

## ***Stylosanthes* species in north-east Thailand: dry matter yields and seed production**

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### **Abstract**

A field experiment was conducted in north-east Thailand between 2003 and 2006 to compare dry matter yields of *Stylosanthes hamata* cv. Verano, *S. guianensis* cvv. Tha Phra, Ubon stylo and Temprano, and *S. scabra* cv. Seca. A second field experiment in 2004 compared the effect of closing date defoliation on subsequent seed yields of Tha Phra and Ubon stylo.

Ubon stylo and Tha Phra produced at least 90% more dry matter than Verano over 3 years, with advantages particularly obvious in the dry season, when they produced 2–6 times the yield of Verano. Ubon stylo and Tha Phra produced 13, 18 and 17 t/ha/yr DM, in the first, second and third years of production, respectively. Temprano produced intermediate dry matter yields, being generally inferior to Ubon stylo and Tha Phra, but superior to Verano and Seca. Both Ubon stylo and Tha Phra would be suitable replacements for Verano.

Ubon stylo produced 2.6 times the seed yield of Tha Phra (959 vs 365 kg/ha). Closing in September doubled seed yield over closing in October.

Management of perennial stylo pastures to increase the commercial use of stylo pastures is discussed. Ubon stylo's stronger resistance to anthracnose, better grazing persistence and higher seed yields than Tha Phra, suggest it would be a better replacement for Verano.

### **Introduction**

The first commercial use of *Stylosanthes* spp. in Thailand, in the mid 1960s, was based upon the importation of seed of Australian cultivars, chiefly Townsville stylo (*Stylosanthes humilis*). Following the devastating outbreak of the fungal disease anthracnose (*Colletotrichum gloeosporioides*) in 1976, Verano (*S. hamata*), which was more resistant than Townsville stylo, replaced it and large-scale pasture development ensued.

Verano has been the backbone of pasture legume development for animal production in north-east Thailand for 30 years. Since 1976, the Department of Livestock Development (DLD) has improved nearly 320 000 ha of communal grazing land by oversowing Verano (Phaikaew and Hare 2005). Over 4000 t of Verano seed has been harvested in north-east Thailand since 1976 and annual production has stabilised at about 50–100 t/yr.

Perennial stylo (*S. guianensis* var. *guianensis*) has been used to a lesser extent than Verano in Thailand for cut-and-carry forage systems. Cultivars Cook, Schofield and Endeavour were evaluated by the DLD in the 1970s, but were replaced by the better performing cv. Graham in the 1980s and 1990s. Anthracnose destroyed Graham in 1996, and the more resistant and higher-producing CIAT 184 (Tha Phra stylo) was substituted (Phaikaew and Hare 2005). It performs very well in the humid tropics (Miles and Lascano 1997).

With the future possibility of both Verano and Tha Phra stylos succumbing to anthracnose, there was a need to evaluate anthracnose-resistant species. In November 1999, we received seed of a blend of lines derived from *S. guianensis* var. *vulgaris* × var. *pauciflora* (ATF 3308) selected for long-term resistance to anthracnose (Grof *et al.* 2001). This blend has been released in Australia as cultivar Nina (Cook *et al.* 2005). However, in Thailand, farmers growing seed and forage call it

“Ubón stylo” and we have continued to use this name since its release in 2002.

In 2001, we received seed of another anthracnose-resistant blend (GC 1576 plus GC 1524), deriving from *S. guianensis* var. *vulgaris* × var. *pauciflora* hybrid material (ATF 3309 cv. Temprano). This selection is less erect than Ubón stylo, similar to Graham, and more suited to dry environments (B. Grof, personal communication).

The hypothesis of this research was that stylos could be identified, which were more productive in Thailand than cv. Verano. The objective was to compare wet and dry season forage production of the new stylos with that of Verano.

Since stylo adoption in Thailand has been a result of successful seed production (Hare and Phaikaew 1999), any new stylo species would need good seed production characteristics. Hence, an additional objective was to compare seed yields of Ubón stylo and Tha Phra.

