo leaf meal.

### **China: current utilisation**

Kexian (2002) has recently outlined the three main ways that stylo has been used very successfully in China.

**(a) In natural grassland or sown pastures for grazing animals** The Xishui Cattle Farm, Baisha County in Hainan province is a good example, where 2433 ha have been improved by sowing a mixed pasture of stylo and *Brachiaria*. This has improved yield and stocking rate of these pastures by three to four times that of native pasture. At the same time this has increased crude protein in forage dry matter from 6% to 12.2%; animal weight gain/day/head from 238g to 359g, sometimes up to 500–600g; calf birth rate from 63% to 93%; and calf survival rate from 85% to 98%; and has reduced the cattle raising period from birth to market from 3–4 years to 1.5–2 years (China National Agriculture Museum 2000).

**(b) As high quality hay and leaf meal** Hainan province has taken the lead in establishing two factories for the commercial production of stylo hay and leaf meal. In 1996 a stylo leaf meal production factory using a commercial quick drying technology at high temperature (600°C) was built by the Santian Agriculture Development Company in Lingshui





A commercial stylo leaf meal factory in Lingshui county in Hainan, China; (inset) the leaf meal (photo: Bai Changjun).

county. Because of the large potential market, another larger factory was built in 1999 in Changjiang county. When fully operational, the combined production of the two factories will be 15,000 t per year. Using this rapid drying process, the current cost of planting, harvesting, processing and transporting one tonne of stylo leaf meal is about 1000 Yuan, and the sale price is 1400 Yuan/t in the domestic market. This generates a 400-Yuan profit (\$US50) per tonne.

The nutritional quality of stylo leaf can be somewhat low if rain and other spoilage occur during sun drying of the cuttings. However, the protein content of the meal is still > 10% and can be readily used to partially replace grains in compound or mixed feeds. Experiments conducted by CATAS show that the optimum stylo leaf meal content of various feed formulations are: pig, 5–10%; broiler, 2–5%; duck, 8–12%; goose, 15–20%; rabbit, 30–40%; cattle, goat and deer, 40–60% (Guodao 2000). Stylo leaf meal in broiler feed can significantly improve meat quality, avoid feather pecking and improve yolk colour.

**(c) As a cover crop** Stylo has been widely used as an intercrop in fruit plantations and young tree farms for ground cover, green manure and forage in recent years. This intercropping system increases land use efficiency and productivity. By providing income from the forage crop, benefit can flow from the first year of planting, overcoming the shortcomings of a longer investment horizon in tree and plantation farming. Its perennial habit enables income generation over several years, and the relatively low technology input necessary for its growth and

management makes it an attractive proposition for local government, private enterprise and farmers alike.

China has an estimated 67 million ha upland area, 13.3 million ha coastal area and 6.66 million ha crop farming area, all of which are suitable for forage production. New varieties with high yield and protein and tolerance of cool temperatures, combined with better harvesting and drying technology, will help expand the use of stylo to many of the underdeveloped areas, improve its quality and lower the processing costs. If smallholder farmers are to lead the way, more attention must be given by government and non-government agencies to the financial and other needs of these resource-poor groups.

### India: use in different production systems

In India natural pastures do not meet the nutritional needs of grazing ruminants, and supplementary feeding of crop residues and concentrates, including commercial feed formulations, is necessary for profitable animal production. Often most of the liveweight gained in the wet season is lost in the dry season, resulting in low net annual growth. An example is the western Ghat region of North Canara District of Karnataka, where the soil is extremely infertile and devoid of any legumes. Animals suffer from calcium deficiency, leading to abnormal molar development which hampers jaw movement. This leads to feed refusal, often resulting in death from starvation.

Although several species of *Stylosanthes* have been introduced to India over the years (Ramesh et al 1997), so far Verano and a locally adapted *S. scabra*, possibly a derivative of the cultivar Fitzroy, are most widespread. In addition, several accessions of the newly described *S. seabrana* from Australia are rapidly expanding in peninsular India. Stylo has a long history of research and development in India and has been used in many production systems, including as a fodder crop, in ley farming and intercropping, in agroforestry and silvipasture, and in wasteland development and soil conservation. These have been described in more detail by Ramesh et al (1997). This section summarises stylo development from personal observations of C.R. Ramesh during extensive travel in India over the last six years.

**Stylo as a fodder crop** There is a significant amount of information on all aspects of stylo research and development in India, largely based on work at experimental stations of government agencies such as the Indian Grassland and Fodder Research Institute and State Government Departments of Animal Husbandry (Ramesh et al 1997). Recent developments in the commercial utilisation of stylo as a fodder crop have mostly been in the peninsular Indian states of Maharashtra, Karnataka, Tamil Nadu, Andhra Pradesh and Kerala. These include a private farm in Tamil Nadu that has developed significant areas of *S. seabrana* to raise Mecheri goats; and the excellent results from large-scale goat feeding trials with two *S. seabrana* and two *S. scabra* accessions used as cut-and-carry fodder at the Nimbkar Agricultural Research Institute near Pune in Maharashtra. Near Coimbatore in Tamil Nadu nearly 100 acres of stylo pastures have been developed for sheep and goats by livestock owners on individual farms, 2–10 acres each. While sheep are allowed to graze, goats are stall fed using cut-and-carry fodder comprising stylo, *Brachiaria* and *Pennisetum* hybrids.



A commercial goat farm in Tamil Nadu in India based on *Stylosanthes seabrana* (photo: C.R. Ramesh).

**Stylo in silvipasture** There has been a resurgence in interest on stylo as a multipurpose legume and a renewed optimism at the grass root level for stylo usage in plantation forestry. This is combined with enthusiasm for improvements in soil and water resources to increase forage, fuel and timber production, and to generate additional income for villagers. Consequently, active participation by villagers in the planning and development process and sharing of income have been key factors. This has been matched by a strong renewed interest from federal and state government departments and, in particular, non-government organisations. At many sites there has been a spectacular transformation of arid and degraded landscapes. Salboni in West Bengal is one such spectacular example. The area has a red lateritic acid (pH 5.4) soil with average fertility in a subtropical climate. Here 1800 of the total 2500 acres available have been planted to native tree species such as *Dalbergia* and introduced *Eucalyptus* with a healthy undergrowth of grass and legumes including *S. guianensis*, *S. scabra* and *S. hamata*. In this government-sponsored scheme villagers receive a proportion of all produce from the land and have managed access to fuel wood and cut-and-carry fodder.

Some members of the paper pulp industry have used the technology of growing *S. hamata* in *Eucalyptus mangium* plantations to improve soil fertility and provide additional income through the sale of fodder and seeds. At a site near Dharwad, over 200 out of a total area of 800 acres have already been established with Verano.

**Stylo in watershed and soil conservation** In the past few years the biggest utilisation of stylo has been by the watershed/wasteland schemes, which are the largest buyers of commercially produced seed in India. In the past these developments were largely government funded and often did not last once the areas were opened for communal grazing. Recent developments include many partnerships between cooperatives of smallholder farmers and non-government organisations (NGOs). Often, because the watershed forms part of the land owned by villagers themselves, along with areas of crown land, the commercial interest of farmers has made these schemes successful.

As an NGO, the Watershed Organization Trust (WOTR) is at the forefront of watershed development in India using stylo to stabilise contour bunds and as a pioneering species. Since 1992 WOTR has worked with over 55 voluntary organisations and the watersheds have covered some 100,000 ha in Maharashtra alone. At one of its sites in 'Pimpalgaon Wagha' there has been a spectacular transformation of 840 ha of an arid and degraded landscape following successful implementation of such a watershed development program between 1989 and 1994, benefiting a population of about 880 people. The impact of the program can be felt in the availability of drinking water all year round, replacement of scrub cattle almost entirely by cross-bred cattle, increased milk production from 150 L/day to 1400 L/day, and a doubling of income through increased grain and oilseed production.





**Rural women near Hyderabad in India cleaning stylo seeds grown by the women labourers on wastelands. Seeds are sold to government authorities for sowing other wastelands (photo: S. Chakraborty).**

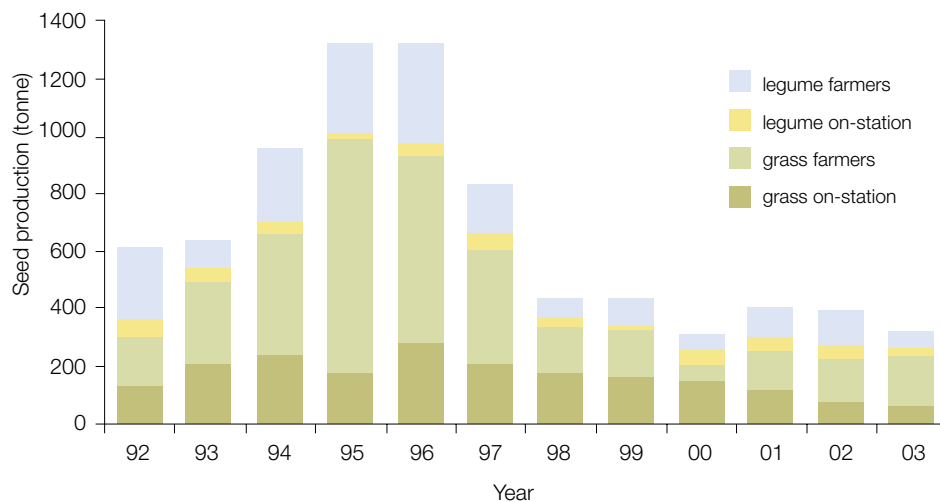
Established in 1967, the Bharatiya Agro Industries Foundation (BAIF) is one of the oldest NGOs in India with a program covering a million families in 8000 villages. At one of its serviced villages 'Manjunathpura', a 70 ha area has been improved through water harvesting, agroforestry and mixed cropping in a region of 600 mm annual rainfall, mostly received within a few days, often over 100 mm/day. To encourage soil organic matter build-up, each property/land parcel has been planted with a mixture of trees such as *Dalbergia latifolia*, *Sesbania sesban*, *Melia azedarach*, *Calliandra calothyrsus*, *Eucalyptus camaldulensis*, *Gliricidia sepium* and *Leucaena leucocephala*, and shrubs such as *Acacia rugata*, *Agave sisalana*, *Agave vera-cruz*, *Caesalpinia crista*, *Erythrina suberosa*, *Jatropha cascus* and *Parkinsonia odoratissimus* along its border, and with legumes including stylo for soil enrichment. The 'Surashetty Kopa' cluster of villages in Karnataka is one of the best examples of stylo significantly improving smallholder income and wellbeing through a BAIF-mediated participatory program. Started in 1998, these villages, covering only 42 acres of near barren land, have used *S. scabra*, *S. hamata* and more recently *S. seabrana* to improve soil fertility and provide income through seed sales. Some farmers in this area have intensified livestock production using forage crops including stylo as the main feed resource, enabling them to cope better with droughts rather than relying solely on cropping.

Government funded initiatives have continued to improve large tracts of wasteland using stylo as the pioneering species. A program of the Hyderabad Urban Development Authority (HUDA), spanning several thousand hectares near Hyderabad, has combined wasteland development with income generation for rural women from selling stylo seeds. The women are paid as labourers for all farming operations and in turn sell stylo seed to HUDA for further sowing in wastelands.

### Seed production of stylo

Commercial seed production of *Stylosanthes* species is carried out mainly in China, India and Thailand, although small quantities of CIAT 184 seed have been produced in northern Malaysia and northern Philippines in the last few years. Likewise, small amounts of CIAT 184 are produced on government stations in Indonesia for on-farm testing.

In Thailand more than 3700 t of legume seed, mostly Verano, was produced during 1976–95. Production declined from a high of 430 t/year in 1989 to 150 t/year in 1995. During 1972–75 Khon Kaen University undertook most of the *S. humilis* seed production. The best treatments yielded between 1420 and 1850 kg/ha (Wickham et al 1977). Encouraged by these yields, a pilot seed production project was established with seven farmers in 1975 (Hare 1993; Wickham et al 1977) who hand harvested 1831 kg of clean seed in early 1976 from only 4 ha. These results showed that northeastern Thailand was well suited to large-scale production of *S. humilis* seed. Although the impact of anthracnose in late 1976 prevented any further development of *S. humilis*, Verano imported from Australia in 1976 by DLD formed the basis of the seed industry (Phaikaew 1994). Starting with five farmers producing 500 kg in 1978 at an average yield of 790 kg/ha, the industry has expanded rapidly and, in 1981, 187 t of seed was produced by 1131 farmers at an average yield of 910 kg/ha (Hare & Phaikaew 1998).



**Figure 5.1** Forage grass and legume seed produced by farmers and forage stations in Thailand during 1992–2002 under government quota.

**Table 5.3 Stylo seed (tons) bought by the Thai Government from DLD forage stations and farmers in Thailand between 1994 and 2003.**

	Verano stylo		Stylo CIAT 184	
	Station	Farmer	Station	Farmer
1994	4	285	0	0
1995	24	125	0	0
1996	18	179	0	0
1997	7	56	0	0
1998	4	58	1	1
1999	2	45	2	7
2000	3	49	4	15
2001	23	37	8	25
2002	2	34	6.4	9.6
2003	1	29	10	12
2004	2	37	8	18

Seed purchased by the Thai government varies from year to year depending on demand by various government projects. The amount of forage seed (including grasses and other legumes) purchased by the Thai government has decreased since 1996 (Figure 5.1), due to decreasing demand from government projects and the government policy to gradually transfer the production and marketing role to the private sector (Phaikaew & Stür 1998).

The main region for producing stylo seed is in northeastern Thailand: Khon Kaen, Buriram, Mahasarakham, Sakonnakorn and Udonthani Provinces. The amount of stylo seed purchased between 1994 and 2003 is shown in Table 5.3. Farmer seed production of CIAT 184 started in 1997. The amount of stylo seed produced was high in 2002 because of a high demand by the government's Fodder Production and Marketing Encouragement Project. This project supports 200 farmer groups (more than 6000 farmers in 32 provinces) to cultivate pasture grasses and legumes for sale. The amount of seed sold directly from farmer to farmer or from farmer to trader is not known. It is estimated that seed producers sold approximately 2–3 t of CIAT 184 and 8–9 t of Verano stylo in 2002 and 2003. The proportion of direct selling is expected to increase with

time. The quality of stylo seed produced is high, with purity of 80–90% for both CIAT 184 and Verano, and a germination percentage of 95% for CIAT 184 and 60–70% for Verano. In 2003 mean farmer seed yields were 260 kg/ha for CIAT 184 and 460 kg/ha for Verano, with many farmers achieving much higher yields. Seed yields of up to 1.4 t/ha of CIAT 184 were recorded in experiments (Table 5.4). In 2003 pasture seed producing farmer groups from many regions of Thailand formed the 'Thai club of pasture seed producers'. This group will develop plans for seed production each year, including the amount of seed produced, the kind of grass and legume seed needed, and marketing of the seed. For the first few years, the government (DLD) will help the group in: control of seed quality through field inspection and provision of laboratory seed testing; seed marketing within the country and for export; and seed packaging and storage. More details of the seed industry in Thailand have been described by Guodao et al (1997) and Phaikaew and Hare (1998).

In India commercial seed of *S. scabra* and *S. hamata* have been produced since the late 1970s, when an estimated 4 ha area was used for seed production in Anantpur in Andhra Pradesh. Seed of *S. seabrana* has been grown commercially only in the last few years near Dharwad



***Stylosanthes guianensis* as an understorey in commercial Eucalyptus plantations in China offers regular income from fodder sales (photo: Ian Willett).**

**Table 5.4 Pure seed yield of Thapra (CIAT 184) stylo at various locations in Thailand.**

Site	Topography & soil type	Annual Rainfall (mm)	Pure seed yield (t/ha)	Treatments	Reference
Khon Kaen	Upland; infertile sandy loam	1100	1.1 and 1.1	Cutting height 20 and 30 cm	Keowthong et al 2002
			0.6, 1.0, 1.4 and 1.4	Cutting 60, 75 and 90-day intervals before 50% flowering and uncut	
Buri Ram	Grey podsolic soil	1057	0.4, 0.6 and 0.7	Cutting height 20, 30 and 40 cm	Sisomporn, pers. comm. 2002
			0.7, 0.7 and 0.3	Cutting at 90, 120 and 150 days after planting	

and Palladam Tamil Nadu. An estimated area of 450–500 ha was under stylo seed production in the 2000–01 season. Commercial seed crops are either left ungrazed, or the crop is grazed from June to August before being closed, when the seeds are allowed to form and mature; the second method is particularly suitable for Verano. Weed management is very important in stylo seed crops. Some growers manage their seed crop as annuals, planting from seed each year. Despite the simple technology, seed yields in farmers' fields for Verano vary from 0.8–1.2 t/ha in dryland conditions to 1.2–1.4 t/ha in irrigated conditions, and compare favorably to the 1.5 t/ha achieved at government experimental stations that use the latest technology (C.R. Ramesh, unpublished information).

In tropical China seed production centres on CIAT 184 and Verano, with smaller quantities of Seca and Graham. Seed crops are managed as annual crops; seedlings are raised like rice in seedbeds, transplanted into the field (or orchard), weeded regularly, fertilised and left to mature. Plants are cut at ground level and threshed, and seed is picked up from the ground for cleaning. Details on the seed industry, management practices and production trends have been described by Guodao et al (1997). Seed yield varies with cultivars: Verano has a mean yield of 390 kg/ha (ranging 240–1020 kg/ha); followed by CIAT 184 with 290 kg/ha (75–520 kg/ha); Seca with 280 kg/ha (100–520 kg/ha); and Graham with the lowest yield of 250 kg/ha (40–470 kg/ha) (Guodao et al 1995).

## Constraints and Future Prospects

The threat of serious anthracnose damage, the poor persistence of *S. guianensis* varieties when cut rather than grazed and the low seed yields at low latitudes are the main factors that have limited more widespread use of stylo.

Anthracnose has damaged several promising *S. guianensis* cultivars like Schofield and Cook. Although a very worthy replacement for the anthracnose-affected cultivars was found in CIAT 184, this variety is



*Stylosanthes seabrana* planted in a coconut plantation in Tamil Nadu in India (photo: C.R. Ramesh).



Windmill farms in Tamil Nadu in India with potential for stylo planting for commercial use (photo: S. Chakraborty).

currently the 'last' line of defence for countries such as China and Thailand. While it is impossible to predict if and when the currently observed tolerance of CIAT 184 to anthracnose will break, it is important to have replacements ready. Any new cultivar must have durable resistance or tolerance to anthracnose as frequent replacement of cultivars is expensive and impractical for a forage species. Two composite varieties with broad resistance to anthracnose have been developed by Bert Grof and colleagues at CIAT and EMPRAPA, and seed has been multiplied by DPI Australia. In Australia the two composites are registered as ATF 3308 and ATF 3309, with ATF 3308 being a 'grazing' type while ATF 3309 is more upright and later flowering. Both of these composites and some of their 'ingredient' accessions are under evaluation in China and Thailand, with promising early results.

In China and South-East Asia *S. guianensis* is used increasingly for leaf meal production, fallow improvement, cover crops and as a fresh cut-and-carry feed for both ruminants and non-ruminants. Many smallholder farmers are feeding freshly cut stylo to pigs, chicken, ducks and fish. In these systems, stylo tends to behave as an annual or semi-annual plant and, although long-term persistence is not usually required, regrowth of *S. guianensis* is often poor after cutting, particularly when cut at a mature stage for leaf meal and seed production.

Low seed yield, particularly in the humid tropics near the equator, has been a major limitation for more widespread use of stylo. Often, smallholder farmers prefer to produce their own seed (or propagate from cuttings) and any species that cannot easily be propagated has a reduced chance of adoption.

Despite these limitations, the use of stylo in agricultural systems in South-East Asia and China is expanding. *Stylosanthes guianensis* continues to be the most productive and broadly adapted legume for the acid infertile soils of humid and subhumid Asia. In recent years CIAT 184 has been grown increasingly by smallholders feeding stylo to monogastric animals but the question of seed supply for these farmers has not yet been resolved satisfactorily. Its ability to suppress weeds has been demonstrated by its use as a cover crop in tree plantations. There are good prospects for using it also to suppress unwanted grasses such as *Imperata cylindrica* in rehabilitation of degraded grasslands. The use of CIAT 184 for leaf meal production in tropical China has great potential for other countries in the region and is under investigation in several countries. For example, research in Thailand is directed to developing pelleting techniques for legume leaf meal, which might open new markets for stylo.

Similarly, Verano stylo continues to be used widely in the seasonally dry tropics of South-East Asia, particularly in Thailand where it continues to be the most important forage legume.

With 15% of the global livestock population supported on only 2% of the world's land area, India has a formidable task in providing adequate forage resources. In addition to grazing pressure, the land resource is exposed to a burgeoning human population and attendant urbanisation. As a result, over 50% of the total area of 329 million ha has been degraded to wasteland through loss of productive vegetation and increased soil erosion.

At the same time, ever increasing demands for food, fodder and fuel wood are being placed on the reduced area of productive land. The estimated requirement of concentrates and green and dry fodder to support livestock population is far greater than what the land resource can sustain using current technology. Stylo has the potential to significantly contribute to this fodder shortage through its multipurpose use in addition to more traditional usage, including citrus, coconut and mango orchards in humid and subhumid areas, mainly in Tamil Nadu and as roadside plantings.

