# ILC2018 Plenary paper\*

# **Leucaena cultivars – current releases and future opportunities** *Cultivares de leucaena – estado actual y oportunidades futuras*

SCOTT A. DALZELL

Leucaena Research and Consulting Pty Ltd, Port Macquarie, NSW, Australia.

# Abstract

The Leucaena genus is made up of 24 different species (19 diploid and 5 tetraploid species). However, early use of the Leucaena genus in agricultural systems was based entirely upon a very narrow germplasm base. A single genotype of Leucaena leucocephala ssp. leucocephala ('common' leucaena) was spread pantropically from its center of origin in Mexico over 400 years ago. Genetic improvement of Leucaena leucocephala began in the 1950s, when vigorous 'giant' leucaena (L. leucocephala ssp. glabrata) was identified in Australia and Hawaii. Cultivars such as Hawaiian Giant K8, Peru and El Salvador were selected and promoted for grazing in Australia and multipurpose agroforestry uses throughout the tropics. Plant breeding for improved forage production resulted in the release of cv. Cunningham in 1976 in Australia. These cultivars of 'giant' Leucaena leucocephala displayed broad environmental adaptability, with the exception of poor tolerance of cold temperatures (and frost) and acid soils. The outbreak of the psyllid insect pest (*Heteropsylla cubana*) from Cuba during the 1980s devastated both 'common' and 'giant' leucaena all around the world. This challenge resulted in renewed interest in lesser-known Leucaena spp. that exhibited tolerance to the pest and in interspecific hybridization as a means of developing new cultivars. Some 'giant' leucaena lines exhibited excellent agronomic traits and a degree of tolerance to the psyllid pest and this resulted in the release of new cultivars in Australia (cvv. Tarramba and Wondergraze) and Hawaii (cv. LxL). Since the 1990s, plant breeding programs have sought to develop cultivars with greater psyllid tolerance using interspecific hybridization. This has resulted in the release of cv. 'KX2-Hawaii' for timber and forage production, and a backcrossed forage cultivar cv. Redlands (Australia). Both cultivars are based upon interspecific hybridization between L. pallida and L. leucocephala ssp. glabrata. Cold-temperature and acid-soil tolerance have been pursued in South American breeding programs based upon L. diversifolia, without commercial success. The development of sterile Leucaena spp. cultivars is currently underway to nullify the environmental weed potential of all current commercial cultivars. Tolerance to cold temperatures (L. diversifolia, L. pallida, L. pulverulenta and L. trichandra), frost (L. greggii and L. retusa) and psyllids (L. collinsii) exists within the Leucaena genus and may be exploited in future hybridization programs. New genetic analyses and molecular plant breeding techniques have the potential to facilitate further gene transfer between Leucaena spp. for the development of the next generation of multipurpose cultivars.

Keywords: Hybridization, plant breeding, psyllid resistance, tree legumes.

# Resumen

El género *Leucaena* está compuesto por 24 especies diferentes (19 diploides y 5 tetraploides). Sin embargo, en su primera fase el uso del género *Leucaena* en sistemas agropecuarios se basó exclusivamente en una estrecha base de germoplasma. Un solo genotipo de *Leucaena leucocephala* ssp. *leucocephala* (leucaena 'común') fue el que que hace más de 400 años se dispersó pantropicalmente desde su centro de origen en México. El mejoramiento genético de *Leucaena leucocephala* comenzó en la década de 1950, cuando se identificó una vigorosa leucaena 'gigante' en Australia y Hawái, *L. leucocephala* ssp. *glabrata*. Cultivares como Hawaiian Giant K8, Peru y El Salvador fueron seleccionados y promovidos para pastoreo en Australia y usos agroforestales múltiples en todo el trópico. Un programa de fitomejoramiento buscando mayor rendimiento de forraje resultó en la liberación del cv. Cunningham en 1976 en

Australia. Los cultivares del tipo 'gigante' de Leucaena leucocephala mostraron una amplia adaptabilidad a las condiciones ambientales, con excepción de tolerancia a temperaturas bajas (incluyendo heladas) y suelos ácidos. El brote del insecto plaga Heteropsylla cubana (Psyllidae) durante la década de 1980 tuvo un efecto devastador en las leucaenas 'común' y 'gigante' en todo el mundo. Este desafío dio lugar a un renovado interés en especies menos conocidas de Leucaena que mostraran tolerancia a la plaga, y en la hibridación interespecífica como medio para desarrollar nuevos cultivares. Algunas líneas de leucaena 'gigante' exhibieron excelentes características agronómicas y cierta tolerancia a la plaga de los psílidos, lo que dio lugar a la liberación de nuevos cultivares en Australia (cvv. Tarramba y Wondergraze) y Hawái (cv. LxL). Desde la década de 1990, programas de fitomejoramiento han buscado desarrollar cultivares con mayor tolerancia a los Psyllidae utilizando la hibridación interespecífica. Como resultado se liberó el cv. 'KX2-Hawaii' para la producción de madera y forraje, y cv. Redlands en Australia, un cultivar forrajero retrocruzado. Ambos cultivares están basados en la hibridación interespecífica entre L. pallida y L. leucocephala ssp. glabrata. En Sudamérica se llevaron a cabo proyectos de mejoramiento basados en L. diversifolia buscando tolerancia a temperaturas bajas y suelos ácidos, sin embargo sin éxito comercial. Proyectos actualmente en curso tienen como objetivo desarrollar cultivares de Leucaena spp. estériles para eliminar el potencial de maleza ambiental de los actuales cultivares comerciales. Dentro del género Leucaena sí existen características como tolerancia a temperaturas bajas (L. diversifolia, L. pallida, L. pulverulenta y L. trichandra), a heladas (L. greggii y L. retusa) y a los psílidos (L. collinsii) y se podrán explotar en futuros programas de hibridación. Las nuevas técnicas disponibles de análisis genético y reproducción molecular de plantas tienen el potencial de facilitar la transferencia de genes entre especies de Leucaena con el fin de desarrollar la próxima generación de cultivares multipropósito.

