Original Research Article

Studies on Maturity Indicators of Leucaena leucocephala in Northern India

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A B S T R A C T

Introduction

Leucaena leucocephala (Mimosoideae), commonly known as subabool is a medium-sized, multipurpose, deciduous tree species. It is a valuable source of pulpwood, small timber, fuel wood, fodder and green manure. It is a tree of the tropics and subtropics and within this region grows up to an elevation of about 500 m. Genus Leucaena consists of 22 identified species (Brewbaker, 2016). Botanical literature claims a sum of 55 species. The only valid ones appear to be L. collinsii, L. diversifolia, L. esculenta, L. lanceolata, L. leucocephala, L. macrophylla, L. pulverulenta, L. retusa, L. shannonii and L. trichodes. All these species have value for the tropics but only L. leucocephala (Lam.) de Wit has been exploited extensively so far (National Research Council, 1984) [21]. Leucaena leucocephala have been observed to exhibit physical dormancy due to presence of hard seed-coat and thereby results poor seed germination. Raising the seedlings from such seeds results a problem due to delayed and poor germination. Availability of mature and viable seeds is a pre-requisite for raising the quality seedlings at massive scale. The

Leucaena leucocephala is a medium-sized, multipurpose tree which is a valuable source of pulpwood, small timber, fuel wood, fodder and green manure. Poor and delayed seed germination is reported due to seed collection at improper maturity stage coupled with physical dormancy. Hence, the present study was carried out to determine the exact maturity stage for seed collection at three locations of northern India i.e. Agra (S1), Mathura (S2) and Hathrus (S3). Among the different pod/seed collection periods, 1st week of June (8th collection) was found best across all the sites as maximum germination 58.7±0.25% at S1, 55.1±0.43% at S2, 54.2±0.28% at S3 was recorded during this stage. The results revealed that moisture content decreased with advancement of maturity. The brown colour of pod and dark brown colour of seed were identified as seed maturity indicators.

Keywords
Moisture content, Pod colour, Seed colour, Seed germination, Seed maturity

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collection of seeds at proper maturity stage and pre-treatments can give good germination, and more vigorous seedlings of *Leucaena leucocephala* can be obtained by inoculating with *R. irregularis* (Kumar, 2018). Thus, the knowledge of proper stage and time of maturity of seeds is essential for collection of abundant quantity of healthy and vigorous seeds. Keeping in view the importance of species and the lack of information on maturity indicators of this species in semi-arid conditions of northern India, the present study was carried out.

**Materials and Methods**

The present study was conducted in Agra (S\(_1\)), Mathura (S\(_2\)) and Hathrus (S\(_3\)) sites of northern India which lies between 27° 10’ N to 26° ‘4 N’ latitude and 78°02’ E to 79°7’E longitude between elevation 165 and 179.8 m amsl. Five trees with clear bole, compact crown, having fair number of pods were selected for the study at each site. The pods of *Leucaena leucocephala* were collected from the all three sites at different dates. First pods collection was made on 2\(^{nd}\) week of February, 2004 and subsequent collections were made at bi-weekly intervals, until the completion of natural seed fall. Pods from different tree species were separately sealed in plastic bags and brought to the laboratory.

Before the seed extraction and cleaning, pod parameters like length, width, moisture content (%), weight of 100 pods, number of pod per 100 g were measure for each collection dates. The size dimensions were recorded with manual Vernier’s caliper and digital electronic balance was used for weighing of pod weight. Moisture content (%) was determined on fresh weight basis by drying (over drying) the material at 103±2°C for 16±1 hrs (ISTA, 1981). The pods were dried for 4 to 5 days and thereafter beaten with a stick and cleaned by winnowing to release seeds. After the process of seed extraction and cleaning, 3 replicates of 10 seeds were measured for seed length, width and size using Vernier’s calliper and weight parameters like number of seed/g, weight of 100 seeds. Moisture content was recorded on fresh weight basis by drying seeds at 103±1°C for 16± 1 hrs (ISTA, 1981) and moisture content was calculated as:

\[
\text{Moisture content} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Fresh weight}} \times 100
\]

