# Morphoagronomic and Isoenzymatic Characterization of 23 Accessions of *Leucaena* spp

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**Key words:** *Leucaena*, genetic resources, variability, isoenzymes

**Antecedents and objectives** Leucaena leucocephala (Lam.) de Witt, is the species that has been most used throughout the world and has been widely studied in animal feeding. In Cuba, the main studies of characterization and identification of this species have focused mainly on morphoagronomic characteristics under field conditions, or in isolated trials, including only the four varieties recorded as commercial (cv. Ipil-Ipil; cv. Perú; cv. Cunningham and cv. CNIA-250). The electrophoresis of isoenzymes favored the use of more efficient genetic markers than the morphological markers in many occasions. In spite of being influenced by the environmental action and depending on the tissue and developmental stage of the plant that is evaluated, these relatively simple, cheap and co-dominant markers, allow us to distinguish the homozygous and heterozygous genotypes, and thus develop studies of mapping and binding, population genetics, etc. (Bretting and Widrlechner, 1995). The objective of this work was the morphoagronomic and isoenzymatic (esterases, peroxidases and alcohol dehydrogenases) characterization of a sample of 23 accessions that included five species of the Leucaena genus (L. leucocephala, L. lanceolada, L. diversifolia, L. macrophylla y L. esculenta).

**Methodologies** The study was conducted in areas of the Experimental Station of Pastures and Forages Indio Hatuey", in plants that were six years old. Plant height and stem diameter at 130 cm above the soil surface were measured, and the number of branches was counted monthly, on four plants. The number of pinnas per leaf, pinnulas per pinna, dimension and form of the foliole; type and position of the glands; and forthe fruit: number of fruits per glomerule, length and width, opening, texture, length and width of the seeds, color and form; and for the flowers: position of the glomerule, quantity and color, were also determined. The isoenzymatic analyses were conducted on new regrowth of each of the accessions evaluated, by the standard methodology of Chamberlain, Hughes and Galway (1996). The specific stains were carried out for peroxidases (*Prx EC. 1.11.1.7*), α-and β-esterases (*Est EC. 3.1.1*), and alcohol dehydrogenases (*Adh EC. 1.1.1.1*), according to Álvarez, Fuentes, Deus, Duque and González (2000). The electrophoretic runs were repeated at least three times and only the consistent and reproducible bands were recorded. The isoenzymatic phenotypes of each accession were recorded as presence/absence of each band (0/1, respectively). The analyses were conducted with the statistical program **SPSS version 11.5.** The binary matrix of isoenymatic data (eliminating the redundant bands) was used to generate a matrix of genetic distances among all the genotype pairs, expressed as the complement of Dice's coefficient (Dice, 1945), using the program SIMQUAL of the statistical pack NTSYS-pc.

**Results and Discussion** The qualitative analysis of the electrophoretic composition of α-and β-esterases and peroxidases in the leaf tissue, allowed us to observe a total of 22 bands in the 23 accessions studied. In the electrophoretic diagram of isoenzymes *Prx*, the existence of a high degree of polymorphism was detected regarding the percentage of polymorphic bands, although only 4 isoenzymatic patterns were detected in the whole sample. This system showed a total of nine well-defined bands, being different the accessions *L. macrophylla* CIAT-17232 (19), belonging to pattern 3 and *L. esculenta* CIAT-17225 (22). Similar isoenzymatic polymorphism was observed in the composition of α-y β-*Est*, which allowed, to differentiate the presence of bands in the accession *L. macrophylla* CIAT-17232 (19) with regards to the other accessions. In this isoenzymatic system a total of 13 bands were observed, of which number one is common for all the accessions. Likewise, it could be observed that no isoforms *Adh* appeared in leaf tissue, this is in agreement with the statements by Loulakakis and Roubelakis (1990); Menezes, Donizeti and Mota (1995), who reported that under normal conditions, the enzymatic activity of these systems disappears in very early stages of plant development, although they can be induced when conditions of anaerobiosis are present (Iglesias, 1994).

**Conclusions and Recommendations** There is variability among the species and accessions of *Leucaena* regarding the evaluated indicators. The analysis of genetic diversity allowed us to differentiate *L. leucocephala* with higher clarity than the other species studied, although there was no gain regarding differentiation within the species. The isoenzymatic systems (esterases y peroxidases) turned out to be polymorphic in the sample studied, mainly the esterases. To conduct other studies with the selected accessions is recommended, correlating the isoenzymatic polymorphism to molecular markers and the main morphoagronomic characteristics evaluated.