Palabras clave: Fitomejoramiento, Heteropsylla cubana, hibridación, leguminosas arbóreas, resistencia a plagas.

#### History

Utilization of multipurpose trees from the 24 species of the Leucaena genus (Abair et al. 2019) has been occurring for millenia in subsistence agricultural systems in seasonally dry forest areas throughout their native range extending from southern Texas, USA to northern Peru (Hughes 1998). In the 16<sup>th</sup> century, Spanish colonists in Central America recognized the potential of leucaena as an animal forage and began the spread of 'common' weedy leucaena (L. leucocephala ssp. leucocephala) throughout the tropics (Gray 1968; Brewbaker 2016). 'Common' leucaena is a small branchy tree with low biomass yield, poor form, early flowering and heavy seed production (Gray 1968). This remarkable plant has wide adaptability to a range of soil types and climatic conditions, where it has become established in disturbed environments (Campbell et al. 2019; Idol 2019). 'Common' leucaena has been utilized by subsistence smallholder farmers pantropically to produce fuelwood, timber, green manure, shade, animal forage and human food. It has also been trialled for use in commercial agriculture as a fodder for ruminant animals (Takahashi and Ripperton 1949; Kinch and Ripperton 1962).

During the 1950s agronomists and plant breeders in Australia and Hawaii began programs to identify and develop superior leucaena cultivars for adoption in commercial agricultural systems (Gray 1968; Brewbaker 2016). Seed of 'common' leucaena was collected from

disparate areas and evaluated. It soon became apparent that 'common' leucaena lacked diversity in key agronomic characteristics (Gray 1968), indicating that this phenotype was genetically identical all around the world, having originated from a narrow genetic base. This was later confirmed by molecular genetic analysis (Sun 1992).

Advances in genetic improvement followed the identification and commercialization of 'giant' types of leucaena (L. leucocephala ssp. glabrata) in Hawaii (Hawaiian Giant K8 - 1975, K28, K29, K67 and K72; Brewbaker et al. 1972) and Australia (El Salvador - 1962, Peru – 1962, Tarramba – 1997; Gray 1968; Oram 1990). The 'giant' types had superior vigor/yield and less precocious seed production (Hutton and Gray 1959; Brewbaker et al. 1972; Brewbaker 1975). Tree form varied within the 'giant' types, with some accessions being arboreal (Hawaiian Giant K8, El Salvador and Tarramba), while others had a greater degree of basal branching (Peru). These early cultivars of 'giant' leucaena have been widely distributed around the world for use in tropical agroforestry systems. A comprehensive, authoritative review of the history of genetic improvement of the Leucaena genus has been compiled by Professor J.L. Brewbaker, University of Hawaii (UH) (Brewbaker 2016).

Plant breeding programs have combined the superior attributes of different accessions of 'giant' leucaena, with breeding objectives including: increased forage yield and branched tree form suitable for direct grazing; and more recently, tolerance of the psyllid insect. Cultivar Cunningham (public domain cultivar in Australia) is an intraspecific hybrid based upon cv. Peru and was released as a forage type by the Commonwealth Scientific and Industrial Research Organisation (CSIRO) within Australia in 1976 (<u>Oram 1990</u>). It was selected for superior forage yield and branched form (<u>Hutton and Beattie 1976</u>). Cunningham has been widely planted in Australia and around the world.

The emergence of a devastating pantropical psyllid insect pest (*Heteropsylla cubana*) during 1983–1990 (Bray 1994), triggered a second phase of genetic evaluation and cultivar development. A number of accessions of 'giant' leucaena had moderate tolerance to the psyllid. These formed the basis of the following cultivars:

- cv. LxL: A synthetic line of 6 intraspecific hybrid forage breeding lines released by the University of Hawaii (UH) in 1996 (Austin et al. 1998). Despite having superior forage yield (~15% heterosis), cv. LxL has had limited commercial utilization in the USA (Brewbaker 2016).
- cv. Tarramba (protected by PBR in Australia): A bred line (UH) from accession K636 collected from highlands in Coahuila, Mexico (Brewbaker 2016) and released in Australia in 1997 (Anonymous 1997). Key attributes of cv. Tarramba are: erect arboreal habit; excellent biomass and forage production; some cool-temperature tolerance; moderate tolerance of the psyllid insect pest; and reduced seed production. Cultivar Tarramba has been readily adopted in smallholder ruminant feeding systems in Indonesia (Kana Hau and Nulik 2019), where its erect stems are valued for fuelwood and construction timber.
- cv. Wondergraze (protected by PBR in Australia): Selfed progeny (S4) from an intraspecific cross between accession K584 and cv. Tarramba bred by UH and released in Australia in 2010 (Anonymous 2008). Key attributes of cv. Wondergraze are: moderate tolerance of the psyllid insect pest; good forage yield; branched tree form; and excellent seedling vigor.