## Materials and methods

Two field experiments were conducted from 2003 to 2006 in Ubón Ratchathani province, north-east Thailand (15°N, 104°E; 130 m asl; AAR 1593 mm) on the Ubón Ratchathani University farm in a 0.15 ha field. The sites were on an upland sandy low humic gley soil (Roi-et soil series) (Mitsuchi *et al.* 1986). Soil samples to 10 cm, taken at sowing in May 2003, showed that the soil was acid (pH 4.3; water method), and low in organic matter (1.5%), N (0.03%), P (3.0 ppm; Bray II extraction method), S (9 ppm) and

K (54.5 ppm). Prior to cultivation, the site had been planted to *Digitaria milanjiana* cv. Jarra for 3 years and ruzi grass, mixed with Verano stylo, for 6 years (Hare *et al.* 2004b).

### Trial 1 – Dry matter yields

Five *Stylosanthes* cultivars (*S. hamata* cv. Verano, *S. guianensis* var. *guianensis* cv. Tha Phra, *S. guianensis* var. *vulgaris* × var. *pauciflora* cv. Ubón stylo, *S. guianensis* var. *vulgaris* × var. *pauciflora* cv. Temprano and *S. scabra* cv. Seca) were compared in a 6-replicate, randomised complete block, field experiment. The site was ploughed in March and April 2003 and then rotary hoed to produce a fine seed-bed the day before planting in May 2003. Details of field management are summarised in Table 1.

Seed was tested for germination immediately before sowing and all accessions were sown at a rate of 10 kg/ha germinable seed. The exception was Seca, which, because of limited seed supply, was sown at 5 kg/ha.

At each sampling date, herbage was cut to 10 cm from ground level, weighed fresh and sorted into stylo and weeds. A 300 g stylo sub-sample was taken and dried at 70°C for 48 hours to determine moisture content. Dried samples were stored for analysis for crude protein, acid detergent fibre (ADF) and neutral detergent fibre (NDF) concentration. After each sampling cut, the remaining herbage was cut to 10 cm and removed.

**Table 1.** Field crop management of stylo evaluation trial (Trial 1).

Field cultivation	Ploughing × 2, discing × 1, harrowing × 1
Plot size	8 m × 5 m
Sowing date	May 16, 2003
Sowing method	Broadcast
Establishment plant counts	6 weeks after sowing; four 0.25 m <sup>2</sup> quadrats/plot
Sampling cuts	Eight 0.25 m <sup>2</sup> quadrats/plot
First wet season	Aug 20 & Oct 29, 2003
First dry season	Feb 20 & Apr 28, 2004
Second wet season	Jun 28, Aug 30 & Oct 29, 2004
Second dry season	Mar 4 & Apr 25, 2005
Third wet season	Jun 29, Aug 29 & Oct 28, 2005
Third dry season	Jan 24 & Apr 25, 2006
Fertiliser	
At sowing	20 kg/ha P, 50 kg/ha K, 20 kg/ha S
After cutting	20 kg/ha P, 50 kg/ha K, 20 kg/ha S
	2003 Aug 20 & Oct 30
	2004 Apr 28, Aug 30 & Oct 29
	2005 Apr 25, Aug 29 & Oct 28

### Trial 2 – Seed production

Seed production from *S. guianensis* cultivars (cvv. Tha Phra and Ubon stylo) was compared using 3 closing date defoliation treatments (no cutting from sowing, cutting on September 1 and cutting on October 1, 2004) in a 5-replicate, randomised complete block field experiment. The field was ploughed and disced into a fine seed-bed in April and May, 2004 and the trial was planted on June 11, 2004. Seed was tested for germination (Ubon stylo 67% and Tha Phra 47%), and was broadcast at a rate of 5 kg/ha germinable seed on to plots measuring 3 m × 3 m (1 m walkways between plots) and raked into the soil. Fertiliser (P 20 kg/ha, K 50 kg/ha, S 20 kg/ha) was applied at sowing and again in October.

At each closing date (September 1 and October 1), four 0.25 m<sup>2</sup> quadrats were cut at 10 cm above the ground and weighed fresh. A 300 g subsample was taken to determine moisture content and crude protein. The remaining herbage on the plots was cut to 10 cm and removed.