The seeds were surface sterilized with 0.1 HgCl\(_2\). Seeds were rinsed thoroughly to remove traces of mercuric chloride before placing for germination. For germination, 3 replicates of 100 seeds each were used. The germination was carried out in Petri dish at laboratory (room temperature) for each collection date. Germination was counted when visible radicle develops. Germination percent was calculated as follow:

\[
\text{Germination (\%)} = \frac{\text{Number of germinated seed (visible redicle of seeds)}}{\text{Total number of seeds put in petridish}} \times 100
\]

Data recorded for different characters on different sites have been tested for their significance by using statistical technique of analysis of variance with Randomized block design suggested by Panse and Sukhatame (1961).

**Results and Discussion**

The results presented in Table 1 revealed that the pod colour of *L. leucocephala* changed from the green colour (2\(^{nd}\) week of February) to brown (1\(^{st}\) week of June) across all the collection sites.
**Table 1** Physical parameters of pods of *Leucaena leucecephala* over the collection period from February to Starting June in different sites

| Site | Date | Day of collection | Pod colours | Pod length (cm) | Pod width (cm) | Wt. of 100 pod (gm) | No. of pod/100 gm | Pod moisture content (%) |
|------|------|-------------------|-------------|----------------|---------------|----------------------|---------------------|-------------------------|
| S₁   | 20/2 | D₁                | Green       | 18.7±0.24      | 1.4±0.03      | 258.1±1.15          | 39.0±0.63          | 52.9±0.44               |
|      | 7/3  | D₂                | Green       | 19.2±0.20      | 1.8±0.05      | 350.7±1.29          | 30.1±0.31          | 55.2±0.75               |
|      | 22/3 | D₃                | Light Green | 21.3±0.23      | 1.8±0.06      | 501.5±1.16          | 19.4±0.37          | 60.6±0.68               |
|      | 6/4  | D₄                | Light Green | 20.9±0.22      | 1.8±0.06      | 504.1±3.34          | 25.4±0.37          | 59.1±0.31               |
|      | 21/4 | D₅                | Light Yellow | 20.4±0.57     | 1.9±0.02      | 511.1±2.30          | 19.8±0.63          | 58.0±0.52               |
|      | 6/5  | D₆                | Yellowish Green with Brown spot | 20.5±0.52 | 1.98±0.02 | 529.6±2.15 | 24.1±0.48 | 56.8±0.25 |
|      | 21/5 | D₇                | Light Brown | 20.5±0.54      | 2.0±0.04      | 526.0±7.72          | 26.3±0.47          | 50.2±0.43               |
|      | 5/6  | D₈                | Brown       | 20.3±0.37      | 2.0±0.07      | 303.0±1.57          | 33.2±0.99          | 40.6±0.25               |
| Mean |      |                   |             | **20.26**      | **1.85**      | **435.51**          | **27.16**          | **54.2**                |
| S₂   | 21/2 | D₁                | Green       | 18.5±0.24      | 1.3±0.03      | 250.6±2.29          | 42.9±1.13          | 54.5±0.32               |
|      | 8/3  | D₂                | Green       | 19.2±0.16      | 1.7±0.06      | 352.2±1.78          | 30.7±0.42          | 57.0±0.36               |
|      | 23/3 | D₃                | Light Green | 20.9±0.24      | 1.8±0.03      | 497.0±0.79          | 21.7±0.41          | 63.4±0.31               |
|      | 7/4  | D₄                | Light Green | 20.9±0.22      | 1.8±0.03      | 500.8±2.87          | 24.8±0.25          | 61.5±0.35               |
|      | 22/4 | D₅                | Light Yellow | 20.6±0.41     | 1.9±0.03      | 498.8±2.47          | 21.6±0.52          | 59.9±0.47               |
|      | 7/5  | D₆                | Yellowish Green with Brown spot | 20.5±0.41 | 1.8±0.03 | 457.4±5.88 | 25.4±0.52 | 59.0±0.37 |
|      | 22/5 | D₇                | Light Brown | 20.4±0.39      | 1.8±0.04      | 395.1±0.70          | 29.2±0.49          | 53.0±0.83               |
|      | 6/6  | D₈                | Brown       | 20.3±0.35      | 1.9±0.04      | 295.7±0.66          | 36.7±0.41          | 39.2±0.41               |
| Mean |      |                   |             | **20.18**      | **2.0**       | **405.95**          | **29.08**          | **56.1**               |
|   |   |   |   |   |   |
|---|---|---|---|---|---|
| S3 | 22/2 | D1 | Green | 18.1±0.25 | 1.3±0.04 | 253.3±4.67 | 44.8±1.16 | 56.2±0.41 |
|   | 9/3  | D2 | Green | 19.5±0.16 | 1.6±1.03 | 358.7±1.96 | 33.5±0.45 | 59.0±0.27 |
|   | 24/3 | D3 | Light Green | 20.1±0.57 | 1.7±0.03 | 485.6±1.03 | 27.0±0.30 | 65.9±0.40 |
|   | 8/4  | D4 | Light Green Golden | 20.8±0.41 | 1.8±0.03 | 486.9±0.76 | 28.9±0.31 | 62.2±0.47 |
|   | 23/4 | D5 | Light Yellow Green | 19.7±0.62 | 1.9±0.03 | 490.0±1.18 | 24.9±0.31 | 61.1±0.47 |
|   | 8/5  | D6 | Yellowish Green with Brown spot | 20.6±0.51 | 1.8±0.02 | 527.1±2.68 | 28.2±0.39 | 60.2±0.28 |
|   | 23/5 | D7 | Light Brown | 20.4±0.42 | 1.9±0.03 | 537.9±1.60 | 29.2±0.39 | 54.5±0.28 |
|   | 7/6  | D8 | Brown | 20.4±0.33 | 1.8±0.05 | 286.0±4.01 | 37.6±0.34 | 42.7±0.35 |
| Mean |   |   |   | **19.98** | **1.7** | **428.19** | **31.76** | **57.73** |
|     |    |    | Average of Across all site | 20.14 | 1.85 | 423.22 | 29.33 | 56.01 |
|    |     | SEM+ for site |   | 0.19 | 0.21 | 4.66 | 0.28 | 0.22 |
| F-test |    |    |   | NS | NS | * | * | * |
| CD |     |    |   | - | - | 9.22 | 0.55 | 0.44 |
|    | SEM+ for Date/day |   |   | 0.31 | 0.34 | 7.60 | 0.45 | 0.36 |
| F-test |    |    |   | * | * | * | * | * |
| CD for day |   |   |   | 0.61 | 0.67 | 15.06 | 0.895 | 0.71 |
| SEm+ for SxD |   |    |   | 0.54 | 0.59 | 13.17 | 0.78 | 0.623 |
| F-test |    |    |   | NS | NS | * | * | * |
| CD for SxD |   |    |   | - | - | 26.08 | 1.55 | 1.23 |

NS- Non Significant, *- Significance at 5% (P<0.05)
**Table 2** Physical parameters of seeds sites of *Leucaena leucocephala* over the collection period from end of February to Starting June in different sites

