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ORIGINAL PAPER

Seed germination and seedling development of *Prunus armeniaca* under different burial depths in soil

GUO Cai-ru • WANG Zhen-long • LU Ji-qi

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Abstract: A semi-greenhouse study was conducted to understand the effects of soil burial depth on seed germination and seedling development. The seeds of wild apricot (*Prunus armeniaca*) were buried at the soil depths of 0-cm, 4-cm, 8-cm, and 12-cm, respectively, to simulate the seed hoarding behavior of rodents in the field. The results revealed that the rates of seed germination and established seedlings from buried seeds were both the highest in 4-cm burial depth group, and then decreased with increasing soil depth. The number of rotten seeds increased in deeper burial depth. It is unfavourable for seed germination at 0-cm burial depth (i.e., seeds were laid on soil surface). There was insignificant effect of burial depth on growth of established seedlings. The results from this study indicated that proper burial depth in soil would be helpful for the seed germination and seedling growth. The seedlings derived from buried seeds at shallower depth (4 cm) in this research have advantage in their early development.

Keywords: wild apricot (*Prunus armeniaca*); seed; soil; germination rate; seedling growth

Introduction

Seed and seedling stages are two of the most critical phases during a plant's life history, while seed germination is an irreversible process with high-risk, wrong timing, and location of germination may cause the death of the individual, impacting population recruitment (Harper 1977). Seed germination and seedling growth are influenced by many abiotic and biotic factors. Burial by soil is thought to be very important factor for the seed germination of many plant species (Barnett 1977). Seeds buried at different soil depths may experience different environmental conditions affecting seed germination and seedling development, such as the concentration of oxygen and carbon dioxide, temperature, water availability, nutrition availability, and others (Borchert et al. 1989). The shallower seed is buried in soil under natural conditions, the easier it may be found by predators, whereas deeply buried seeds have to invest more reserves into stems for their elongation before shoot apices can reach and emerge soil surface. Burial depth is a key factor for the fate of seed (Chen and Maun 1999; Guo et al. 2001; Guo et al. 2009).

Wild apricot (*Prunus armeniaca*) is a common fruit crop in hills in temperate deforested regions of China. This plant species is either the dominant species in wild apricot shrub, or the subdominant or co-dominant species in other types of shrubland on dry-sunny slopes (Ma et al. 1997), and usually be a pioneer species in the course of community succession in deforested area (Zhang and Wang 2001). Owing to their high nutritional value, wild apricot seeds are the preferred food resources for small rodents (Chen et al. 2002), and they often were removed and larder hoarded and / or scatter hoarded by rodents in the field for the future when food supply is poor in winter and early spring (Lu and Zhang 2004b, 2008; Li and Zhang 2007; Zhang et al. 2007). Some of those scatter-hoarded seeds might eventually escape from predation by hoarders or other predators, and germinate, establish new seedlings successfully (Lu and Zhang, 2004b; Zhang et al. 2007). Wood mice and squirrels, as seed dispersers, especially play an important role during seed germination and seedling establishment via their scatter-hoarding behavior, in which one or more seeds are separately buried at shallow surface in the soil (several centimeter in soil) (Vander Wall 1990 1993b; Lu and Zhang 2004b; Zhang et al. 2005). For a comprehensive understanding of the recruitment process, it is, therefore, necessary to reveal how rodents impact on the mortality of wild apricot juveniles at each stage.

It was demonstrated that depth of seeds buried by scatter-hoarding rodents will facilitates seedling establishment (Jansen and Forget 2001; Lu and Zhang 2004a; Vander Wall 1990; Will-

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GUO Cai-ru • WANG Zhen-long • LU Ji-qi (✉)
Institute of Biodiversity and Ecology, Department of Bioengineering, Zhengzhou University, Zhengzhou 450001, P. R. China.
E-mail: lujq@zzu.edu.cn

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son and Traveset 2000; Zhang et al. 2005; Zhang et al. 2007). The reason is possible that the water availability for buried seeds is adequate and stable for seed germination (Shaw 1968) during periods of drought (Forget 1990; Kollmann and Schill 1996), and the latter is especially important for recalcitrant seed species. Furthermore, soil burial can promote seed germination and seedling establishment by reducing the difficulty of radicle's inserting to soil (Griffin 1971). Consequently, roots of germinated seed would penetrate the soil surface more easily, and emerging seedlings would root more firmly.

In this study, we specifically intend to investigate the effects of burial depths on germination of wild apricot seeds and their seedling growth. By burying seeds in different depths in soil, we try to know the effects of burial depth on seed germination and seedling recruitment, and to understand the interaction between scatterhoarding behavior of rodents and regeneration of forest tree. We predicted that shallow burial depth (about 4 cm) will promote seed germination and seedling development of *Prunus armeniaca*.