Plots were harvested for seed from February 21–23, 2005. At harvest, the herbage was beaten with sticks to make all remaining seed fall to the ground before all herbage on each plot was cut and removed. Each plot was swept with brooms and the collected material sieved and winnowed to collect the stylo seed. The seed was further cleaned in the laboratory and a purity test performed on each seed lot. Seed yields were corrected to 10% seed moisture and 100% pure seed.

Data from each trial were analysed by conventional analysis of variance and treatment means compared by LSD at the  $P = 0.05$  probability level.

## Results

### Rainfall

Rainfall at the trial site was below the 14-yr mean of 1538 mm/annum in the first two years of the study but similar to the mean in the third year (Table 2). The second year, 2004, was particularly dry, with rainfall 30% below the mean and wet season rain finishing early, in mid-September. In contrast, very heavy rainfall (433 mm) fell in September 2005 and 144 mm of rain fell during January–April 2006.

### Trial 1 – Dry matter yields

All stylo cultivars, except Seca, established well, with seedling counts >250/m<sup>2</sup> (Table 3). Seca plant numbers were much lower at 46/m<sup>2</sup>.

In the first wet season, Tha Phra produced more dry matter ( $P < 0.05$ ) than Verano, Temprano and Seca (Table 3). Weeds, mainly *Zornia* spp. constituted 80% of the herbage in the Seca plots. In the following seasons, volunteer seedlings of Verano replaced the weeds and by the third year, Verano constituted nearly 90% of the herbage in the Seca plots.

Dry matter yields of Tha Phra and Ubon stylo were higher ( $P < 0.05$ ) than those of other cultivars in the following seasons, except for Temprano in the third year (Table 3). Temprano outyielded Verano and Seca in most seasons.

Crude protein concentrations of Verano were higher than those of Ubon stylo, Temprano and Seca in the first and third wet seasons and

**Table 2.** Rainfall at Ubon Ratchathani University during the study and the 14-yr mean (1992–2005).

Month	Rainfall				
	Mean	2003	2004	2005	2006
			(mm)		
Jan	1	0	0	0	0
Feb	12	42	0	0	7
Mar	30	55	1	5	29
Apr	81	99	53	87	108
May	216	249	143	152	
Jun	249	234	206	278	
Jul	254	112	150	229	
Aug	278	335	297	285	
Sep	295	276	188	433	
Oct	90	28	4	26	
Nov	29	0	3	50	
Dec	4	0	0	0	
Total	1539	1430	1045	1545	

**Table 3.** Plant counts 6 weeks after sowing and dry matter yields of 5 stylo cultivars.

Cultivar	Plant counts	Dry matter yield							
		Wet 2003	Dry 2003–04	Wet 2004	Dry 2004–05	Wet 2005	Dry 2005–06	3 year means Wet	Dry
	(no/m <sup>2</sup> )	(kg/ha)							
Verano	253	5580	2800	8336	536	4280	3669	6065	2335
Tha Phra	306	7746	5863	14423	3718	10638	6245	10936	5275
Ubon stylo	381	6993	5857	14899	3459	10673	6396	10855	5236
Temprano	264	5521	4363	12692	2724	8963	5383	9059	4157
Seca	46	1443	3293	9035	1562	4738	1925	5072	2260
LSD (P<0.05)	62	1816	1282	1380	556	1896	1381	1690	1084

higher than those of all cultivars in the second wet season (Table 4). In the dry season, CP concentrations in Verano tended to be lower than those of other cultivars but differences were not always significant. Crude protein concentrations of all cultivars were, on average, 4.4 (Seca) to 8.0 (Verano) percentage points higher in the wet season than in the dry season.

In the second and third wet seasons, ADF (30.8 and 31.7%) and NDF (40.9 and 32.5%) concentrations in Verano were lower than in other cultivars. In the dry season, ADF concentrations in Verano (39.3–45.0%) tended to be higher than those of other cultivars but NDF concentrations in Verano (49.6–53.8%) were similar to those of other cultivars.

#### Trial 2 – Seed yields

Seed yields of Ubon stylo were 2.6 times those of Tha Phra, but with slightly lower 1000-seed weight (Table 5). Seed crops cut in September had twice the seed yield of crops cut in October.

Ubon stylo produced more dry matter than Tha Phra at closing for seed production, but this forage was 1 percentage point lower in crude protein concentration than Tha Phra (Table 6).