The excellent adaptation of the highly productive *S. seabrana* to a wide range of environments in India (see Ramesh et al, this volume) without the need to apply specific *Bradyrhizobium* strains is going to further enhance the use of stylo. Several thousand hectares in Tamil Nadu is under windmill farming. Private entrepreneurs who own the land are keen to establish stylo and grass pastures as a source of additional income. Perhaps the biggest development for India has been the demonstration of the suitability of stylo leaf meal as a cost-effective addition to poultry rations (see Guodao et al, this volume).

## Conclusions

*Stylosanthes guianensis*, *S. seabrana* and *S. hamata* are likely to be used increasingly in agricultural systems in Asia. Despite the short-lived nature of *S. guianensis* when used in a cut-and-carry situation, farmers in the more humid areas of the region are increasingly using CIAT 184. Alternative composite varieties are available but still need to be tested more widely and 'geared up' to be available as alternative and potential replacements for CIAT 184.



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## Chapter 6

# The role of *Stylosanthes* spp. in mixed crop–livestock systems in Africa and Australia

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### Summary

A range of *Stylosanthes* spp. (stylo) have been evaluated for their role in providing nitrogen in cropping systems. Their use has ranged from short-term legume fallows to situations where stylos are used as longer-term pastures in delivering improved forages through fodder bank technology. Both *S. hamata* and *S. guianensis* provide significant N accumulation to a range of subsequent cereal crops such as maize, millet and upland rice on acid, light textured soils on smallholder systems in subhumid West Africa. The nitrogen delivered to the subsequent crop has often been in excess of the equivalent of a fertiliser application of 30–40 kg N/ha. These same species have been evaluated in interrow cropping systems in the same regions; the experimental results have demonstrated that any benefits from such systems are highly dependent on managing the competition between cereal crop and stylo. The use of *S. hamata* in semi-arid cropping systems in northern Australia also benefits subsequent cereal crops and there is some evidence that Caatinga stylo (*S. seabrana*) can play a role in overcoming soil N decline on heavy textured clay soils in subtropical eastern Australia. Despite the potential benefits of using stylos in smallholder and commercial scale cropping systems, there has not been widespread adoption because of a range of socioeconomic factors. However, increases in the demand for cereals in West Africa may encourage future adoption, especially if that demand is accompanied by expected increased demand for animal products.

### Introduction

Mixed crop–livestock enterprises are the basis of many agricultural production systems throughout the tropics. They are practised in a wide range of climatic and edaphic environments and are extremely variable in their operation, the crops and animals grown and the products sold. The majority of sheep, goats and cattle in the tropics are grown in mixed systems (Gardiner & Devendra 1995) where crop and livestock production are closely integrated. This integration may include the use of crop residues as a source of animal feed during the dry season, intercropping or undersowing with various forage and dual purpose legumes, and crops and forages grown in rotation systems of various types.

In the tropics and subtropics mixed crop–livestock farming systems are being influenced by a range of external factors including climate change, increasing population density, shifting demographics and changes in household income, and parallel changes in demand for livestock products. In West Africa the rapid changes in human and livestock populations over the past 20 years have seen the traditional shifting agriculture and long natural fallow periods give way to more intensive agricultural systems and major changes in land use and environment (Tarawali et al 1999). The population of sub-Saharan Africa is expected to grow from 0.65 billion in 2000 to 1.1 billion by 2025, with the population of urban dwellers across Africa changing from 37% to 50% in that time (Anon. 2004). The growing population of urban dwellers will undoubtedly lead to a greater demand for food from the rural sector and inevitably more intensive use of resources, especially land. The past decade has also seen a rapid increase in livestock product consumption and this is predicted to continue. It has been estimated that milk consumption across the tropics will increase by about 3.2% per annum until the year 2020 (Delgado et al 1999) and beef

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consumption is expected to double in developing countries between 1993 and 2020. The mixed crop–livestock farming systems of the tropics and subtropics will be most affected by this increase in demand for livestock products as more than 85% of the world's cattle, sheep and goats are held in these mixed systems in the tropics (Gardiner & Devendra 1995).

In most regions where continuous cropping is used, reduced soil fertility is now a major constraint to grain production. With over 50 years of cultivation in the northern grain belt of Australia, on what were once highly fertile soils, soil N has declined to a level where premium wheat protein levels are no longer attainable without substantial N inputs (Dalal et al 1991). Similarly, in sub-Saharan Africa, long periods of cultivation have resulted in serious reductions in soil organic matter and soil N (Tarawali et al 1999). This decline plus the expansion of cropping onto less fertile soils and the inability to maintain long-fallow practices has led to an expanded soil fertility research program in Australia and sub-Saharan Africa over the past 20 years. Of particular importance is the program of research into the potential role of forage legumes in mixed cropping systems, to enhance both livestock and crop production.

A wide range of legumes have been tested for use in mixed crop–livestock systems and a much smaller number have been widely adopted. *Mucuna* (*Mucuna pruriens*) had been adopted by about 10,000 farmers in Benin by 1996 (Tarawali et al 1999), and *Stylosanthes hamata* has been widely adopted by a total of 27,000 adopters for use as fodder banks by livestock producers in West Africa (Elbasha et al 1999). Some of the crop–livestock producers have taken advantage of the improved soil fertility in these fodder bank lands and subsequently grown cereal crops (Tarawali et al 1998).

In Australia, Butterfly pea (*Clitoria ternatea*) has been adopted by >100 mixed crop–livestock producers in central Queensland, with the total area occupied by the legume increasing from almost none in 1995 to about 100,000 ha currently (M. Conway, pers. comm.). Lablab (*Lablab purpureus*) has also become a widely used tropical annual ley legume throughout the northeastern Australian cropping zone.

While legumes such as lablab and butterfly pea are used predominantly on heavy textured soils, the genus *Stylosanthes* has attracted a significant research focus in West Africa and northern Australia because of its known adaptation to low fertility soils, particularly soils with low phosphorus levels. A number of *Stylosanthes* spp. (referred to as stylo in this paper), including *S. guianensis*, *S. hamata*, *S. fruticosa* and *S. seabrana*, have been tested for their potential as short- or long-term fallow pastures, or for intercropping with cereals in tropical and subtropical farming systems. This research has demonstrated some benefits of stylo fallows and other stylo-based practices with respect to yields of subsequent maize, rice and millet crops.

## The Impact of Stylo on Cereal Crops in Africa

Stylos have been integrated into the farming systems of Africa in a range of ways. The most common is a system where small areas of stylo are sown with superphosphate fertiliser to produce fodder banks, which are fed to livestock during the dry season. After several years, this same land can be used for cropping (Bayer & Waters-Bayer 1989). Other options for incorporating stylo into the cropping system have focused on shorter fallow periods. Muhr et al (2002) described a potential farming system where the legume fallow was only about 12 months, and Becker and Johnson (1998) outlined a system of incorporating stylos into the upland rice systems of West Africa (Ivory Coast) in which the legume fallow period would be only 9 months. Other research has focused on integrating stylo into a cropping system through intercropping or undersowing rather than the use of rotations or fallows (Mohamed-Saleem 1985). Some, like that of Becker and Johnson (1998), used undersowing of stylo within a rice crop for pasture establishment, and then a fallow after the removal of the crop. Whatever the method, the overriding outcome of these studies show that stylo fallows and/or stylo intercropping can improve soil fertility sufficiently to increase yields of subsequent maize and other cereal crops. Not only is there evidence that soil fertility is improved through the use of legumes, including stylos, there is strong evidence of benefits to the physical attributes of the soil (Tarawali & Mohamed-Saleem 1995).

A number of authors have reported a significant contribution of biologically fixed N from stylo fallows to subsequent cereal crops. Mohamed-Saleem and Otsyina (1986) showed that two- and three-year fallows of *S. hamata* and *S. guianensis* contributed up to the equivalent of 100 kg N/ha (Table 6.1). The capacity for stylo to regenerate from its soil seed reserves was also considered to be an advantage. In later work Tarawali (1991) similarly showed that maize following *S. guianensis* cv. Cook grown for two to four years as fodder banks in the subhumid zone of Nigeria produced a grain yield which was equivalent to that obtained from fertilising natural fallow with 45 kg N/ha.

Muhr et al (1999a, 1999b) investigated the potential of a short-term legume fallow system in subhumid southwestern Nigeria where forages were grown during the wet season and harvested for fodder. The plants were allowed to regrow until the mid dry season before being turned into the soil as green manure for maize sown in the following wet season. In the dry season crop, *Aeschynomene histrix*, *Centrosema pubescens* and *S. guianensis* all produced in excess of 10 t DM/ha. At one of the sites in this study, where the maize grain yield following *S. guianensis* was highest, 90 kg/ha of N was contained in the residues of the green manure crop.

Benefits of a stylo fallow have also been demonstrated in cereal crops other than maize. Working in the savanna and forest zones of the Côte d'Ivoire, Becker and Johnson (1998) showed that a single dry-season *S. guianensis* fallow of six months sown in the latter half of a wet season

**Table 6.1 Impact of stylo fallows on the yields of subsequent maize crops and the equivalent N required by a 3-year cropped area to attain similar maize yields.**

Previous land use	Maize grain yield (kg/ha)	N-contribution (kg/ha)
Cropped 3 years	460	–
Natural fallow	1270	30
<i>S. hamata</i> , 2 years	1330	32
<i>S. hamata</i> , 3 years	2510	90
<i>S. guianensis</i> , 1 year	1640	44
<i>S. guianensis</i> , 2 years	2700	110

Source: Tarawali & Mohamed-Saleem (1995) using data from Mohamed-Saleem & Otsyina (1986)

rice crop increased the subsequent yield of a rice crop sown after the fallow by at least 50% over the alternative weed fallow. The effectiveness of *S. guianensis* in terms of total N accumulation during the fallow period in this study was comparable with the most efficient species, namely *Mucuna* and *Canavalia* spp. Legume DM yields for *S. guianensis* ranged from 1.4 t/ha to 16.6 t/ha, and total N accumulation from 40 kg N/ha to 201 kg N/ha across the regional zones where the study was undertaken, with a consistent 60–70% of that total N being derived from nitrogen fixation. On the basis of these results, Becker et al (1998) suggested that the use of stylo or other legumes was a feasible option for upland rice producers and that legume fallows provide significant increases in rice yields. In a region with high population growth and increasing demand for land, such practices might provide an alternative to high-cost fertiliser usage as a means to combat a rapid decline in soil fertility. Although a range of legumes was effective in delivering N benefits to rice crops, stylos and other small seeded legumes have an advantage over the faster growing and more vigorous species (eg *Canavalia* spp. and *Mucuna* spp.) in undersowing and relay sowing, as they were less competitive with the maturing rice crop and so more suitable for undersowing (Akanvou et al 2001).

Southern and eastern Africa have seen far less research targeting the role of stylos in cropping systems. On higher fertility soils and in more subtropical environments in Africa, such as in Rwanda, several legumes other than stylos have provided greater dry matter and soil nitrogen. In the Rwandan Highland, stylo (probably *S. guianensis*) contributed the equivalent of about 109 kg N/ha as a green manure cover crop (Yamoah & Mayfield 1990). In Malawi MacColl (1990) showed that maize yields following a two-year stylo fallow (probably *S. guianensis*) were significantly greater than those without fertiliser but less than those obtained with an application of 40 kg N/ha.

### Intercropping

The potential role of stylos in intercropping systems has also been investigated in a subhumid zone of West Africa with varying outcomes. Shehu et al (1997) found little evidence of net benefit from using *S. hamata* as an intercropping species in maize systems in subhumid Nigeria as the decline in maize yield was usually far greater than the

compensatory stylo yield at most reasonable row spacings. In Niger Kouame et al (1993) found that intercropping millet and *S. hamata* had no beneficial effect on millet in the first year, but that competition from the stylo in the following year when millet was sown into the same area was such that millet yields were reduced by 40–70%. Similarly, Hulugalle (1989) recorded a reduction in maize yields due to the undersowing of *S. hamata* in a dry year, but in two wetter years intercropping actually resulted in greater total yield (maize plus stylo) and no reduction in maize grain. Benefits other than N accumulation have been recorded through intercropping, with Hulugalle (1989) reporting better root depth and density of maize in maize–*S. hamata* treatments compared to maize-alone treatments.

Although these results suggest that intercropping with stylo may provide benefits in some years and under some conditions, they also demonstrate that benefits are dependent upon successfully managing the competition between stylo and the crop for water and other resources. One of the key management issues is the timing of stylo sowing relative to sowing of the crop. Mohamed-Saleem (1985) found that it was necessary to delay stylo sowing by three to nine weeks to avoid a significant reduction in maize yield in this subhumid environment, although when sown at nine weeks the stylo yield was severely compromised and could not be expected to deliver significant forage or N benefits.

### The Impact of Stylo on Cereal Crops in Australia

Some of the earliest work on the use of stylo in cropping systems was in northern Australia where, in the 1960s, Townsville stylo (*S. humilis*) was shown to benefit animal production and, when grown in rotation, provide fixed N for sorghum and rice farming systems. Sorghum grown on land that had been under Townsville stylo for some years yielded about 50% more than when grown in virgin cropping soil (Chapman et al 1996). The susceptibility of Townsville stylo to anthracnose and changes in fertiliser and commodity prices saw the use of stylo in cropping systems cease for some years. However, research at Katherine in the Northern Territory during the early 1980s investigated the role of *S. hamata* in mixed crop–livestock systems in that semi-arid tropical environment. Sorghum was sown into herbicide-sprayed stylo pastures, and cattle were grazed

on native pastures during the growing season and transferred to the cropping area in the dry season to graze the sorghum stubble and the regenerated stylo pasture. This work demonstrated that the benefit of a legume pasture to two successive sorghum crops was equivalent to about 30 kg N/ha for a one-year legume pasture and 55 kg N/ha for a three-year legume phase (Jones et al 1996). In a similar environment Cameron (1996) showed that a range of legumes, including *S. hamata*, provided the equivalent of 40–80 kg N/ha to subsequent sorghum crops. Although *S. hamata* cvv. Verano and Amiga were considered to be useful fallow legumes in northern Australia, Cameron (1996) believed *Centrosema pascuorum*, a self-seeding annual in the monsoonal semi-arid climate, to be more productive.

In addition to the work with *S. hamata* in cropping systems in semi-arid tropical Australia, there has also been more recent evaluation of stylos in the more intensive crop–livestock farming systems of subtropical eastern Australia. *Stylosanthes seabrana* cvv. Unica and Primar were released as legumes that could be sown into native pastures on the clay soils of southern Queensland (Edye et al 1998) to improve feed quality and beef production. In recent years these cultivars have been evaluated under a range of grazing systems. Their potential for producing high forage yield and persistence suggest that, as components of long-term ley pastures, they could contribute to soil fertility amelioration on rundown cropping soils.

Although *S. seabrana* persisted for at least three years in small-scale evaluation trials on a range of heavy clay cropping soils in the semi-arid cropping belt of southern and central Queensland (B.C. Pengelly, unpublished data), there are few data available to estimate biologically fixed nitrogen. T.J. Hall (unpublished data) demonstrated that *S. seabrana* cv. Unica increased the total soil N status as effectively as a lucerne (*Medicago sativa*) and Bambatsi (*Panicum coloratum*) pasture (which is one widely used in the region), and to levels greater than those recorded from a soil with a recent history of almost continuous cropping (Figure 6.1).

In another study in southern Queensland *S. seabrana*-green panic (*Panicum maximum* var. *trichoglume* cv. Petrie) pastures grazed at 1.25 ha/steer for between 250 and 320 days per year over five years produced up to 0.71 kg/year/steer (R.L. Clem, unpublished data). Legume density increased over the five years of the experiment and there was a rapid increase in soil seed reserves, which together suggest that such a grazing system should be sustainable for several years (Table 6.2). However, the slow increase in stylo composition and yield of stylo, despite good initial establishment, indicates that *S. seabrana* pastures would not meet the objectives of short-term leys, where a high yield of legume in the early life of the ley is desirable to maximise the amount of biologically fixed N in the system. The increase in legume composition

**Table 6.2 Changes in legume density and soil seed in *S. seabrana* pastures under grazing in southeastern Queensland.**

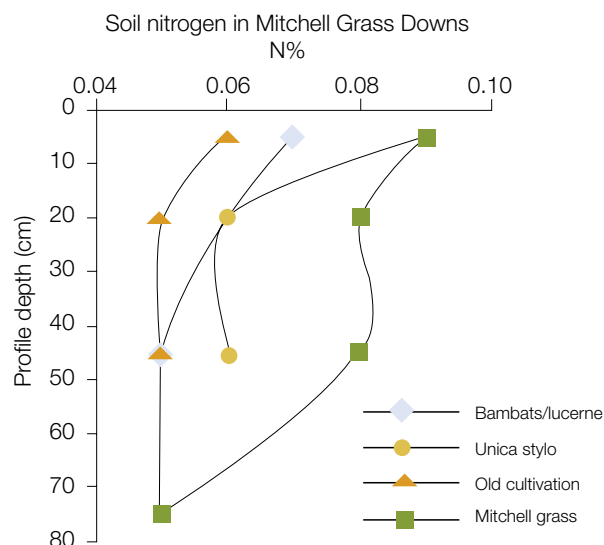
Year	Legume density (plants/m <sup>2</sup> )	Soil seed (kg/ha)
1998	16	-
1999	11	-
2000	12 (35)	38
2001	7 (71)	90
2002	37 (16)	85

Source: B. Clem, unpublished data

**Table 6.3 Changes in end of season forage yields and liveweight gain of steers grazing *S. seabrana* pastures.**

Year	End of season yield		Steer liveweight gain (kg/day/steer)	Grazing days
1998	1960	100	–	–
1999	5490	385	0.39	290
2000	3850	1155	0.62	320
2001	3890	935	0.66	290
2002	4330	1860	0.71	270
2003	n.a.	n.a.	0.62	250

Source: B. Clem, unpublished data



**Figure 6.1** Total soil nitrogen in the top 75 cm of soil in four land-use systems on heavy textured Mitchell Grass Downs (Vertisol) soils in southern Queensland (T.J. Hall, unpublished data). Mitchell grass results are from a native pasture which has been extensively grazed over the past 7 years; old cultivation data are from an area which had been cropped for 6 years, followed by 2 years of lucerne and, most recently, 3 further years of cropping; the Unica stylo paddock had been under continuous cropping for 6 years and then 5 years of pastures; and the lucerne–Bambatsi pasture had 8 years of continuous cultivation followed by 3 years of pasture.

and yield by the third year of the study and the high animal growth rates (which were 20% higher from *S. seabrana* pastures than from grass-only pastures; Table 6.3) suggest that, when included in a longer-term pasture phase, *S. seabrana* has the potential to make a valuable contribution to soil nitrogen. This is partly a reflection of the need to inoculate Primar and Unica seeds with a specific strain of *Bradyrhizobium*, which is otherwise lacking in most Australian soils (Date et al 1996). But *S. seabrana* does not require inoculation with specific *Bradyrhizobium* strains for establishment at sites within Brazil (Date 2000) or India (Ramesh et al, this volume), where native strains in the soil can adequately nodulate this species. With much better initial establishment success than *S. scabra* Seca, *S. seabrana* may be a useful species in targeted cropping systems in countries where it nodulates with native *Bradyrhizobium* strains.

## Adoption and the Future

Despite the experimental evidence that using stylos in short- or long-term fallows or phase pastures have favourable economic implications for both livestock and crop production in mixed farming systems in both commercial scale and smallholder farming systems in the tropics, there appears to have been limited adoption of these practices. Most of the literature refers to potential adoption rather than providing actual figures, although Tarawali et al (1998) state that maize producers have adopted the practice of sowing crops in areas of land previously used for fodder

banks, but provides no information on the number of adopters. Stylo fodder banks are widely used in the subhumid zone of West Africa, with over 27,000 farmers having adopted the technology (Elbasha et al 1999). However, the main driver in fodder bank technology adoption has been the supply of higher quality forage for cattle in the dry season, not benefits to crop production. Similarly, in northern Australia the adoption of stylo technologies is almost invariably associated with overcoming dry season forage constraints in beef production. It appears that nowhere has there been widespread, sustained adoption of technologies which take advantage of the improved soil N status under stylo pastures through cropping.

The reasons for not adopting legume fallow technologies are complex and include both socioeconomic and biophysical factors. In West Africa the constraints to adoption include the availability and cost of increased labour to undertake new farming practices, the lack of capital and supplies of seed, fencing costs and the ingress of nitrophilous weeds (Balasubramanian & Blaise 1993; Becker et al 1998; Tarawali et al 1998; Tarawali & Mohamed-Saleem 1995). Although Tarawali and Peters (1996) showed an economic benefit where maize yield increased substantially, they also warn that the variability in climate, the sometimes necessary late planting of maize, the inherently poor nature of soils, the quality of the pasture fallow, and the weediness to crops all have the potential to greatly reduce projected benefits. With these constraints and risks, Becker and Johnson (1998) suggested that legume fallow technology would have to provide advantages other than higher crop yields to become adopted. They suggested that technologies would also have to contribute to a reduction in labour inputs and deliver better returns on labour investment to be successful.

The story in northern Australia is similar. McCown (1996) suggested that although ley farming was technically possible, the high risk of cropping in a semi-arid tropical environment was a major constraint to adoption. Other constraints were largely economic factors, such as the lower returns and risks in the northern Australian semi-arid tropics, costs of transport, and the comparative advantages in other enterprises such as horticultural crops. Higher beef prices in recent years have been accompanied by a very buoyant live-cattle export industry operating from this region, which would also have reduced the attractiveness of cropping compared to beef production.