Methods

Seeds collection

Wild apricot, *P. armeniaca*, fruits in June annually. We collected the fruits in June, 2007 from Mt. Wangwushan area, Jiyuan, Henan Province, China, and then took out the seeds for experimental use. Seed of wild apricot is ovoid in shape with biconvex sides, 22.1 mm×16.0 mm (long diameter × wide diameter) in size (n = 50), and weighs for (0.66 ± 0.16) g, (n=200) in fresh seeds.

Seed planting

Plastic flower pots (30 cm in height and 20 cm in diameter, and with 3 small holes at the bottom) were employed and filled with mixture of fertile soil and fine sand (2:1). Total 200 perfect seeds of wild apricot, similar in size and mass, were selected for experiments, and were divided into four groups (i.e., 50 in each group). The first 50 seeds were then planted at 50 flower pots with a soil depth of 0-cm. Similarly, the other three groups of seeds were planted in soil with depths of 4-cm, 8-cm, and 12-cm, respectively.

All the flower pots with seeds were placed outdoor, and watered with an interval of 3-5 days. Once the first seedling emerged, we investigated and recorded the emerging time of seedlings and seedling growth status including leaf number and seedling height every two days.

The seedling growth was divided into a few stages, such as pre-germination, rooting, subterranean stem, and seedling. Based on these categories, we recorded the number of emerged seeds, seedlings, and rotten seeds, then measured the stem height, and axial root length of seedlings. At the end of investigation, all the seedlings were excavated from flower-pot, and cleaned with fresh water. Each seedling was dried in an oven to constant weight under (120±5)˚C. Finally, for all seedlings, data including dried weights of leaves, stem above ground, stem below ground and root were collected. This experiment was carried out from March to June, 2008.

Data analysis

SPSS for Windows (version 13.0) was employed in data statistics and analysis. Based on normal distribution of data, one-way ANOVA was used in determining the effects of burial depth on seed germination, and seedling growth. LSD was employed in determining the differences of seedling biomass between every two burial depth groups. Chi-square test was used for analyzing the differences of seed germination rates between seeds in four burial depths, and seed germination rates among burial depth groups.

Results

In general, unsuitable burial depths of seeds produced a negative effect on seed germination and root growth in seeds of wild apricot, while the seeds at shallower burial depths took more advantages.

Seed germination among different burial depths

The seed germinating results of wild apricot seeds buried in all four burial-depth groups were investigated and recorded (Table 1). The result revealed that seed germination rate was the highest at 4-cm depth group (32%), while the seed germination rate was 5% in 0-cm depth group. For 8-cm and 12-cm depth groups, the rates of seed germination decreased sharply with increasing burial depths. The differences of rates of established seedlings among four burial-depth groups were significant (χ^2=20.933, *df*=3, P<0.001). The number of rotten seeds increased with the increasing burial depths (Table 1).

Table 1. Germination result of buried seeds of *Prunus armeniaca* among four burial-depth groups

Burial depth (cm)	Rotten seeds	Un-germinated seeds	Incomplete germination seeds	Seedling number
0	0	46	2	2
4	4	19	11	16
8	9	20	10	11
12	14	23	12	1

Time budget of seedling emergence

On 24th day after seed planting, seedlings emerged from 0 cm, 4-cm and 8-cm burial groups, whereas the first seedling derived from 12-cm burial-depth group was found at 27th day after beginning of this experiment (Fig. 1). There were no new seedlings emerged on 44th day after seeds planting in each burial-depth group.

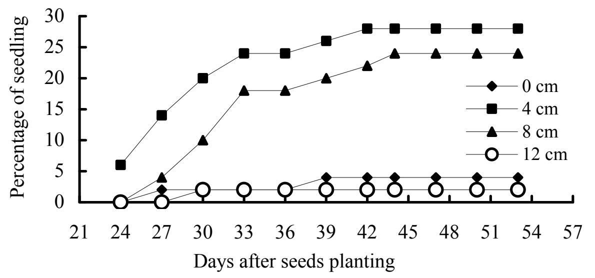

Fig. 1 Time budget of seedling emergence in four burial-depth groups of seeds of *Prunus armeniaca* after planting

Influence of soil burial depths on seedling development

Seedling heights and leaf number

In the course of our investigation, the developed status of seedlings was measured and recorded (Table 2). Seedlings from 4-cm burial-depth group grew rapidly in the early stage (before 45th day). There was proximate significant difference in seedling height (Figs. 2, 3), and leaf number (Fig. 4) among four burial burial-depth groups, while the difference in seedling heights was insignificant with time.