Closing in October produced 3 times as much forage, but with lower crude protein concentrations, as closing in September.

#### Discussion

This research has shown that 2 perennial stylo cultivars (Ubon stylo and Tha Phra) produced higher dry matter yields than Verano in both wet and dry seasons, 90% more over 3 years. These advantages were especially pronounced in the dry season, when Ubon stylo and Tha Phra produced 2–6 times the yield of Verano. These large differences in dry matter yields were because the perennials stayed green and productive throughout the year, whereas Verano, a biennial, dried off or died in the dry season. This is particularly important as stylo cultivars that have superior dry season production and broad adaptability and are more suitable for cut-and-carry forage than Verano, are of special interest to farmers in Thailand.

Dry matter production from Ubon stylo and Tha Phra in this trial was excellent. Ubon stylo and Tha Phra produced 13, 18 and 17 t/ha/yr DM in the first, second and third years of production, respectively. These yields are far greater

**Table 4.** Crude protein concentrations of 5 stylo cultivars.

Cultivar	Crude protein					
	Wet 2003	Dry 2003–04	Wet 2004	Dry 2004–05	Wet 2005	Dry 2005–06
	(%)					
Verano	20.0	12.4	20.9	12.6	22.1	13.9
Tha Phra	19.2	14.3	19.6	15.5	22.0	15.6
Ubon stylo	18.4	14.5	19.7	16.4	20.9	14.6
Temprano	18.2	13.9	19.4	14.9	20.8	14.5
Seca	17.1	13.2	16.9	14.1	19.6	13.1
LSD (P<0.05)	1.2	ns	1.1	1.2	1.0	0.9

**Table 5.** Effect of cultivar and time of closing on seed yields and seed weights of 2 stylo cultivars.

Treatment	Seed yield	TSW
<i>Cultivar</i>	(kg/ha)	(g)
Ubon stylo	959	2.50
Tha Phra	365	2.59
LSD (P<0.05)	425	0.06
<i>Time of final closing cut</i>		
Control (not cut)	618	2.60
September	921	2.53
October	453	2.50
LSD (P<0.05)	449	0.08

than those of Tha Phra in other trials in Thailand (Phaikaew *et al.* 2004). Of special interest was that the dry matter yields of Ubon stylo and Tha Phra were equal to yields produced by the grasses *Brachiaria* spp. (Hare *et al.* 2005) and *Digitaria milanjiana* cv. Jarra (Hare *et al.* 2004b) in earlier trials on the same site. Moreover, the stylos produced twice the amount of crude protein as the grasses and with no applied fertiliser nitrogen.

Dry matter yields of Temprano, which is also a perennial, were generally lower than those of Ubon stylo and Tha Phra but higher than Verano and Seca. Temprano was selected as a more prostrate perennial stylo, suitable for grazing in low rainfall areas (AAR 700–1000 mm) (B. Grof, personal communication). It has multi-gene resistance to anthracnose and could be a good replacement for Verano in drier environments, should the single-line resistance of Verano (Iamsupasit *et al.* 1995) succumb.

While production from Seca was lower than that of the other 3 perennial stylos, it did produce 4.7, 10.6 and 6.7 t/ha/yr DM, in the first, second and third year of production, respectively. These yields are above those normally produced by Seca on dry, infertile soils and equal to yields obtained on more fertile soils under lenient cutting (Cook *et al.* 2005). Since seed supply was limited, Seca was sown at 1.15 kg/ha germinable seed, compared with the other stylo cultivars, which were sown at 10 kg/ha germinable seed. If Seca had been sown at the higher sowing rate, it may have been more productive. Seca is normally grown in association with perennial sown and native grasses, which are grazed. It is not well adapted to cut-and-carry management due to slow establishment, low palatability and poor persistence in cut-and-carry systems.

