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Effect of Methods of Seed Collection on Quality and Storability of *Prinsepia utilis* Royle (Bhekal) Seed

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ABSTRACT

The effect of different methods of seed collection on seed viability and vigour of *Prinsepia utilis* Royle (bhekal) were studied. Bhekal fruits were collected using two methods: (a) directly from plant branches (green, purple-green, and violet fruits) and (b) fruits collected from the ground under the plant (fallen fruits and fruits eaten by birds). Data collected were arc-sin transformed before being subjected to a two-way Analysis of Variance (ANOVA). The Least Significant Difference (LSD) was used to compare the significantly different treatment means at a 5% probability level. Seeds collected directly from the branches showed significant differences from fruits collected from the ground under the plant in terms of germination percentage (G%), mean germination time (MGT days), and germination value (GV). The fruits directly collected from branches don't show any significant difference regarding germination percentage and mean germination time. Violet fruits showed maximum germination value as compared to green and purple-green fruits. There were no significant differences in fruits collected from the ground and fruit eaten by birds in terms of germination percentage, mean germination time, and germination value. Seeds collected directly from branches- violet showed a maximum mean viability period (P_{50}) of 72 days compared to all.

Keywords *Prinsepia utilis,* Seed, Collection, Extraction, Storage, Viability.

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INTRODUCTION

*Prinsepia utili*s Royle (bhekal) is a moderate-sized deciduous shrub of Rosaceae primarily confined to continental eastern Asia. Its distribution ranges from western Pakistan and Northwest India to Southern and central China, Eastern Mongolia, Russia, Manchuria, and Korea. In India, this species is distributed throughout the Himalayan region. Generally, occurs from 1600 to 2500m altitude ranges of Uttarakhand, Jammu and Kashmir, Himachal Pradesh, Sikkim, and Khasi - Jayantia hills (Maikhuri *et al.*1994). It has varying habitats, including the hill slopes to the vertical cliffs and mountain valleys to the barren land. It grows naturally on slopes near the edges of the forest area, in well-drained and water-deficient areas on hill slopes, in open and degraded forests, and at the margins of agriculture fields, villages, and habitations.

This plant is also reported to have been colonized in the degraded areas in Garhwal Himalayas (Maithani *et al.* 1986). Therefore, the species could be helpful in the reclamation of the degraded wastelands in the Himalayas (Bhagat and Singh 1991). Bhekal fruits are edible, and their seed kernels yield about an edible oil having rubefacient and many other medicinal properties. The local people use plant root, stem, and leaves to cure skin alignment and diseases. The seed cake is applied for fungal treatments. Besides its medicinal and ecological importance, the plant plays a crucial role in holistic and ritualistic practices in havan-pooja, baptism ceremonies, cremations, and other magico-religious ceremonies.

In bhekal, the fruit matures mid-May in Chakrata hills, and all fruits are shed within a month. Fruits at different stages of maturity can be seen during the fruiting season. Collecting bhekal seed of the desired maturity is difficult as the fruit grows in bunch. Thus, the fruits collected are of different colors, containing seeds at various stages of maturity. The method of seed collection and physiological maturity of seed is a significant factor that directly affects the vigour of nursery stock. Several methods for the collection of fruits and seeds have been developed. The methods range from simple collection from the ground after natural seed fall to advanced techniques using sophisticated and expensive equipment such as elevated platforms, mechanical shakers, balloons, or helicopters. Seed collection is often the most labor and cost-intensive part of all the seed handling operations, and one may be tempted to cut down labor costs and choose the cheapest possible method. Knowing the seed's developmental stages and exact time of seed maturity is crucial for collecting an abundant quantity of vigours and healthy seeds. Physiologically pre-harvest sprouts or immature seeds are a constrained problem resulting in poor quality of seed (Wang *et al*. 2018, Finch-Savage and Bassel 2016, Fu *et al.* 2017, Lee *et al*. 2016).

A perusal of the literature revealed that no in-depth study carried out on the seed collection method was available. Present investigations were carried out to examine whether seeds collected directly from branches, viz., green fruit, purple-green fruit, and violet fruit, were better than those collected from under the plant (over-matured fallen fruits and fruit eaten by birds) regarding germination and storability.

