

International Journal of Advanced Biochemistry Research



ISSN Print: 2617-4693
 ISSN Online: 2617-4707
 IJABR 2024; 8(3): 301-307
www.biochemjournal.com
 Received: 16-12-2023
 Accepted: 19-01-2024

Jai Kumar
 Faculty of Forestry, BAU,
 Ranchi, Jharkhand, India

Hiranmayee Nayak
 College of Forestry, OUAT,
 Bhubaneswar, Odisha, India

Animesh Sinha
 Institute of Forest
 Productivity (ICFRE),
 Ranchi, Jharkhand, India

Screening of suitable Kalmegh genotypes through its seed germination test

Jai Kumar, Hiranmayee Nayak and Animesh Sinha

DOI: <https://doi.org/10.33545/26174693.2024.v8.i3d.732>

Abstract

Percentage of germination is the most important characteristics of the seed to be used for cultivation. Since Kalmegh is a seed propagated crop, hence it is imperative that detailed information regarding its seed germination parameters may provide the much-required stimulus to utilize this plant in generating and sustaining a possible means of livelihood by integrating medicinal plants cultivation in different farming system. Keeping the importance of seed germination behavior, an experiment was undertaken during 2018-19 and 2019-20, to study the seed germination behavior of Kalmegh genotypes to screen out most promising genotypes of Kalmegh suited for the climatic and edaphic conditions of Jharkhand at BAU, Ranchi. Mean number of days taken for initiation of seed germination varied from 2.00 to 3.00 days with grand mean as 2.74 days, while mean number of days taken for 50% of final seed germination varied from 2.00 to 5.00 days. Mean number of days taken for completion of seed germination varied from 3.33 days (OAP₆) to 13.67 days (JHAP₃). Germination period of different Kalmegh genotypes varied from 6.33 days (OAP₆) to 15.67 days (JHAP₂, JHAP₃ and CHAP₁) with pooled mean as 11.93 days. Out of the days taken to complete germination period, nearly 43% of the time was required for seed germination initiation and 62% time to achieve 50% of final germination count. Once the seed started germination, it took 8.82 days to complete seed germination i.e. 57% time was required from initiation to completion of germination. Germination trend of seeds shows that most of the seeds germinated between third to sixth days. Mean rate of germination was varied from 0.017 (GAP₁) to 0.303 (OAP₆) with its pooled mean as 0.080, while mean germination energy varied from 5.33% (OAP₁) to 64.67% (IC471890). Mean germination percentage varied from 21.00% (OAP₁) to 97.67% (MPAP₃) with pooled mean as 61.09%. Altogether eight genotypes namely JHAP₁, OAP₆, MPAP₂, MPAP₃, MPAP₄, IC111286, IC471890 and GAP₁ showed excellent germination percentage having value more than 90.00%. Three Kalmegh genotypes (MPAP₃, OAP₆, JHAP₁) gave highest germination percentage (94.00, 93.00 & 90.67%) respectively. The germination percentage was not found correlated with its geographical distribution.

Keywords: Kalmegh, *Andrographis paniculata*, germination period, germination energy, germination percentage

Introduction

Germination of a seed involves the reactivation of the metabolic pathways that lead to growth and the emergence of the radicle and plumule (ISTA, 2006) ^[8]. Since germination is a complex biological process and several factors have to enact simultaneously on it; the resultant effect is reflected in the form of emergence of seedling, after a certain period of time (Black and Halmer, 2006) ^[2]. Only those seeds which germinate rapidly and vigorously under favorable & controlled conditions are likely to be capable of producing vigorous seedlings in field conditions. Thus, it would be desirable to have information regarding germination parameters for producing good quality seedlings. A systematic study on seed germination parameters is essential for any seed propagated crop to ensure crop stand and yield. Since Kalmegh is seed propagated, it is essential to assess the seed quality for ensuring the crop stand and herb yield which depend on quality seed (Kumar *et al.*, 2011) ^[12]. Seed germination is a big problem in Kalmegh due to its wild nature. Internal factors affecting seed germination are seed viability, genotype, seed maturation and seed dormancy. Low seed germination is one of the main problems in Kalmegh because of dormancy (Saraswathy *et al.*, 2004; Kumar *et al.*, 2011b; Talei *et al.*, 2012) ^[18, 13, 20]. Seed dormancy in Kalmegh is mainly caused by two different mechanisms consists of: a hard seed-coating layer and

Corresponding Author:
Jai Kumar
 Faculty of Forestry, BAU,
 Ranchi, Jharkhand, India

existence of unknown inhibitor proteins in the seed and seed coat. The hard seed-coating layer in Kalmegh seeds provides a combination of physical and physiological dormancy that prevents water uptake by the embryo resulting in lower germination percentage. The information available on seed quality and germination of Kalmegh is very meager. Therefore, enhancing seed germination to produce healthy and vigorous seedlings is crucial to meet current requirements (Kohli & Kumari, 1986; Kumari *et al.*, 2012) [11, 14].

