Experimental sand burial affects seedling survivorship, morphological traits and biomass allocation of *Ulmus pumila* var. *sabolusa* in Horqin Sandy Land, China

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Abstract As a native tree species, *Ulmus pumila* var. *sabolusa* (sandy elm) is widely distributed in Horqin Sandy Land, China. However, seedlings of this species have to withstand various depths of sand burial after emergence because of increasing soil degradation, which is mainly caused by overgrazing and climate change. An experiment was conducted to evaluate the changes in its survivorship, morphological traits and biomass allocation when seedlings buried at different burial depths : unburied controls and seedlings buried vertically up to 33, 67, 100 or 133% of their initial mean seedling height. The results showed that partial sand burial treatments (i.e., less than 67% burial) did not reduce seedling survivorship, which still reached 100%. However, seedling mortality increased when sand burial was equal to or greater than 100%.In comparison with the control treatment, seedling height and stem diameter increased at least by 6 and 14% with partial burial, respectively. In the meantime, seeding taproot length, total biomass, and relative mass growth rates at least enhanced by 10%, 15.6%, and 27.6%, respectively, with the partial sand burial treatment. Furthermore, sand burial decreased total leaf area and changed biomass allocation in seedlings, partitioning more biomass to aboveground organs (eg., leaves and stems) and less to belowground parts (roots). Complete sand burial after seedling emergence inhibited its re-emergence and growth, even leading to its death. Our findings indicated that seedlings of sandy elm had some resistance to partial sand burial and were

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adapted to sandy environments from an evolutionary perspective. The negative effects of excessive sand burial after seedling emergence might help to understand failures in recruitment of sparse elm in the study region.

Keywords: sand burial, seedling, sandy elm, morphological, biomass allocation, Horqin Sandy Land

Introduction

Horqin Sandy Land, shaped in the middle the Pleistocene period, is located in the southeast of the Mongolia Plateau, China (Qiu, 1989). Because of climatic changes (rainfall distribution and global warming) and excessive human disturbances (i.e., over-utilization of renewable natural resources), vegetation degradation and land desertification have become more obvious in the past 50 years (Cao et al., 2008; Jiang et al., 2003; Zhang et al., 2004). Sand moves fast in the horizontal or vertical space because of the effects of strong winds during spring and summer, leading to different burial depths, which might range from 0.5cm to 56.0cm (Liu et al., 2014).

Soil genesis is the pivotal process that determines the evolution of the soil system, and offers services and resources to mankind (Berendse et al., 2015; Niu et al., 2015). Simultaneously, disturbances (such as land-use intensification and overgrazing) have a profound impact on the soil genesis process because of the increasing population and consumption (Brevik et al., 2015; Verheijen et al., 2009; Wang et al., 2016). Excessive human interferences change soil hydrological, geochemical and biological cycles, inducing serious land degradation such as acidification, salinization and desertification (Bellamy et al., 2005; Foley et al., 2005; Gabarrón-Galeote et al., 2013; Smith et al., 2015). It is well known that vegetation plays an important role in controlling soil genesis and degradation in fragile ecosystems such as estuarine, desert and sandy lands (Berendse et al., 2015; Cerdà, 1998; Miao et al., 2014). Moreover, vegetation functional traits (e.g., morphology and establishment) in response to environmental stress (e.g., nutrient deficiency, water deficit, high irradiance, extreme

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temperatures) might be important adaptive life history adaptive strategy (Miner et al., 2005; Wang et al., 2014). Plants usually have to face a trade-off between survival and growth in response to environmental changes by regulating their phenotypic plasticity (e.g., biomass allocation, relative growth rate) and/or physiological traits (e.g., antioxidant enzyme activities, membrane permeability, contents of osmotic substance) (Li et al., 2015; Qu et al., 2012; Tian et al., 2015; Wu et al., 2013).

In sandy ecosystems, instability of the soil surface is one of the most damaging factors to biological activity. Furthermore, a sparse vegetation cover and a loose soil texture are highly susceptible to sand movements (Liu and Guo, 2005; Yan et al., 2005). Sand movement, the most direct evidence caused by land degradation, is regarded as a selective force determining colonization, establishment and establishment of vegetation (Maun, 1994; Maun and Lapierre, 1986). Plants might respond differently to various degrees of sand burial, and evolve different regenerative adaptations during the periods of soil seed bank formation, seed germination, and seedling emergence and development (Li et al., 2014; Qian et al., 2015; Tang et al., 2016).