| Site | Date | Day of collection | Seed colour | Seed length (mm) | Seed width (mm) | Seed size (mm) | No. of seed/100 g | Seed wt/100 seed | Seed (mm) diameter | No. of seed/pod | Seed moisture content (%) | Germination (%) |
|------|------|-------------------|-------------|------------------|-----------------|----------------|------------------|-----------------|-------------------|----------------|--------------------------|-----------------|
| S₁   | 20/2 D₁ | Dark Green        | 6.4±0.22    | 3.7±0.15         | 41.4±2.74       | 1030.3±0.45    | 88.7±0.22       | 1.0±0.10        | 22.6±0.54         | 53.1±0.39       | 0.0±0.0                   | 3640            |
|      | 7/3 D₂  | Light Green       | 6.7±0.15    | 4.2±0.13         | 45.1±1.99       | 1091.4±1.42    | 113.8±0.21      | 1.1±0.10        | 23.6±0.34         | 53.0±0.96       | 0.0±0.0                   | 3640            |
|      | 22/3 D₃ | Bright Green      | 7.0±0.28    | 4.8±0.20         | 49.6±3.62       | 1124.2±8.84    | 178.1±0.07      | 1.6±0.16        | 25.7±0.45         | 50.9±0.52       | 6.5±0.21                  | 3640            |
|      | 6/4 D₄  | Bright Green      | 7.2±0.13    | 4.8±0.20         | 52.6±2.00       | 813.1±6.62     | 96.1±0.10       | 1.5±0.16        | 24.8±0.42         | 48.3±0.35       | 35.5±0.31                 | 3640            |
|      | 21/4 D₅ | Yellowish Bright Green | 7.6±0.16   | 4.9±0.23         | 58.0±2.45       | 551.3±3.49     | 180.7±0.12      | 1.4±0.16        | 23.5±1.16         | 45.4±0.27       | 49.1±0.42                 | 3640            |
|      | 6/5 D₆  | Yellow Brownish Green | 7.5±0.16   | 4.8±0.20         | 56.5±2.50       | 1012.2±3.45    | 150.3±0.13      | 1.4±0.16        | 22.8±1.02         | 44.5±0.35       | 52.0±0.52                 | 3640            |
|      | 21/5 D₇ | Bright Light Brown | 7.2±0.13    | 4.7±0.15         | 52.0±2.00       | 1413.3±3.55    | 126.3±0.14      | 1.4±0.16        | 22.1±0.92         | 43.5±0.38       | 56.3±0.22                 | 3640            |
|      | 5/6 D₈  | Dark Brown        | 6.8±0.20    | 4.6±0.16         | 46.6±2.74       | 1935.2±2.69    | 52.7±0.03       | 1.6±0.60        | 21.7±1.81         | 42.2±0.47       | 58.7±0.25                 | 3640            |
| Mean |      |                  | 7.05        | 4.5              | 50.15           | 1121.4         | 12.34           | 1.38            | 23.3              | 47.6            | 32.27                     |                 |