MATERIALS AND METHODS

Sample collection, seed extraction, and processing

The samples were collected in May 2017 from Chakrata forest, India, located between 30°31' to 31°3' N and 77°42' to 78°05' E. The mean maximum and minimum temperatures were -4±5°C and 25±5°C, and humidity between 10-70% were recorded during the experiment. Bhekal fruits were collected using two methods: (a) directly from plant branches (green fruit, purple-green fruit, and violet fruits) and (b) fruits collected from the ground under the plant (fallen fruits and fruits eaten by birds).

Fruits directly collected from the branches were kept overnight in gunny bags at ambient temperature to soften the pulp. All green and purple-green fruits became soft within a day, while some green fruits (approx. 15%) remained firm and were discarded from the lot. Seeds were de-pulped completely from the endocarp by macerating them into cotton bags (30x30 cm). Processed clean seeds were shade-dried till EMC (Equilibrium Moisture Content) for three days. The seed lot identity was maintained during drying, and the percent moisture content and initial germination percentage were determined as per ISTA (2021).

Germination study

All the seed samples were stored at ambient room temperature in open containers. Seeds were examined under controlled conditions for assessing viability and vigour at different storage periods viz 0 days (fresh), 30, 60, 90, 120, and 150 days. Germination tests were performed between blotting paper (BP) using 4 replications of 100 seeds, incubated at 25±1 °C and 90±5 °C R.H. (Relative humidity). Seed germination data were recorded daily for up to 21 days, and when the radicle became 1 cm in length, the seed was considered germinated. Mean germination time (MGT days) was calculated as described by Rawat and Thapliyal (2003), while the germination value (GV) was calculated as per Djavanshir and Pourbeik (1976). The pattern of seed germination declines in

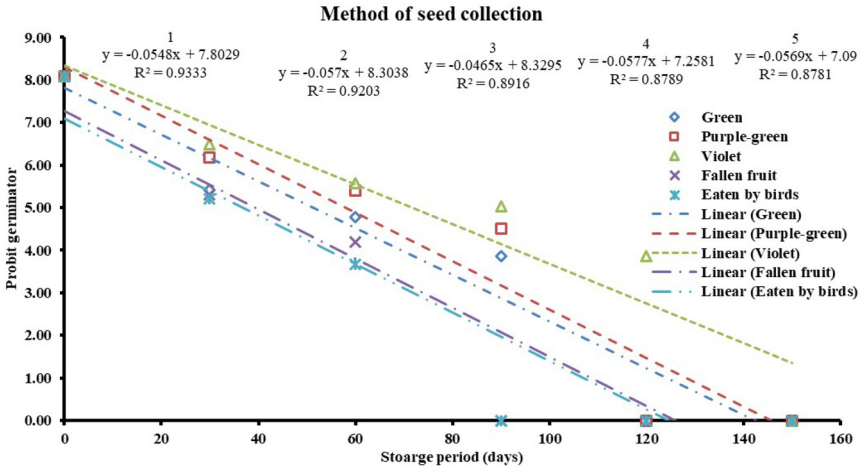

Fig. 1. Regression between probit germination and seed storage period (days) obtained from different collection methods.

each seed lot was subjected to probit analysis (Fig. 1). Finney (1952), however, the survival curves were drawn (Roberts 1973) between expected germination percent and storage period (days) for examine the mean viability period, i.e., P_{50} (time taken to death of 50% seeds) as described by Ellis and Roberts (1980) and Ellis (1982). The germination %, mean germination time (MGT days), and germination value (GV) were analyzed through analysis of variance (ANOVA) using the SPSS package. The p value <0.05 was taken as significant.

RESULTS

Effect of method of seed collection on quality of fresh seeds

Table 1 indicates the effect of the method of seed collection, mean germination time (MGT), and germination value (GV). Seeds collected directly from plant branches (green fruit, purple-green fruit, and violet fruits) significantly showed higher germination percentage, lower mean germination time, and maximum germination value as compared to seeds collected from the ground under the plant (i.e., fruit eaten by birds and fallen fruit). Seed procured from violet fruit showed a significantly higher germination percentage (95.50), followed by purple-green and green fruits with 93.00 and 91.50%, respectively, as compared to fallen fruits (65.25) and fruit eaten by birds (63.50). However, there was a non-significant difference between fruits collected from branches (Table 1).