As an indigenous tree species, *Ulmus pumila* var. *sabolusa* (sandy elm) has been widely distributed in the leeward slope of fixed and semi-fixed sand dunes and became the main component of sparse woodlands of the Horqin Sandy Land (Jiang et al., 2013; Tang et al., 2014; Tang et al., 2013). Since prehistoric times, it has been closely relate to human life, providing hardwood for farming tools and furniture, fuel for nomad, and fodder from its tender leaves, young fruits and edible bark (Ma, 1989; Schlütz et al., 2008). In addition, the sparse-elm woodlands, not only offered shelter for wildlife and domestic animals and a suitable environment for psammophytes; but also protected soil from wind erosion and burial,

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providing a very important ecological and social function in these arid and semi-arid lands (Yang et al., 2003). Despite we realize of the effects of sand burial on establishment of sandy elm, much of our comprehension and recognition to date come from ocular observations at the field rather than from controllable experiments (Maun, 1997). For example, we observed that even though many plant of non-dormant, dispersed seeds germinated and seedling emergence occurs in the late spring, few surviving seedlings were detected in the following field surveys in the degraded sparse woodlands.This phenomenon is hampering its recruitment and will have negative effects on future community structure of these woodland ecosystems.

Studies on the effects of sand burial have been widely reported in the field of seedlings survival (Belcher, 1977; Cheplick and Grandstaff, 1997; Harris and Davy, 1987; Li et al., 2015; Liu et al., 2008; Perumal and Maun, 2006), physiological characteristics (Shi et al., 2004; Wang et al., 2012; Zhao et al., 2015), and reproductive strategies (Liu et al., 2014; Sun et al., 2014; Zhao et al., 2007) of coastal marshes and wetlands plants. In general, it appears to be a threshold sand burial depth for each plant species to matain vigor and the subsequent sustain growth (Maun, 1997). Below that burial level, plant emergence and development have been promoted by increasing sand burial depth (Qu et al., 2012; Yang et al., 2007). Once the threshold have been exceeded, a deterioration of seedling vigor and reduced growth has occur, and even leaded to seedling death (Maun, 1997; Maun and Lapierre, 1986). However, no research has been conducted on the effects of continual sand accumulation on sandy elm seedlings after emergence to date, because of the limited area of sparse elm woodland. So we investigated the effects of experimental sand burial on seedling survivorship and growth of sandy elm. The main objectives of this study were: (1) to evaluate the effect of sand burial on seedling survivorship; (2) to access the changes on seedling

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morphological traits and biomass allocation in response to sand burial, and (3) to explain the failure of sandy elm regeneration, and provide a theoretical basis for achieving a successful recruitment and vegetation establishment on sandy elm woodlands.

Materials and methods

Study site

The experiment was conducted at the Wulanaodu Desertification Experimental Station of the Institute of Applied Ecology, Chinese Academy of Sciences (43°02′N,119°39′E, 480 m a.s.l.) located in western Horqin Sandy Land, China (Figure 1). This site experiences temperate continental climate. Mean annual temperature and precipitation are 7.3℃ and 315 mm, respectively. Almost 75% of precipitation occurs from June to September during the growing season. Annual average wind speed is 4.4 m s^{-1}; the windy season is from March to June (Liu et al., 2012; Miao et al., 2014). The landscape is characterized by sparse woodlands, sand dunes and lowland areas. The dominant soils are aerolian soils, and major plant species include some shrubs (e.g., *Salix gordejevii* and *Caragana microphylla*), and perennial and annual herbs (e.g., *Bassiadasyphylla*, *Agriophyllum squarrosum*) (Cao et al., 2011) .

Experimental methods

In mid-May 2015, sandy elm seeds were first collected from multiple, mature individuals and then mixed altogether. After careful selection, uniform and intact seeds were chosen and sowed in plastic pots (45 cm diameter, 30 cm height). Sandy soil was taken from nearby woodlands, and it was sieved to remove debris and branches. All seeds were covered by sand to a

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depth of 0.5-1.0 cm. In a parallel study, we found that that depth was the most suitable promoting the greatest percentage and speed of seedling emergence for sandy elm (Tang et al., 2016). Holes in the bottom of the pots were covered with nylon mesh to prevent soil loss, while allowing drainage of water. All pots were watered every three days to keep the soil moist. Twenty days after sowing, 8 to 12 seedlings emerged; eight similar seedlings were retained in each pot, and the rest were removed. Mean seedling height (5.4±0.5 cm) was obtained after measuring the height of each seedling in every pot. Afterwards, seedlings were experimentally buried to either 0 (T0, no burial, control treatment) or 33% (T33; 1.8 cm), 67% (T67; 3.6 cm), 100% (T100; 5.4 cm) or 133% (T133; 7.2 cm) soil depth of the original, overall mean seedling height. For this purpose, sandy soil was added to the pots according to the different burial depths. Each seedling was kept vertical while buried. Six replicates were used per treatment, so there was a total of 30 pots in this experiment. Meanwhile, 15 randomly selected seedlings were harvested to determine the original measurements for growth analysis before sand burial.