#### Environmental limitations to 'giant' leucaena

The following environmental constraints restrict the productivity of 'giant' leucaena: defoliation by frost; poor growth under cool temperatures; and lack of tolerance of acid soils (<u>Hutton 1983</u>). While 'giant' leucaena can survive severe frost by regrowing from the root crown during spring (<u>Felker et al. 1998</u>), minor frost (0 to -3 °C) burns the leaves from plants and moderate frost (<-3 °C) kills stems to ground level (<u>Dalzell et al. 1998a</u>;

Middleton and Clem 1998;). This restricts the ability of farmers in subtropical areas to utilize 'giant' leucaena forage during the winter protein feed gap, when it would be of tremendous benefit to livestock production. Growth of 'giant' leucaena slows significantly when average daily temperatures drop below 25 °C and average monthly minimum temperatures drop below 22 °C (Mullen et al. 2003c), restricting forage production during spring and autumn in subtropical areas and year-round production in the elevated tropics. 'Giant' leucaena thrives on neutralalkaline calcareous soils. It grows poorly on acid soils (pH water 1:5 < 5.2) due to calcium and phosphorus deficiency and aluminum toxicity adversely impacting root growth, rhizobium nodulation and nitrogen fixation (Hutton 1983). There are large tracts of acid soils in tropical areas that would otherwise be suitable for 'giant' leucaena development.

#### Interspecific hybridization

Many of the lesser-known Leucaena spp. have agronomic traits that address the limitations to adaptation of L. leucocephala ssp. glabrata, including: psyllid resistance (L. collinsii, L. esculenta and L. pallida) (Mullen et al. 2003b); cold tolerance (L. diversifolia, L. pallida and L. trichandra) (Mullen et al. 2003c); and frost tolerance (L. greggii, L. pulverulenta and L. retusa) (Hughes 1998). However, these species cannot be commercialized directly in agroforestry systems because they have other serious limitations to utility such as low biomass/forage yield (L. greggii and L. retusa) (Mullen et al. 2003a), poor forage quality (L. diversifolia, L. esculenta, L. greggii, L. pallida, L. pulverulenta and L. trichandra) (Dalzell et al. 1998b; Jones et al. 1998) and potentially a lack of longevity or tolerance of regular defoliation (L. esculenta, L. greggii, L. pallida and L. retusa) (Mullen et al. 2003a).

A high degree of interspecific cross compatibility has been identified within the *Leucaena* genus (Gonzalez et al. 1967; Sorensson and Brewbaker 1994). Interspecific hybridization enables plant breeders to combine superior traits from different species to form the basis of populations for further selection and genetic improvement. Hybridization programs have been undertaken to develop new cultivars of *Leucaena* with the following characteristics:

#### Low mimosine forage

Variability in concentration of the toxic amino acid mimosine in foliage exists within the *Leucaena* genus. Species with lower concentrations of mimosine include: *L. pulverulenta* (Gonzalez et al. 1967; Brewbaker et al. <u>1972</u>; <u>Bray et al. 1984</u>); and *L. collinsii*, *L. diversifolia*, *L. shannonii* and *L. trichandra* (Brewbaker and Kaye <u>1981</u>; <u>Saunders et al. 1987</u>). Hybridization to produce low-mimosine forage cultivars for feeding to monogastric animals has been attempted. Hybrid lines of *L. pulverulenta*  $\times$  *L. leucocephala* ssp. *glabrata* were developed with low mimosine concentration in Australia; however, the programs were unsuccessful as lowmimosine breeding lines (~25% reduction in mimosine) had significantly lower forage yields than existing commercial cultivars (Bray et al. 1984).

# Acid soil tolerance

In general, little specific tolerance to acid soils was identified within the Leucaena genus by a comprehensive environmental adaptation study (Mullen et al. 2003c). However, acid soil tolerance has been reported within accessions of L. diversifolia and L. trichandra (Hutton 1983). Hybrid breeding programs (× L. leucocephala ssp. glabrata) in South America (CIAT/EMBRAPA) and Southeast Asia (MARDI) have been undertaken to develop psyllid-tolerant and acid soil-tolerant forage cultivars (Wong et al. 1998). Two hybrid cultivars were released in Malaysia in 1998 (Aminah and Wong 2004), cv. Bharu (L. trichandra × L. leucocephala ssp. glabrata breeding line 40-1-18) and cv. Rendang (L. diversifolia  $\times$ L. leucocephala ssp. glabrata breeding line 62-6-8). The commercial success of these cultivars is unknown and wider assessment of their agronomic performance and forage quality (palatability and digestibility) is required. Concentrations of condensed tannins in both cultivars have been reported to be high and to adversely impact rumen function (Khamseekhiew et al. 2000; Kok et al. 2013; Saminathan et al. 2015, 2017). These hybrid cultivars need to be compared with alternative multipurpose shrub legumes with known acid soil tolerance, e.g. Calliandra calothyrsus, Cratylia argentea and Flemingia macrophylla.

# Psyllid resistance

Hybrid cultivars have been developed from *L. pallida* × *L. leucocephala* ssp. *glabrata* (designated KX2 hybrids) for forage and biomass/timber. These hybrids have shown high yield with broad environmental adaptation (Mullen et al. 2003c), psyllid resistance (Mullen et al. 2003b), cool tolerance (Austin et al. 1997; Mullen et al. 2003c) and intermediate forage quality (Dalzell et al. 1998b).