In summary, ley farming or fallow farming with stylos will inherently be difficult. Most stylos are adapted to soils of low fertility, which are often marginal for crop production. They are also best adapted to the subhumid and semi-arid regions, which are often associated with high rainfall variability, and their use in fallows and ley/phase pastures require quite complex management.

The future role of forage and green manure legumes in tropical cropping systems, including stylo fallows and stylo intercropping, may well depend on the rapidly changing socioeconomic environment, especially



in West Africa. The predicted increase in the population of the region and its growing urbanisation are likely to lead to greater demand, and perhaps higher prices, for cereals. Those increased prices may provide farmers with financial incentives to adopt technologies that can significantly increase their crop production. If there is a parallel increase in demand for animal products, as suggested by Delgado et al (1999), stylo technologies may then become more attractive for mixed livestock farmers, as their use can enhance both crop and livestock productivity. Other constraints, including those associated with land tenure and the risks of growing a legume pasture and then capturing the N benefits in the cropping phase of a farming system, will remain as challenges. Alternatives to legume technologies, such as greater use of fertilisers, may also become more attractive in such scenarios.

The past 20 years of research has delivered a wealth of information on the benefits and risks associated with the use of stylos in commercial and smallholder mixed crop–livestock farming systems in the tropics. Translating that information to appropriate adoption will now depend on matching the technologies to farmers' needs, especially in the rapidly changing socioeconomic environment of Africa.

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## Chapter 7

# *Stylosanthes* in the reclamation and development of degraded soils in India

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### Summary

The use of *Stylosanthes* spp. (stylo) has made a tremendous impact on the agricultural systems in India, especially in improving natural grazing lands, rehabilitating degraded forests and community lands, and rejuvenating rainfed crop and livestock production systems. It supports the livelihood of >70% of the nomadic and rural population, where the livestock is dependent on these resources. Since 1968 the Indian Grassland and Fodder Research Institute has made a major contribution to stylo research and development, including recent on-station and on-farm operational research to identify and utilise new stylo for the various production systems. Stylo is used as a pioneering coloniser to stabilise degraded and problem soil and watersheds in extensive areas spanning most of the Indian states. Both government and non-government sectors have actively participated in this work, often following participatory approaches. The availability of new and more suitable stylo has generated a renewed enthusiasm for all such development programs where the rehabilitation of degraded sites is desired. Combining environmental remediation with the forage and commercial needs of communities that rely on stylo would assure a sustainable bright future for stylo.

### Introduction

The increasing population of livestock in India calls for strategies to manage excessive grazing to protect vulnerable areas, while at the same time increase forage resources to meet the demand for fodder, dry forage and feed. Currently there is a wide gap between the demand and supply of these items (22% deficit in dry fodder and >50% in green fodder), which accelerates the problem of land degradation.

In high rainfall zones perennial vegetation is the major resource that conserves the soil, while in semi-arid climates sown pastures and multitier silvipastoral systems can minimise runoff and soil loss. Similarly, on sloping lands contour farming and agroforestry practices like SALT (Sloping Agricultural Land Technology) and alley cropping can potentially reduce soil erosion. However, proper planning of land use, including an appropriate selection of vegetation, is required when developing watersheds, catchments, flood plains and riverbanks to check the progress of land degradation.

Development of sustainable production systems is a current objective of agricultural research and development in many countries including India (Paroda 2001). Edye (1984) has reported the role of *Stylosanthes* in different production systems. Sustainability aims at maintaining production, the environment, economic gains and social equity in both temporal and spatial dimensions. This goal can only be attained when environmental protection is balanced with social and economic sustainability. Achieving food and income security is a prerequisite for poor farmers, who are often unable to focus beyond their immediate survival needs, if they are to make decisions and investments affecting the sustainable use of natural resources. Balancing intensive production with natural resource management is likely to be the only way to reverse degradation, alleviate poverty and improve food and income security of resource-poor farmers.

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Many forage species, in particular legumes, are multipurpose plants and desirable candidates for land management. They offer direct and indirect benefits to crop production including nitrogen enrichment of soils; the suppression or reduction of weeds, insect pests and diseases; addition of green manure; improvement of fallow; and use as cover crops and live barriers, among others. They reduce production costs by lowering the need for external inputs such as fertilisers and pesticides, and offer additional environmental benefits including reduced pesticide residues in crops, soil and water; conservation of fossil energy; and soil conditioning through nitrogen fixation. Improvements in soil fertility can support an increased intensification of agricultural production systems. The increased land use efficiency resulting from intensification can lead, in its turn, to protection of areas unsuitable for agricultural production by implementing policies which favour maximising returns from labour and land rather than clearing new land for agriculture (Schultze-Kraft & Peters 1997). Increased feed production on agricultural land marginal for cropping and recuperation of degraded land also contribute to increased land use efficiency (Schultze-Kraft & Peters 1997).

Traditionally in India, legumes have been used as cover crops to improve soil fertility and in turn provide forage for livestock, but their importance declined at a time when the price of mineral fertilisers fell and the focus shifted to high-value soybean crops. Farmers have reacted to changing commodity prices by changing crops and farming practices to maximise profitability, but some have contributed to the already severe problems of land degradation in India. Many government-funded programs of watershed development, wasteland reclamation and revegetation have been started to reverse the trend. Most, if not all, of these programs use the tropical legume stylo as the pioneering coloniser as part of the soil stabilisation component of these developments. During the past three decades non-government organisations and farmer self-help groups have actively joined these initiatives to improve the sustainability of their land and water and assure the supply of quality forage and fuel. This paper outlines the extent of land degradation in India and, using case studies, summarises the use of stylo in the reclamation and development of degraded lands in India.

## Land Degradation

Of the total earth's surface, 78% of the land area is unsuitable for agriculture. Of the remaining 22%, about 9% suffers from physical, chemical and biological constraints and requires special management practices to achieve sustained economic agricultural production (Dagar & Singh 1994). Land degradation is a global phenomenon; dry lands in arid, semi-arid and subhumid climates, covering some one-third of the land surface, are where more than 750 million people live. Human impacts from overpopulation on this fragile land have resulted in desertification in some instances. On the other hand, salinisation and waterlogging have been the main land degrading forces where irrigation is provided. It is estimated (Anon. 1992) that about 2 billion ha of land that were once biologically productive are now degraded. The current rate of land degradation is estimated at 5–7 million ha/year. Globally, land degradation affects about one-sixth of the world's population, 70% of all dry lands (amounting to 3.6 billion ha), and one-quarter of the total land area of the earth (Anon. 1992).

### The extent of land degradation in India

Estimates of degraded/wasteland areas in India differ due to differences in definition and the criteria used (Table 7.1). For instance, Abrol and Sehgal (1992) contended that over 50% (175 million ha) of the total geographical area in India (329 million ha) might have been affected in one way or another. Consequently, about 6600 million t of soil is lost to oceans annually. Because the loss of topsoil depends on the soil type and the extent of degradation (Table 7.2), there are specific soil types that require most attention. Considering the burgeoning human and livestock population in India and the limited land area available, land degradation and the loss of soil productivity are serious concerns, with far reaching socioeconomic and ecological consequences. Sehgal and Abrol (1994) have prepared a land degradation map of India on a 1:4.4 million scale using the Global Assessment of Soil Degradation (GLASOD) criteria. Their study revealed that out of 329 million ha, some 187.7 million ha, representing 57% of the land area, are suffering from problems associated with land degradation (Table 7.3). Of these,

**Table 7.1 Estimates of degraded/wasteland areas in India.**

Estimated area of degraded land / wasteland (million ha)	Percentage of total geographical area (329 million ha)	Source of information and reference
95.0	28.9	Department of Environment, Govt. of India (Vohra 1980)
175.0	53.2	Ministry of Agriculture and Cooperation, New Delhi (1982)
53.3	16.2	National Remote Sensing Agency, Dept. of Space, Govt. of India, Hyderabad (1985)
93.7	28.5	Society for Promotion of Wastelands Development (Bhumbla & Khare 1984)
123.0	37.5	National Wasteland Development Board, Ministry of Environment and Forest, New Delhi (1985)
187.7	57.0	Sehgal & Abrol (1994)

**Table 7.2 Predicted loss of soil productivity due to water erosion of different soil types (Sehgal & Abrol 1994).**

Soil erosion class	Soil loss (t/ha)	Loss of productivity (%)		
		Alluvium-derived soils (Inceptisols)	Black soils (Vertisols)	Red soils (Alfisols)
Nil to very slight	<5	nil	<5	<10
Slight	5–10	<5	5–10	10–15
Moderate	10–20	5–10	10–25	25–50
Strong	20–40	10–5	25–50	>50
Severe	>40	25–50	>50	n.a.

**Table 7.3 Severity of soil degradation and its extent (million ha) (Sehgal & Abrol 1994).**

Type of degradation	Severity of degradation				Total area
	Low	Medium	High	Very high	
Water erosion	5.0	24.3	107.2	12.41	148.91
Wind erosion	–	–	10.8	2.7	13.50
Chemical deterioration	2.8	2.0	9.0	–	13.8
Physical deterioration	6.4	5.2	–	–	11.6
<b>Total</b>	<b>14.2</b>	<b>33.2</b>	<b>124.3</b>	<b>16.1</b>	<b>187.81</b>

15.1 million ha suffers very high and 127.0 million ha high severity of degradation, indicating the need for immediate steps to check further degradation (Table 7.3). Water erosion affects the largest proportion of land, followed by salinisation and terrain deformation. Human induced chemical deterioration is evident in 14 million ha, evident as salinisation in 10 million ha and a loss of nutrients and organic matter in about 4 million ha. The severity of degradation varies depending upon the site characteristics, eg slope, aspect, topographic position, wind speed, velocity of rainfall etc.

### Problems of degraded lands

Soil degradation means a decline in soil quality which brings about unsustainable production systems with erratic and poor production. The visible signs are severe erosion, flooding, siltation, salinisation and desertification. These are mostly man made and can be rectified with different levels of human effort.

Hot spots of land degradation are often associated with human activities such as:

- environmental damage due to settlements, industry and transport – waste disposal; pollution etc;
- intensive agriculture – land, air and water pollution; badly managed irrigation (waterlogging); salinisation; groundwater depletion; saline water intrusion; weed infestation; pest proliferation;
- accelerated agricultural development – loss of rural landscape, biodiversity, wildlife habitats and groundwater quality; siltation of wetlands, inland waters and coastal zones; pesticide load;

- improper management of areas with conflicting demand for scarce water resources;
- Land tenure conflict and unsustainable farming systems due to socio-politico-economic reasons;
- faulty land use practices in major watersheds causing land degradation due to change in hydrological regimes of surface water and groundwater, silting of water bodies and disruption of ecological balance.

### Use of *Stylosanthes* in Revegetation of Degraded Lands

Revegetation of degraded lands requires plant species that can tolerate abiotic and biotic stresses, are easy to establish, vigorous in growth habits, provide easy and early land cover, and produce products of economic value. In India degraded lands are increasingly being targeted for the supply of forage and grazing resources. During the past three decades many species of multipurpose trees, grasses and legumes have been tried in different parts of the country. Some have met with limited success while others have proved very successful and are highly sought after in development programs. Among the successful species, *Cenchrus ciliaris*, *Stylosanthes hamata*, *Leucaena leucocephala* and *Pennisetum pedicellatum* have become essential components of many degraded land development programs. In this chapter discussion will mainly focus on *Stylosanthes* species and other species will be dealt with as appropriate.



## Why *Stylosanthes*?

*Stylosanthes* as a genus (stylo) has received the most attention worldwide as a tropical pasture legume and several cultivars have been released in a number of countries (many chapters in this volume provide lists). The genus is well adapted to the tropics and subtropics (Maass & Sawkins; Andrade et al, this volume) and is particularly suited to areas of low soil fertility with low P content and pH, although forms adapted to alkaline soils are available in the Caribbean, Central America and Mexico (Rai & Shankar 1996). Stylo performs well under drought conditions (Patil & Pathak 1986). In contrast to most other tropical pasture species, stylo usually has a high N content combined with a very low P content; the P content decreases as the plants age, especially under water stress situations. Although the amount of P is inadequate for the nutrition of grazing animals, other minerals seem to be available in sufficient amounts. In addition to improving natural rangeland and animal performance, stylo has shown particular promise as a ley and cover crop in plantation agriculture (Niranjan et al 1994).

**Wasteland development and soil conservation** The biggest expansion in the use of stylo in India has occurred in the development and stabilisation of watershed and wastelands, mainly through government funded or initiated schemes. The program is now well established and has generated a tremendous interest among non-government organisations (NGOs) and farmer groups, who are now improving watersheds and developing wastelands using private resources (see Phaikaew et al, this volume for more details). As a component of agroforestry and/or silvipastoral systems, stylo plays a significant role in the stabilisation and sustainable utilisation of degraded lands. For instance, *S. hamata* is used with *C. ciliaris*, *C. setigerus* and *Panicum antidotale* in soil conservation and land stabilisation in sand dunes in Haryana, and *S. guianensis* is used for stabilising terraces in the foothills and valleys of Manipur (Gupta et al 1989). Government organisations such as the National Wasteland Development Board, agricultural universities and NGOs have used stylo to restore wastelands and develop them as forage, fuel wood, forest products and horticultural resources.

Introduction of *S. hamata* with *C. ciliaris* and *D. annulatum* grasses in rainfed situations can produce quality forage and crude protein if the mixed stand is fertilised with 30 kg P<sub>2</sub>O<sub>5</sub>/ha. This yield can only be achieved through the application of 60 kg N + 30 kg P<sub>2</sub>O<sub>5</sub>/ha in pure *D. annulatum* (Rai 1988) and *C. ciliaris* stands (Rai 1989). Growing *C. ciliaris* and *S. hamata* in alternate rows gives 29.3% higher dry forage yield and 67.2% higher crude protein yield over the grass alone (Singh et al 1983).

At least three species have the potential to be used in wasteland development. (1) *S. fruticosa* (wild lucerne) is mainly distributed in dry localities of Orissa, Andhra Pradesh, Karnataka, Tamil Nadu and Kerala at altitudes up to 900 m and on the coastal areas. Tribal people use it for medicinal purposes against diarrhoea and cold. (2) *S. guianensis* is distributed in high rainfall areas of Assam, West Bengal, Bihar, Uttar

Pradesh, Madhya Pradesh, Maharashtra, Karnataka, Andhra Pradesh, Tamil Nadu and Andaman and in the Nicobar Islands, and some are adapted to acidic, saline-alkaline and waterlogged areas. Cultivars IGFRI S-4214 and IGFRI S-96-1 grow well in wet areas with para grass, setaria, Rhodes, guinea, Dinanath (*Pennisetum pedicellatum*) and thin Napier. (3) *S. hamata* is by far the most widely used species in wasteland development in India since it is well adapted to infertile soil and can be grown on a wide range of soil types from shallow sandy clay-loam to heavy black cotton and red chalka soil. It is suitable for semi-arid tracts with around 425 mm annual rainfall. Other species including *S. humilis*, *S. scabra* and *S. seabrana* have more specialised niches in the targeted environment. For instance, *S. seabrana* is suited to heavy soils and *S. scabra* can be grown in areas with about 325 mm rainfall.

The commercial seed industry in India continues to produce mostly *S. scabra* (most likely Fitzroy and Seca) and *S. hamata* (most likely Verano). Often, seed used for revegetation of wasteland is bought as a generic 'stylo' seed without any attempt to match species or cultivars to the target area. Therefore, details on species and cultivars are not available for much of the 'environmental' usage of stylo. In one study conducted by IGFRI in Madhya Pradesh (Anon. 1992) contoured hill slopes were seeded with stylo and run-off was measured during a heavy rainfall period. The vigorous growth of stylo provided a good ground cover and reduced peak flow to reduce run-off by up to 97.3% (Table 7.4).

One of the difficulties associated with the development of wastelands, which are also common grazing lands, is the ability to control grazing. Consequently, many government run programs have been unsuccessful in maintaining a productive grazing resource where land has been fenced off for two to three years. Overexploitation of such community resources once they are opened up for grazing has been the principal reason for the failure of many such schemes, and therefore the total land area improved through such schemes, which exists on paper, is no longer available. We have chosen to summarise the successful development schemes through a few selected recent case studies.

**Case study 1** Through the setting up of a 'Rajiv Gandhi Watershed Mission' by the Government of Madhya Pradesh, a variety of wastelands have been reclaimed (Anon. 2001). With 3800 watersheds and 101 collaborating NGOs, Madhya Pradesh has India's largest watershed management program, covering 7002 villages in the Jhabua district alone. A total of 147,066 ha has been covered under this integrated watershed development to allow more intensive cropping including intercropping, and developments using pasture and silvipastoral species, particularly on the transition and discharge zones. Intensive soil and water conservation treatment work has been completed on 13,430 ha. Fodder development has been promoted on private as well as government land with an emphasis on grass beds, silvipasture and pastures development. Fodder production now occurs on 2950 ha with *S. hamata*, Dinanath, Sukli and Batodi as the major fodder species. This has provided

**Table 7.4 Peak discharge of water from a micro-watershed during heavy rainfall at Datia in Madhya Pradesh stabilised with stylo on contour trench mounds on a hill (Anon. 1992).**

Date	Storm rainfall (mm)	Peak discharge (m <sup>3</sup> )		% decrease over control
		Control	Treated	
4.8.91	5.8	0.12	0.031	74.2
6.8.91	34.8	0.045	0.031	31.3
7.8.91	32.0	0.045	0.0132	70.7
8.8.91	26.2	0.076	0.0021	97.3
9.8.91	22.2	0.118	0.056	52.05
10.8.91	9.6	0.118	0.042	64.4
11.8.91	18.0	0.022	0.013	40.9
14.8.91	17.6	0.55	0.042	92.4
23.8.91	29.0	0.012	0.0021	82.8
24.8.91	15.2	0.08	0.02	75.0
25.8.91	87.4	1.80	0.27	85.0

immediate returns to villagers and has generated biomass to enhance soil fertility and retain soil cover as aids in soil conservation. Many watershed villages have become self-sufficient in fodder and forage. The value of stylo for regeneration of such lands and for provision of forage for grazing and cut-and-carry fodder has been realised. Consequently, the mission now aims to cover an enhanced target of over 3.1 million ha under 702 mini watersheds spread over 45 districts.

**Case study 2** In the Chattisgarh region lateritic and sandy loam soils with poor water retention are prone to heavy erosion and leaching. Perennial, tree-based land use and silvipastoral systems with multipurpose tree species like *Dalbergia sissoo*, *Pongamia pinnata* and *Terminalia bellerica* have been particularly useful in this region. These have been planted with legume and grass species like *S. hamata* and *Pennisetum* spp. Alternatively, agricultural crops could be rotated with leguminous, fodder grass species in a two- or three-year cycle. Successful intercropping of *S. hamata* between rows of sorghum has been demonstrated in different locations (Hazra & Rawat 1991; Singh & Hazra 1988; Wani & Umrani 1993). Oversowing *S. hamata* in maize, sorghum and bajra under rainfed conditions on the red soils of Jhansi has increased grain yield of cereals by 6–26% (Singh et al 1986).

Stylo has been used as a ley in various cropping and forage production schemes. Growing *S. hamata* for three years as a ley supplied an estimated 80–100 kg/ha nitrogen to the soil (Reddy et al 1989). At Jhansi and Hyderabad four years of ley cropping with *S. hamata* significantly increased subsequent grain yield of sorghum and pearl millet (Niranjan et al 1994)

**Case study 3** Substantial portions of the drylands are usually not suitable for crop production. The Central Research institute for Dryland Agriculture (CRIDA), Hyderabad has developed a number of alternate land use systems where such marginal land can be put to productive use for augmenting fodder and fuel needs. In a system integrating crops, trees, fruit trees and pastures to optimise production in Southern and Central

India, production of *S. hamata* on degraded marginal land has been found very profitable. Structural degradation of soil is widely perceived as being a major problem in the cropping of Alfisols in the semi-arid tropics, where poor structural stability is the result of low and inactive clay content and low amounts of decomposed organic matter.

A study conducted by CRIDA using *S. hamata* in a crop rotation with castor and sorghum revealed that two to three years of legumes in a four-year rotation added 30–35 kg N per ha to the soil and was economical (Joshi 2002) (Table 7.5). Further, ley with *S. hamata* reduced disease problems in the grain and oil seed crops.

**Case study 4** In Auroville (south India) an international community located in the Vanur Block of VPR District, Tamil Nadu, has regenerated degraded lands in two different soil zones, a red laterite and a saline-alkaline shallow black cotton soil. The aim was to regenerate the land for planting of timber wood (*Dalbergia latifolia*, *Pterocarpus santalinus*, *Kigelia pinnata*), non-edible oil seeds such as neem (*Azadirachta indica*) and minor fruits such as *Diospyros melanoxydon*. Attempts at direct reforestation with these species had been made earlier but were unsuccessful. Untreated seeds of *A. holosericea* and *S. hamata* were mixed and broadcast on the barren land during summer months, while *S.*

**Table 7.5 Economics of ley farming (Joshi 2002).**

Treatments	Net return per ha (Rs)	Benefit:cost ratio
Castor sorghum stylo sorghum	10465	2.85
Castor stylo stylo sorghum	9684	3.25
Sorghum stylo stylo sorghum	11073	3.83
Stylo castor stylo sorghum	9171	2.64
Fallow fallow stylo sorghum	2779	2.95
Fallow stylo stylo sorghum	9005	4.71
Stylo stylo stylo sorghum	15769	7.15

*hamata* was sown at the onset of monsoon rains. *Stylosanthes hamata* was the first to germinate and its growth rate was excellent considering the soil condition. A fairly dense stand of plants developed, reaching 10–15 cm in height after six months of growth. After three years a dense carpet of *S. hamata* covered the entire area, except in places where water logging occurred. Stylo was left uncut and quickly spread to adjacent areas to form a lush, self-seeding soil cover. After suitable growth *S. hamata* cuttings and lopped leaves of *Acacia holosericea* were used as mulch for establishing trees, and no additional watering was needed in the area.