Species selected for the investigations was *Andrographis paniculata* Wall. ex Ness, commonly known as Kalmegh used as a wonderful drug in all prevailing systems of medicine viz. Ayurvedic, Siddha, Unani, Homeopathic, traditional and modern system as well as in tribal medicine of India and South East Asian countries (Chadha, 1985) [3]. Kalmegh is a member of the Acanthaceae family, is an important indigenous medicinal plant found throughout tropical & sub-tropical Asia (Katakya and Handique, 2010) [9]. It is an annual, erect, stem acutely quadrangular, with many four-angled branches, grows to a height of around 30-110 cm in moist, shady places. Fruit is a linear-oblong capsule, 1-2 cm long and 2-5 mm wide, compressed

loculicidal, nearly glabrous, longitudinally furrowed on the broad faces. Capsule contains 6-12 small, subquadrate, yellow to dark brown, slightly translucent seeds (Norman and Bunyapraphatsara, 1992; Department of Medical Sciences, 1995) [16, 6]. It thrives well in tropical climatic conditions i.e. hot and humid. However, it can be cultivated in subtropical regions during the monsoon season (Lattoo *et al.*, 2008) [15].

Materials and Methods

The experimental site was Birsa Agricultural University, Kanke, Ranchi, located at 23°26'30" N latitude and 85°18'20" E longitude in Chhota Nagpur plateau, situated in north eastern part of India and at an altitude of between 646 m above the mean sea level in Jharkhand. The experimental materials comprised of twenty-five genotypes of Kalmegh, for which seeds were collected from its natural habitat across six states of India and NBPGR, New Delhi including wild and cultivated varieties. Out of 25 Kalmegh genotypes, 4 each were collected from Jharkhand, Chhattisgarh, Madhya Pradesh, Karnataka each, 6 from Orissa, 2 from NBPGR and one from Gujarat.

Table 1: Details of *Andrographis paniculata* genotypes used as experimental materials

Sl. No.	State / Institution	Genotypes & location
1.	Jharkhand	T ₁ - JHAP ₁ (Ranchi East Forest Division), T ₂ - JHAP ₂ (Hazariabag Forest Division), T ₃ - JHAP ₃ (Ramgarh Forest Division), T ₄ - JHAP ₄ (Khunti)
2.	Orissa	T ₅ - OAP ₁ (Angul), T ₆ - OAP ₂ (Nayagarh), T ₇ - OAP ₃ (Puri), T ₈ - OAP ₄ (Cuttack), T ₉ - OAP ₅ (Dhenkanal), T ₁₀ - OAP ₆ (Keonjhar)
3.	Chhattisgarh	T ₁₁ - CHAP ₁ (Dhamtari), T ₁₂ - CHAP ₂ (Sonpur Road, Dhamtari), T ₁₃ - CHAP ₃ (Bastar), T ₁₄ - CHAP ₄ (Rajnandgaon)
4.	Madhya Pradesh	T ₁₅ - MPAP ₁ (Amarkantak), T ₁₆ - MPAP ₂ (Vindhyan Range, Mandwa), T ₁₇ - MPAP ₃ (Rewa), T ₁₈ - MPAP ₄ (Kanha)
5.	Karnataka	T ₁₉ - KAP ₁ (Bijapur), T ₂₀ - KAP ₂ (Mysore Madikeri Road), T ₂₁ - KAP ₃ (Mysore Ooty Road), T ₂₂ - KAP ₄ (IIHR, Bangalore)
6.	NBPGR, New Delhi	T ₂₃ - IC 111286 (New Delhi), T ₂₄ - IC 471890 (New Delhi)
7.	Gujarat	T ₂₅ - GAP ₁ (Junagarh)

Seeds of Kalmegh were collected in the month of May, 2018 and trail was conducted using seed germinator to evaluate seed germination parameters. Experiment was conducted in Completely Randomized Design with 25 treatments replicated thrice at constant controlled temperature between 25 °C to 26 °C (Valdiani *et al.*, 2012c) [22] with relative humidity varied between 70% to 80% in seed germinator and 16 h light & 8 h dark regime. For germination tests, a total of 100 air-dried seed samples of each *Andrographis paniculata* genotypes were sterilized by soaking in 10% NaOCl solution for 10 minutes in separate petri plates (Talei *et al.*, 2011) [21]. Whatman No. 2 filter papers were chosen as the seedbeds to support a better germination. 10 ml of water per dish was given to moisten dry seeds and begin the imbibing process. Each plate was labeled appropriately with masking tape and wax pencil. Dissection scope was used to magnify samples to get a better look for accurate data collection. To avoid any contamination and humidity loss, all petri plates (16 cm diameter × 3 cm deep) were sealed with parafilm to prevent any water loss and infection during germination process (Talei *et al.*, 2012) [20].

Data on germination trend was recorded from initiation to completion of germination and based upon this, different germination parameters were calculated. Parameters studied were initiation of germination (days), 50% of final seed

germination (days), completion of germination (days), germination period (days), rate of germination (Bewley and Black, 1982) [1], germination energy (%) (Khanna, 1993) and germination percentage (%) by using the formula cited by Czabator (1962) [5]. Collected data on seed germination parameters was subjected to analysis of variance (ANOVA) and significant difference at 5% and 1% level was used to compare the means of different test parameters for all the genotypes as described by Panse and Sukhatme (1985) [17]. Standard error of mean, coefficient of variation and critical difference was computed by the procedure given by Singh and Chaudhary (1985) [19]. Germination percentages (original values) were transformed into arc sine root transformation used for statistical analysis.