| S₂   | 21/2 D₁ | Dark Green        | 6.1±0.23    | 3.5±0.17         | 37.7±2.83       | 1033.2±1.56    | 83.0±0.30       | 1.0±0.10        | 24.2±0.89         | 53.1±0.32       | 0.0±0.0                   | 3640            |
|      | 8/3 D₂  | Light Green       | 6.2±0.20    | 3.9±0.18         | 38.8±2.47       | 1097.7±1.58    | 86.2±0.29       | 1.1±0.10        | 24.3±0.49         | 54.0±0.23       | 0.0±0.0                   | 3640            |
|      | 23/3 D₃ | Bright Green      | 6.8±0.25    | 4.5±0.17         | 46.8±3.46       | 1166.8±12.61   | 92.0±0.11       | 1.3±0.15        | 25.6±0.50         | 52.3±0.24       | 5.0±0.44                  | 3640            |
|      | 7/4 D₄  | Bright Green      | 7.1±0.23    | 4.5±0.17         | 50.9±3.16       | 894.0±12.81    | 138.0±0.11      | 1.3±0.15        | 24.5±0.43         | 49.0±0.31       | 30.8±0.49                 | 3640            |
|      | 22/4 D₅ | Yellowish Bright Green | 7.4±0.16   | 4.5±0.22         | 56.5±2.50       | 556.8±2.35     | 174.5±0.12      | 1.2±0.13        | 23.3±0.73         | 47.1±0.41       | 45.1±0.28                 | 3640            |
|      | 7/5 D₆  | Yellow Brownish Green | 7.2±0.25   | 4.4±0.16         | 52.6±2.00       | 1034.9±2.30    | 159.5±0.12      | 1.4±0.16        | 23.1±0.48         | 46.3±0.53       | 51.9±0.48                 | 3640            |
|      | 22/5 D₇ | Bright Light      | 7.0±0.101   | 4.4±0.16         | 49.0±0.16       | 1513.9±2.21    | 137.5±0.30      | 1.5±0.16        | 22.2±0.47         | 45.9±0.33       | 52.1±0.38                 | 3640            |
|          | Brown          | 6/6 D₈ | Dark Brown | 6.6±0.22 | 4.4±0.16 | 44.0±3.02 | 1992.0±5.31 | 51.9±0.10 | 1.5±0.16 | 20.7±0.34 | 44.0±0.38 | 55.1±0.43 |
|----------|----------------|--------|------------|---------|----------|-----------|------------|----------|----------|----------|----------|----------|
| Mean     |                |        |            | 6.8     | 4.2      | 46.9      | 1161.2     | 11.53    | 1.28     | 23.48    | 48.96    | 30.01    |
| S₃       |                | 22/2 D₁ | Dark Green | 5.9±0.23 | 3.4±0.16 | 35.3±2.78 | 1040.4±2.18 | 83.0±0.31 | 1.0±0.10 | 24.7±0.56 | 57.2±0.27 | 0.0±0.0  |
|          |                | 9/3 D₂  | Light Green| 6.1±0.18 | 3.8±0.20 | 37.5±2.20 | 1000.5±2.39 | 84.3±0.28 | 1.1±0.10 | 24.5±0.64 | 56.6±0.33 | 0.0±0.0  |
|          |                | 24/3 D₃ | Bright Green| 6.5±0.17 | 4.3±0.15 | 42.5±2.17 | 1117.1±9.93 | 87.3±0.06 | 1.3±1.5  | 26.2±0.36 | 56.9±0.39 | 5.4±0.77 |
|          |                | 8/4 D₄  | Bright Green| 6.7±0.15 | 4.4±0.16 | 45.1±1.81 | 942.1±2.60  | 138.5±0.07| 1.3±0.15 | 25.7±0.15 | 49.8±0.44 | 32.0±0.64 |
|          |                | 23/4 D₅ | Yellowish Bright Green | 7.2±0.25 | 4.5±0.17 | 52.4±3.53 | 561.4±1.79  | 170.5±0.12| 1.2±0.13 | 23.5±0.69 | 47.9±0.32 | 46.2±0.55 |