In black cotton soils, planting vegetative barriers along the contours, rather than building earth bunds, is recommended to control erosion. For instance, Vetiver grass or valuable fodder grasses such as *Cenchrus* species can be planted along the contour lines (Anon. 2001). Once established, the plants prevent erosion, the roots help in water percolation, and the continuous build-up of organic matter improves the general soil structure. Untreated seeds of *Sesbania aculeata* (common name: dhaincha) at a rate of 5 kg/ha, *Stylosanthes scabra* at 12 kg/ha and *Sesbania bispinosa* at 750 gm/ha were broadcast during the summer rains (July–August, 360 mm) when most of the cracks had filled up, but without tilling the soil. Minor soil and water conservation measures were taken. The area was fully fenced off so that no grazing or trespassing could take place. From the 2 ha of land, only half a hectare of degraded land (category 2) grew a dense stand of *S. scabra* plants. The *Sesbania bispinosa* stand was sparse, and the dhaincha plants never really grew and withered away soon after the end of the rainy season. The *S. scabra*, however, grew up to 1.5 m tall and, in some cases, taller after two years. Grass started growing more abundantly between the *S. scabra* plants, and controlled grazing by cattle was allowed on the grasses and *S. scabra*.

In red lateritic soils, *Acacia holosericea* and *S. hamata* were the pioneering colonisers and in black cotton soils *Dodonaea viscosa* was the most important species. The overall conclusion of the above experiments is that selected plant species can indeed play a major role in reversing erosion and restoring soil health and it is possible to bring back to productivity even the most degraded soils.

**Case study 5** In a study around Jhansi on a large degraded forest area and community wastelands, *S. hamata* has made its mark in the recovery of fertility and productivity. The land, originally producing <1 t/ha biomass was able to produce 10 t/ha in a ten-year rotation to become economically viable, offering long-term employment for local villagers (Pathak et al 1995).

**Case study 6** The Indo German Watershed program operated by an NGO, the Watershed Organization Trust (WOTR) located at Ahmednagar in Maharashtra, is at the forefront of watershed development in India, using stylo to stabilise contour bunds and as a pioneering species. WOTR works primarily as a program manager, offering training for other

NGOs and producing regular publications including planning manuals and kits. During the period 1993–2003, WOTR has developed from 7 watersheds, working with 7 NGOs, to 158 watersheds involving 76 NGOs. The area covered has increased from 15,500 ha to 162,000 ha stretching across two states (WOTR 2004). In the 'Bhoyare' watershed, which covers 1124 ha in a <550 mm annual rainfall zone, a 'ridge to valley' development strategy is used to stop soil erosion and arrest the flow of silt. The primary emphasis is on stabilisation and improvement of the soil resource and conservation of moisture. Cropping of sunflower and small grain cereals is practised on terraces bound by contour ridges. Forage grasses and stylo are planted on the contour ridges in between tree seedlings selected by the villagers.

**Case study 7** As one of the oldest NGOs in India, the Bharatiya Agro Industries Foundation (BAIF) operates in the federal states of Maharashtra, Karnataka, Gujarat, Rajasthan, Uttar Pradesh, Madhya Pradesh and Andhra Pradesh spread over 10,000 villages covering a million families. While there are too many development activities to list in detail, wasteland developments based on stylo and other fodders are prominent at 'Mylana hally' (700 ha), 'Manjunathpura' (70 ha), 'Lakhihali' (497 acres) and 'Surashetty Kopa' (42 acres) in Karnataka, among others (Hegde et al 1988). BAIF aims for integrated rural development through sustainable management of degraded land, livestock water and vegetation resources. It is well known for its livestock development program, which covers upgrading of cattle breeds.

**Case study 8** A program of the Hyderabad Urban Development Authority (HUDA), spanning 700 ha in Mamidipally near Hyderabad, has combined wasteland development with income generation for rural women from selling stylo seeds. The women are paid as labourers for all farming operations and in turn sell stylo seed to HUDA for further sowings in wastelands. The women's group has successfully cultivated *S. hamata* in large areas combined with multipurpose tree species for seed production.

### Other related uses of *Stylosanthes*

**In-situ moisture conservation** The Vetiver grass-based in-situ moisture conservation practices tailored for different soil types and rainfall patterns prevalent in India have contributed to a 10–35% improvement in dryland crop yields (Anon. 2001). The most notable examples are contour farming in shallow red soils, compartmental bunding in shallow black soils, inter-plot water harvesting in alluvial soils in high rainfall regions, and raised bed and sunken systems in deep Vertisols. Adopted in combination with other practices, eg organic manuring and fertiliser use, these have led to a substantial improvement in yields. Vegetative barriers are also an effective inter-terrace land treatment in place of earthen barriers. This technology using stylo seeded on the bunds has provided additional nutritious forage and soil and water conservation value in the watersheds (Pathak 2002).

**Table 7.6 Current annual increment of height and diameter growth in multipurpose trees in association with stylo (Anon. 1993).**

Tree species	Height growth (cm)		Diameter growth (cm)	
	Control	Treated	Control	Treated
<i>Acacia tortilis</i>	23.8	63.4	0.14	0.44
<i>Albizia amara</i>	30.5	37.3	0.20	0.52
<i>Anogeissus pendula</i>	13.7	14.7	0.10	0.27
<i>Azadirachta indica</i>	37.0	21.6	0.12	0.14

**Stylo as a nurse crop** The potential of stylo for soil improvement through nitrogen fixation and carbon sequestration provides added nourishment to the associated vegetation. In a study on degraded lands the tree growth of several multipurpose trees was improved when in association with *S. hamata* (Table 7.6). This also produced sufficient nutritious forage for livestock. Such associations are exploited, with increasing popularity, in many systems including: i) in the social forestry programs run by the state governments as a mandate, stylo is recommended and used to provide an effective ground cover and forage for the livestock; ii) in the Telengana region of Andhra Pradesh, *S. hamata* sown in a *Leucaena leucocephala* plantation gives better yield (4.5 t/ha/year) and dry matter accumulation by *L. leucocephala* than with grasses like *C. ciliaris* (Neelam-Seharan et al 1989); iii) in West Bengal, fuel wood species such as *Eucalyptus* and *Acacia auriculiformis* perform well with sowings of *S. guianensis*, *S. scabra*, *S. hamata* and *S. humilis* (Lahiri 1992); iv) in the acid soils, *Acacia auriculiformis* with *S. humilis* help to improve pasture yield and fuel wood production (Gill 1992); and v) in forestry plantations of the eastern Himalayas, *S. hamata* in bamboo plantations and teak seed nurseries improve soil fertility (Singh et al 1992).

In plantation and orchard crops such as *Psidium guajava*, *Eugenia jambolana* and *Annona squamosa*, stylo is used to suppress weeds, enrich soil and provide fodder (Dasthagir & Suresh 1990). There are similar reports for custard apple and gooseberry (*Phyllanthus emblica*) (Gill & Gangawar 1992); *Alnus nepalensis* and pineapple on hilly terrain of Meghalaya (Chauhan et al 1993); coconut in Kerala (Pillai et al 1995); and tea gardens in Kurseong (Macalpine 1956).

**Intercropping with rainfed crops** In one IGFRI study productivity improved by 50% of the expected yield of the mixture when stylo was grown with *Panicum maximum* in different proportions under rainfed conditions (<800 mm rainfall) (Table 7.7, Bhatt 2003). Similar intercropping with other rainfed crops such as sorghum, pearl millet and maize has improved yield and quality of the cereal crop, improved fertility, and protected soil and nutrients loss (Table 7.8, Singh et al 1986). In one study soil nutritional status and physical and chemical characteristics changed for the better using *S. hamata* as an intercrop with sorghum or pearl millet (Table 7.9, Hazra 1997). Increase in soil fertility, especially available nitrogen, with stylo used in a rotation with cereals has translated

**Table 7.7 Forage dry matter production (t/ha) of *Panicum maximum* and *Stylosanthes hamata* under different proportions in a mixture (Bhatt 2003).**

Treatments	Grasses	Stylo	Total
<i>P. maximum</i>	8.01	–	8.01
<i>S. hamata</i>	–	3.05	3.05
<i>S. hamata</i> with rhizobium	–	4.25	4.25
<i>P. maximum</i> + <i>S. hamata</i> (1:1)	5.59	2.84	8.43
<i>P. maximum</i> + <i>S. hamata</i> (1:1) + rhizobium	5.40	3.75	9.15
<i>P. maximum</i> + <i>S. hamata</i> (1:2)	5.82	3.40	9.22
<i>P. maximum</i> + <i>S. hamata</i> (1:2) + rhizobium	5.96	3.44	9.38
<i>P. maximum</i> + <i>S. hamata</i> (2:1)	6.20	3.42	9.62
<i>P. maximum</i> + <i>S. hamata</i> (2:1) + rhizobium	6.33	3.48	9.82
CD at 5%	0.414	0.624	–

**Table 7.8 Effect of intercropping of *Stylosanthes* in cereal fodder crops (Singh et al 1986).**

Treatment	Dry forage yield (t/ha)			Crude protein yield (t/ha)			Land equivalent ratio (LER)*
	Cereal	Stylo	Total	Cereal	Stylo	Total	
<b>Stylo in between rows of:</b>							
pearl millet	5.70	1.50	7.20	0.46	0.28	0.74	1.27
sorghum	8.97	1.36	10.35	0.80	0.25	1.05	1.36
maize	8.74	1.97	10.71	0.84	0.37	1.21	1.55
<b>Stylo broadcast amongst:</b>							
pearl millet	5.17	2.59	7.76	0.42	0.48	0.90	1.44
sorghum	8.49	1.88	10.37	0.76	0.35	1.11	1.42
maize	7.65	2.25	9.90	0.73	0.42	1.15	1.46
<b>Monoculture of:</b>							
pearl millet	–	–	6.19	0.49	–	0.49	
sorghum	6.19	–	8.63	0.76	–	0.76	
maize	8.63	–	8.12	0.76	–	0.76	
stylo	8.12	4.31	4.31	–	0.81	0.81	

\*LER = ratio of dry matter production under mixture and sole crop.

**Table 7.9 Improvement of soil physical and chemical properties and fertility status through intercropping of *Stylosanthes* in cereal fodder crops (Hazra 1997).**

Stylosanthes treatments	pH	Conductivity (mm hos/cm)	Organic carbon (%)	Available nutrients (kg/ha)			Bulk density (g/cc)	Water-stable aggregates (>0.25 mm)	Field capacity (%)	Pore space (%)
				N	P	K				
<b>Between rows of:</b>										
pearl millet	7.1	0.14	0.58	202	15.0	275	1.50	18.8	15.0	43
sorghum	7.1	0.11	0.67	193	14.5	271	1.47	19.7	15.3	44
maize	7.0	0.11	0.67	191	14.2	269	1.45	20.1	15.4	45
<b>Broadcast amongst:</b>										
pearl millet	7.1	0.13	0.63	218	14.7	272	1.45	19.5	15.2	45
sorghum	7.1	0.10	0.68	207	14.2	267	1.42	21.2	15.6	46
maize	7.0	0.11	0.70	205	14.0	263	1.41	21.5	15.8	46
<b>Monoculture of:</b>										
pearl millet	7.5	0.20	0.53	195	15.6	280	1.51	18.8	14.8	42
sorghum	7.4	0.17	0.55	187	14.8	272	1.48	19.5	15.0	44
maize	7.4	0.18	0.56	183	14.5	275	1.46	19.7	15.0	44
stylo	7.1	0.12	0.67	237	13.2	267	1.38	22.2	15.6	47
<b>Before rotation</b>	7.6	0.28	0.43	167	11.3	281	1.57	17.5	14.5	40



**Table 7.10 Improvements in soil fertility and sorghum grain yield following various rotational regimes with *Stylosanthes* and other crops under rainfed conditions at Hyderabad (Singh & Singh 1987).**

Crop rotation				Soil fertility before sorghum cultivation		Sorghum grain yield in year 4 (t/ha)
Year 1	Year 2	Year 3	Year 4	Organic carbon (%)	Available N (kg/ha)	
Castor	Sorghum	Castor	Sorghum	0.22	109.9	1.8
Castor	Sorghum	<i>S. hamata</i>	Sorghum	0.31	112.9	2.6
Castor	<i>S. hamata</i>	<i>S. hamata</i>	Sorghum	0.22	125.4	2.9
Sorghum	<i>S. hamata</i>	<i>S. hamata</i>	Sorghum	0.35	141.1	3.4
<i>S. hamata</i>	Castor	<i>S. hamata</i>	Sorghum	0.25	135.7	3.2
Fallow	Fallow	<i>S. hamata</i>	Sorghum	0.28	90.9	1.9
Fallow	<i>S. hamata</i>	<i>S. hamata</i>	Sorghum	0.28	97.2	2.7
Castor	Sorghum	Fallow	Sorghum	0.21	108.2	1.1
Castor	Fallow	Fallow	Sorghum	0.15	87.7	1.0
<i>S. hamata</i>	<i>S. hamata</i>	<i>S. hamata</i>	Sorghum	0.56	141.1	3.9

to increased cereal yield (Table 7.10). In the fourth year of a rotation, the organic carbon (0.56%) and available N (144.7 kg/ha) were optimum in treatments where *S. hamata* was grown continuously for three years followed by a sorghum – *S. hamata* – *S. hamata* – sorghum rotation (0.35% organic carbon and 141.1 kg/ha available N) (Rai & Shankar 1996; Singh & Singh 1987). Improvements in soil fertility can potentially extend the benefit beyond the immediate cropping season.

**Regeneration of mine sites** The Nagari Mines near Dandeli have successfully used a mixture of *S. hamata* and *S. scabra* to revegetate a 17 sq km area of mine dumps within the thick evergreen tropical forest region of Karnataka. The revegetation started during 1998 and by 2000 the whole area had been covered by stylo. The Indian Aluminum Industries is using *S. hamata* and *S. seabrana* along with *Chloris*, *Brachiaria decumbens*, *B. brizantha* and *B. mutica* to effectively cover the mud slurry to prevent it spreading to neighboring agricultural lands at its aluminum producing factory near Belgaum in Karnataka. This mix of species has effectively colonised the barren wasteland within a year.

## Future Prospects

The expansion of the Joint Forest management program to about 28 million ha of degraded forests in the coming decade provides a great scope for using stylo as a nurse crop to benefit the land and livestock. Recently identified *S. seabrana* with enhanced adaptation increases the potential areas and prospects for the use of stylo in India. So far, the native *S. fruticosa* has not been used to any extent in any economic or environmental programs. If community concerns over exotic species overpowering native species gain momentum, encouraging the native species may offer a good compromise. If resource-poor farmers are to take an interest in the sustainable development of the land resource, ecological improvement to arrest land degradation must deliver immediate and significant economic benefit for what may appear a

long-term development. Recent studies in India have established the economic feasibility of using dried stylo leaf meal as a supplement for commercial poultry feed ingredient (Guodao et al, this volume), and this can be an additional incentive to wasteland development activities. Better coordination between government, farmer groups, poultry feed manufacturers and NGOs needs to occur for the commercial production of leaf meal from wastelands to progress. Matching ecological sustainability with economic gain and livelihood support are some of the prospective areas for the future.

## Acknowledgments

Authors thank Dr Sukumar Chakraborty of CSIRO Plant Industry for providing the opportunity to contribute to this volume and for a critical review of the manuscript; and Crispino Lobo (WOTR), G.N.S. Reddy (BAIF) and Dr K. Bhaskar (Indian Administrative Service, Member KAWADA (Karnataka Watershed Development Authority)) for information on respective watershed programs.

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# Chapter 8

## Biotic and abiotic constraints to *Stylosanthes* production

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### Summary

The genus *Stylosanthes* Sw. has provided ample germplasm for a wide variety of agroecological situations in the tropics. The most important attributes of successful *Stylosanthes* cultivars used as pasture or ley in the tropics are resistance to diseases, high seed yield and adaptation to poor soils. This chapter reviews work conducted on the biotic and abiotic limitations to *Stylosanthes* production, and discusses management strategies to combat some of these limitations.

### Introduction

The South American leguminous genus *Stylosanthes* Sw. (stylo) is the source of several species with agronomic usefulness as pasture, forage and green manure. One prime attribute of *Stylosanthes* species that attracted initial agronomic interest was their generally high level of tolerance of both biotic and abiotic stresses. For example, Tuley (1968) noted that stylo is 'largely free from serious pests and diseases'. While efforts at domestication have uncovered severe and widespread susceptibility to anthracnose, a disease caused by the fungus *Colletotrichum gloeosporioides* (Penz.) Sacc., and several insect pests, broad adaptation to drought and 'problem' soils are notable in some of the commercial *Stylosanthes* species.

Reliable world production figures are unavailable, but varieties of *Stylosanthes* have been eagerly adopted in the pastoral areas of northern Australia ever since the value of *S. humilis* Kunth was recognised early in the 1900s. By the late 1990s at least 600,000 ha were contributing about US\$20 million annually to beef production through higher turn-off weights, improved weaner and heifer nutrition and reduced drought risk (Miller et al 1997). Stylo adapts best to lighter textured soils with an annual average rainfall of more than 500 mm. It establishes easily; spreads naturally; resists drought, fire and overgrazing; and increases beef production. Technologies in seed production, sowing and management are well developed, and an extra 50,000 ha of stylo are being sown annually in northern Australia alone.

The relative importance of different abiotic constraints on the growth, survival and productivity of *Stylosanthes* depends on the physiological adaptation of the different species. Studies include Williams and Gardner's (1984) review of the literature on environmental constraints (water deficit, temperature, water excess, frost, fire); Fisher and Ludlow's (1984) documentation

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of adaptation responses to water deficits; and Lenné and Calderón's (1984) review of the available information on biotic constraints. In this chapter we review additional information on both biotic and abiotic stress adaptations of *Stylosanthes*.

## Biotic Constraints

### Anthracnose disease

Anthracnose, caused by the fungus *Colletotrichum gloeosporioides* (Penz.) Sacc., is a disease of major importance to *Stylosanthes* production worldwide (Lenné et al 1984). The disease dramatically reduces dry matter (DM), forage quality (Cardozo et al 1983) and seed yields in all three species (*S. humilis*, *S. guianensis* (Aubl.) Sw. and *S. hamata* (L.) Taub.) grown in Australia (Davis et al 1987).



(a) **Symptoms of anthracnose on *Stylosanthes* species: (a) contrasting reactions of susceptible (left) and resistant (right) genotypes to inoculations with *Colletotrichum gloeosporioides* in greenhouse tests; (b) anthracnose lesions in the field.**

***Colletotrichum gloeosporioides*** This fungus is a heterogeneous and highly complex species, comprising several host-specific populations and exhibiting both morphological (Cox & Irwin 1988; Davis et al 1992; Munaut et al 2001) and pathogenic variability (Chakraborty et al 1996; Kelemu et al 1996, 1999; Lenné & Burdon 1990). Two distinct biotypes of *C. gloeosporioides*, designated A and B, have been described as causing disease symptoms on *Stylosanthes* species in Australia (Irwin & Cameron 1978), although the pathogen population is much more complex in South America (Kelemu et al 1996). Biotype A isolates have a wider host range than biotype B ones (Vinijsanun et al 1987), which are mostly restricted to *S. guianensis*.

The centre of origin of the host *Stylosanthes*, and thus the presumed centre of genetic diversity of its pathogen *C. gloeosporioides*, is in South America. Studies support the hypothesis that the South American isolates of the pathogen infecting *S. guianensis* exhibit a wide range of genetic and pathogenic diversity (Chakraborty et al 2002; Kelemu, Badel,

Moreno et al 1997; Kelemu et al 1996, 1999; Weeds et al 2003). Isolates of the pathogen collected elsewhere also exhibited genetic diversity (Munaut et al 1998).

**Pathogen's features** The pathogen has been extensively studied using both molecular techniques and virulence quantification methods (Braithwaite et al 1990; Chakraborty et al 2002; He et al 1996; Kelemu et al 1996; Masel et al 1993; Stephenson et al 1997). Masel and co-workers (1996) presented molecular evidence for chromosome transfer between biotypes A and B of *C. gloeosporioides*. Biotype A isolates carry a 2-Mb chromosome while some biotype B isolates carry what appears to be a dispensable 1.2-Mb chromosome. A new field isolate collected in Australia was shown to contain both the 2-Mb and 1.2-Mb chromosomes. After detailed molecular analysis, the researchers concluded that the 2-Mb chromosome in the new isolate is perhaps a horizontal transfer from a biotype A to a biotype B isolate (Masel et al 1996). This transfer may be a mechanism for genetic variability among pathogen isolates.