Results and Discussion

Germination parameters of Kalmegh seeds: Seed germination parameters like number of days taken for initiation of germination, 50% of final germination and completion of seed germination by different Kalmegh genotypes is presented in Table 2.

Number of days for initiation of seed germination: During 2018-19, the mean number of days taken for initiation of seed germination varied from 2.00 to 3.00 days and highly significant difference was observed among

different Kalmegh genotypes. 16 Kalmegh genotypes took 3 days to initiate seed germination while eight had taken 2 days for it. Grand mean recorded for initiation of seed germination was calculated as 2.68 days in 2018-19, while in 2019-20, little higher value (2.79 days) was calculated. During 2019-20, almost same trend was observed *i.e.* the number of days taken to initiate the seed germination varied from 2.00 to 3.00 days with highly significant difference. The minimum number of days was observed for three Kalmegh genotypes namely JHAP₄, OAP₆ and CHAP₄ (2.00 days), while the maximum number of days (3.17 days) was

recorded in case of 5 Kalmegh genotypes namely OAP₃, OAP₅, CHAP₁, CHAP₂ and CHAP₃. In general, seeds of Jharkhand and Orissa had taken lesser number of days, while seeds of Chhattisgarh, Madhya Pradesh, Karnataka and Gujarat took higher number of days to initiate seed germination. Three genotypes JHAP₄, OAP₆ and CHAP₄ took less than two days for initiation of seed germination and genotypes of Chhattisgarh (CHAP₁, CHAP₂ and CHAP₃) and Orissa (OAP₃ and OAP₅) had taken highest number of days for initiation of seed germination (3.17 days).

Table 2: Mean number of days for initiation of germination, 50% of final germination and completion of seeds germination taken by different Kalmegh genotypes

Treatments	Days taken for initiation of seed germination		Days taken for 50% of final seed germination		Days taken for completion of seed germination	
	1 st year	2 nd year	1 st year	2 nd year	1 st year	2 nd year
T ₁ (JHAP ₁)	2.00 ^c	3.00 ^{ab}	2.00 ^f	3.33 ^{si}	7.00 ^{ijk}	6.67 ^{ij}
T ₂ (JHAP ₂)	2.67 ^b	2.67 ^{abc}	2.00 ^f	5.33 ^{cde}	12.67 ^{ab}	10.00 ^{cde}
T ₃ (JHAP ₃)	2.00 ^c	2.33 ^{bc}	3.00 ^c	5.67 ^{bcd}	13.67 ^a	10.00 ^{cde}
T ₄ (JHAP ₄)	2.00 ^c	2.00 ^c	3.67 ^b	6.67 ^a	7.00 ^{ijk}	11.00 ^{abc}
T ₅ (OAP ₁)	3.00 ^a	3.00 ^{ab}	2.67 ^{ce}	5.67 ^{bcd}	7.33 ^{ijk}	6.67 ^{ij}
T ₆ (OAP ₂)	2.00 ^c	2.33 ^{bc}	3.00 ^c	5.00 ^{de}	11.67 ^{bc}	9.67 ^{def}
T ₇ (OAP ₃)	3.00 ^a	3.33 ^a	3.00 ^c	5.67 ^{bcd}	6.33 ^{jk}	7.33 ^{hi}
T ₈ (OAP ₄)	3.00 ^a	3.00 ^{ab}	2.00 ^f	4.67 ^{ef}	12.67 ^{ab}	11.67 ^a
T ₉ (OAP ₅)	3.00 ^a	3.33 ^a	3.00 ^c	5.33 ^{cde}	7.00 ^{ijk}	9.67 ^{def}
T ₁₀ (OAP ₆)	2.00 ^c	2.00 ^c	3.00 ^c	3.00 ⁱ	3.33 ^l	6.00 ^j
T ₁₁ (CHAP ₁)	3.00 ^a	3.33 ^a	3.00 ^c	6.00 ^{abc}	12.67 ^{ab}	8.00 ^{gh}
T ₁₂ (CHAP ₂)	3.00 ^a	3.33 ^a	3.67 ^b	6.00 ^{abc}	11.00 ^{cd}	10.67 ^{abcd}
T ₁₃ (CHAP ₃)	3.00 ^a	3.33 ^a	3.00 ^c	5.00 ^{de}	7.67 ^{hij}	8.67 ^{fg}
T ₁₄ (CHAP ₄)	2.00 ^c	2.00 ^c	5.00 ^a	6.33 ^{ab}	10.33 ^{cdef}	11.33 ^{ab}
T ₁₅ (MPAP ₁)	3.00 ^a	3.00 ^{ab}	2.00 ^f	6.33 ^{ab}	8.33 ^{ghi}	11.00 ^{abc}
T ₁₆ (MPAP ₂)	3.00 ^a	3.00 ^{ab}	4.00 ^b	4.00 ^{fg}	6.67 ^{jk}	10.33 ^{bcd}
T ₁₇ (MPAP ₃)	3.00 ^a	2.67 ^{abc}	3.00 ^c	4.00 ^{fg}	6.33 ^{jk}	10.33 ^{bcd}
T ₁₈ (MPAP ₄)	3.00 ^a	3.00 ^{ab}	3.00 ^c	4.00 ^{fg}	6.00 ^k	9.67 ^{def}
T ₁₉ (KAP ₁)	3.00 ^a	3.00 ^{ab}	3.00 ^c	4.67 ^{ef}	4.00 ^l	7.33 ^{hi}
T ₂₀ (KAP ₂)	3.00 ^a	3.00 ^{ab}	3.00 ^c	4.00 ^{fg}	10.00 ^{def}	10.00 ^{cde}
T ₂₁ (KAP ₃)	3.00 ^a	3.00 ^{ab}	3.00 ^c	4.00 ^{fg}	9.00 ^{fgh}	11.00 ^{abc}
T ₂₂ (KAP ₄)	2.00 ^c	2.33 ^{bc}	2.00 ^f	4.00 ^{fg}	10.00 ^{def}	9.67 ^{def}
T ₂₃ (IC 111286)	2.00 ^c	2.33 ^{bc}	3.00 ^c	4.67 ^{ef}	10.67 ^{cde}	10.00 ^{cde}
T ₂₄ (IC 471890)	3.00 ^a	2.67 ^{abc}	3.00 ^c	4.00 ^{fg}	9.33 ^{efg}	10.67 ^{abcd}
T ₂₅ (GAP ₁)	3.00 ^a	3.00 ^{ab}	3.00 ^{cd}	4.00 ^{fgh}	7.00 ^{ijk}	9.33 ^{ef}
Grand Mean	2.66	2.79	2.96	4.85	8.71	9.47
S.E.(m)	0.07	0.23	0.12	0.23	0.49	0.35
C.D.5%	0.19	0.66	0.33	0.66	1.40	1.01
C.D.1%	0.25	0.86	0.43	0.86	1.81	1.31
C.V. (%)	4.27	14.09	9.09	8.24	9.75	6.45