 Cultivar KX2-Hawaii was bred by UH and was released in 2007 (<u>Brewbaker 2008</u>). This cultivar was developed by 6 cycles of recurrent selection from advanced generations of the original F1 hybrid *L. pallida* K376  $\times$  *L. leucocephala* ssp. *glabrata* K8. It was selected under regular cutting/coppicing for psyllid resistance, forage/biomass yield and self-sterility. To date, there has been limited commercial utilization of cv. KX2-Hawaii.

Cultivar Redlands (protected by PBR in Australia) was bred by the University of Queensland (Anonymous 2015) and was released in 2017. This hybrid cultivar was developed using 5 elite KX2 F1 hybrids bred by UH. These parents were open-pollinated (panmixia) and F2 seed planted for intense selection (5-10% retention) under the criteria of psyllid resistance, yield, tree form (high degree of basal branching) and selfsterility. After another cycle of recurrent mass selection, elite F3 trees were backcrossed (BC) (handpollinated) to L. leucocephala ssp. glabrata cv. Wondergraze. Elite psyllid-resistant BC progeny were backcrossed again to produce breeding lines that were effectively 87.5% cv. Wondergraze and 12.5% L. pallida. The best BC2 breeding lines were then selfpollinated 3 times. Selfed breeding lines were assessed for in vitro forage quality (digestibility plus crude protein and condensed tannin concentrations) and their palatability determined under direct grazing. Cattle had a preference for cvv. Cunningham and Wondergraze plots ahead of cv. Redlands, but cv. Redlands was readily eaten (Shelton et al. 2019). A trial comparing hedgerow pastures of cvv. Wondergraze and Redlands and measuring cattle liveweight gain is currently underway in north Queensland (Lemin et al. 2019). Cultivar Redlands is recommended for humid (average annual rainfall >800 mm) psyllid-prone areas.

# Cold tolerance

Hybrids based upon L. diversifolia (KX3) and L. pallida (KX2) with L. leucocephala ssp. glabrata have been developed by UH and distributed for evaluation throughout the tropics (Brewbaker 2016). These hybrids are vigorous, psyllid-resistant/tolerant and have superior growth under cool temperatures during autumn and spring in the subtropics (Middleton and Clem 1998; Mullen et al. 2003a, 2003c) and year-round in the elevated tropics (Austin et al. 1997). The forage quality of these hybrids requires careful evaluation, as it is likely to be lower than 'giant' leucaena owing to higher concentrations of condensed tannins inherited from L. diversifolia and L. pallida (Austin et al. 1997; Dalzell et al. 1998b). With the exception of cv. KX2-Hawaii, no cultivars from this breeding program have been commercialized. KX3 hybrids have been developed and evaluated in southern

Brazil (<u>Austin et al. 1998</u>) and Argentina (<u>Goldfarb and</u> <u>Casco 1998</u>) for frost and cold tolerance; however, no known commercial cultivars have been released from these programs.

#### Wood/biomass/pulp production

Fast-growing Leucaena spp. hybrids have great potential for high-value timber, biomass (bioenergy) and paper pulp production (Brewbaker 2016). Cultivar KX4-Hawaii is a male-sterile triploid hybrid between L. leucocephala ssp. glabrata K636 and L. esculenta K838 developed by UH (Brewbaker 2013). This hybrid is vegetatively propagated, psyllid-tolerant, arboreal, vigorous and cooltolerant. Significant areas (>18,000 ha) of 'giant' leucaena in Gujarat, Maharashtra and Madhya States in India are managed for wood production to supply paper pulp mills (Khanna et al. 2019). Genetic improvement of 'giant' leucaena germplasm has been undertaken through intense selection and mutagenesis to improve biomass yield. A triploid L. collinsii  $\times$  L. leucocephala ssp. glabrata hybrid has been developed by JK Paper Ltd (Khanna et al. 2019) and is currently being vegetatively propagated for evaluation of biomass yield and paper pulp characteristics. This hybrid also has potential as a forage plant and requires wider evaluation for environmental adaptation, forage production and animal feeding.

#### Sterility

'Common' and 'giant' leucaena have the potential to become environmental weeds of disturbed ruderal habitats in the absence of grazing animals (Campbell et al. 2019; Idol 2019). Breeding programs within Australia are currently developing sterile cultivars for use in extensive grazing systems in jurisdictions where the promotion of 'giant' leucaena is not sanctioned. Strategies are focussing on developing sterility (male or female) in commercial cultivars via mutagenesis (McMillan et al. 2019) or gene editing to prevent flowering (Real et al. 2019). Interspecific hybridization to develop sterile triploids is also being explored (Real et al. 2019). In addition to reducing or eliminating the weed potential of Leucaena spp. cultivars, sterility may confer a significant yield (forage or biomass) advantage as plant resources are not diverted from vegetative growth to seed production.

# Future directions for cultivar development

Superior accessions of lesser-known *Leucaena* spp. (Table 1) have been identified in extensive germplasm

evaluation trials (<u>Mullen et al. 2003a</u>; 2003b; 2003c). These could be utilized to develop new interspecific hybrids to overcome the lack of cold, frost and acid soil tolerance in current commercial cultivars. Other accessions within these lesser-known taxa are held within international germplasm collections and require further agronomic evaluation (consult the World Leucaena Catalogue; <u>Bray et al. 1997</u>).

As understanding of the genetic base of the *Leucaena* genus improves, new tools have become available for plant breeding. Phylogenetic studies of the evolutionary history of the *Leucaena* genus have identified the parents of the 5 allotetraploid species (Govindarajulu et al. 2011a) and enabled the definition and elucidation of relationships between the 19 diploid species (Govindarajulu et al. 2011b; Abair et al. 2019). Sequencing of the *L. trichandra* genome has been completed and will enable genetic markers to be identified for traits of interest in breeding programs (Abair et al. 2019). Application of molecular marker-assisted selection should accelerate rates of genetic gain in traditional and molecular plant breeding programs.