The forage quality of Verano was generally better than that of Ubon stylo and Tha Phra (higher CP) in the wet season because of fresh new forage

**Table 6.** Dry matter yield and crude protein concentration in stylo forage at time of closing seed crops.

Treatment	Dry matter	Crude protein
<i>Cultivar</i>	(kg/ha)	(%)
Ubon stylo	2562	21.3
Tha Phra	2114	22.4
LSD (P<0.05)	353	0.8
<i>Time of final closing cut</i>		
September	1054	22.4
October	3622	21.3
LSD (P<0.05)	353	0.8

produced by young plants, which grew from fallen seed. However, in the dry season, the crude protein concentration of Verano decreased rapidly as it senesced and died. In contrast, the crude protein concentrations in Ubon stylo and Tha Phra remained good throughout the dry season. Both Ubon stylo and Tha Phra seem to be superior to Verano for use by smallholder farmers, with the higher yields of better quality fodder in the dry season being a critical advantage.

However, a major limitation of perennial stylos in Thailand has been their lack of persistence and decline in productivity beyond 2 years (Hare *et al.* 2003; 2004a), because of continual hard grazing or close to ground level, persistent cutting. Since cutting height was 10 cm and there was a good recovery period in our trial, all stylo species were still growing very well at the end of the third year and, except for Seca, maintaining a very strong plant density in the plots.

Farmers in Thailand use sown pastures for cut-and-carry forage rather than for grazing. They prefer to plant grasses rather than legumes, because of higher forage yields (Phaikaew and Hare 2005). However, if Ubon stylo and Tha Phra produce the yields in villages that we achieved in our research plots, there may be more farmer interest in sowing stylos, especially given the higher forage quality of the legumes and the savings in applied nitrogen fertiliser.

In order to increase stylo adoption by farmers, particular attention must be given to promoting correct cutting management to maintain stylo stand density and persistence. Cutting after plants have become tall and woody can kill the plants, since there are few growing points close to the ground on mature plants (Cook *et al.* 2005). We stimulated lower branching early, by cutting to 10 cm 3 months after sowing and then cutting at the same height at 2-month intervals in the wet

season and at longer periods in the dry season. After 3 years, the Ubon stylo, Tha Phra and Temprano plots were still dense and almost weed-free.

While Ubon stylo and Tha Phra produced similar dry matter yields in this trial, the single-line resistance of Tha Phra to anthracnose may cause it to succumb to anthracnose in the future. The multi-line resistance to anthracnose of Ubon stylo may make it persist better than Tha Phra. Tha Phra also has the tendency to become more woody with age than Ubon stylo, which makes it more vulnerable to dying out. Ubon stylo stems are a lot finer than Tha Phra stems and may be part of the reason for its persistence in grazed pastures. Cattle graze both leaves and stems of Ubon stylo, but with Tha Phra they prefer leaves, leaving the stems to become large and woody (M. Hare, unpublished data).

To be adopted by farmers, new stylo species need to be good seed producers. Ubon stylo is a prolific seeder, producing over 950 kg/ha, nearly 3 times the seed yield of Tha Phra. Phaikaew *et al.* (2004) also reported that Ubon stylo produced more seed than Tha Phra.

This study has reinforced earlier findings on the importance of closing strategy on both seed and forage production of perennial stylos. Excessive vegetative growth in perennial stylo seed crops can produce moisture stress during flowering, resulting in lower seed yields (Hare 1985). Cutting to 40 cm in August produces shorter plants at flowering and reduces moisture stress, allowing a heavier seed set (Hare 1985). Loch *et al.* (1976) showed that the timing of perennial stylo defoliation was critical and was best carried out about 4 weeks before first-flower initiation, although timing may differ by cultivar. In South America, cutting too low or too late in the wet season has been a frequent cause of failure of perennial stylo seed crops (Ferguson *et al.* 1999).

The major advantage of defoliation compared with not defoliating for smallholder farmers is that forage can be fed to livestock. In the current study, cutting Ubon stylo and Tha Phra in October, after the first flowers appeared, decreased seed yields to half that produced by defoliating in September. The undefoliated plots did not suffer from moisture stress, even though they produced a considerable bulk between flowering and harvest.

If farmers can develop the skills to manage perennial stylos as a sown pasture and a legume protein bank for dry season forage, there will be

more widespread use of perennial stylo. With proper cutting management, Ubon and Tha Phra perennial stylos can produce excellent regrowth and can persist strongly for at least 3 years and could be ideal replacements for Verano. Ubon stylo's stronger resistance to anthracnose, better grazing persistence and higher seed yields than Tha Phra, make it the better choice.

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