It can be observed that all of the Kalmegh genotypes initiated seed germination activities between 2 to 3 days, and after initiation of germination, most of the seeds germinated between 3 to 5 days and after that very little increase in germination percentage was observed *i.e.* it becomes more or less constant. So, this period may be considered as very critical period for higher germination percentage of Kalmegh seeds. Above results are in accordance of Talei *et al.*, (2012) [20], they recorded initiation of seed germination of Kalmegh seeds as three days. Kumar *et al.*, (2011) [12] also found days to initiation of seed germination of Kalmegh as 3rd days from seed sowing at 30 °C. It was observed that the genotypes showed early initiation of germination had light colored seeds. Harvais and Hadley (1967) [7] demonstrated that light-coloured seeds can swell and germinate, while brownish or brown seeds remained without any symptoms of swelling.

Number of days for 50% of final seed germination:

During 2018-19, mean number of days taken for 50% of final seed germination varied from 2.00 to 5.00 days. Most of the genotypes attained 50% germination simultaneously with initiation of seed germination. Genotypes of Chhattisgarh (CHAP₄) and Madhya Pradesh (MPAP₂) took higher number of days than other genotypes to attain 50% germination. In 2019-20, the mean number of days was found more than the mean number of days for 50% germination during 2018-19 for almost all the genotypes. It might be because of physiological dormancy of freshly collected Kalmegh seeds. The grand mean for 50% germination during 2019-20 for all the Kalmegh genotypes was 4.85 days, which was almost double than of 2018-19 (2.96 days). In 2019-20, mean number of days for 50% germination was varied from 3.00 days (OAP₆) to 6.67 days (JHAP₄) and Kalmegh genotypes of Chhattisgarh and Madhya Pradesh took higher number of days than other

genotypes. The minimum number of days for 50% germination was recorded for JHAP₁ (2.67 days) and the maximum for JHAP₄ (5.17 days). Days taken for 50% of final seed germination was achieved in just after one days of initiation of seed germination, however two genotypes of Jharkhand (JHAP₃ and JHAP₄) took nearly 2 to 3 days to achieve it.

Number of days for completion seed germination: During 2018-19, mean number of days taken for completion of seed germination varied from 3.33 days (OAP₆) to 13.67 days (JHAP₃) and highly significant difference was noticed between different Kalmegh genotypes. Two genotypes from Madhya Pradesh (MPAP₃ and MPAP₄), one of Karnataka (KAP₁) and one of Orissa (OAP₃) had taken lesser number of days than other genotypes for completion of seed germination. Grand mean for all the Kalmegh genotypes in 2018-19 recorded lesser number of days (8.71 days) than of 2019-20 (9.47 days). In 2019-20, number of days for completion of seed germination was varied from 6.00 days (OAP₆) to 11.67 days (OAP₄) and highly significant difference was observed between different Kalmegh genotypes. Almost all the genotypes of Orissa took lesser number of days for completion of seed germination, however most of the genotypes of different states took more than 10 days to complete seed germination. The maximum value of seed germination days was observed for OAP₄ (12.17 days) and the minimum for OAP₆ (4.67 days). In some genotypes, like OAP₆, KAP₁ and OAP₃, the completion of germination period was observed as only 2 to 3 days, but in general, the days taken for completion of seed germination for most of the genotypes lied between 8-9 days. In general, initiation of seed germination started in

Kalmegh seeds between 2 to 3 days, and after its initiation, it completes their germination in 9.09 days.