Chromosome/ploidy doubling has been successfully undertaken in a number of Leucaena spp. (Shi 2003). Diploid species could be doubled, which may enhance cross-compatibility for desired interspecific hybrids. Leucaena collinsii (2n) is of particular interest as a forage plant as it is psyllid-resistant (Mullen et al. 2003b), has moderate forage yield (Mullen et al. 2003a; 2003c), excellent in vitro forage quality (Dalzell et al. 1998b) and has proved productive under cattle grazing (Jones et al. 1998). Producing and evaluating artificial tetraploid L. collinsii lines could deliver valuable new forage cultivars. Similarly, halving ploidy levels of the tetraploid Leucaena spp. would generate diploid (2n) lines that could be used to develop sterile triploid cultivars. Gametophytic self-incompatibility systems could be used to produce F1 interspecific hybrid seed (Brewbaker 2016).

New genetic technologies have potential to modify the *Leucaena* genome, including transgenic improvement, e.g. suppressing mimosine synthesis by the transfer of a gene from *Rhizobium* sp. into *L. leucocephala* ssp. *glabrata* K636 via agrobacterium (Jube and Borthakur 2010), or gene deletion using CRISPR technology (Real et al. 2019). Mutagenesis has been used to alter the genome of *L. leucocephala* ssp. *glabrata* to increase plant yield (Khanna et al. 2019) or induce sterility (McMillan et al. 2019).

Modern vegetative propagation techniques can be used for embryo rescue of F1 interspecific hybrid seeds that are prone to abort and to mass produce elite sterile germplasm for commercial application.

Breeding objective	Constraint	Taxon	Accession
Forage	Cold tolerance	L. diversifolia	K778, K784, K806, OFI104/94, CPI33820
		L. pallida	K748, K802, K376, CQ3439
		L. trichandra	OFI53/88, OFI35/88
	Frost tolerance	L. retusa	_1
	Sterility (2n	L. collinsii	OFI51/88, OFI52/88
	parent)	L. magnifica	OFI1984, OFI58/88
Timber/biomass/	Cold tolerance	L. diversifolia	K778, K784, K806, OFI104/94, CPI33820
paper pulp/shade		L. pallida	K748, K802, K376, CQ3439
		L. trichandra	OFI53/88, OFI35/88
	Frost tolerance	L. pulverulenta	_1
		L. greggii	_1
		L. retusa	_1
	Sterility (2n	L. cruziana	OFI51/87
	parent)	L. esculenta	_1
	-	L. magnifica	OFI1984, OFI58/88
		L. macrophylla ssp. istmensis	OFI47/85
		L. macrophylla ssp. macrophylla	OFI55/88
		L. multicapitula	OFI81/87
		L. pulverulenta	_1
		L. salvadorensis	_1
		L. trichandra	OFI53/88, OFI35/88

**Table 1.** Superior accessions of key *Leucaena* spp. for use in future hybridization programs [adapted from Mullen et al. (2003a; 2003b; 2003c)].

<sup>1</sup>Superior accessions not identified – evaluation of a diverse array of accessions required.

# Challenges and opportunities for future cultivar development

Many of the lesser-known Leucaena taxa have been identified only within the last 30 years and are represented by few accessions/provenances, e.g. L. confertiflora, L. cuspidata, L. involucrata, L. lempirana, L. magnifica, L. matudae and L. pueblana, from limited geographical areas in international germplasm collections (Hughes 1998; Brewbaker 2016). Further germplasm collection, conservation, multiplication and evaluation of these taxa are required. In addition, recent advances in Leucaena taxonomy (Abair et al. 2019) and the use of molecular markers will enable the accurate description of germplasm currently held (often misidentified and/or duplicated) in international collections and facilitate a much-needed update of the World Leucaena Catalogue (Bray et al. 1997). The World Leucaena Catalogue could be promoted as a 'source of truth' for the identification of Leucaena spp. accessions exchanged for use in future breeding programs. Germplasm collections are expensive to maintain, as seed needs to be refreshed and multiplied. Seed of some species, e.g. L. esculenta, appears to have a shorter lifespan under long-term storage.

A number of important practicalities must be considered when formulating *Leucaena* spp. breeding programs, including: focussing on forage quality for multipurpose tree legumes to ensure the forage produced fattens animals; long-term field testing of interspecific hybrids or elite lesser-known species to ensure longevity under frequent cutting or heavy grazing; determining the promiscuity of new cultivars for *Rhizobium* spp. to facilitate effective nodulation and adequate rates of biological nitrogen fixation (Mullen et al. 1998); estimating the cost of producing propagules (seed vs. vegetative planting material) at a commercial scale suited for adoption in target farming systems; and understanding the environmental requirements and establishment practices (seed vs. vegetative planting material) required for rapid widespread adoption of new cultivars.

Finally, a key challenge to breeding *Leucaena* is the long time-frame (>10 years) and significant resources (financial and human) required to develop new cultivars. Collaboration between international breeding programs would make the most of these limited resources. Such collaboration may include: the exchange of successful breeding technologies/techniques and elite germplasm; and undertaking coordinated  $G \times E$  trials of advanced breeding lines and emerging cultivars. The spirit of such collaboration has been epitomized by Professor James L. Brewbaker (University of Hawaii), who for over 50 years has generously shared his vast knowledge of *Leucaena* spp. collection, genetics and breeding plus elite germplasm with plant breeders around the world.