Table 2. Growth status of seedlings from seeds of *Prunus armeniaca* among four burial-depth groups

Burial depth (cm)	Stem length (cm)	Root length (cm)	Leaf number	Leaf weight (mg)	Root weight (mg)	Root growth rate (g·d^{-1})	Dry weight of seedling (mg)
0	13.11±3.86	16.2±1.7	15.5±5.5	894±41	2065±75	16.52	3266±208
4	20.09±0.91	14.06±0.83	16.93±1.61	1031±81	1565±135	12.52	3166±219
8	22.33±1.45	9.8±0.75	16.9±1.58	1016±121	1150±175	9.20	2791±296
12	27.1	11.2	17	990	1370	10.96	3022

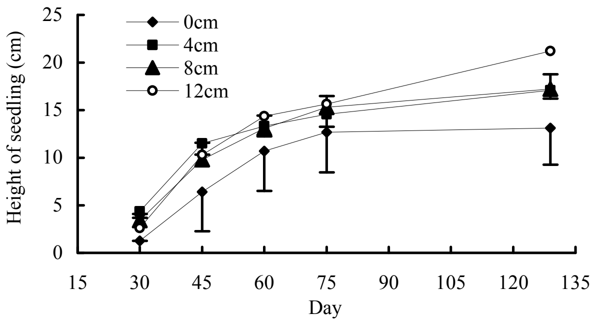

Fig. 2 Mean seedling heights among four burial-depth groups in different developing stages

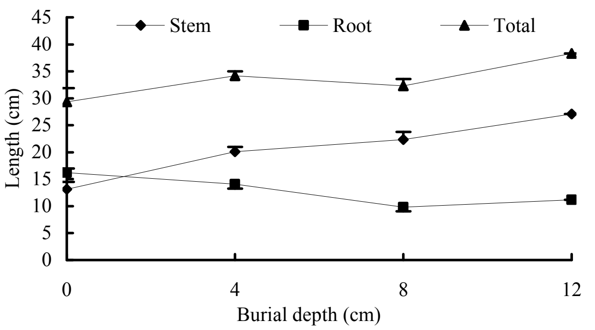

Fig. 3 Mean lengths of different parts of seedling

Dry weight of different parts of seedling

The weights in three parts of established seedlings were measured for each burial-depth group (Fig. 5). The mean biomasses of leaves were 894 g, 1 031 g, 1 016 g, and 990 g at burial-depth of 0 cm, 4 cm, 8 cm, and 12 cm, respectively, and there were insignificant differences among them (F=0.092, df=3, 26, p=0.964). For root, although the mean biomass in 0-cm group was larger than that in three other burial-depth groups, the differences among them were not significant (F=2.173, df=3, 26, p=0.115). The mean weights for whole seedling in four burial groups were respectively 3 266 g, 3 166 g, 2 791 g, and 3 022 g, and also exhibited insignificant differences (F=0.421, df=3, 26, p=0.739). This indicated that development of an emerged seedling was insignificantly affected by seed burial depth in soil.

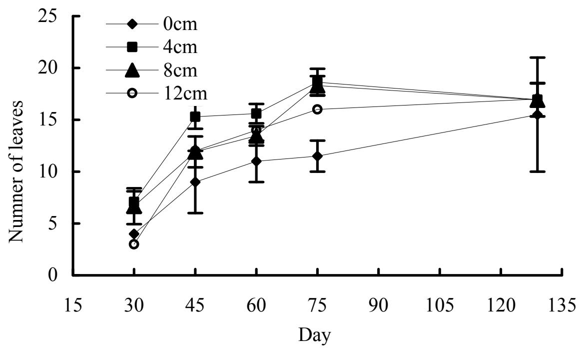

Fig. 4 Comparison of leaf number of seedlings derived from four burial-depth groups

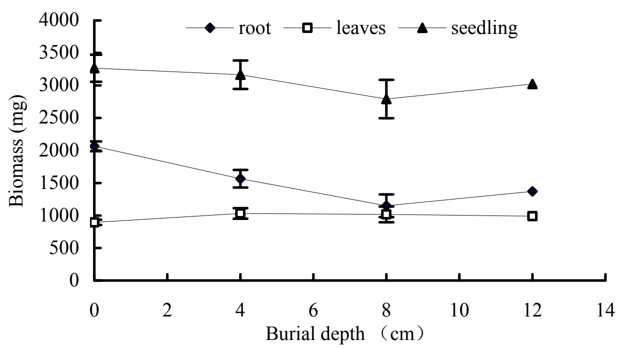

Fig. 5 Comparison of weights of root, leaves and whole plant of established seedlings among different burial-depth groups