Seed germination parameters like mean number of days taken for germination period and rate of germination of seeds by different Kalmegh genotypes is presented in Table 3.

Number of days for germination period of seeds: In 2018-19, the germination period of different Kalmegh genotypes varied from 6.33 days (OAP₆) to 15.67 days (JHAP₂, JHAP₃ and CHAP₁) and highly significant difference was observed among them. 16 genotypes of Kalmegh showed more than 10.00 days of germination period, while 9 genotypes took less than 10 days for it. Higher grand mean values of germination period of Kalmegh seeds were observed in 2019-20 (12.45 days) than of 2018-19 (11.41 days). In 2019-20, the range of germination period was varied from 9.00 days (OAP₁ and OAP₆) to 14.67 days (OAP₄). Only three genotypes namely JHAP₁, OAP₁ and OAP₄ took less than 10 days to complete the germination period while 22 genotypes took more than 10 days for it. In 2019-20 also, highly significant difference was observed between different Kalmegh genotypes. The range of germination period varied from 7.67 days (OAP₆) to 15.17 days (OAP₄). The germination period of few genotypes was less than 10 days (JHAP₁, OAP₁ and OAP₆) but three genotypes (JHAP₂, JHAP₃ and OAP₄) had taken more than 2 weeks to complete it and highly significant difference was noticed between these two groups. 12 genotypes took 10 to 12 days for complete their germination period, while in the case of OAP₆, minimum number of days (less than 8 days) was observed.

Table 3: Mean number of days for germination period and rate of germination of seeds taken by different Kalmegh genotypes

Treatments	Germination period of seeds (days)		Rate of germination of seeds	
	1 st year	2 nd year	1 st year	2 nd year
T ₁ (JHAP ₁)	9.00 ^g	9.67 ^{hi}	0.142 ^{cdefg}	0.15 ^{ab}
T ₂ (JHAP ₂)	15.67 ^a	13.67 ^{abcd}	0.078 ^{hi}	0.10 ^{bc}
T ₃ (JHAP ₃)	15.67 ^a	13.00 ^{bcde}	0.073 ⁱ	0.10 ^{bc}
T ₄ (JHAP ₄)	9.00 ^g	14.00 ^{abc}	0.142 ^{cdefg}	0.09 ^{bc}
T ₅ (OAP ₁)	9.33 ^g	9.00 ⁱ	0.172 ^{cd}	0.15 ^{ab}
T ₆ (OAP ₂)	13.67 ^{cd}	13.00 ^{bcde}	0.085 ^{ghi}	0.10 ^{bc}
T ₇ (OAP ₃)	9.33 ^g	10.67 ^h	0.161 ^{cde}	0.14 ^{abc}
T ₈ (OAP ₄)	15.67 ^{ab}	14.67 ^a	0.078 ^{hi}	0.09 ^c
T ₉ (OAP ₅)	10.00 ^{fg}	12.67 ^{cde}	0.142 ^{cdefg}	0.10 ^{bc}
T ₁₀ (OAP ₆)	6.33 ^h	9.00 ⁱ	0.303 ^a	0.17 ^a
T ₁₁ (CHAP ₁)	15.67 ^{ab}	12.00 ^{efg}	0.081 ^{ghi}	0.13 ^{abc}
T ₁₂ (CHAP ₂)	14.00 ^{ac}	13.67 ^{abcd}	0.090 ^{fghi}	0.09 ^{bc}
T ₁₃ (CHAP ₃)	11.33 ^{ef}	12.00 ^{ef}	0.125 ^{cdefghi}	0.12 ^{abc}
T ₁₄ (CHAP ₄)	12.33 ^{cde}	14.33 ^{ab}	0.097 ^{fghi}	0.09 ^{bc}
T ₁₅ (MPAP ₁)	11.33 ^{ef}	14.00 ^{abc}	0.137 ^{cdefgh}	0.09 ^{bc}
T ₁₆ (MPAP ₂)	9.67 ^{fg}	12.67 ^{cde}	0.150 ^{cdef}	0.10 ^{bc}
T ₁₇ (MPAP ₃)	9.33 ^g	13.00 ^{bcde}	0.161 ^{cde}	0.10 ^{bc}
T ₁₈ (MPAP ₄)	9.00 ^g	12.67 ^{cde}	0.178 ^c	0.10 ^{bc}
T ₁₉ (KAP ₁)	7.00 ^h	10.00 ^{hi}	0.250 ^b	0.14 ^{abc}
T ₂₀ (KAP ₂)	13.00 ^{cde}	13.00 ^{bcde}	0.100 ^{efghi}	0.10 ^{bc}
T ₂₁ (KAP ₃)	12.00 ^{de}	13.67 ^{abcd}	0.114 ^{defghi}	0.09 ^{bc}
T ₂₂ (KAP ₄)	12.00 ^{de}	12.33 ^{de}	0.102 ^{efghi}	0.10 ^{bc}
T ₂₃ (IC 111286)	12.67 ^{cde}	13.00 ^{bcde}	0.093 ^{fghi}	0.10 ^{bc}
T ₂₄ (IC 471890)	12.33 ^{cde}	13.33 ^{abcde}	0.108 ^{efghi}	0.09 ^{bc}
T ₂₅ (GAP ₁)	10.00 ^{fg}	12.33 ^{de}	0.142 ^{cdefg}	0.02 ^d
Grand Mean	11.41	12.45	0.13	0.11
S.E.(m)	0.54	0.47	0.01	0.01
C.D.5%	1.54	1.33	0.02	0.02
C.D.1%	2.01	1.73	0.03	0.02
C.V. (%)	8.22	6.49	11.11	8.48

Rate of germination of seeds: The rate of germination of seeds of different Kalmegh genotypes showed highly significant difference among them and its maximum value was recorded for OAP₆ (0.303) and minimum in JHAP₃ (0.073) 2018-19. Only two genotypes namely OAP₆ and KAP₁ had excellent germination rate *i.e.* more than 0.20. Rest 23 genotypes showed medium or less rate of germination.