The seeds extracted from violet fruits showed significantly lower mean germination time (7.66 days), followed by purple-green and green fruits with 9.07 days and 9.20 days, respectively, as compared to the seeds extracted from fallen fruits (12.70 days) and fruit eaten by birds with 13.49 days. However, no significant difference existed between seeds obtained directly from branches (Table 1). Seeds obtained from violet fruits showed the highest GV (66.42), followed by seeds procured from purple-green fruits and green fruits with 55.00 and 53.98, respectively, as compared

Table 1. Effect of different methods of seed collection on germination of fresh seeds.

Parameters	Methods of seed collection				
	Fruits collected from branches			Fruits collected from ground	
	Green	Purple-green	Violet	Fallen fruit	Eaten by birds
Germination (%)	91.50 (75.67)[a]	93.00 (75.47)[a]	95.50 (81.33)[a]	65.25 (53.93)[b]	63.50 (53.04)[b]
MGT (days)	09.20[b]	09.07[a]	07.66[b]	12.70[a]	13.49[a]
GV	53.98[b]	55.00[b]	66.42[a]	28.21[c]	24.60[c]

In each row the values not followed by the same letter are significantly different (p> 0.05).

to the fallen fruit (28.21) and fruit eaten by birds (24.60). Germination value showed a non-significant difference between the seeds obtained from green fruits and purple-green fruits (Table 1).

Effect of method of seed collection on quality of stored seeds

Seeds obtained from various methods of seed collection were stored in open containers for upto 150 days at ambient room temperature. Germination shows a decline with the storage period increased in all types of seeds. The rate of decrease till 60 days of storage was higher in seeds extracted from fruits eaten by birds (57.50%), followed by green fruits (53.50%), fallen fruit collected from the ground (51.25%), and purple-green fruit with a 32.25% decline. Seeds obtained from violet fruit show the least reduced germination activity (26.25%). However, when seeds were stored for a more extended period (upto 90 days), seeds obtained from fruit eaten by birds and fallen fruit failed to show any germination. The seeds obtained from green fruit showed 79.25% decline activity, followed by purple-green fruits and violet fruit with 64.00 and 46.25% decline activity, respectively. All kinds of seeds were failed to show germination when stored for a more extended period of 150 days. The rate of decline in germination percentage was found to be lowest in violet seeds during this storage period. The pattern of reduction in germination is presented in Fig. 2.

Fruits directly collected from plant branches showed a higher mean germination percentage (43.54) than those collected from the ground and under the plant (18.88). Seeds extracted from violet fruit showed a significant maximum mean germination percentage (52.71), followed by purple-green and green fruits with 44.13 and 33.79 percentage upto 150 days of storage as compared to fallen fruit (19.96) and fruit eaten by birds (17.79) (Table 2). The MGT was observed to increase (poor) significantly (Fig. 3a), while GV decreased with increasing storage period (Fig. 3b). Fruits directly collected from branches exhibited lower mean MGT (15.82 days) than fruits collected from the ground under the plant (19.40 days). Violet fruit showed significantly lowest mean MGT (14.14 days) followed by the seeds obtained from purple-green fruits (15.98 days), green fruit

Table 2. Effect of seed storage on mean germination (%), mean MGT and mean germination value (GV) of different methods of seed collection.

Parameters	Methods of seed collection				
	Fruits collected from branches			Fruits collected from ground	
	Green	Purple-green	Violet	Fallen fruit	Eaten by birds
Germination (%)	33.79 (30.91)^{c}	44.13 (37.36)^{b}	52.71 (45.72)^{a}	19.96 (19.24)^{d}	17.79 (17.42)^{d}
MGT (days)	17.35^{c}	15.98^{d}	14.14^{e}	19.14^{b}	19.66^{a}
GV	14.78^{c}	21.08^{b}	30.30^{a}	6.83^{d}	5.43^{d}
P_{50} (days)	**51.15**	**57.96**	**71.60**	**39.14**	**36.73**

In each row the values not followed by the same letter are significantly different ($p > 0.05$).