#### References

(Note of the editors: All hyperlinks were verified 17 April 2019.)

- Abair A; Hughes CE; Bailey CD. 2019. The evolutionary history of *Leucaena*: Recent research, new genomic resources and future directions. Tropical Grasslands-Forrajes Tropicales 7:65–73. doi: <u>10.17138/TGFT(7)65-73</u>
- Aminah A; Wong CC. 2004. Dry matter productivity and nutritive quality of leucaena hybrid lines for high protein feed production. Journal of Tropical Agriculture and Food Science 32:251–256. goo.gl/5VT2MF
- Anonymous. 1997. *Leucaena leucocephala*: 'Tarramba' syn K636. Plant Varieties Journal 10(1):19. goo.gl/2VEBD6
- Anonymous. 2008. Leucaena leucocephala ssp glabrata: 'Wondergraze'. Plant Varieties Journal 21(2):201–203. goo.gl/nwy1Ji
- Anonymous. 2015. Leucaena pallida × Leucaena leucocephala: 'BL-12'. Plant Varieties Journal 28(2):262–265. goo.gl/ g8NVAd
- Austin MT; Early RJ; Brewbaker JL; Sun W. 1997. Yield, psyllid resistance and phenolic concentration of *Leucaena* in two environments in Hawaii. Agronomy Journal 89:507–515. doi: <u>10.2134/agronj1997.00021962008900030022x</u>
- Austin MT; Sun W; Brewbaker JL; Schifino-Wittmann MT. 1998. Developing *Leucaena* hybrids for commercial use. In: Shelton HM; Gutteridge RC; Mullen BF; Bray RA, eds. Leucaena – adaptation, quality and farming systems. Proceedings of a workshop held in Hanoi, Vietnam, 9–14 February 1998. ACIAR Proceedings 86. ACIAR, Canberra, ACT, Australia. p. 82–85. purl.umn.edu/135197
- Bray RA. 1994. The leucaena psyllid. In: Gutteridge RC; Shelton HM, eds. Forage tree legumes in tropical agriculture. CAB International, Wallingford, UK. p. 283–291. <u>hdl.handle.net/</u> <u>10568/49375</u>
- Bray RA; Hutton EM, Beattie WM. 1984. Breeding *Leucaena* for low-mimosine: Field evaluation of selections. Tropical Grasslands 18:194–198. goo.gl/XvKRsv
- Bray RA; Hughes CE; Brewbaker JL; Hanson J; Thomas JB; Ortiz A. 1997. The World Leucaena Catalogue. Department of Agriculture, University of Queensland, Brisbane, Australia. goo.gl/WG7GGU
- Brewbaker JL. 1975. Registration of Hawaiian Giant K8 Leucaena (Reg. No. 16). Crop Science 15:885–886. doi: 10.2135/cropsci1975.0011183X001500060049x
- Brewbaker JL. 2008. Registration of KX2-Hawaii, interspecifichybrid *Leucaena*. Journal of Plant Registrations 2:190–193. doi: <u>10.3198/jpr2007.05.0298crc</u>
- Brewbaker JL. 2013. 'KX4-Hawaii', seedless interspecific hybrid *Leucaena*. HortScience 48:390–391. doi: <u>10.21273/</u><u>HORTSCI.48.3.390</u>
- Brewbaker JL. 2016. Breeding *Leucaena*: Tropical multipurpose leguminous tree. Plant Breeding Reviews 40:43–120. doi: <u>10.1002/9781119279723.ch2</u>
- Brewbaker JL; Plucknett DL; Gonzalez V. 1972. Varietal variation and yield trials of *Leucaena leucocephala* (Koa Haole) in Hawaii. Research Bulletin 166. Hawaii Agricultural Experiment Station, University of Hawaii, Honolulu, HI, USA. <u>hdl.handle.net/10125/40996</u>