During 2019-20, again all the genotypes showed highly significant difference among them and its range varied from 0.017 (GAP₁) to 0.169 (OAP₆). In 2019-20, none of the Kalmegh genotypes showed good germination rate, however only three genotypes namely JHAP₁, OAP₁ and OAP₆ showed satisfactory rate of germination having value more than 0.150. As far as the grand mean of both the years is concerned, lesser rate of germination was observed in 2019-20 (0.105) than of 2018-19 (0.132). Grand mean for both the years for rate of germination was calculated as 0.080 and different Kalmegh genotypes showed highly significant difference among them. The maximum rate of germination was observed OAP₆ (0.236) and the minimum for GAP₁ (0.080). The genotypes having low germination period showed higher rate of germination and *vice-versa*. Perusal of seed germination parameters of Kalmegh seeds revealed that out of the days taken to complete germination period (11.93 days), nearly 23% of the time was required for seed germination initiation (2.74 days) and 33% time (3.91 days) to achieve 50% of final germination count. Once the seed started germination, it took 9.09 days to complete seed germination *i.e.* 77% time was required from initiation to completion of germination.

Seed germination parameters like mean germination energy and germination percentage of different Kalmegh genotypes is presented in Table 4.

Germination energy of seeds: During 2018-19, the germination energy of Kalmegh seeds varied from 12.67% (JHAP₄) to 64.67% (IC471890) and different Kalmegh genotypes showed highly significant difference. Genotypes such as JHAP₄, OAP₁, OAP₄, CHAP₁, and MPAP₁ showed less than 20% germination energy. During 2019-20, significantly lesser germination energy was calculated (22.00%) with respect to 2018-19 (37.08%) as revealed by the grand mean of respective year. In 2018-19, maximum value of germination energy was noticed for MPAP₃ (50.00%) and minimum for OAP₁ (5.33%). In 2019-20, all genotypes showed considerably less germination energy and JHAP₄, OAP₁, OAP₅, CHAP₁, MPAP₁ showed less than 10% germination energy.

More than 40.00% germination energy was observed in seven genotypes namely JHAP₁, OAP₆, MPAP₂, MPAP₃, MPAP₄, IC 471890 and GAP₁, while very lower germination energy (less than 10%) was observed in JHAP₃ and MPAP₁. Nearly 12% of the total seeds germinate daily and at the peak of germination nearly 30% of the seed germinates. Two genotypes OAP₆ and KAP₁ showed high rate of germination, while nearly 16 Kalmegh genotypes had their rate of germination of 0.100. Kumar *et al.*, (2011) [12] also found maximum germination energy on 7th day at 25 °C. They found high rate of germination of Kalmegh seeds around 25 °C with its range varied from 10 °C to 30 °C.

Table 4: Mean germination percentage and 100 seed weight of different Kalmegh genotypes