No strong association between random amplified polymorphic DNA genotypes and pathogen races is detected (Chakraborty et al 1999; Kelemu et al 1999). This apparent lack of correlation is neither unexpected nor surprising. The differences that separate the pathogen races are expected to be overshadowed by the magnitude of genetic diversity among isolates within individual races.

**Components of resistance** Advances in molecular biology have enhanced knowledge of defence mechanisms operating in *Stylosanthes* species. Host peroxidase isogenes were expressed in *S. humilis* after inoculation with *C. gloeosporioides* and, during the early stages of host-pathogen interactions, before penetration of the epidermal cell wall (Harrison et al 1995). As much as four-fold increases in chitinase activity have been reported in *S. guianensis* leaves inoculated with *C. gloeosporioides*, systemically protecting the plant against the pathogen (Brown & Davis 1992). After examining compatible and incompatible interactions between *C. gloeosporioides* and *Stylosanthes* species, Sharp et al (1990) concluded that the deposition of callose was closely associated with race-specific resistance.

**Disease management** Host resistance is the cheapest and most practical method of controlling anthracnose. Moderate to high levels of resistance exist among genotypes of *Stylosanthes* spp. (Chakraborty et al 1990; Fernandes et al 1993; Iamsupasit et al 1991, 1995; Kelemu et al 1996). However, building durable host resistance remains a challenge because of the pathogen's complex population structure.

The role of *Stylosanthes* genotype mixtures in reducing anthracnose has been studied (Chakraborty et al 1991, 1995; Davis et al 1994). In one study three accessions and two cultivars with varying levels of anthracnose resistance to four races of the pathogen were used to evaluate disease development during three summer seasons. The results indicated that plant survival in individual accessions did not significantly

improve in mixed stands compared with that in pure stands, although, in some years, significant reduction occurred in the area under the disease progress curve (AUDPC) in certain mixtures compared with pure stands of the components (Chakraborty et al 1991).

Naturally existing conditions such as phylloplane bacteria may play a role in disease management in certain ecological zones. For example, population density of spore-forming bacilli on the phylloplane of *S. scabra* negatively correlated with anthracnose severity, indicating a possible antagonism of these bacteria to *C. gloeosporioides* (Hetherington et al 1995). Lenné and Brown (1991) reported that bacteria, particularly *Pseudomonas* spp. and *Bacillus* spp., collected from the phylloplane of *S. guianensis* in Peru, inhibited both mycelial growth in vitro and conidial germination of the anthracnose pathogen on leaves of *S. guianensis*. A *Bacillus subtilis* isolate from the phylloplane of *S. guianensis* inhibited the growth of *C. gloeosporioides* in culture (Kelemu & Badel 1994).

The pathogen can be seed borne, and treatment of seeds with Benlate® (500 g/kg benomyl), applied as a dry seed treatment at rates of up to 6 g/kg seed, can effectively prevent seedborne infections (Davis 1987).

### **Rhizoctonia foliar blight**

Foliar blight, caused by *Rhizoctonia solani* Kühn (teleomorph: *Thanatephorus cucumeris* (Frank) Donk) and reported in many parts of the world (Lenné 1990), affects *Stylosanthes* species in tropical America wherever the annual rainfall is more than 1500 mm (Lenné & Calderón 1984; Olaya & Lenné 1986). The disease initially appears as water-soaked spots on infected leaves and, under favourable conditions such as prolonged humidity, the spots progress into rotting and extensive foliar death. The pathogen has an extensive host range and can produce substantial foliar damage on susceptible genotypes (Baker 1970), including various tropical and subtropical forage legumes and crops (Galindo et al 1983; Hepperly et al 1982; Lenné 1990).

The pathogen's sclerotia can survive in soil or on plant debris for long periods. They first appear as white masses on infected plant tissues and, as they mature, turn brown and become loosely attached. They shed easily, becoming primary sources of inoculum.

Control over the disease can be elusive. However, Kelemu et al (2001) transformed *S. guianensis* CIAT 184 plants with a rice chitinase gene, producing transgenic plants with resistance to *Rhizoctonia* foliar blight.



**Reactions of transgenic *Stylosanthes guianensis* CIAT 184 (left) and control untransformed *S. guianensis* CIAT 184 plants to *Rhizoctonia solani* in greenhouse tests 8 days after inoculation. Arrow shows blighted leaves in the lower part of the control plant.**

### **Wilt and dieback disease**

Kelemu, Badel and Fernandes (1997) described a severe wilt and dieback disease affecting *Stylosanthes* stands in Colombia and Brazil. The causal agent of this disease is identified as *Lasiodiplodia theobromae* (Pat.) Griff. & Maubl., a fungus that is a virulent, unspecialised, facultative wound pathogen with worldwide distribution and more than 500 different host plants. *Stylosanthes guianensis* genotypes vary in their reactions to the pathogen, and sources of resistance such as *S. guianensis* CIAT 10136 are available (Kelemu, Badel & Fernandes 1997).



(a)

**Symptoms of wilt and dieback disease caused by *Lasiodiplodia theobromae* on inoculated *Stylosanthes* plants (a) in the greenhouse and (b) under natural conditions in the field (Caquetá, Colombia).**



(b)

### **Other diseases**

*Stylosanthes* is host to various phytoplasmas, expressing symptoms such as little leaf, witches' broom and floral abnormalities (Rue et al 2001; Schneider et al 1999). Little leaf disease was found to have limited or no effect on seed yield if the plants have been flowering for about eight weeks before symptom expression (Rue et al 2002). Viral diseases such as those caused by peanut stripe virus (Mishra et al 1993), alfalfa mosaic virus (Mih & Hanson 1998) and potyviruses related to peanut mottle virus (Morales et al 1991) also infect *Stylosanthes* species.

For a list of other pathogens infecting *Stylosanthes*, see Lenné and Trutmann (1994) and Lenné (1990).

### **Insects**

*Stylosanthes* species can be attacked by various pests (Lenné & Calderón 1984), including insects such as stemborers (*Caloptilia* Hübner sp.; Thomas & Diaz 1989), budworms (*Stegasta bosquella* Chambers) and leaf-eaters (Edye 1987).

## Abiotic Constraints

### Drought

Many perennial forms of *Stylosanthes* use various adaptive strategies to cope with very severe water deficits (Williams & Gardner 1984). They exhibit substantial osmotic adjustment that contributes to the maintenance of turgor at low tissue water potentials, and their tissues are insensitive to considerable desiccation. The perennials are also able to root deeply and extract water at water potentials considerably lower than the  $-1.5$  MPa normally associated with wilting point. Thus, the perennial species *S. hamata* cv. Verano and *S. scabra* Vogel cv. Seca are suited to survive dry seasons in the tropics. The annual species *S. humilis*, in contrast, escapes the dry season by completing its life cycle before tissue dehydration becomes critical for its survival (Fisher & Ludlow 1984). It postpones dehydration and its effects through physiological mechanisms such as leaf movement, deeprootedness and leaf shedding.

A comparative study evaluated the accumulation of low molecular weight solutes in water-stressed grain (*Cicer arietinum* L., *Cajanus cajan* (L.) Millsp., *Lablab purpureus* (L.) Sweet, *Vigna radiata* L., *V. mungo* (L.) Hepper, *V. unguiculata* L. and *Glycine max* (L.) Merrill) and pasture legumes (*S. hamata*, *S. scabra*, *Psoralea eriantha* Benth., *Rhynchosia minima* (L.) DC, *Macroptilium atropurpureum* (DC) Urban, *G. tomentella* Hayata and *Centrosema pubescens* Benth.) (Ford 1984). The major compounds that accumulated in the youngest, fully expanded leaves with water stress were *O*-methyl-inositols; 2-methyl-2,3,4-trihydroxybutanoic acid-1,4-lactone; and proline. Concentration of inorganic ions, sugars and organic acids decreased or were unchanged in most of the species under water stress. The betaines glycinebetaine, trigonelline and stachydrine were detected in low concentrations in most of the legumes but did not accumulate to any degree during water stress. All the legumes that tolerated low leaf water potentials accumulated *O*-methyl-inositol pinitol. The other species, except *M. atropurpureum*, contained ononitol or *O*-methyl-scyllo-inositol but no pinitol. This study suggested that pinitol accumulation may be an effective indicator of legume tolerance of low leaf water potentials.

Kitamura and Abe (1984) conducted field experiments in irrigated and dry soils and determined the relative drought sensitivity of *M. atropurpureum* cv. Siratro, *S. humilis* cv. Townsville, *S. guianensis* cv. Schofield, *Desmodium intortum* (Miller) Urban cv. Greenleaf, *D. uncinatum* (Jacq.) DC cv. Silverleaf, *Neonotonia wightii* Wight & A. Lackey cv. Tinaroo and *C. pubescens*. The percentage of total plant DM in roots increased under dry conditions in all species except *C. pubescens*. Siratro showed the largest increase and Greenleaf the largest decrease in root DM per plant under wet and dry conditions respectively. The differences in leaf water potential in irrigated and dry soils showed that the legumes Siratro, Townsville, Schofield and *C. pubescens* have high relative drought tolerance.

In plot experiments 17 *Stylosanthes* accessions (CIAT) belonging to three species were assessed for drought tolerance, herbage production and quality, regenerative ability and self-seeding character, and anthracnose resistance (Mohamed-Saleem & Otsyina 1984). Total DM yield of the *S. capitata* Vogel, *S. guianensis* 'tardio' and *S. macrocephala* Ferr. & Sousa Costa accessions were 2.5–4.2, 7.1–7.6 and 3.3–4.9 t/ha, respectively. Seed yields ranged from 40 to 182 kg/ha, except for *S. guianensis* 'tardio', which did not seed. *Stylosanthes guianensis* 'tardio' retained all its leaves during the dry season, whereas *S. macrocephala* shed all its leaves.

Peters et al (1997) determined the productivity and nutritive value of *Chamaecrista rotundifolia* (Pers.) Greene (syn. *Cassia rotundifolia*) cv. Wynn, *Centrosema pascuorum* Benth. cv. Cavalcade, *S. guianensis* cv. Pucallpa and *S. hamata* cv. Verano after applications of single superphosphate (SSP) and weed control during the dry season in subhumid Nigeria. Wynn, when growing in competition with native vegetation, had the highest DM yields. During the dry season, legume yields were more stable than those of the associated grasses and herbs. Applications of SSP at low levels of available soil phosphorus increased legume productivity but native grasses and herbs did not respond to the fertiliser applied. The nutritive value of all legume species was low for most of the dry season, the decline being largely a function of changes in the leaf:stem:litter proportions. These authors suggested that drought tolerance and capacity to retain leaf should receive more attention when evaluating forage species for use in the dry season.

In a greenhouse trial Bailey et al (1983) determined the effect of soil water status on the critical phosphorus concentration (CPC) in apices and whole tops of *S. hamata* cv. Verano. The plants were grown with six rates of P and three ranges of soil water potential, and were harvested at 10 and 14 weeks after germination. The CPC of whole tops and apices declined between the two harvests. At the first harvest the CPC of whole tops and apices increased as the soil water potential decreased, but at the second harvest no effect of soil water potential was seen on the CPC. Probably, the earlier harvest water stress delayed physiological development, resulting in a CPC characteristic of chronologically younger tissue but, by the second harvest, the decline in CPC with age had ceased for all water treatments.

Under drought conditions the responses of *Andropogon gayanus* Kunth and *S. guianensis* to variations in mycorrhizal inoculation (natural soil, soil without mycorrhizae and soil inoculated with *Glomus etunicatum*) and application of different doses of P (five levels) to soil were evaluated (Souza et al 2000). Plants were cultivated in pots in a greenhouse experiment, and results for shoot and root development showed that increasing P rates significantly increase DM production and improve the plants' drought tolerance. Response to mycorrhizal inoculation was marked in *S. guianensis*, particularly when P rates were intermediate.

Carvalho and Schank (1989) conducted a comparative study on drought adaptation of two species of *Stylosanthes*. They grew plants of *S. hamata* cv. Verano and *S. guianensis* cv. Schofield separately in pots under greenhouse conditions, subjecting them to recurring cycles of soil drying and wetting. They assessed the effect on plant growth by using standard growth analysis techniques, taking six harvests at ten-day intervals and starting 45 days after planting. As expected, the water stress treatment reduced the DM yield of both species, influenced DM distribution in the plants and reduced various growth parameters. *Stylosanthes guianensis* yielded significantly more DM per pot than did *S. hamata* under both conditions (Table 8.1). Flowering was delayed by about eight days in the water-stressed *S. hamata* plants compared with the non-stressed treatment; *S. guianensis* did not flower at all under any of the conditions.

Negative relationships are known to exist between transpiration efficiency (TE) and carbon isotope discrimination ( $\Delta$ ) and between TE and specific leaf area (SLA). Thumma et al (1998) conducted a greenhouse experiment, using eight accessions of *S. scabra* to identify whether cv. Seca (a drought-resistant cultivar) differs in TE from other accessions of *S. scabra* and to determine the relationship between TE and  $\Delta$  under both well-watered and water-stressed treatments.

Seca maintained the highest TE (lowest  $\Delta$ ) under both control and stress treatments, and leaf  $\Delta$  correlated significantly and negatively with TE under both control and stress conditions. These authors also found a significant and negative relationship between  $\Delta$  and DM production under the stress treatment. The interaction between accession and watering treatment was not significant for either TE or  $\Delta$ . Significant agreement was found between performance in the field and that in the laboratory for these eight accessions. TE may, in fact, significantly contribute to drought

resistance in Seca. Furthermore,  $\Delta$  and/or SLA may be useful as selection criteria in breeding programs to identify lines with high TE.

Thumma et al (2001) also evaluated three cuttings (maintained through vegetative propagation) from each of 120 F<sub>2</sub> genotypes from a cross between two *S. scabra* genotypes (CPI 93116 and cv. Fitzroy) chosen on the basis of degree of polymorphism and differences between traits such as TE, SLA and  $\Delta$ . They subjected the plants to water stress (40% field capacity) for 45 days and evaluated for biomass productivity (BP) traits (total, shoot and root DM), transpiration, TE, SLA,  $\Delta$ , relative water content (RWC) and content of the osmoprotectant trans-4-hydroxy-N-methyl proline (MHP), which accumulates under water stress conditions. They developed a linkage map consisting of 151 random amplified polymorphic DNA markers for QTL (quantitative trait loci) analysis.

Biomass productivity traits correlated positively with TE and negatively with  $\Delta$  and SLA, whereas transpiration correlated positively with BP traits and negatively with SLA. Values of  $\Delta$  correlated significantly and negatively with TE. Several markers were found to be common between the different traits.

### Temperature and photoperiod

Ison and Humphreys (1984) reviewed the juvenile stages of *Stylosanthes* species and morphological changes at flowering. They covered aspects related to photoperiod responses of several *Stylosanthes* species; induction of flowering at suboptimal temperatures or photoperiods; effects of temperature interactions with photoperiod responses; and effects of moisture stress on flowering in terms of flower development, timing and duration of flowering and seed yield components. In a related study Argel and Humphreys (1983) determined the effects of variation in

**Table 8.1 Effect of water stress on dry matter production and distribution among plant parts in *Stylosanthes hamata* and *S. guianensis* (adapted from Carvalho & Schank 1989).**

Days after planting	Total plant dry matter (g/pot)		Root dry matter (g/pot)		Root-to-shoot ratio	
	No stress	Stress	No stress	Stress	No stress	Stress
<b><i>Stylosanthes hamata</i></b>						
45	1.30	–	0.64	–	0.97	–
55	3.34	1.76	1.13	0.74	0.51	0.71
65	5.28	1.97	0.91	0.61	0.21	0.45
75	8.28	3.20	1.09	1.09	0.15	0.52
85	11.0	3.61	1.38	1.26	0.14	0.54
95	11.2	3.23	1.26	0.61	0.13	0.23
<b><i>Stylosanthes guianensis</i></b>						
45	2.80	–	0.66	–	0.31	–
55	2.91	2.50	0.57	0.52	0.75	0.26
65	4.72	2.50	0.75	0.49	0.19	0.24
75	7.84	3.34	1.21	1.03	0.12	0.45
85	13.0	4.07	1.86	1.29	0.17	0.46
95	14.4	3.85	1.66	0.62	0.13	0.19



temperature, soil moisture supply and illuminance on seed dormancy and seed formation of *S. hamata* cv. Verano. They grew plants in the open in successive seasons in both controlled-temperature cabinets and in the greenhouse, where shading and watering treatments were varied after the onset of flowering.

The results of both studies indicated that hardseededness was strongly developed and was positively related to temperature during seed formation. After seed maturation, seed moisture content was the main factor governing hardseededness, and this quality was negatively associated with temperature during seed formation. Seeds formed under high temperatures had higher contents of lignin (which was concentrated in the counter-palisade cells) and hemicellulose, lower contents of cellulose and shorter palisade cells; cutin content was independent of temperature. Dormancy was more strongly developed in the hooked upper articulation than in the hookless lower articulation. Short durations of soil moisture stress that reduced leaf water potential to minimum values of about  $-2.5$  to  $-2.8$  MPa, and shading treatments that reduced radiation to about 20% of full daylight, reduced seed production, but hardseededness was not consistently related with these treatments.

Flowering date, rate of floret appearance, total floret differentiation, duration of flowering, floret abortion, lower articulation formation and time to pod maturity were found to be sensitive to temperature. Levels of seed production and hardseededness in cv. Verano increased under warm conditions during flowering, thus favouring, possibly even restricting, the cultivar's adaptation to dry tropical environments.

Changes in water potential affects germination of *S. guianensis* seeds (Delachiave et al 1994). In one experiment *S. guianensis* seeds were given osmotic pretreatment with mannitol or polyethylene glycol (PEG) in the imbibition phase (14 h) and then germinated on filter paper moistened with distilled water. In a second experiment, after the imbibition phase, seeds were kept at water potentials between 0 and  $-1.8$  MPa. The seeds pretreated during imbibition had high germination percentages, the

highest being with the PEG treatment. In the second experiment the PEG solutions significantly reduced the germination percentage compared with mannitol. From  $-1.2$  MPa onwards, germination ceased in the PEG treatments.

### Mineral deficiency and toxicity

Most mineral deficiencies and toxicities suffered by *Stylosanthes* forages will produce visible symptoms that are useful for diagnostic purposes (Table 8.2).

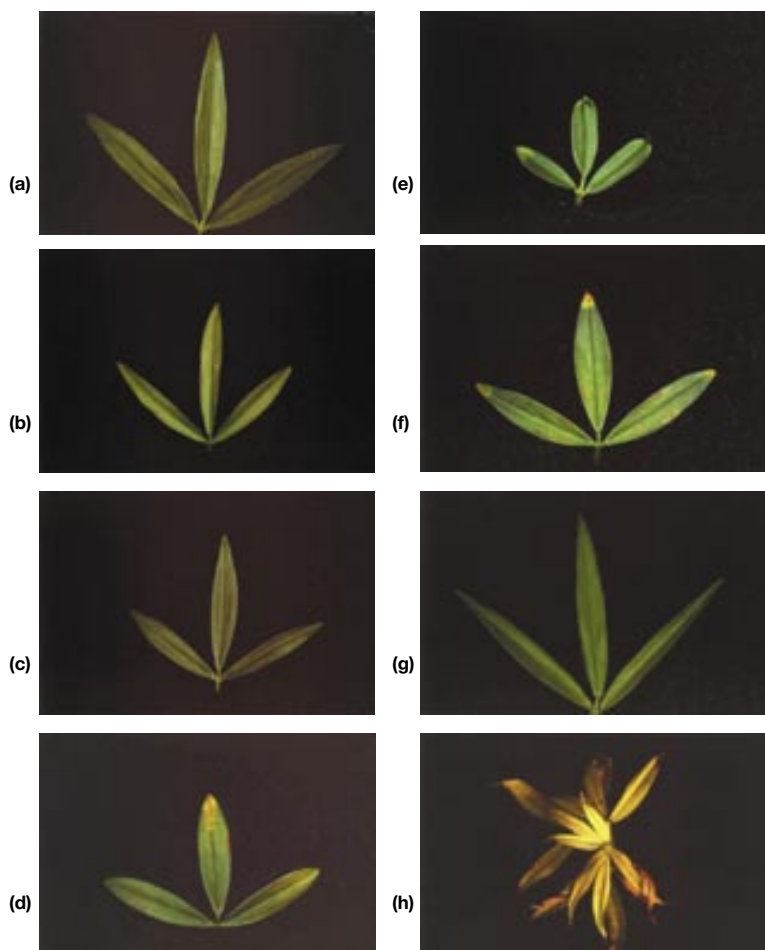
Gilbert et al (1992) used greenhouse experiments to evaluate screening techniques for their usefulness in rapidly characterising new introductions of forage legumes. Characteristics evaluated were growth and nodulation in different soil types, reaction to waterlogging, response to P, growth on acid soils with low Ca and P, and reactions to high Mn, Al and salinity. Legumes tested were *Leucaena leucocephala* (Lam.) de Wit. cv. Cunningham, *Macroptilium lathyroides* (L.) Urban cv. Murray, *M. atropurpureum* cv. Siratro, *Centrosema schiedeanum* (Schlecht.) Will. & Clem. cv. Belalto, *S. guianensis* cv. Cook, *S. hamata* cvs. Verano and CPI 33205, and *S. scabra* cvs. Seca and Fitzroy. The screening techniques used in this study provided a reasonable assessment of the nutritive value of the legumes, judging by previous findings. Responses to soils and fertilisers generally agreed well with previous experience with these legumes, but assessments of responses to acidity, low Ca and salinity were not so successful, possibly because the species were tolerant of the conditions.