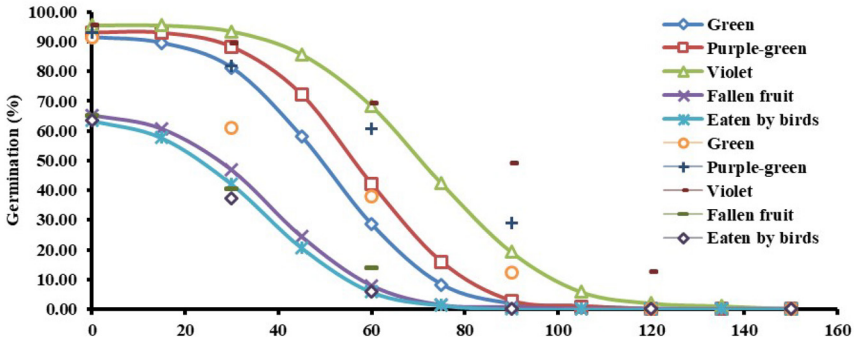

Fig. 2. The decline in germination percentage during storage of seeds, obtained from different collection methods. The lines show survival curves and points show original germination values.

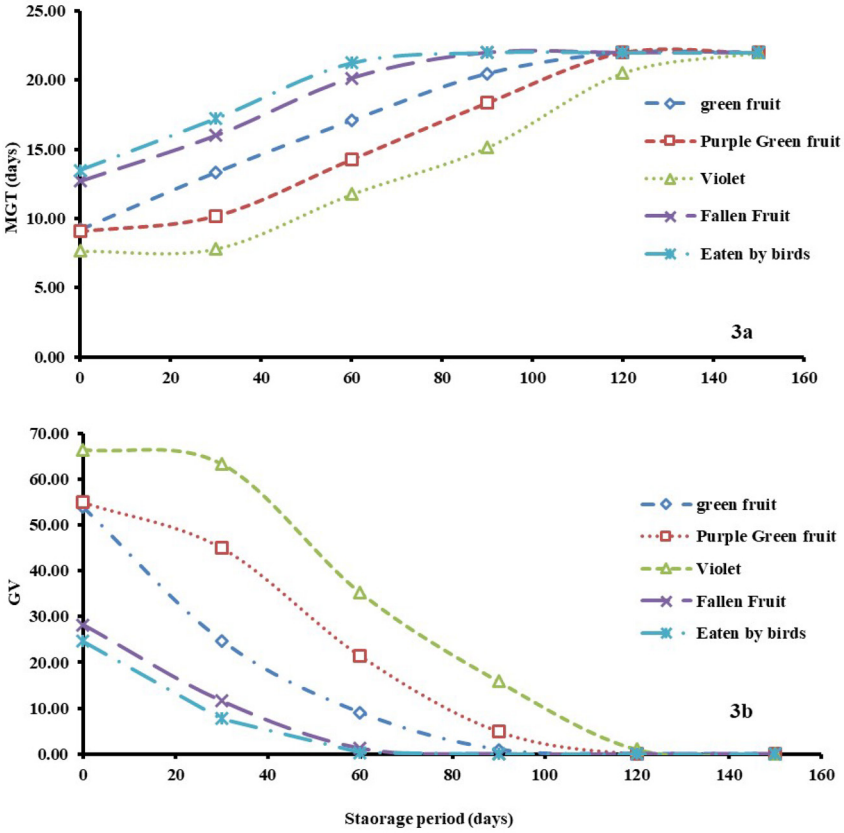

Fig. 3. The change in MGT (3a) and GV (3b) during storage in seeds obtained from different methods of seed collection.