Surviving seedlings were counted after 45 days of treatment initiation. They were considered alive when there was fresh phloem occurred in both stem and roots, and green tissue on leaf blades. Seedling height was first measured from the new soil surface level to the seedling apex, and then marked immediately. Stem diameter was measured close to the burial surface using a vernier caliper. In the meantime, 15 randomly selected seedlings were dug out in each treatment; roots were picked up as intact as possible from the sandy soil. Taproot lengths were measured, and total leaf area was obtained using a Portable Area Meter (Li-Cor3000A, Lincoln, Nebraska, USA). Finally, plant organs (i.e., leaves, stems and roots) were dried at 80 °C , and weighed thereafter reaching a constant mass for each seedling in the laboratory.

Calculations

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The (1) relative height growth rate (RHGR, mm cm^{-1}d^{-1}), and (2) relative mass growth rate of seedlings (RMGR, mg mg^{-1}.d^{-1}) were calculated according to the following equations (Walck et al., 1999; Zhao et al., 2007):

$$\mathrm{RHGR}=\frac{H_2-H_1}{H_1(T_2-T_1)} \qquad (1);$$

$$\mathrm{RMGR}=\frac{\ln M_2-\ln M_1}{T_2-T_1} \qquad (2);$$

where H_2 and H_1 were seedling heights at the end and beginning (i.e., immediately before sand burial) of the experiment, respectively; M_2 or M_1 were the total dry biomass of seedlings either after 45 days from study initiation or just before sand burial, respectively; ln was the natural logarithm, and T_2-$T_1$was time from sand burial (i.e., 45 days).

Statistical analysis

All data were tested for normality and homogeneity of variance prior to analysis. Data were log-transformed if necessary (Sokal and Rohlf 1995). The effects of experimental sand burial on seedling height, RHGR, plant stem diameter, total leaf area, RMGR, dry biomass and percentage biomass allocation were evaluated by one-way ANOVA. Whenever F tests were significant, Tukey's test was used to compare treatment means at $P<0.05$. All statistical analysis used SPSS 21.0 (SPSS Inc., Chicago, USA), and drawing was made using Origin Pro 9.0 (Origin Lab Corp, USA).

Results

Effects of permanent sand burial on seedling survival

The effect of sand burial depth on seedling survival was significant ($F_{4,25}$=38.339, $P<0.001$). During the whole study, seedling survival was 100% on the unburied (T0) and partial burial treatments (T33, T67). Simultaneously, seedling survival

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(84.48±8.8%) was significantly lower in the completely sand burial treatment (T100) than in the control treatment. No seedling survived after burial depth exceeded the overall mean, original height of seedlings (i.e., on T133).

Changes of morphological seedling traits in response to sand burial

Seedling height was significantly affected by sand burial depths after 45 days of burial ($F_{3,56}$=139.978, $P<0.001$). The highest seedling height was observed in the T33 treatment, which was significantly greater than that in the T67 treatment (Figure 2A; $P<0.05$). Height of seedlings in the control treatment was significantly lower than that in the T33 and T67 treatments, but higher than that in the T100 treatment (i.e., 10.66±0.66 cm; Figure 2A, $P<0.05$).

The relative height growth rate of seedlings (RHGR) was significantly affected ($F_{3,56}$= 286.877; $P<0.001$) after 45 days ofsand burial (Figure 2B). Highest (0.057±0.004 mm·cm^{-1}d^{-1}) and lowest (0.023±0.006 mm·cm^{-1}d^{-1}) relative growth rates for seedling height were shown in the T33 and T100 treatments, respectively (Figure 2B). The pattern of change with burial depth was similar to that described for seedling height (Figure 2A); values were greater in the control than 100% covered by sand (Figure 2B; $P<0.05$).