- Brewbaker JL; Kaye S. 1981. Mimosine variations in species of the genus *Leucaena*. Leucaena Research Reports 2:66–68. goo.gl/39kFQF
- Campbell S; Vogle W; Brazier D; Vitelli J; Brook S. 2019. Weed leucaena and its significance, implications and control. Tropical Grasslands-Forrajes Tropicales 7 (in press).
- Dalzell SA; Miller DR; Miller BC. 1998a. Frost tolerance of *Leucaena* spp. in subtropical Australia. In: Shelton HM; Gutteridge RC; Mullen BF; Bray RA, eds. Leucaena adaptation, quality and farming systems. Proceedings of a workshop held in Hanoi, Vietnam, 9–14 February 1998. ACIAR Proceedings 86. ACIAR, Canberra, ACT, Australia. p. 174–177. purl.umn.edu/135197
- Dalzell SA; Stewart JL; Tolera A; McNeill DM. 1998b. Chemical composition of *Leucaena* and implications for forage quality. In: Shelton HM; Gutteridge RC; Mullen BF; Bray RA, eds. Leucaena – adaptation, quality and farming systems. Proceedings of a workshop held in Hanoi, Vietnam, 9–14 February 1998. ACIAR Proceedings 86. ACIAR, Canberra, ACT, Australia. p. 227–246. purl.umn.edu/135197
- Felker P; Sorensson CT; Ueckert D; Jacoby P; Singer E; Ohm R. 1998. Growth, cold-hardiness, protein content, and digestibility of 70 *Leucaena* seedlots on three sites in Texas, USA. Agroforestry Systems 42:159–179. doi: <u>10.1023/</u> A:1006125624985
- Goldfarb MC; Casco JF. 1998. Selection and agronomic characterisation of *Leucaena* genotypes for cold tolerance. In: Shelton HM; Gutteridge RC; Mullen BF; Bray RA, eds. Leucaena adaptation, quality and farming systems. Proceedings of a workshop held in Hanoi, Vietnam, 9–14 February 1998. ACIAR Proceedings 86. ACIAR, Canberra, ACT, Australia. p. 172–173. purl.umn.edu/135197
- Gonzalez V; Brewbaker JL; Hamill DE. 1967. *Leucaena* cytogenetics in relation to the breeding of low mimosine lines. Crop Science 7:140–143. doi: <u>10.2135/cropsci1967.</u> 0011183X000700020014x
- Govindarajulu R; Hughes CE; Alexander PJ; Bailey CD. 2011a. The complex evolutionary dynamics of ancient and recent polyploidy in *Leucaena* (Leguminosae; Mimosoideae). American Journal of Botany 98:2064–2076. doi: <u>10.3732/</u> <u>ajb.1100260</u>
- Govindarajulu R; Hughes CE; Bailey CD. 2011b. Phylogenetic and population genetic analyses of diploid *Leucaena* (Leguminosae; Mimosoideae) reveal cryptic species diversity and patterns of divergent allopatric speciation. American Journal of Botany 98:2049–2063. doi: 10.3732/ajb.1100259
- Gray SG. 1968. A review of research on *Leucaena leucocephala*. Tropical Grasslands 2:19–30. <u>goo.gl/XwZJgD</u>
- Hughes CE. 1998. Monograph of *Leucaena* (Leguminosae-Mimosoideae). Systematic Botany Monographs 55:1–244. doi: <u>10.2307/25027876</u>
- Hutton EM. 1983. Selection and breeding of leucaena for very acid soils. Leucaena research in the Asian Pacific region. Proceedings of a workshop held in Singapore, 23–26 November 1982. IDRC, Ottawa, Canada. p. 23–26.
- Hutton EM; Gray SG. 1959. Problems in adapting *Leucaena* glauca as a forage for the Australian tropics. Empire Journal of Experimental Agriculture 27:187–196. goo.gl/nKuvAW

- Hutton EM; Beattie WM. 1976. Yield characteristics in three bred lines of the legume *Leucaena leucocephala*. Tropical Grasslands 10:187–194. goo.gl/Ro5UWB
- Idol T. 2019. A short review of leucaena as an invasive species in Hawaii. Tropical Grasslands-Forrajes Tropicales 7 (in press).
- Jones RJ; Galgal KK; Castillo AC; Palmer B; Deocareza A; Bolam M. 1998. Animal production from five species of *Leucaena*. In: Shelton HM; Gutteridge RC; Mullen BF; Bray RA, eds. Leucaena – adaptation, quality and farming systems. Proceedings of a workshop held in Hanoi, Vietnam, 9–14 February 1998. ACIAR Proceedings 86. ACIAR, Canberra, ACT, Australia. p. 247–256. purl.umn.edu/135197
- Jube SLR; Borthakur D. 2010. Transgenic *Leucaena leucocephala* expressing the *Rhizobium* gene *pyd*A encoding a meta-cleavage dioxygenase shows reduced mimosine content. Plant Physiology and Biochemistry 48:273–278. doi: <u>10.1016/j.plaphy.2010.01.005</u>
- Kana Hau D; Nulik J. 2019. Leucaena in West Timor, Indonesia: A case study of successful adoption of cv. Tarramba. Tropical Grasslands-Forrajes Tropicales 7 (in press).
- Khamseekhiew B; Liang JB; Wong CC; Jalan ZA. 2000. Ruminal and intestinal digestibility of *Leucaena leucocephala* and *Arachis pintoi* in zebu cattle. Asian-Australasian Journal of Animal Sciences 13 Supplement A:333–334.
- Khanna NK; Shukla OP; Gogate MG; Narkhede SL. 2019. Leucaena for paper industry in Gujarat, India: Case study. Tropical Grasslands-Forrajes Tropicales 7:200–209. doi: <u>10.</u> <u>17138/TGFT(7)200-209</u>
- Kinch DM; Ripperton JC. 1962. Koa Haole: Production and processing. Research Bulletin 129. Hawaii Agricultural Experiment Station, University of Hawaii, Honolulu, HI, USA. <u>hdl.handle.net/10125/53867</u>
- Kok CM; Sieo CC; Tan HY; Saad WZ; Liang JB; Ho YW. 2013. Anaerobic cellulolytic rumen fungal populations in goats fed with and without *Leucaena leucocephala* hybrid, as determined by real-time PCR. Journal of Microbiology 51:700–703. doi: 10.1007/s12275-013-2540-z
- Lemin C; Rolfe J; English B; Caird R; Black E; Dayes S; Cox K; Perry L; Brown G; Atkinson R; Atkinson N. 2019. Comparing the grazing productivity of 'Redlands' and 'Wondergraze' leucaena varieties. Tropical Grasslands-Forrajes Tropicales 7:96–99. doi: <u>10.17138/TGFT(7)96-99</u>
- McMillan HE; Liu G; Shelton HM; Dalzell SA; Godwin ID; Gamage H; Sharman C; Lambrides CJ. 2019. Sterile leucaena becomes a reality? Tropical Grasslands-Forrajes Tropicales 7:74–79. doi: <u>10.17138/TGFT(7)74-79</u>
- Middleton CH; Clem R. 1998. Evaluation of *Leucaena* germplasm on clay soils in Central and Southern Inland Queensland. In: Shelton HM; Gutteridge RC; Mullen BF; Bray RA, eds. Leucaena – adaptation, quality and farming systems. Proceedings of a workshop held in Hanoi, Vietnam, 9–14 February 1998. ACIAR Proceedings 86. ACIAR, Canberra, ACT, Australia. p. 154–156. purl.umn.edu/135197
- Mullen BF; Frank VE; Date RA. 1998. Specificity of rhizobial strains for effective N<sub>2</sub> fixation in the genus *Leucaena*. Tropical Grasslands 32:110–117. goo.gl/B5aTNN