(17.35 days), and the fruits collected from the ground (19.14 days). The poorest mean MGT was recorded in fruit eaten by birds with 19.66 days after 150 days of seed storage (Table 2). Fruits directly collected from plant branches also showed higher mean GV (17.93) than those collected from the ground under the plant (6.13). Violet fruits showed a significant maximum GV (30.30), followed by purple-green (21.08), green fruit (14.78), fruit collected from the ground (6.83), and fruit eaten by birds (5.43). However, a non-significant difference was found between fruits collected from the ground and fruit eaten by birds (Table 2). Survival curves showed that seeds collected directly from plant branches retained the highest mean viability compared to those collected from the ground (fallen fruit and fruit eaten by birds). Seeds procured from violet fruit retained a maximum period of mean viability (P_{50}) with 71.60 days as compared to the seeds obtained from purple green fruits (57.96 days), green fruit (51.15 days), fallen fruit (39.14 days), and fruit eaten by birds (36.73 days) (Table 2).

DISCUSSION

Seed collection is often a labor and cost-intensive part of the seed-handling operation. Appropriate methods may reduce labor costs. Any cheapest method can be used if seed viability and longevity are unaffected. Rajabi *et al.*(2020), Baskin and Baskin (2014) and Xu *et al.* (2014), reported a wide range of variations in germination. This is generally due to seed source (Footitt *et al*. 2020), environmental conditions (Penfield and MacGregor 2017), seasonal dormancy (Finch-Savage and Footitt 2017), but much of the reported variation may be due to the method of collection and seed handling (Schmidt 2011, Chancy and Knudson 1988).

The collection of *P. utilis* seeds can be made either directly from plant branches or from fallen fruits. The fallen fruits consist of two categories, i.e., whole fruits and stones, which birds ate. Fallen fruits are easy to collect without harm and will also cut the labor cost. But they are prone to contamination with soil-borne pathogens (Terborgh 2012, Schafer and Kotanen 2003, Schafer and Kotanen 2004, Blaney and Kotanen 2001, Beckstead *et al.* 2007). Fallen fruits are generally matured. Collection of seeds directly from the plant is more labor and cost-intensive. But it can be collected at the right time of maturity as per our desire without losing seed viability and vigour, which is an important aspect.

Seeds collected from branches, viz., green fruit, purple-green fruit, and violet fruit, showed significantly better germination than seeds collected from the ground under the plant (fallen fruit and fruit eaten by birds). Similarly, the mean germination time (MGT) and germination value (GV) follow the same trend. Seed collected directly from plant branches when stored showed the slowest gradual decline in germination value and increase in MGT. While fallen fruits (over-mature) or fruit eaten by birds had lost rapid germination ability during storage. Seeds collected from branches also remained viable for a longer period (P_{50}) (72 days) as compared to seeds of purple-green (58 days), green fruit (51 days), fallen fruit (39 days), and fruit eaten by birds (37 days). Our findings align with the previous reports by Susanto *et al.* (2016). They stated that fallen fruits tend to show low viability. These findings duly supported by the observation of Srimathi *et al.* (2013) in *Pongamia pinnata,* Gurunathan *et al.* (2009), Kathiravan (2004), Silva *et al.* (2017) in *Jatropha curcas,* Srimathi *et al*. (2001) in Jamun. Ferreira *et al*. (2017), Hay and Whitehouse (2017), Tetteh *et al.* (2018) also found a swift and significant reduction of germination percentage in the seeds obtained from immature fruits (Yellow and Green). Bhekal seeds seem to attain peak of maturity when the fruit turns violet as it showed significantly minimum MGT and maximum GV and performed best during storage. In disparity to our findings, Hathi *et al.* (2020), Gupta (2019), and Muthu and Selva (2017) advocated that post-harvest of maturity indices stored better than dark fruit color. Murrinie *et al.* (2021) also showed that *Feronia limonia* L. Swingle collected from the ground showed the highest seed germination and seedling emergence with or without post-harvest maturation storage. However, *F. limonia* seeds are different and require preconditioning before showing.

CONCLUSION

On the basis of the experiment, it is concluded that *P. utilis* fruits of any color, capable of softening within overnight, will give better germination percentage and viability than the fruits collected from the ground, and violet fruit may be preferred for longer storage of seeds rather than seeds extracted from the ground under the plant (fallen fruits and fruit eaten by birds) should not be collected in any case.

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