Treatments	Germination energy of seeds (%)		Germination percentage of seeds (%)	
	1 st year	2 nd year	1 st year	2 nd year
T ₁ (JHAP ₁)	45.00 ^{de}	37.33 ^c	94.67 ^{ab}	86.67 ^{abc}
T ₂ (JHAP ₂)	45.67 ^{de}	23.33 ^{gh}	72.67 ^e	66.33 ^{fg}
T ₃ (JHAP ₃)	35.67 ^f	14.33 ^{jk}	48.33 ⁱ	51.33 ⁱ
T ₄ (JHAP ₄)	12.67 ^k	6.00 ^m	21.67 ^m	38.67 ^j
T ₅ (OAP ₁)	17.33 ^l	5.33 ^m	21.00 ^m	22.33 ^m
T ₆ (OAP ₂)	33.00 ^f	20.00 ^{hi}	60.67 ^g	55.33 ^{hi}
T ₇ (OAP ₃)	24.67 ^{hi}	15.00 ^j	29.67 ^k	30.33 ^{kl}
T ₈ (OAP ₄)	45.67 ^{de}	19.67 ^{hi}	79.67 ^d	69.33 ^f
T ₉ (OAP ₅)	14.67 ^{jk}	5.67 ^m	26.67 ^{kl}	21.67 ^m
T ₁₀ (OAP ₆)	48.33 ^{cd}	45.33 ^b	96.33 ^a	89.67 ^{ab}
T ₁₁ (CHAP ₁)	17.33 ^l	9.67 ^{lm}	26.33 ^{kl}	26.67 ^{lm}
T ₁₂ (CHAP ₂)	44.00 ^e	13.67 ^{jk}	66.67 ^f	56.33 ^h
T ₁₃ (CHAP ₃)	45.67 ^{de}	17.00 ^{ij}	69.67 ^{ef}	57.67 ^h
T ₁₄ (CHAP ₄)	29.33 ^g	10.67 ^{kl}	43.67 ^j	40.67 ^j
T ₁₅ (MPAP ₁)	13.33 ^k	6.33 ^m	24.67 ^{lm}	31.67 ^k
T ₁₆ (MPAP ₂)	58.00 ^b	37.00 ^c	89.67 ^c	83.67 ^{cde}
T ₁₇ (MPAP ₃)	63.67 ^a	50.33 ^a	97.67 ^a	90.33 ^a
T ₁₈ (MPAP ₄)	51.33 ^c	32.33 ^{de}	95.00 ^{ab}	85.00 ^{bcd}
T ₁₉ (KAP ₁)	21.67 ⁱ	19.00 ⁱ	45.33 ^{ij}	51.33 ⁱ
T ₂₀ (KAP ₂)	26.33 ^{gh}	25.67 ^{fg}	66.67 ^f	64.67 ^g
T ₂₁ (KAP ₃)	36.67 ^f	20.00 ^{hi}	57.00 ^h	54.33 ^{hi}
T ₂₂ (KAP ₄)	35.67 ^f	20.67 ^{hi}	61.67 ^g	62.33 ^g
T ₂₃ (IC 111286)	34.00 ^f	28.67 ^{ef}	89.67 ^c	79.33 ^e
T ₂₄ (IC 471890)	64.67 ^a	33.00 ^d	92.33 ^{bc}	81.33 ^{de}
T ₂₅ (GAP ₁)	62.67 ^a	34.00 ^{cd}	94.00 ^{ab}	86.33 ^{abc}
Grand Mean	37.08	22.00	62.85	59.33
S.E.(m)	1.27	1.30	1.21	1.53
C.D.5%	3.61	3.71	3.46	4.35
C.D.1%	4.69	4.82	4.49	5.65
C.V. (%)	5.93	10.26	3.88	5.18

Germination percentage of seeds: During 2018-19, the maximum germination percentage was noticed for MPAP₃ (97.67%) and the minimum for OAP₁ (21.00%) and different Kalmegh genotypes showed highly significant difference among them. Altogether eight genotypes namely JHAP₁, OAP₆, MPAP₂, MPAP₃, MPAP₄, IC111286, IC471890 and GAP₁ showed excellent germination percentage having value more than 90.00%. The grand mean of all the 25 Kalmegh genotypes was calculated as 62.85%, while in 2019-20, slightly lesser germination percentage (59.33%) was recorded. In 2019-20, the range of germination percentage varied from 21.67% (OAP₅) to 90.33% (MPAP₃) and again highly significant difference was observed among them. Three genotypes namely OAP₁, OAP₅ and CHAP₁ gave less than 30.00% germination percentage. The range of germination percentage was varied from 21.67% (OAP₁) to 94.00% (MPAP₃) and different Kalmegh genotypes showed highly significant difference in between them. On the average basis, germination percentage of Kalmegh seeds was nearly 60.00%. From the findings, it can be inferred that the eight genotypes like MPAP₃, OAP₆, JHAP₁, GAP₁, MPAP₄, IC 471890, MPAP₂ and IC 111286 can be screened out as best to obtain maximum germination percentage (more than 80.00%). Chauhan *et al.*, (2009) found 72, 75 and 78 germination % of Kalmegh seeds in filter paper, soil and sand respectively at 25 °C. They observed maximum germination percentage as 95% at 25 °C, while Kumar *et al.*, (2011)^[12] found 95% germination potential of Kalmegh seeds. Talei *et al.*, (2012)^[20] recorded the germination percentage Kalmegh seeds between 34.33 to 94.33%. Kumari *et al.*, (2012)^[14] also observed highest germination (99.2% and 88.3%) in Kalmegh variety CIM-Megha and wild collection. The germination percentage was not found correlated with its geographical distribution, and in each state, some germplasm gave higher germination percentage, while some gave lower germination percentage. Valdiani *et al.*, (2012a)^[20] reported high contamination and low germination are two prevalent problems in *Andrographis paniculata*.

Conclusion

Mean number of days taken for initiation of seed germination varied from 2.00 to 3.00 days and most of the genotypes attained 50% germination simultaneously with initiation of seed germination. Minimum number of days for 50% germination was recorded for JHAP₁ (2.67 days). Mean number of days taken for completion of seed germination varied from 3.33 days (OAP₆) to 13.67 days (JHAP₃). Out of the days taken to complete germination period (11.93 days) by Kalmegh seeds, nearly 23% of the time was required for seed germination initiation (2.74 days) and 33% time (3.91 days) to achieve 50% of final germination count. Once the seed started germination, it took 9.09 days to complete seed germination i.e. 77% time was required from initiation to completion of germination. The range of germination period varied from 7.67 days (OAP₆) to 15.17 days (OAP₄), while the rate of germination of seeds varied from 0.303 (OAP₆) to 0.073 (JHAP₃). The range of germination energy varied from 5.33% (OAP₁) to 64.67% (IC471890), and the range of germination percentage varied from 21.00% (OAP₁) to 97.67% (MPAP₃). The germination percentage was not found correlated with its geographical distribution, and in each state, some germplasm gave higher germination percentage,

while some gave lower germination percentage. Altogether eight genotypes namely JHAP₁, OAP₆, MPAP₂, MPAP₃, MPAP₄, IC111286, IC471890 and GAP₁ showed excellent germination percentage. So, it may be concluded that the genotypes like JHAP₁, OAP₆, MPAP₂, MPAP₃, MPAP₄, IC 111286, IC 471890 and GAP₁ may be screened out as superior for higher germination and better stocking thus higher yield in the field and may be adopted for commercial cultivation under the climatic and edaphic conditions of Jharkhand.