- Mullen BF; Gabunada F; Shelton HM; Stür WW. 2003a. Agronomic evaluation of *Leucaena*. Part 2. Productivity of the genus for forage production in subtropical Australia and humid-tropical Philippines. Agroforestry Systems 58:93– 107. doi: 10.1023/A:1026040631267
- Mullen BF; Gabunada F; Shelton HM; Stür WW. 2003b. Psyllid resistance in *Leucaena*. Part 1. Genetic resistance in subtropical Australia and humid-tropical Philippines. Agroforestry Systems 58:149–161. doi: <u>10.1023/A:</u> <u>1026092424732</u>
- Mullen BF; Shelton HM; Gutteridge RC; Basford KE. 2003c. Agronomic evaluation of *Leucaena*. Part 1. Adaptation to environmental challenges in multi-environment trials. Agroforestry Systems 58:77–92. doi: <u>10.1023/A:1026068</u> <u>215337</u>
- Oram RN. 1990. Register of Australian herbage plant cultivars. 3rd Edn. CSIRO Division of Plant Industry, Melbourne, Victoria, Australia.
- Real D; Han Y; Bailey D; Vasan S; Li C; Castello M; Broughton S; Abair A; Crouch S; Revell C. 2019. Strategies to breed sterile leucaena for Western Australia. Tropical Grasslands-Forrajes Tropicales 7:80–86. doi: <u>10.17138/TGFT(7)80-86</u>
- Saminathan M; Sieo CC; Abdullah N; Wong CMVL; Ho YW. 2015. Effects of condensed tannin fractions of different molecular weights from a *Leucaena leucocephala* hybrid on *in vitro* methane production and rumen fermentation. Journal of the Science of Food and Agriculture 95:2742–2749. doi: 10.1002/jsfa.7016
- Saminathan M; Gan HM; Abdullah N; Wong CMVL; Ramiah SK; Tan HY; Sieo CC; Ho YW. 2017. Changes in rumen protozoal community by condensed tannin fractions of different molecular weights from a *Leucaena leucocephala* hybrid *in vitro*. Journal of Applied Microbiology 123:41–53. doi: 10.1111/jam.13477
- Saunders JA; Oakes AJ; Wiser JW. 1987. The relationship of mimosine and protein in *Leucaena leucocephala*. Leucaena Research Reports 8:68–74.
- Shelton HM; McMillan H; Halliday MJ; Rolfe J; Keating M; Saunders T&C. 2019. Grazing preference by cattle for the psyllid-resistant leucaena inbred cv. Redlands compared with the commercial *L. leucocephala* cvv. Cunningham and Wondergraze. Tropical Grasslands-Forrajes Tropicales 7 (in press).
- Shi X. 2003. Genetic improvement of *Leucaena* spp. and *Acacia koa* (Gray) as high-value hardwoods. Ph.D. Thesis. University of Hawaii, Honolulu, HI, USA. <u>hdl.handle.net/10125/56593</u>
- Sorensson CT; Brewbaker JL. 1994. Interspecific compatibility among 15 *Leucaena* species (Leguminosae: Mimosoideae) via artificial hybridizations. American Journal of Botany 81:240–247. doi: <u>10.1002/j.1537-2197.1994.tb15435.x</u>
- Sun WG. 1992. Isozyme polymorphism in the leguminous genus Leucaena. M.Sc. Thesis. University of Hawaii, Honolulu, HI, USA. <u>hdl.handle.net/10125/56225</u>
- Takahashi M; Ripperton JC. 1949. Koa Haole (*Leucaena glauca*), its establishment, culture and utilisation as a forage crop. Research Bulletin 100. Hawaii Agricultural Experiment

Station. University of Hawaii, Honolulu, HI, USA. p. 6–44. hdl.handle.net/10125/31059

Wong CC; Chen CP; Hutton EM. 1998. Development of acid/psyllid tolerant *Leucaena* hybrids for ruminant production. In: Shelton HM; Gutteridge RC; Mullen BF;

Bray RA, eds. Leucaena – adaptation, quality and farming systems. Proceedings of a workshop held in Hanoi, Vietnam, 9–14 February 1998. ACIAR Proceedings No. 86. ACIAR, Canberra, ACT, Australia. p. 132–135. <u>purl.umn.</u> edu/135197

(Accepted 24 March 2019 by the ILC2018 Editorial Panel and the Journal editors; published 31 May 2019)

#### © 2019



*Tropical Grasslands-Forrajes Tropicales* is an open-access journal published by *International Center for Tropical Agriculture (CIAT)*. This work is licensed under the Creative Commons Attribution 4.0 International (CC BY 4.0) license. To view a copy of this license, visit <u>https://creativecommons.org/licenses/by/4.0/</u>.