Mclvor et al (1988) determined the nutrient requirements of *S. hamata* cv. Verano on a eucrozem near Charters Towers, northern Queensland. In pot experiments responses to S, P and Cu were obtained, together with a lime-sulphur interaction. In the field, however, the only response obtained was to S. These soils offer good prospects for legume development.

**Table 8.2 General description of key visual symptoms of nutrient disorders in *Stylosanthes guianensis* CIAT 184.**

Nutrient disorder	Visual symptoms
<b>Deficiency</b>	
N	Chlorosis developing in older leaflets, followed by general chlorosis of the whole plant
P	Stunted growth; new leaflets are dark green to purple (instead of remaining dark green) and smaller than those of normal plants
K	Older leaflets show interveinal chlorosis with marginal and apical necrosis
Ca	New leaflets become white; growing points die and curl; older leaflets remain dark green
Mg	Interveinal chlorosis, developing into marginal chlorosis, occurring first on older leaves
S	Chlorosis of younger leaflets; older leaflets initially remain dark green. In severe deficiency, symptoms are similar to those of N deficiency
<b>Toxicity</b>	
Mn	Leaf margins of fully expanded leaves develop necrosis; leaflet veins become dark brown





**Visual symptoms of nutrient disorders in leaves of *Stylosanthes guianensis* CIAT 184 under greenhouse conditions at CIAT-Palmira, Cali, Colombia: (a) control; (b) N deficiency; (c) P deficiency; (d) K deficiency; (e) Ca deficiency; (f) Mg deficiency; (g) S deficiency; and (h) Mn toxicity.**

Rao et al (1996) compared the shoot and root growth responses of four tropical forages: *Brachiaria dictyoneura* (Fig. & De Not.) Stapf (grass), *Arachis pintoii* Krapov. & Greg., and *S. capitata* and *Centrosema acutifolium* Benth. The forages were grown in monoculture or in grass + legume associations at different levels of soil phosphate supply. The grass, grown either in monoculture or in association, responded more to P supply than did the three legumes in terms of both shoot and root production. At 50 kg/ha of P, the grass's yield per plant in association was greatly enhanced compared with that of the grass in monoculture. Among the three forage legumes, *S. capitata* produced the least amount of shoot biomass in association. The increase in the size of grass plants in association, compared with that in monoculture, may have been caused by reduced competition from the legumes, particularly from *S. capitata*. This study pointed out the need to take into account the differences in P responses between grasses and legumes when improving legume persistence in grass-legume associations.

Mohamed-Saleem and Otsyina (1986) used the subtractive technique in micronutrient trials on a ferruginous soil in the subhumid zone of Nigeria. They showed that DM yields of *S. hamata* cv. Verano from treatments lacking Cu or P were 64% less than that of the control, which received all nutrients. Yields were reduced by 13–23% with no applications of B, Mo, Zn, Mg, Ca and K; and 30–40% when Co or S was eliminated. Subtraction of S, Zn and Mo resulted in more severe yield reductions in the second year than the first. Nutrient composition of herbage varied widely according to treatment. Zinc levels were well below the critical limit required for plants and animals (12–18 ppm and 10–30 ppm, respectively). Positive interaction between P and Cu in terms of DM and yield was observed.

Thomas et al (1997) measured, over a three-year period, the proportion of N derived from fixation by  $^{15}\text{N}$  isotope dilution in three tropical grass-legume pastures on two Oxisols of different texture. Amounts of N fixed ranged from 0.3 to 40 kg N/ha every 12 weeks during the wet season and were greatest with *S. capitata*, followed by *C. acutifolium* and *A. pintoii*, mainly as a result of greater legume biomass in the first compared with the other two species. The percentage of N derived from the atmosphere (%Ndfa), ie from the fixation of atmospheric nitrogen, was generally greater than 80% in all legumes on both soil types. Results indicated that, in tropical pastures sown on low-fertility acid soils, the amounts of N fixed by forage legumes are dependent on legume growth and persistence. Based on these results, the authors recommended that approximate amounts of N fixed may be estimated by simply multiplying legume biomass N by 0.8.

Rao et al (1992) evaluated somaclonal variation in plant adaptation to acid soils in *S. guianensis*, and found significant differences in partitioning of fixed carbon between the shoot and roots, root biomass production, and uptake of N and phosphorus. Valarini et al (1997) found significant differences among somaclones of *S. scabra* in  $\text{N}_2$  fixation-related traits and suggested that somaclonal variation should be a useful source of nodulation-related variants.

### Other abiotic constraints

Growth and regeneration of summer-growing pasture legumes were studied on a black earth site near Dalby on the Darling Downs during 1982–85 (Keating & Mott 1987). Pure swards of some annual and perennial species (*M. atropurpureum* cv. Siratro, *M. lathyroides* cv. Murray, *S. scabra* cv. CPI 55856 and *Rhynchosia minima* cv. CQ2970) were productive (rainfed, 2–3 t/ha; irrigated, 4–6 t/ha) over three years. A moderately severe summer drought (to be expected on a 1-in-6 frequency), while severely limiting DM production relative to irrigated plots, did not adversely affect plant survival or regeneration in the following season. Waterlogging associated with normally high autumn and winter rainfall did prevent perennation, but regeneration from soil seed reserves was excellent and sward survival was not affected (4 t/ha after three years). The soil studied had a moderate level of subsoil salinity

(ECe of 3.6 dS/m at 115 cm) but the levels of chloride found in legume tissues (0.5–1.5%) were unlikely to limit growth or prejudice survival. Observations taken over the same period on pure grass plots (*Panicum coloratum* L. var. makarikariense cv. Bambatsi) revealed that the grass had a deeper and more extensive root system (459 cm/cm<sup>2</sup>) than did any of the legume species (eg Siratro 111–118 cm/cm<sup>2</sup>), was more effective in extracting soil water during dry periods, and gave DM yields of 4–16 t/ha and 9–12 t/ha with and without irrigation, respectively.

## Management Strategies for Biotic and Abiotic Constraints

Ensuring *Stylosanthes* growth and persistence can involve either cultural practices or genetic modification or a combination of the two.

Perhaps the most severe biotic constraint on *Stylosanthes* spp. is the grazing animal. In semi-arid Australia, where inherent biological productivity is limited by available moisture and stocking rates are consequently low, adapted *Stylosanthes* spp. (eg *S. scabra* and *S. hamata*) can naturalise and increase over time in associated pastures under grazing. In the humid tropics, which are more common in tropical America, where inherent biological productivity and hence stocking rates are higher, persistence of *Stylosanthes* in associated grazed pastures has been consistently disappointing, probably as a result of both grazing and trampling, as well as more vigorous growth of the associated grass under better moisture conditions. In many cases this lack of legume persistence may have little to do with the conventional biotic constraints of disease and pest susceptibility.

Genetic improvement and cultivar development in *Stylosanthes* have most commonly involved evaluation and selection of genotypes from diverse germplasm collections (Edye 1997; Miles & Lascano 1997). *Stylosanthes* breeding based on genetic heterogeneity arising from controlled hybridisation or opportunistic natural outcrossing has been more limited and less productive to date.

The first major review of *Stylosanthes* breeding was published in 1984 (Cameron et al 1984). Subsequent reviews (Cameron et al 1997; Miles & Grof 1997) paid special attention to breeding for anthracnose resistance, and Pereira et al (2001) recently reviewed *Stylosanthes* breeding from a Brazilian perspective. Not much applied plant breeding has been carried out recently, apart from B. Grof's persistent efforts to develop genetically heterogeneous cultivars of *S. capitata*, *S. macrocephala* (Grof et al 2001a) and *S. guianensis* (Grof et al 2001b).

### Breeding for disease resistance

A major focus of *Stylosanthes* improvement programs has been stable (durable) resistance to anthracnose. Breeding strategies have been consciously based on assumption of a highly heterogeneous pathogen population, a decision that has been amply confirmed by recent molecular data (Weeds et al 2003). These strategies involve deployment of one or more of the following:

- multiple anthracnose resistance genes in heterogeneous populations of *S. scabra* (Cameron et al 1997; Chakraborty et al 1991), *S. guianensis* (Grof et al 2001b; Miles & Grof 1997) and *S. capitata*-*S. macrocephala* (Grof et al 2001a; Miles & Grof 1997).
- multiple major resistance genes in a single genotype ('gene pyramiding') in *S. scabra* (Cameron et al 1997)
- quantitative ('polygenic' or 'partial') resistance in *S. scabra* (Cameron et al 1997; Chakraborty et al 1990) and *S. guianensis* (Miles & Grof 1997)

Wide inter- and intraspecific variation for reaction to anthracnose among *Stylosanthes* germplasm accessions has been observed (White et al 2001). Major resistance genes have been reported for *S. guianensis* and *S. scabra* in the Australian breeding programs (Cameron & Irwin 1986; Cameron et al 1997). Much of the observed phenotypic variation among accessions is probably quantitative. Evidence for segregation of major resistance genes was not observed in a natural *S. guianensis* population (Miles & Lenné 1984) nor in segregating generations in the *S. guianensis* breeding program conducted at CIAT (1985).

Highly anthracnose-resistant genotypes are readily identified, either in collections of natural germplasm or in hybrid breeding populations. Resistance 'breakdown' has been reported for selected progenies from the *S. capitata* breeding program (Miles & Grof 1997), but the observed susceptibility of selected lines at the late seed multiplication stage could have had other causes (Edye 1997; Miles & Grof 1997).

Despite success in isolating anthracnose-resistant lines, the success of the commercial cultivars released from *Stylosanthes* breeding programs has so far been limited because of cultivar defects (eg poor seed yield or poor persistence) that have no relationship with disease resistance. The recent release of cv. Campo Grande, a bi-specific and genetically heterogeneous population of *S. capitata* and *S. macrocephala*, appears to be meeting with at least some initial success (B. Grof, pers. comm., September 2003). Two *S. guianensis* cultivars developed by B. Grof from hybrid material provided by CIAT (Grof et al 2001b) are being commercialised in Australia (B. Grof, pers. comm., September 2003; Southedge Seeds Pty Ltd. 2003). It is too early to make a definitive judgment on the durability of anthracnose resistance in these new *Stylosanthes* cultivars, or of their ultimate commercial success.

### Breeding for tolerance of abiotic constraints

Abiotic constraints that have received attention in *Stylosanthes* breeding programs include low soil fertility (mainly low P), low pH, frost, flooding and drought (Cameron et al 1984). Tolerances of individual constraints are readily identified in diverse germplasm collections, so the challenge is to combine either required tolerances or tolerance(s) of one or more constraints with other attributes (eg forage yield and quality, seed yield) desirable in a successful cultivar. The task is formidable; for example, a long-term breeding project aimed at retaining desirable attributes (frost avoidance and drought resistance) of subtropical fine-stem stylo

(*S. guianensis* var. *intermedia*) while improving forage yielding ability and broadening environmental adaptation was eventually abandoned (Cameron et al 1997).

Thumma et al (2001) investigated associations among several physiological traits (TE,  $\Delta$  and SLA) that were supposedly related to drought resistance of *S. scabra*. Additionally, they identified quantitative trait loci (QTLs) for the physiological and production traits in an F<sub>2</sub> population in a greenhouse experiment. The authors suggest that the use of QTLs would permit 'pyramiding' of the different physiological components of drought resistance in a breeding program, but this hypothesis has not yet been confirmed.

### Genetic engineering approaches for *Stylosanthes* improvement

Transformation and regeneration protocols have been developed for *Stylosanthes* species and can be routinely done (Kelemu et al 2001; Manners 1987, 1988; Manners & Way 1989; Quecini et al 2002; Sarria et al 1994), making molecular manipulation of the genus feasible. The controversy over genetically modified plants aside, the advent of recombinant DNA technology and gene transfer systems allows the isolation and introduction of various genes from a potpourri of sources for several desirable agronomic traits. Genetic engineering not only widens the pool of useful genes (by removing the species barriers encountered in traditional plant breeding methods) for use in biotic and abiotic stress management, but also allows the use of several desirable genes in a single event, thus shrinking the time needed to incorporate novel genes into an elite plant background.

Resistance genes of plant origin, as well as those from other organisms, continue to be cloned and introduced into various plants to enable them to defend themselves from causal agents of biotic stresses (Hulbert et al 2001; Mourgues et al 1998; Rossi et al 1998; Schuler et al 1998; Tang et al 1999). Genomic approaches are increasing our understanding of the genetic basis of plant disease resistance by enabling us to better understand resistance genes themselves, as well as other defence-related genes, and the pathways they regulate. As our understanding of the molecular basis of host-pathogen interactions increases, so will our ability to modify plants with durable resistance to a wide range of pathogens (Cohn et al 2001; Dixon et al 1996; Martin et al 2003).

Because pests and pathogens possess several mechanisms for overcoming resistance, genetically engineered plants, like those developed through traditional plant breeding methods, will, eventually succumb to pest or pathogen pressures. Nevertheless, combining different forms of resistance from different organisms through modern technologies may offer an advantage in constructing a formidable barrier more difficult for the pest or pathogen to overcome.

## Contribution to Crop-Pasture-Fallow Systems in the Tropics

Countries in sub-Saharan Africa urgently need simple and self-reliant innovations to raise food and fodder productivity. Livestock suffer from protein deficiencies, particularly during the dry season. Dzowela and Baker (1993) reviewed advances in forage legume research in sub-Saharan Africa. In systems with minimal fertilisers, forage legumes have reduced soil fertility decline; in pastoral systems they contributed to rehabilitating the range and improving grazing quality; and in agropastoral systems they have improved the quality and thus the use of crop residues, natural pastures and fallow lands. Lack of seed and a variety of socioeconomic reasons comprise the major impediments to the adoption of forage legumes.

In the three broad categories of agroecosystems in sub-Saharan Africa, the legumes identified as being the most successful are:

- for humid and high rainfall areas—*Stylosanthes* spp., *Calopogonium* Desv. spp., *Centrosema pubescens*, *Desmodium* Desv., *Pueraria phaseoloides* (Roxb.) Benth. and *Neonotonia wightii*
- for drier conditions—*Macroptilium atropurpureum*, *Stylosanthes* spp. and *Medicago sativa* L.
- for the highlands and subtemperate areas—*Trifolium* Zohary & Heller spp.

Peters et al (1994) evaluated five accessions, comprising *Aeschynomene hystrix* Poirlet, *Centrosema acutifolium*, *C. pascuorum*, *S. guianensis* and *S. hamata* cv. Verano, over two years for use in fodder banks in subhumid Nigeria. The most promising accession identified was *A. hystrix* I12463, with yields of more than 6 t/ha DM in the second growing season, good drought tolerance, ability to compete with the native vegetation and high nutritive value.

Management options of *S. hamata* cv. Verano-based pastures were reviewed by de Leeuw and Mohamed-Saleem (1994). They also explored the changes needed to manage small-scale leys or fallows so they can serve as livestock feed and function as improved fallows to raise soil fertility in smallholder crop-livestock systems. Because it tolerates anthracnose, Verano stylo has become the most widespread species in West Africa and remains the mainstay of fodder-bank development in Nigeria, Mali and Côte d'Ivoire. In semi-arid Niger, it proved most promising for intercropping with millet on deep Arenosols. Management of large-scale Verano-based pastures has relied on, above all, intensive research originally developed for *S. humilis* in the monsoonal rangelands in Australia.

Similarly, in Nigeria low-input oversowing of Townsville stylo (*S. humilis*) was seen as a panacea for poor quality rangelands in the semi-arid and subhumid zones, adapting Australian technology to West African circumstances. *Stylosanthes hamata* was introduced in the early 1970s and proved superior to Townsville stylo when used for rangeland

improvement. Although the adoption rate of fodder banks in Nigeria has been encouraging, the concept is more adapted to medium-scale agropastoralists with as many as 50 head of cattle than to smallholders for whom cropping is more important than livestock production.

Research conducted to test the impact of forage legumes on livestock productivity in the subhumid zone of Nigeria showed that cattle grazing *Stylosanthes*-based pastures in the dry season produced more milk, lost less weight and had shorter calving intervals, and their calves survived better, than cattle grazing natural pastures (Tarawali & Mohamed-Saleem 1995). Goats grazing legume pastures had significantly less weight loss in the wet season. Both observations were attributed to the greater nutritive value of the forage legume relative to the natural pasture.

The N recycled by legume leys to subsequent crops was also assessed in bioassays. Results showed that N supplied by *Stylosanthes* to subsequent crops varied from 30 to 80 kg N/ha. Grain yields from areas previously cropped with legumes were always higher and, in some cases, double those from natural pasture. The superior performance of crops following *Stylosanthes* was associated with the legume improving the soil physical and chemical properties. The incorporation of forage legumes into cropping systems shows great potential for maintaining sustainable farming systems.

To enhance soil fertility and dry-season feed for cattle in crop–livestock farming systems in subhumid West Africa, Muhr et al (1999) suggested a short-term improved fallow system based on forage legumes. At two sites in southwestern Nigeria, the authors compared 13 legume species with the natural fallow vegetation. They evaluated establishment, DM yield and nutritive value of leguminous herbage in the year of establishment, and regeneration of legumes after a cropping phase. Only a few species yielded more dry season herbage than the natural fallow, but most had considerably higher N concentrations (0.6–2.4% of DM) and *in sacco* DM digestibility (DMD) (19–65%) than did the natural fallow vegetation (0.7–1.0% N and 28–30% DMD). In mid dry season *S. guianensis*, *Centrosema pubescens* and *Aeschynomene histrix* had the highest yields (10 t/ha DM) of standing herbage plus shed leaves at one site. The same species also showed good self-regeneration after a maize crop but were surpassed by other species with respect to N concentration and digestibility. Taking into account both yield and quality, *S. guianensis* was the most promising, despite fairly low N and P concentrations in the shoots.

Legume cover crops are a potential means of overcoming N depletion in the derived savanna of West Africa. Tian et al (2000) conducted a three-year trial near Ibadan, southwestern Nigeria to measure the N contribution of 13 legume cover crops and compare it with urea N, using a N fertiliser replacement index for a maize test crop. Two series of trials involved the following legume cover crop species: *Aeschynomene histrix*, *Centrosema brasilianum* (L.) Benth., *C. pascuorum*, *Chamaecrista rotundifolia*, *Cajanus cajan*, *Crotalaria verrucosa* L., *Cr. ochroleuca* G.

Don, *Lablab purpureus*, *Mucuna pruriens* (L.) DC, *Psophocarpus palustris* Desv., *Pseudovigna argentea* (Willd.) Verdc., *Pueraria phaseoloides* and *S. hamata*.

Cover crops increased grain yield of the subsequent maize crop by 25–136% over the control without N application. Nitrogen uptake by the maize crop was higher following cover crops than after maize or natural grass. Perennial (*C. brasilianum*, *S. hamata*, *Ca. cajan*, *P. phaseoloides* and *Cr. verrucosa*) and annual (*Ch. rotundifolia*, *M. pruriens*, *Cr. ochroleuca* and *L. purpureus*) species could potentially save 50–100 kg N/ha for maize crops. Results from this comparative study indicated that the N fertiliser replacement indexes can be predicted, using the above-ground biomass amount of cover crops at 20 weeks after planting (in a drier year) or the N concentration at that stage (in a wetter year).

Continued cropping seems impractical in tropical Africa without adequate replenishment of nutrients and organic matter in soils. Mohamed-Saleem and de Leeuw (1994) reviewed the progress on stylo-based pasture development for agropastoral production systems. *Stylosanthes hamata* cv. Verano was found to be particularly adaptable and resilient in agropastoral farming in the subhumid zone of West Africa. In on-station and on-farm trials, stylo has contributed significantly to livestock productivity and soil fertility. It is easily integrated into cropping systems and has improved food and fodder productivity on a sustainable basis. Even so, farmer adoption of forage legumes (eg *Stylosanthes*) into land use systems has been slow. These authors suggested the need for further research to determine whether this is due to ‘adoption lag’ or to a negative farmer evaluation of the benefits.

The performance of *Stylosanthes* species as animal feed or ley crop in tropical America was assessed by Miles et al (1994). They have considered results from both experiment station and on-farm research on selected farming systems of tropical America. Use of *Stylosanthes* spp. in the American tropics is essentially confined to extensive grazing systems, including oversowing into native range in improved grass–legume associations or as legume banks to supplement either native range or improved pure grass pastures. *Stylosanthes* is also being successfully integrated in crop–pasture production systems. The most important attributes of successful *Stylosanthes* cultivars, used as pasture or ley in tropical America, are resistance to disease, high seed yield and adaptation to low-fertility soils.

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# Chapter 9

## Anthracnose disease of *Stylosanthes*

Sukumar Chakraborty<sup>1</sup>

### Summary

The anthracnose disease has been the single most important factor to influence the development, productivity and persistence of *Stylosanthes* worldwide. The disease has rendered ineffective a large number of once promising cultivars that can no longer be grown commercially. Low per hectare monetary return and the need to maintain disease control over a large area for a long time has also restricted the choice of control options. Research and development, much of it by international multidisciplinary teams, has helped alleviate the anthracnose problem and allowed expansion of the use of this important tropical pasture legume over a large geographical area spanning at least five continents. Research has elucidated the genetic structure of the pathogen population through comparisons between centres of diversity and commercial utilisation. Demonstrations that races can arise convergently from different genetic lineages and highly aggressive strains can rapidly evolve under controlled experiments have highlighted the importance of genetic plasticity in this pathogen. Improved knowledge of pathogenic variation has underpinned breeding strategies to develop cultivars with broadly based resistance, mostly through selection. Molecular markers have improved efficiency of selection. Qualitative single-gene resistance has been more prone to breakdown by new pathogen virulence in several countries, and multilines consisting of resistant components and quantitative multi-gene resistance have proved useful. Knowledge of epidemiology, pathogen biology and host–pathogen interaction has also improved. Models based on weather dependence have been validated using data from field sites, often in different continents, and have provided a framework for predicting the risk of serious damage from anthracnose. This has allowed better targeting of *Stylosanthes* germplasm and cultivars to the various agroclimatic zones.