|          |                | 8/5 D₆  | Yellow Brownish Green | 7.0±0.21 | 4.4±0.16 | 49.4±2.96 | 1070.2±1.91 | 150.5±0.12| 1.3±0.15 | 23.4±0.70 | 46.9±0.21 | 52.2±0.73 |
|          |                | 23/5 D₇ | Bright Light Brown | 6.6±0.16 | 4.3±0.15 | 43.8±2.12 | 1619.5±1.94 | 119.5±0.13| 1.4±0.16 | 22.0±0.58 | 46.5±0.33 | 54.0±0.57 |
|          |                | 7/6 D₈  | Dark Brown | 6.4±0.16 | 4.3±0.15 | 41.2±2.12 | 1997.2±5.38 | 50.9±0.11 | 1.3±0.15 | 20.6±0.37 | 45.9±0.38 | 54.2±0.28 |
| Mean     |                |        |            | 6.5±0.22 | 4.2±0.16 | 46.9±3.02 | 1161.2      | 11.53    | 1.28     | 23.48    | 48.96    | 30.01    |
| Average of Across all site | 6.8     | 4.29   | 46.75      | 1150.4   | 11.64   | 1.3       | 23.54      | 49.12    |
| SEM+ for site | 0.096   | 0.086  | 1.27      | 11.17    | 0.883   | 0.07      | 0.352      | 0.20     | 0.19 |
| F-test   | *         | *      | *         | *       | NS      | *         | *         | *       | *       |
| CD       | 0.191     | 0.171  | 2.52      | 36.13    | -       | 0.14      | NS        | 0.40     | 0.38 |
| SEM+ for Date/day | 0.157   | 0.14   | 2.08      | 18.25    | 1.44    | 0.113     | 0.57       | 0.327    | 0.31 |
| F-test   | *         | *      | *         | *       | *       | *         | *         | *       | *       |
| CD for day | 0.312   | 0.27   | 4.11      | 36.13    | 2.85    | 0.223     | 1.137      | 0.649    | 0.61 |
| SEM+ for SxD | 0.273   | 0.24   | 3.60      | 31.6     | 2.496   | 0.195     | 0.995      | 0.567    | 0.538 |
| F-test   | NS       | NS     | NS        | NS      | NS      | *         | *         | *       | *       |
| CD for SxD | -       | -      | -         | 62.57    | -       | 0.387     | -          | 1.12     | 1.06 |
Similarly, the seed colour changed from dark green to dark brown at maturity (Table 2). The change in pod colour with the advancement of maturity was also reported by other researchers, also, in different tree species (Bonner, 1976; Ramakrishnan et al., 1990; Bharathi et al., 1996; Rai et al., 1988 and Shah, 2005). Further, it has been observed that the pod dimensions i.e. length and width also increase with the advancement of collection periods. With the advancement of maturity, the moisture content in pods also decreased. During the first collection in the month of February it was found maximum in all sites and there after it declined with the advancement of collection dates. At Agra site (S1), the pod moisture content decreased from 52.9 to 40.6%, whereas at Mathura (S2) and Hathrus sites (S3) it decreased from 54.5 to 39.2 % and 56.2 % to 42.7 %, respectively (Table 1). The average pod moisture content at (S1), (S2) and (S3) was 54.2%, 56.1% and 57.7 % at all three sites respectively (Table 1). Similarly, the seed moisture content also declined with the advancement of maturity.