Discussion

Seed germination represents a risky transition from the stage most tolerant to environmental conditions (i.e., resting seed) to the weakest and most vulnerable stage in plant development, the seedling (Harper 1977). The result from our research showed that wild apricot seeds germinated better in 4-cm than that in other burial depth groups. It is surprisingly that small rodents usually scatter hoarded (i.e., shallowly buried) seeds of wild apricot in the similar burial depth under field conditions (Lu and Zhang 2004a, b; Zhang and Wang 2001; Zhang and Zhang 2008). Furthermore, some seedlings were established successfully from buried seeds that escaped from predation by hoarders and other seed consumers (Li and Zhang 2007; Lu and Zhang 2004 a, b; Zhang and Wang 2001). As a fact, burial will protect the seeds from draught and predation by other animals (Barnett 1977; Shaw 1968), and is thought to be very important for germination of many plant species (Griffin 1971; Borchert et al. 1989). However, merely two seeds in 0-cm burial depth group germinated and established seedling successfully. This was possibly because the surface soil would provide inadequate humidity for the needs of seeds' germinating. Similar results were reported by several authors (Guo et al. 2001; Su et al. 2007; Yang et al. 2007; Guo et al. 2009). Our result indicated that suitable and shallow burial depth in soil will promote seed emerging and seedling development, while it was not propitious for seeds recruitment either on soil surface or in deeply burial depth (Fig. 6).

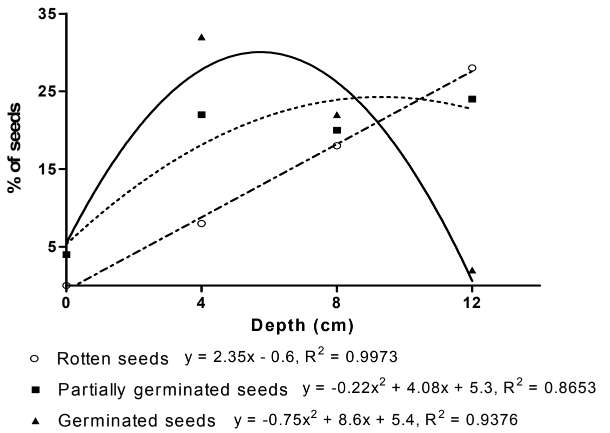

Fig. 6 Relationship between seed burial depth and seed status including seed rot, seed germination and incomplete seed germination

In this research, a positive relationship was found between rates of rotten seeds and burial depth (Fig. 5). In other words, deeply buried seeds would easily suffer from rotting, because soil humidity within flower pot was higher in deeper soil than that in surface and shallower soil. Nonetheless, this protocol would minimize the treated error for data analysis, and was employed in previous researches (Guo et al. 2001; Guo et al. 2009). Soil water saturation over prolonged periods of time generates a negative impact on nearly all terrestrial plants, as a consequence of the slow diffusion rates of gases in water (Jackson 1985; Armstrong 2002), which hampers oxygen supply to roots and reduces respiration and photosynthesis rates (Voesenek et al. 2006). Several studies have shown that soil waterlogging, as a result of periodic-to-continuous flooding of bottomland, is the major factor affecting tree regeneration in many temperate forested wetlands (Kevin and Brooks 2003; Pérez-Ramos and Marañón 2009). In this research, more rotten seeds were found in deeper soil layer, which was possibly due to higher humidity in deeper burial depths.

Seed burial also reduces the risk of seeds being discovered by predators or being attacked by pests. Experiments have shown that, in the field, buried seeds disappear (i.e., removed by seed predators) at far lower rates than those seeds placed on soil surface (Vander Wall 1990; Zhang et al. 2005; Zhang et al. 2007). In temperate zone, experimentally scatterhoarded seeds indeed disappear more slowly under lower densities, indicating that seeds would suffer littler predation (Stapanian and Smith 1984; Clarkson et al. 1986; Zhang et al 2007).

Suitable conditions including temperature and humidity are necessary for seed germination, emergence, and seedling recruitment (Michael and Paul 2000; Kos and Poschlod 2007, 2008). The variance of germination and seedling recruitment might reflect the influence of temperature and humidity among different burial depths. Under the optimal configuration between sunshine and temperature, seeds would germinate better, and more seedlings were established successfully. The result from this study showed that 4-cm depth group had higher seed germination rate, which indicated that the favorite micro-habitat for germinating of seeds of wild apricot was possibly at the 4-cm depth group (Vander Wall 1990, 1993a; Zhang et al. 2005; Zhang et al. 2007).

The mean daily increased weights of root were 16.52, 12.52, 9.20 and 10 ($g\cdot d^{-1}$) at the burial depths of 0 cm, 4 cm, 8 cm, and 12 cm, respectively. Higher measured value in 0-cm group (16.52) was reasonable because seedlings derived from this group occupied in the largest soil depth in pots. However, few seedlings established within this burial depth weakened the above mentioned fitness. Under field conditions, a less developed root system would inevitably decrease the survival rate of the resultant seedling (Lloret et al. 1999) by reducing seedling's ability to absorb water deeper during the dry season (Nicotra et al. 2002).

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