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### Introduction

*Stylosanthes* spp. originating mainly from Central and South America are among the most economically significant pasture and forage legumes in the tropical and subtropical regions. Because of their adaptation to acid and infertile soils in semi-arid environments, they have been introduced to many countries in Africa (de Leeuw et al 1994) and Asia (Guodao et al 1997) including Australia, to improve animal production and to restore soil nitrogen. In a rangeland environment *Stylosanthes* provides nutritious fodder to grazing animals such as cattle, goats and sheep. In Asia and Africa it is more commonly used as a cut-and-carry fodder or as dried hay or leaf meal preparations to feed poultry, pigs, ducks, fish and other farm animals. It has been widely adopted in northern Australia; an estimated 18 million ha is suited to this legume in the state of Queensland alone (Weston et al 1981). Currently nearly one million ha of grazing land in Australia is under *Stylosanthes*.

Anthracnose disease caused by the fungus *Colletotrichum gloeosporioides* is the most serious impediment to the commercial utilisation of *Stylosanthes* worldwide. The disease was first recorded at Deodoro in Brazil in 1937 on *S. humilis* (Anon. 1937) and is now widespread in all countries where this legume is grown. Anthracnose limits the number of otherwise well-adapted cultivars that can be grown in any region. Although some demonstrate a degree of partial resistance, no single cultivar can totally escape damage. The pathogen, which spreads via infected seeds (Davis 1987), can infect all aerial plant parts and has serious impacts on the establishment, production and persistence of all commercially significant species of *Stylosanthes*. Other diseases caused by fungi, bacteria, viruses and mycoplasmas are of relatively minor or regional economic significance.

The ability of *C. gloeosporioides* to quickly develop new races whenever new resistant varieties are deployed makes anthracnose a particularly difficult disease to manage. While many cultivars have been developed, none has resisted serious anthracnose damage for more than a few years. A number of recent international projects have increased knowledge of this disease required to underpin anthracnose management that previous reviews by Chakraborty (1997); Chakraborty, Cameron et al (1996); Lenné (1994); Lenné & Calderon (1984); and Irwin, Cameron & Lenné (1984) do not cover. This paper is intended as a timely summary of existing knowledge, together with new information, to provide an up-to-date treatise on anthracnose.

## Economic Significance of Anthracnose

Anthracnose affects biomass and seed yield, and production losses in affected pastoral areas can be up to 100% (Lenné 1986). Its impact on the seed- and forage-based grazing industry stems from gradual weakening of susceptible plants that are no longer productive or persistent. The Australian industry supplied seeds for the domestic and export markets before the advent of anthracnose, but this has been severely curtailed due to a fear of inadvertently spreading the pathogen via seeds. Anthracnose has reduced the seed production potential and general vigour in cultivars such as Seca, Verano and Amiga, which have some levels of partial resistance. A more insidious impact of anthracnose is a gradual decline of pasture and rangelands productivity and loss of ground cover, which often leads to soil erosion and land degradation. But the potential impact of anthracnose on the productivity and sustainability of a pastoral enterprise is largely unknown due to a lack of established quantitative links between disease severity and sustainable animal production (Chakraborty, Leath et al 1996).

The history of *Stylosanthes* utilisation around the world has been a testament to the devastation caused by anthracnose and nowhere is this more graphic than in Australia. In the early 1970s anthracnose wiped out half a million ha of naturalised *S. humilis* that covered almost 2 million ha in northern Australia, triggering an interest in the improvement and usage of this legume. This occurred soon after anthracnose was first observed in Australia on *S. fruticosa* in 1973 at Kalinga on the Cape York Peninsula (O'Brien & Pont 1977). *Stylosanthes humilis* cultivars Lawson, Gordon and Paterson, released prior to the advent of anthracnose, are all susceptible to the disease. Germplasm introduction, and evaluation and breeding programs in CSIRO and the various state departments of Agriculture have released a total of 15 cultivars from 5 *Stylosanthes* spp. in the past 30 years (Table 9.1). However, none has remained anthracnose resistant, as the pathogen has incessantly adapted to overcome host plant resistance. Anthracnose susceptibility has forced the abandonment of many cultivars and, today, commercial seed is only produced for six or seven cultivars. The two latest releases, Primar and Unica, are among a long list of once-resistant varieties that have succumbed to a new race of *C. gloeosporioides* within a very short time (Davis et al 1984), only two years after release in this instance. At the time of their release, glasshouse and field screening had demonstrated a high level of resistance for both Primar and Unica (Trevorrow et al 1998).

*Stylosanthes* cultivars have suffered a similar fate in South and Central America and many promising accessions and varieties have failed to persist for more than a few years (Miles & Lascano 1997; Trutmann 1994). In addition to many of the early Australian cultivars that were imported in the early 1970s but failed due to high anthracnose susceptibility, at least nine cultivars from three *Stylosanthes* spp. have been released in South America (Grof et al, 2001; Miles & Lascano,

**Table 9.1 Chronology of *Stylosanthes* cultivars released for commercial use in Australia.**

<i>Stylosanthes</i> species	Cultivar	Year of release
<i>S. guianensis</i> (Aubl.) Sw. var. <i>intermedia</i>	Oxley	1965
<i>S. guianensis</i> (Aubl.) Sw. var. <i>guianensis</i>	Schofield	1966
	Cook	1971
	Endeavour	1971
	Graham	1980
<i>S. humilis</i> (H.B.K.)	Lawson	1968
	Gordon	1968
	Paterson	1969
<i>S. hamata</i> (L.) Taub.	Verano	1973
	Amiga	1988
<i>S. scabra</i> Vog.	Seca	1976
	Fitzroy	1979
	Siran	1991
<i>S. seabrana</i> Maass & Mannetje	Primar	1996
	Unica	1996

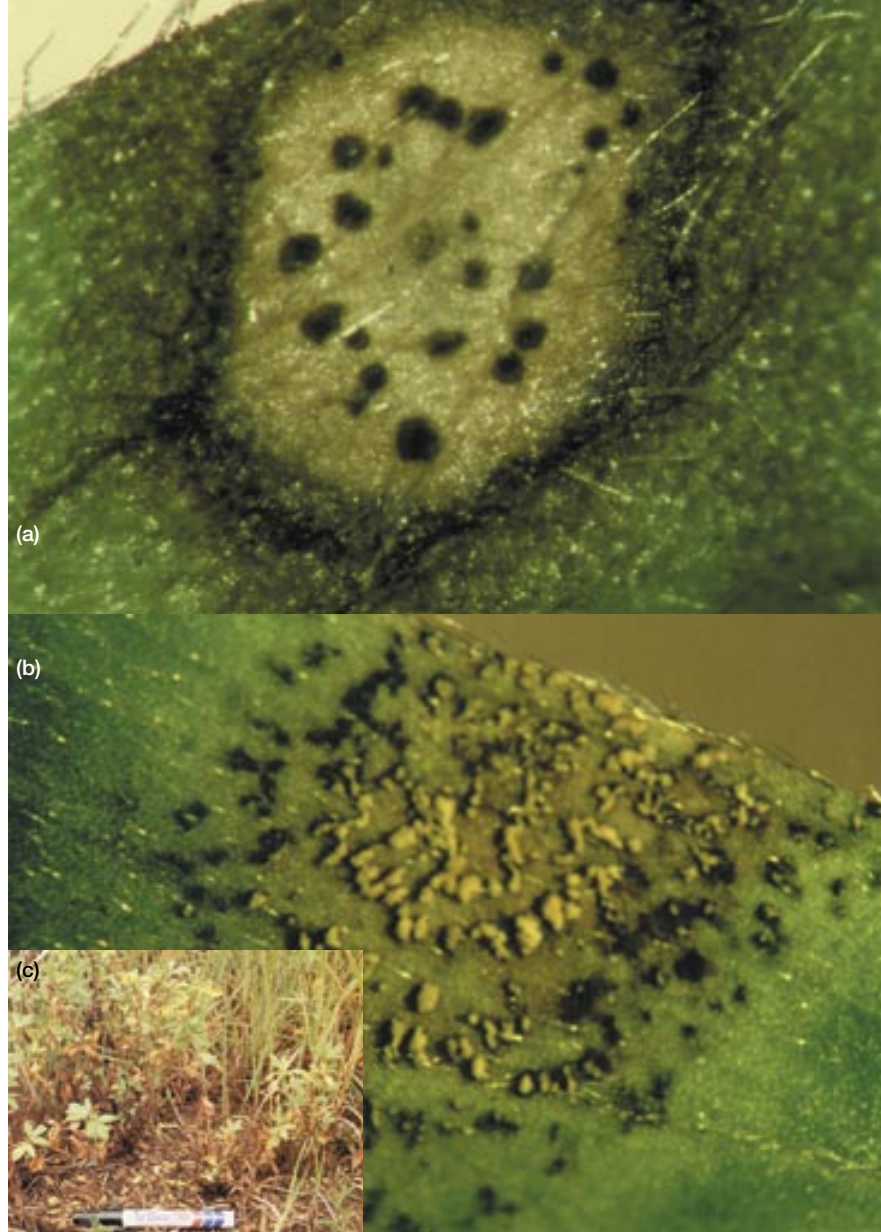
Sources: Cameron et al (1993); Eyles (1989); Oram (1990)

1997). Some did not have any commercial seed produced or much large-scale adoption. Others, including 'Capica', the *S. capitata* blend, and Mineirão, where some commercial seed was produced in the late 1980s, did not persist to support animal production beyond the first few years. However, there has been a recent resurgence in interest for Mineirão among pastoralists in Brazil (Ronaldo Andrade, pers. comm.). The latest release is a multiline cultivar 'Estilosantes Campo Grande' developed directly from natural germplasm accessions and released in 2001 (Grof et al 2001). Although it is too early to assess the success of this cultivar, glasshouse screening of component lines have shown several highly susceptible components (Celso Fernandes, pers. comm.).

In many Asian countries anthracnose seriously impacted on the first cultivars of *S. humilis*, *S. hamata*, *S. guianensis* and *S. scabra* introduced from Australia and other countries (Goudao et al 1997). These were soon replaced by more resistant varieties. For instance, in Thailand several *S. humilis* cultivars were replaced by *S. hamata* cv. Verano, and in China *S. guianensis* cv. Schofield was replaced by Graham, Cook and finally CIAT 184 (Guodao et al 1997). CIAT 184 has been increasingly affected by anthracnose and two new selections of CIAT 184, Reyan No 2 and No 10, were released in 2001. India is unique among countries outside the centre of diversity to have a native *S. fruticosa* population in the southern peninsular regions (Hooker 1879). However, apart from sporadic reports of anthracnose (Ramesh et al 1997), no systematic work had been done until recently (Ramesh et al, this volume). Similarly, no detailed assessment of anthracnose is available for Africa, except for its impact on seed production (Agishi 1994), despite a substantial amount of research and development activity (de Leeuw et al 1994). The economic impact of anthracnose in Asia and Africa is difficult to determine due to a lack of published records.

## The Pathogen

Worldwide *C. gloeosporioides*, teleomorph *Glomerella cingulata* (Stonem.) Spauld & Schrenk, is the predominant anthracnose pathogen, although typical symptoms can result from infection by *C. dematium* f.sp. *truncata* (Lenné & Sonoda 1978). The sexual stage does not play a significant role in the disease epidemiology and asexually produced conidia are the principal inoculum source. Conidia produced in acervuli in a mucilaginous matrix are prevented from dispersal by wind alone and require surface wetness for short distance dispersal through rain splash (Pangga et al, this volume). *Colletotrichum gloeosporioides* is externally seed borne, and the more widespread distribution occurs through infected seeds (Davis 1987) and via windblown rain droplets. Genetic plasticity of *C. gloeosporioides* has allowed founder populations to adapt to previously resistant varieties following its introduction to a new country/area.



Leaf lesions caused by *Colletotrichum gloeosporioides* biotype A on *Stylosanthes scabra* (a) and by biotype B on *S. guianensis* (b). Severe infection by biotype A causes extensive defoliation under suitable weather conditions to affect survival and persistence of infected stands (c).

## Biotypes and races – pathogenic variation

Pathogenic variation in *C. gloeosporioides* was identified in the 1970s with the discovery of two biotypes in Australia, distinguished by their symptoms, cultural and molecular characteristics, and host range (Irwin & Cameron 1978; Irwin, Cameron & Lenné 1984). Biotype A produces discrete lesions with grey centres and dark brown margins on all aerial plant parts, and infects all species of *Stylosanthes*. Biotype B mainly infects *S. guianensis*, causes a general necrosis of the terminal shoots with a blighting of affected plant parts, and produces lesions on leaves which are not clearly delineated. The two biotypes have distinct genetic fingerprints and carry different numbers of supernumerary chromosomes (Irwin & Cameron 1978; Manners et al 1992).



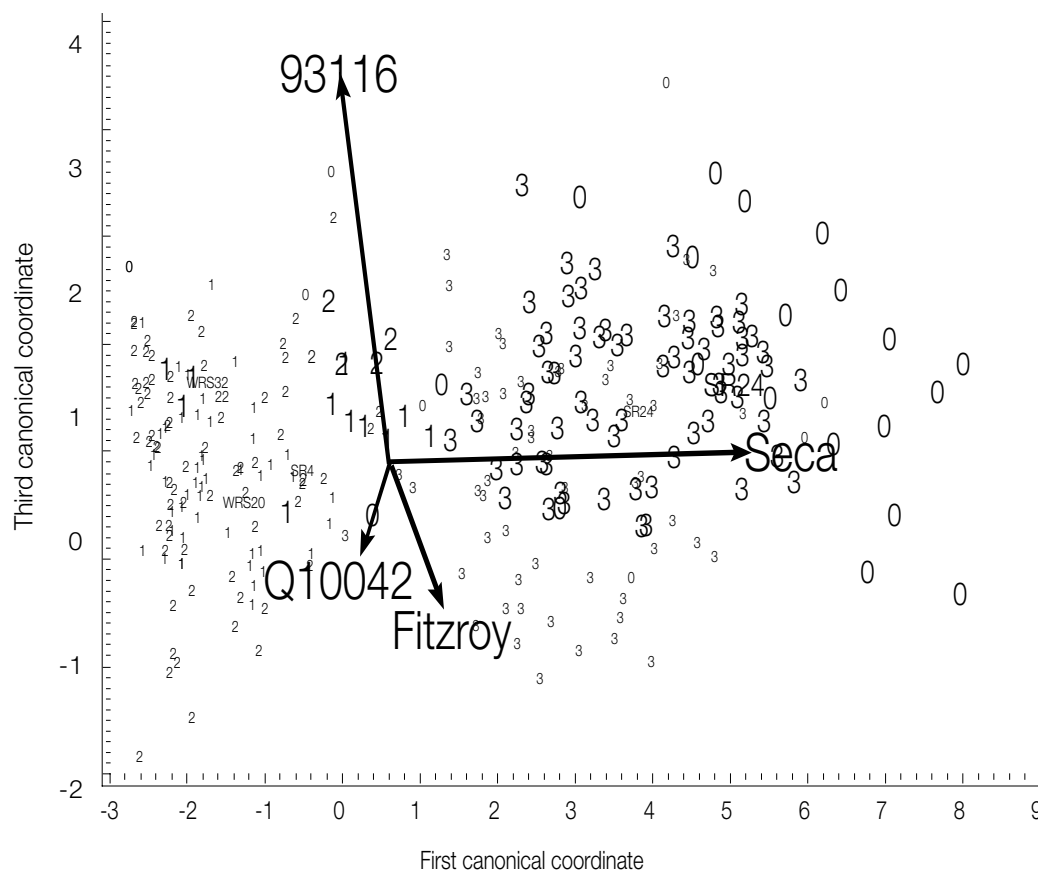
There is further pathogenic specialisation within each biotype in Australia (Davis et al 1987; Irwin, Cameron & Lenné 1984). Within biotype A there are currently six races: four on *S. scabra* and one each on *S. viscosa* and *S. seabrana*. In addition, there are four biotype B races in Australia. All races have evolved in response to the deployment of new resistant varieties; the latest, race 5, has appeared in response to the release of two *S. seabrana* cultivars, Primar and Unica (Trevorrow et al 1998).

Historically, pathogenic variation has been determined by qualitatively assigning resistant and susceptible classes to disease severity data using a threshold value following inoculation of a set of host differentials. Early work in both Australia and overseas used this approach (Chakraborty et al 1988; Davis et al 1984, 1987; Irwin & Cameron 1978). For host-pathogen combinations following a gene-for-gene specificity, such tests determine the virulence or avirulence (Shaner et al 1992) of a pathogen isolate on a given host differential, which is then used for race assignment. However, for pathogens such as *C. gloeosporioides* virulence also measures relative aggressiveness (Shaner et al 1992) of an isolate. Consequently, infection assays typically produce large experimental errors (Chakraborty & Jones 1993) and race assignment is complicated by the variation in the infection assay. Both the number of races and race assignment of individual isolates can change depending on the threshold used to designate virulence, making the identification of

races difficult. A multivariate technique using linear discriminant function analysis overcomes this by incorporating variation between infection assays in determining race clusters and assigning isolates to these clusters (Figure 9.1; Chakraborty, Thomas et al 1996).

At the centre of diversity there has been ample evidence of pathogenic variation in Brazil and Colombia, where previous research (Lenné 1988) has shown variation in anthracnose resistance within the native *S. capitata* population in Minas Gerais, thereby inferring variation in the pathogen population. Similarly, both qualitative and quantitative differences in virulence among isolates infecting the native *S. guianensis* population have been reported from South America (Miles & Lenné 1984). A common assumption in breeding programs has been that diversity in *C. gloeosporioides* in tropical America is immense (Miles & Lascano 1997). However, a rigorous analysis of pathogenic diversity has recently been completed for isolates originating from *S. guianensis* (Kelemu et al 1997, 1999); a set of 12 differentials has been developed to classify 57 pathotypes. More recently, 11 host differentials from five *Stylosanthes* species were used to analyse 296 *C. gloeosporioides* isolates infecting host species other than *S. guianensis* in Brazil (Chakraborty et al 2002). In this study eight race clusters were classified, with isolates remaining unclassified and therefore representing potential additional races.

**Figure 9.1** Classification of Australian isolates of *Colletotrichum gloeosporioides* into race clusters (large numerals) using linear discriminant functions analysis of their severity on a host differential of *Stylosanthes scabra*. Unclassified isolates are represented by a zero (large numeral). The four original axes (arrows) relate the log-transformed severity scores to the two canonical coordinates. The arrows originate from the point representing the overall mean severity and the length represents the change in canonical coordinates given a unit change from the mean score for each differential.



With over 96% of isolates from South-East Asia, Africa, Australia and South America from host species other than *S. guianensis* producing typical biotype A symptoms, and all isolates from *S. guianensis* producing biotype B symptoms, there is strong evidence of the existence of *C. gloeosporioides* biotypes outside Australia (Davis et al 1990). More recent work has confirmed this (Chakraborty et al 2002). However, 11 out of 104 blight-inducing biotype B isolates from *S. guianensis* also infected the *S. scabra* cultivar Fitzroy (Kelemu et al 1999), which is not a host for this biotype in Australia (Irwin & Cameron 1978). This indicates that the distinction between the two biotypes in South America may not be as clear cut and there may be more than two biotypes.

Only limited studies have been made on the pathogen population outside Australia and South America. In India eight race clusters have been detected in 277 presumed biotype A isolates examined so far using eight host differentials (Ramesh et al, this volume). Of these, the isolate population collected from the native *S. fruticosa* contains five of the six races. This suggests that the native host species did not give rise to any specific race, and races have most likely arisen as a consequence of growing introduced varieties. In China five potential races of the putative biotype A pathogen have been detected in 25 isolates using nine host differentials (Kexian et al, unpublished information).

Although morphology, growth optima and pathogenicity data demonstrate a greater pathogen diversity outside Australia (Davis et al 1992), a direct comparison of races between countries is not possible due to different host differential sets having been used to classify isolates in each country. Pathogenic diversity in Brazil is unexpectedly limited and most of the putative biotype A isolates can be grouped into one of only eight race clusters. Interestingly, more complex races are largely distributed at or near research stations where *Stylosanthes* germplasm have been screened for a long time (Chakraborty et al 2002), and mostly simple races are found in wild-host populations, indicating a host directed selection towards more complex races. However, in countries outside the centre of diversity *C. gloeosporioides* has rapidly diversified into a number of races following the release of new cultivars, despite its limited gene pool. Pathogenic diversification is expected to intensify at centres of diversity, with more widespread use of specific cultivars over a wide area.