Maximum seed moisture (S1 53.1±0.39%, S2 53.1±0.32 %, and S3 57.2±0.27 %) was observed in seeds collected during February month in all sites and the minimum (S1 42.2±0.47%, S2 44.0±0.38 %, and S3 45.9±0.38 %) was in the seeds collected during 1st week of June (Table 2). Loss of water during seed maturity is more inherent phase of seed development. The decline in seed moisture content during pod/seed development is often attributed to the continued deposition of storage material in seeds (Ellis et al., 1987). Change in colour and decrease in moisture content were also identified as maturity indicators by many researchers (Grover et al., 1963; Carl and Snaw, 1971; Edward, 1980; Cram and Linquist, 1982; Welbaun and Bradford, 1988; Maideen et al., 1990; Singh, 1998 and Phyartyal et al., 2002). The mean pod/fruit length and width were recorded 20.26 cm and 1.85 cm (S1), 20.18 cm and 2.0 cm (S2) and 19.98 cm and 1.7 cm (S3).

The data on seed germination (Table 2) revealed that the maximum germination (58.7 per cent at S1, 55.1 per cent at S2 and 54.2 per cent at S3) was recorded in the seeds collected during the 1st week of June for all the sites. At this collection date, the pods and seeds were having brown and dark brown, respectively. At this stage the seed moisture content was 42.2 % S1, 44.0 % S2 and 45.9 % S3, the size of seeds was 46.6 mm² (S1), 44.0 mm² (S2) and 41.2±2.12 mm² (S3), seed weight/ 100 seeds was 152.7 g (S1), 51.9 g (S2), 50.9 g(S3). Pandit et al., (2002) also reported the similar observation in Populus ciliata, the drop in moisture content of capsules from 80% to 60% during maturation coincided with the maximum germination in seeds.

From the present study, it may be concluded that colour as well as moisture content of pods and seeds are good indicators of maturity. First week of June is an appropriate period for pod collection for getting maximum germination in L. leucephalain semi-arid conditions of northern India.

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