References

1. Bewley JD, Black BM. Physiology and biochemistry of seed germination, Part- II. New York: Springer Verlag; c1982. p. 32-34.
2. Black MH, Halmer P. The encyclopedia of seeds: science, technology and uses. Wallingford, UK: CABI; c2006. p. 224.
3. Chadha YR. The Wealth of India: Raw Materials, Vol. 1A. New Delhi, India: Council of Scientific and Industrial Research; c1985. p. 264.
4. Chauhan JS, Tomar YK, Singh NI, Ali S, Badoni A, Debarati RA, *et al.* Assessment of compatible substratum for *Andrographis paniculata* standard seed germination testing. J Am Sci. 2009;5:70-75.
5. Czabator FJ. Germination value: an index combining speed and completeness of pine seed germination. Forest Sci. 1962;8:386-396.
6. Department of Medical Sciences, Ministry of Public Health. Thai Herbal Pharmacopoeia. Vol. I. Bangkok, Thailand: Prachachon Co., Ltd.; c1995. p. 152.
7. Harvais G, Hadley G. The development of *Orchis purpurella* in asymbiotic and inoculated cultures. New Phytologist. 1967;66:217-230.
8. International Seed Testing Association (ISTA) Rule. International Rules for Seed Testing. Bassorsdorf, Switzerland: International Seed Testing Association; c2006.
9. Katakya A, Handique PJ. A brief overview on *Andrographis paniculata* (Burm. f) Nees., a high valued medicinal plant: Boon over synthetic drugs. Asian J Sci Technol. 2010;6:113-118.
10. Khanna LS. Principles and Practice of Silviculture. Dehra Dun: Khanna Banhu; c1993. p. 195.
11. Kohli RK, Kumari A. Cause and cure of dormancy in *Cassia occidentalis* L. seeds. In: Workshop on special problems in physiological investigations of tree crops. Kottayam: R.R.I.I.; c1986. p. 75-81.
12. Kumar B, Verma SK, Singh HP. Effect of temperature on seed germination parameters in Kalmegh (*Andrographis paniculata* Wall. ex Nees.). Ind Crops Prod. 2011;34:1241-1244.
13. Kumar RN, Chakraborty S, Nirmal KJI. Methods to break seed dormancy of *Andrographis paniculata* (Burm. f. Nees): An important medicinal herb of tropical Asia. Asian J Exp Biol Sci. 2011;2:143-146.
14. Kumari A, Lal RK, Singh KLB. Comparative study of seed germination and seed vigour test in *Andrographis paniculata* (Acanthaceae). Botanica serbica. 2012;36(1):49-52.
15. Lattoo SK, Dhar RS, Khan S, Bamotra S, Bhan MK, Dhar AK, *et al.* Comparative analysis of genetic diversity using molecular and morphometric markers in

- Andrographis paniculata* (Burm. f.) Nees. Genet Resour Crop Evol. 2008;55:33-43.
16. Norman RF, Bunyapraphatsara N. Thai Medicinal Plants. Thailand; c1992. p. 402.
 17. Panse VG, Sukhatme PV. Statistical methods for agricultural workers. New Delhi: ICAR; c1985. p. 381.
 18. Saraswathy S, Manavalan RSA, Vadivel E, Manian K, Subramanian S. Studies on seed germination in Kalmegh (*Andrographis paniculata* Nees.). J South Ind Hortic. 2004;52:286-290.
 19. Singh RK, Chaudhary BD. Biometrical methods in quantitative genetic analysis. New Delhi: Kalyani Publishers; c1985.
 20. Talei D, Mihdzar AK, Yusop MK, Valdiani A, Puad MA. Physico-protein based dormancy in medicinal plant of *Andrographis paniculata*. J Med Plants Res. 2012;6(11):2170-2177.
 21. Talei D, Saad MS, Khanif YM, Kadir MA, Valdiani A. Effect of different surface sterilizes on seed germination and contamination of King of Bitters (*Andrographis paniculata*). Am-Euras J Agric Environ Sci. 2011;10(4):639-643.
 22. Valdiani A, Kadir MA, Saad MS, Talei D, Omidvar V, Chia SH, *et al.* Intraspecific crossability in *Andrographis paniculata* Nees. A barrier against breeding of the species. Scientific World Journal. 2012;2012:1-9.
 23. Valdiani A, Kadir MA, Tan SG, Talei D, Puad MA, Nikzad S, *et al.* Nain-e Havandi (*Andrographis paniculata*) present yesterday, absent today: A plenary review on underutilized herb of Iran's pharmaceutical plants. Mol Biol Rep. 2012;39(5):5409-5424.