### Genetic variation and its source

The two Australian biotypes are genetically distinct and can be differentiated by restriction fragment length polymorphisms (RFLP) (Braithwaite et al 1990), double stranded RNA (Dale et al 1988) and the number and size of supernumerary chromosomes (Masel et al 1990). Biotype A isolates have five large chromosomes (2–>6 Mb) and 8–10 ‘mini-chromosomes’ (270–600 kb) whilst biotype B isolates have three large chromosomes (4.7–6 Mb) and 2–5 mini-chromosomes (300–1200 kb) (Masel et al, 1990). RFLP analysis using dispersed repeats and low copy sequences from *C. gloeosporioides* and heterologous rDNA

and DNA fingerprinting probes clearly shows the difference between the two biotypes (Manners et al 2000).

There is considerable genetic variation within each biotype, as demonstrated using isozyme (Lenné & Burdon 1990), RFLP (Manners et al 1992), electrophoretic karyotype (Masel et al 1993) and RAPD (Chakraborty et al 1999; Kelemu et al 1999; Weeds et al 2003) markers. Genetic diversity in Brazil and Colombia is extensive, followed by China and India, with the Australian pathogen population being the least diverse, probably due to its geographical isolation and an effective quarantine. There were 87 haplotypes among the 144 isolates from Brazil and Colombia, and 82 haplotypes among the 95 Indian isolates, and all 43 Chinese isolates had unique haplotypes. The extensive diversity in China is unexpected given that organised introduction and evaluation of *Stylosanthes* spp. only started in the 1960s (Guodao et al 1997). In Brazil and India, both with native *Stylosanthes*, a high level of genetic differentiation in the pathogen population was found at sites with a native or naturalised host population. Interestingly, many isolates from germplasm evaluation sites in Brazil that were pathogenically diverse (Chakraborty et al 2002) had identical haplotypes (Weeds et al 2003). Overall, genetic diversity is more extensive at sites where the host has been grown for a long time (Kelemu et al 1999), allowing the pathogen time to diversify.

The mechanisms that generate genetic variation in *C. gloeosporioides* are not well understood. Although perithecia of its sexual stage (*Glomerella cingulata*) are often found on dead *Stylosanthes* stems, highly virulent teleomorphic forms have not been recorded and all virulent isolates are anamorphic (Ogle et al 1986). RFLP and double-stranded RNA data suggest little or no recombination between biotypes (Manners et al 1992). However, transfer of supernumerary chromosomes between the biotypes has been demonstrated in the laboratory (He et al 1998).

Overall, no close relationship has been found between a race and its genetic grouping based on isozyme (Lenné & Burdon 1990) or molecular markers (Chakraborty et al 1999; Kelemu et al 1999; Weeds et al 2003). This indicates that the same race can arise convergently from different genetic lineages, and helps to explain the number of races that have arisen in some countries away from the centre of diversity despite the pathogen being genetically less diverse. Although groupings of isolates from two genetic marker systems can be very similar, these do not match virulence groupings for the isolates (Chakraborty et al 1999). Many avirulence gene families are highly conserved (Leach et al 1995), while selectively-neutral regions can evolve at a fast rate. Thus, a concordance between a genotype, based on selectively-neutral regions, and a race, determined by avirulence genes, can only be obtained by analysing genes which control avirulence or are closely related to these genes.



Consequently, association between molecular markers and virulence patterns in plant pathogens can be perfect, partial or absent (Leung et al 1993), and close association between virulence and selection-neutral molecular markers has only been recorded in a limited number of pathogens (Crowhurst et al 1991).

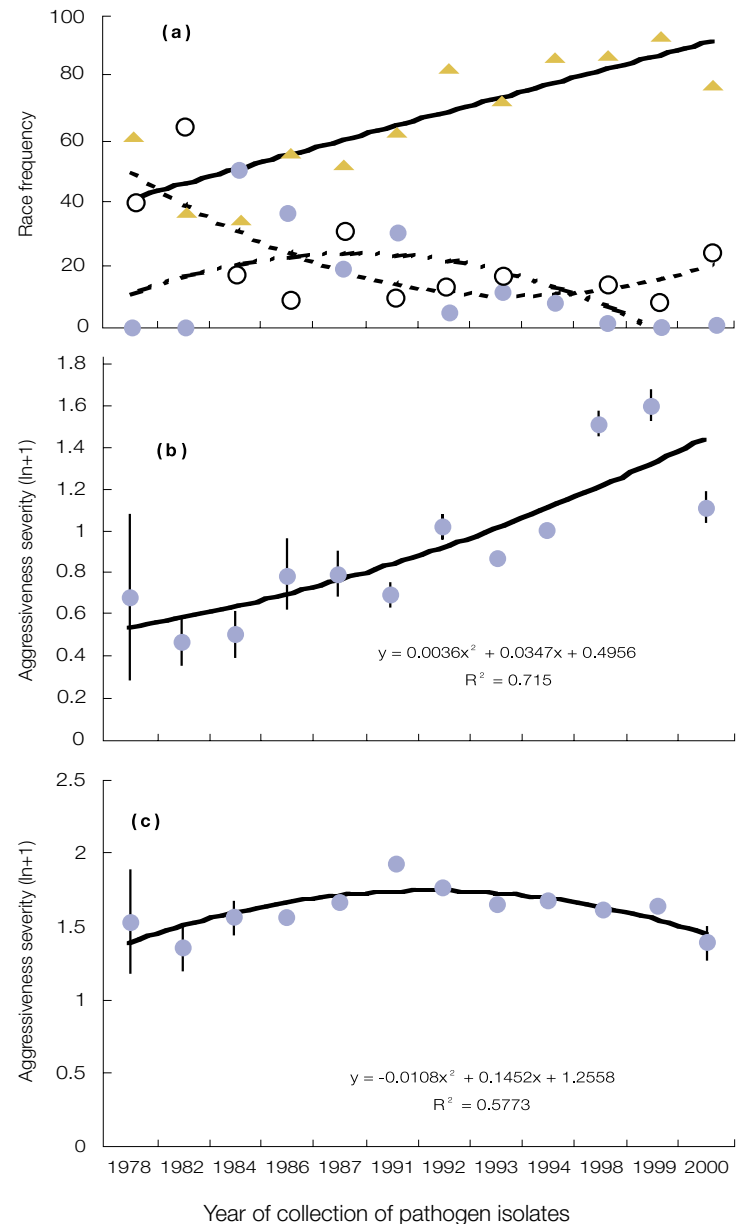
## Pathogen evolution

Virulence, the genetic ability of a pathogen race to overcome genetically determined host resistance, and aggressiveness, a property of the pathogen reflecting the relative amount of damage caused to the host without regard to resistance genes (Shaner et al 1992), together determine the overall fitness of a pathogen population. Some pathogens can become virulent on new cultivars in only five asexual cycles (Alexander et al 1985). Similarly, in some pathogens aggressiveness can increase in only three sexual generations (Kolmer & Leonard 1986). Despite anecdotal evidence on the appearance of new and more aggressive races in Australia (Chakraborty et al 1999; Davis et al 1987; Irwin, Cameron & Lenné 1984; Trevorrow et al 1998), race frequency dynamics and the evolution of highly aggressive strains have only recently been experimentally demonstrated (Chakraborty & Datta 2003). From surveys of commercial pastures spanning 22 years between 1978 and 2000, an analysis of virulence on six host differentials of more than 1700 isolates has shown that the frequency of simple races 1 and 2 has declined over this period, while race 3 frequency has steadily increased (Figure 9.2). During this period pathogen aggressiveness has increased on Seca but not on Fitzroy. Prior to 1987 the population consisted of weak to moderately aggressive isolates, but between 1991 and 1999 most isolates were aggressive to highly aggressive towards Seca (Figure 9.2). The increased aggressiveness on Seca is a reflection of its dominance in commercial pastures in Australia and not an indication of the influence of host resistance. When evolution was studied by inoculating two isolates onto Seca and Fitzroy over 25 sequential infection cycles, both isolates increased their aggressiveness on both cultivars over the generations (Figure 9.3) (Chakraborty & Datta 2003).

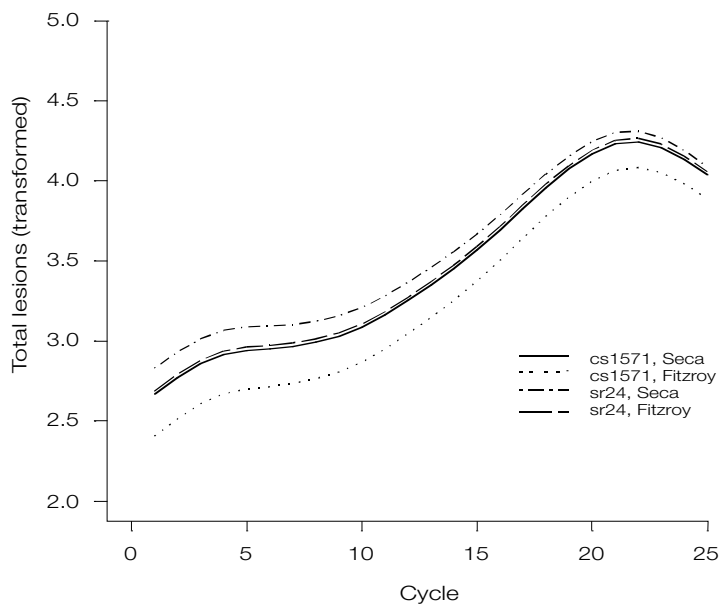
## Epidemiology

The life cycle of this necrotrophic pathogen (Irwin, Cameron & Lenné 1984) is restricted to *Stylosanthes* and a limited number of tropical pasture legumes (Vinijsanun et al 1987), and to saprophytic growth and survival on crop residue on the soil surface (Boland et al 1995). Infection is generally initiated from seed-borne inoculum that can be easily controlled by fungicide application (Davis 1987). *Colletotrichum gloeosporioides* produces conidia in acervuli in a mucilaginous matrix (Louis & Cook 1985) and requires free surface water to suspend the conidia and thereby make them available for splash dispersal. Pangga (2002) used single drops of simulated rainfall in a rain tower and exposed disease-free plants at different distances from infected plants in a field to determine  $d_{50}$ , the distance at which the number of conidia decreases by half. His results showed that  $d_{50}$  was 2–3 cm under single drop

impaction, 6–11 cm under simulated rain and 2–6 m in the field. On occasions, conidia are spread up to 10 m in the field without rain. Lesions are detected up to 10 cm away from inoculum source under simulated rain, but up to 5 m under field conditions. Hourly mean temperature and rainfall intensity significantly influenced dispersal in the field. This study



**Figure 9.2** Changes in race frequency and aggressiveness of *Colletotrichum gloeosporioides* population collected from the field during a 22-year period and tested on *Stylosanthes scabra* cultivars in a glasshouse. (a) Changing frequency of race 1 (○), 2 (●) and 3(▲); (b) changing aggressiveness on the partially resistant cultivar Seca; and (c) changing aggressiveness on the susceptible cultivar Fitzroy.



**Figure 9.3** Changing aggressiveness of *Colletotrichum gloeosporioides* isolates sr24 and cs1571 with 25 sequential infection cycles on *Stylosanthes scabra* cultivars Seca and Fitzroy.

has shown that although splash droplets may spread the pathogen locally, conidia in the field are dispersed to greater distances than expected for dispersal by splash alone, and other mechanisms also play a role in their dispersal. For instance, the pathogen produces conidia on fertile setae in acervuli (Lenné et al 1984) and on hyphal conidiophores (Cox & Irwin 1998), which can be easily dislodged by air movement and therefore available for aerial dispersal.

In controlled environments infection occurs within a temperature range of 20–30°C if surface wetness over 24 hours is available (Irwin, Cameron & Ratcliffe 1984). Although disease develops over a range of surface wetness duration, disease severity increases with increasing wetness duration due to improved infection efficiency (Chakraborty et al 1990). Disease severity is also unaffected by brief interruptions in surface wetness if relative humidity stays above 85%. Following artificial inoculation, disease develops in 3–7 days on leaves (Chakraborty et al 1988) and within 3–12 days on stems (Iamsupasit et al 1993). Lesions start producing secondary conidia by 3–12 days and this continues depending on the host resistance, pathogen isolate and environmental conditions (Iamsupasit et al 1993).

Infection in the field is associated with the availability of water but heavy rainfall reduces severity, possibly due to washing off of conidia (Davis et al 1987). Besides rain, other water-related weather variables such as relative humidity over 95% and net evaporation can also explain a significant amount of variation associated with anthracnose development

(Chakraborty & Billard 1995; Chakraborty & Smyth 1995). Recent work has shown the superiority of artificial neural networks over the more commonly used regression models in predicting weather dependence of anthracnose (Chakraborty et al 2003). Models trained on data from field sites within one continent correctly predicted disease severity at field sites in another continent. Although neural networks can be robust and useful to predict disease severity over a broad range of field sites, they can be difficult to train and still require computing resources not yet widely available. More traditional multiple regression models have formed the basis of weather-based frameworks to target cultivars and germplasm to different agroclimatic zones (White et al 2001).

## Host-Pathogen Interaction

Biotype A and B isolates differ in the timing and percentages of conidial germination and penetration (Irwin, Trevorrow & Cameron 1984; Ogle et al 1990; Trevorrow et al 1988; Vinijsanun et al 1987). Infection proceeds in a similar fashion for anamorphic *C. gloeosporioides* and teleomorphic *G. cingulata* (Ogle et al 1986) but anamorphic isolates produce more successful penetrations than teleomorphic forms. This complements the finding that teleomorphic strains have relatively low virulence (Irwin, Trevorrow & Cameron 1984). Germination and penetration occur in both compatible and incompatible interactions involving specific race host differential combinations, but compatible interactions have higher percentages. Differences in the extent and rate of fungal growth between compatible and incompatible interactions become evident after 48 hours of inoculation. Histological manifestations of resistance and first signs of active host defence are apparent after 24–48 hours in incompatible interactions (Trevorrow et al 1988). These include deposition of apposition layers on the inner surface of epidermal walls, aggregation of granular cytoplasm beneath appressorium, and browning of cell wall and contents (Ogle et al 1990).

Callose deposition is associated with resistance to biotype B infection of *S. guianensis* (Sharp et al 1990). Other host responses, including peroxidase and chitinase activity and genesis of disease-related proteins, were induced during infection but were either unique to compatible interactions or occurred too late in incompatible interactions to be active in resistance. Host-specific peroxidase is induced within 24 hours of inoculation of *S. humilis* and precedes penetration of the epidermal cell wall (Harrison et al 1995). A coordinated expression of specific pathogen genes has been demonstrated for the anthracnose pathosystem in recent years (Manners et al 2000). Of these, a glutamine synthetase gene expressed during early stages of infection is induced by nitrogen deprivation in axenic culture (Stephenson et al 1997). By averting hypersensitive response, this gene plays a role during the biotrophic phase of the pathogen (Stephenson et al 2000).

## Genetics of Resistance

Knowledge of genetics of resistance largely comes from breeding programs conducted in Australia at the CSIRO (Cameron et al 1997) and in South America by CIAT (Miles & Grof 1997). The genetic structure of the pathogen population in these two continents has strongly influenced findings. However, a rigorous analysis of the number of genes controlling resistance to either biotype has not been carried out outside Australia. In the absence of any segregation for major genes in *S. guianensis*, quantitative resistance has been assumed at the centre of diversity in Brazil and Colombia (Miles & Lenné 1984). In Australia at least three loci with dominant alleles for resistance have been identified from reactions of *S. guianensis* against biotype B races 1 and 3 (Irwin, Cameron & Lenné 1984), while some accessions show oligogenic resistance (Cameron & Irwin 1983). Similarly, both quantitative and qualitative (major gene) resistance against the biotype A pathogen has been detected in Australia in *S. scabra* (Cameron & Irwin 1983; Chakraborty et al 1988). *Stylosanthes scabra* Seca carries two dominant resistance genes; one confers complete resistance while the second controls partial resistance (Irwin, Cameron & Lenné 1984; Irwin et al 1986). Some *S. viscosa* accessions also have resistance to certain biotype A races in Australia (Irwin et al 1986). *Stylosanthes hamata* resistance to biotype A in Australia, on the other hand, is predominantly inherited as a quantitative trait, and a large proportion of the genetic difference among accessions is additive (Iamsupasit et al 1995).

## Management of Anthracnose

The spatial heterogeneity in the distribution of pathogen races in Brazil (Chakraborty et al 2002) indicates that complex pathogen races are restricted to certain sites when cultivars with specific resistance genes are not widely deployed. *Colletotrichum gloeosporioides* spreads through splash-dispersed conidia (Pangga 2002) and can be externally seed borne. Seed infestation can be easily treated using common fungicides (Davis 1987) and spread through rain splash is limited to short distances (Pangga et al, this volume). The extensive genetic diversity of *C. gloeosporioides* populations (Weeds et al 2003) poses a greater threat to widely adopted cultivars as new races can evolve from different clonal lineages. Management strategies must consider pathogen diversity and avoid cultivars with single gene resistance to minimise the all too familiar boom and bust cycle.

The use of genotype mixtures has produced variable results, with some trials showing promise for *S. guianensis* mixtures (Lenné 1985) while others developed for *S. scabra* showed no yield or anthracnose advantage over pure stands of components (Chakraborty et al 1991; Davis et al 1994). Similarly, cultivar Capica, a blend of five *S. capitata* accessions, did not persist beyond two years in commercial pastures (Miles & Lascano 1997). This is not unexpected since mixtures generally do not offer any protection against splash-dispersed pathogens (Wolfe 1985). In particular, mixtures made up of susceptible components do

not restrict anthracnose development or spread, but if all components are resistant to one or more races, these can offer long-term protection. Cultivar 'Siran', released in Australia in 1995, is a composite of three bred *S. scabra* lines with superior anthracnose resistance and yield compared to the commercial cultivar Seca (D.F. Cameron, unpublished). The main advantage of *Stylosanthes* mixtures appears to be in reducing the rate of evolution of damaging complex races (Davis et al 1994).

Forms of quantitative resistance that reduce the rate of epidemic development have been successfully used for anthracnose management in Australia in both *S. hamata* (Iamsupasit et al 1991, 1993) and *S. scabra* (Chakraborty et al 1988; Smyth et al 1992). Advanced breeding lines developed using a recurrent selection protocol to incorporate several sources of quantitative resistance against all existing races have been developed as an insurance against a breakdown of resistance in current commercial varieties (Cameron et al 1997). The development and use of molecular markers has greatly helped to better understand phylogenetic relationships and identify different germplasm (Kazan et al 1993; Liu et al 2000).

## Future Prospects and Challenges

*Stylosanthes* anthracnose has arguably become one of the best-studied diseases of pasture and forage plants, mainly as a result of a sustained effort by a team of international researchers focused on important aspects. Strong international collaboration and a multidisciplinary approach have generated useful and new knowledge on the host, pathogen and environmental aspects of the disease triangle to facilitate the development of sustainable management options. The genetic structure, virulence profile and evolution of the pathogen population have been characterised; understanding of anthracnose epidemiology improved; and anthracnose-resistant and high-yielding cultivars developed and shared between countries by virtue of a free and open germplasm exchange program. The exchange of germplasm and an active collaboration between research, development and extension personnel from a number of countries have undoubtedly made the biggest impacts on *Stylosanthes* technology in Australia, Thailand, the Philippines, Indonesia, China and India, among others. All cultivars in these countries have originated from introduced germplasm. Improved understanding of the pathogen population has meant that countries can now assess the risks from exotic races and take steps to minimise their accidental introduction. These linkages must be maintained for obvious reasons, as only regular monitoring can keep pace with the 'arms race' common in most host-pathogen interactions. Unfortunately, in recent years research effort on anthracnose has been severely curtailed or ceased altogether in Australia and at CIAT in South America. The problem is compounded by a declining number of pasture pathologists worldwide (Cameron & Lenné 1994). However, there is now increased emphasis on *Stylosanthes* in Brazil, China and India with recent or imminent release of new cultivars. Plant protection professionals will need to extend their

horizons beyond disciplinary and geographical boundaries to solve new and ongoing challenges and problems.

## Acknowledgments

Much of the Australian research findings come from work by Nipon Iamsupasit, Peter Wilson, Ireneo Pangga, Ross Perrott, Raylee Trevorrow and other postgraduate students; and postdoctoral researchers Drs. Pauline Weeds and Neal White. The Australian Meat and Livestock Corporation, the Australian Centre for International Agricultural Research, the Australian Rural Industries Research and Development Corporation, the Australian Grains Research and Development Corporation, the Australian National Greenhouse Advisory Committee, the Australian Government Department of Industry Science and Technology, the Commonwealth Scientific and Industrial Research Organization and the Cooperative Research Centre for Tropical Plant Protection, among others, funded much of the research reported in this review. I am indebted to Drs Segenet Kelemu, John Miles and Peter Kerridge, from CIAT; Drs Celso Fernandes, Maria Jose Charchar and Ronaldo Andrade from EMBRAPA; Drs C. R. Ramesh, P.S. Pathak, N.P. Melkania, Amaresh Chandra and C.R. Hazra from IGFRRI; Mr Liu Guodao, Yi Kexian and Bai Changjun from CATAS; Mr Bob Davis and Ian Staples from QDPI; Drs Don Cameron, Bob Clements, Raymond Jones, Bruce Pengelly, John Manners, Kemal Kazan, Chunji Liu and other colleagues from CSIRO for input into ideas and discussions. Fiona McKay, Ross Perrott, Toh he Wong, Leonard Ford and John Baisden provided technical assistance. All assistance is gratefully acknowledged.

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