After 45 days from initiation of the study, stem diameter ($F_{3,56}$=26.669, $P<0.001$), taproot length ($F_{3,56}$=30.942, $P<0.001$) and total leaf area ($F_{3,56}$=35.961, $P<0.001$) of seedlings were also affected by sand burial (Figure 3 A, B, C). Stem diameter was 20% greater ($P<0.05$) in the T33 than in the T0 treatment (Figure 3A). However, stem diameters were similar in the control and T67 treatments (Figure 3A, $P>0.05$). Values in the T100 treatment, nevertheless, were 13.4% lower than those in the unburied control (Figure 3A). While taproot length was lowest in the control treatment, it was highest in the T67 treatment (Figure 3B; 35.7% higher than that in the control; $P<0.05$). The total leaf area of seedlings was significantly greater in the

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control than in the T67 and T100 treatments (Figure 3C; $P<0.05$). The lowest total leaf area, however, was found in the T100 treatment (Figure 3C).

Effects of sand burial on biomass growth and relative mass growth rate

There were significant differences in total seedling biomass ($F_{3,56}=129.949$, $P<0.001$) and its component organs[e.g., leaves ($F_{3,56}=93.965$, $P<0.001$) and roots ($F_{3,56}=50.474$, $P=0.002$)] after the experiment. The only exception was for seedling stem biomass ($F_{3,56}=2.017$, $P=0.122$) which was similar in all sand burial treatments (Table 1). Greatest total biomasses were reached in the T33(369.65±17.27 mg) and T67 treatments (372.50±15.74 mg) (Table 1). Total biomass of seedlings was significantly lower in T100 treatment than in those treatments (Table 1). Patterns shown for the biomasses of leaves and roots among treatments were similar t for the total biomass of seedlings (Table 1).

Significant differences were found in allocation of seedling biomass to leaves ($F_{3,56}=12.841$, $P<0.001$), stems ($F_{3,56}=27.579$, $P<0.001$) and roots ($F_{3,56}=7.594$, $P<0.001$). On leaves, percentage biomass allocation was greatest in the T33and T67 treatment, and lowest in the control and T100 treatments (Table 2). Percentage biomass allocation to stems was greatest in the T100 treatment (24.1±1.9%) , values on stems was greater in the control than in the T33 and T67 treatments (Table 2). Finally, percentage biomass allocation to roots showed a slight decreasing trend from the control to T33 and T67 treatments (Table 2); values determined in the T100 treatment for this organ were significantly lower than in the control and T33 treatment (Table 2).

The relative mass growth rate of seedlings was significantly affected by sand burial at the end of experiment ($F_{3,56}=136.370$, $P<0.001$). Greatest relative mass growth rate values of 0.031±0.001 mg • mg^{-1}day^{-1} were shown both in the T33 and T67

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treatments (Figure 4). These values were significantly greater than those found in the control (Figure 4, $P<0.05$). Lowest relative mass rates of growth were determined on seedlings grown in the T100 treatment (0.026±0.001 mg • mg^{-1}day^{-1}; Figure 4).

Discussion

Seedling survivorship in response to permanent sand burial

In sand land regions, seedlings might be buried at different depths between emergence and the end of the windy season, from late spring to early summer (Chen and Maun, 1999) and this was stimulated in our experiment. After the 45-days, partial burial with sand (to 33% or 67% of their height) did not influence the survival of seedling of sandy elm seedling, as there was no mortality. These results agreed with studies of He et al. (2008), Liu et al. (2008) and Qu et al. (2012), which reported that survivorship of *Artemisia halodendron, Corispermum macrocarpum* and *Caragana microphylla* was either maintained or increased by moderate sand burial in Horqin Sandy Land. Survivorship of these shrubs, however, declined among plant species once their seedlings were covered by sand either equal to or more than 100% of their height, and this was also the case for our experiment. Survival decreased sharply by a mean of 15.6%, when seedling were completely buried (to 100% of their height), while deeper sand burial resulted in no survival of sandy elm at all, as seedlings withered and rotted in the soil .This precise threshold offers a clear primary explanation for the absence of sandy elm seedling after relatively deep sand burial. Field survey in recent years has showed that serious land degradation and reduction of vegetation cover has aggravated sand mobility, particularly in the leeward and semi-fixed dunes. Seedlings could successfully complete their

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periodic recruitments only taking advantage of scarce favorable spatio-temporal chances (Tian et al., 2015; Wu et al., 2013) Maun (1981, 1997) and Disraeli (1984) also indicated that a certain tolerance to partial sand burial was an effective strategy for survival and subsequent establishment of seedlings in sandy environments. Most seedlings of the grass *Distichlis spicata* died when completely covered by sand in North America (Brown, 1997; Li et al., 2015), while some *Artemisia squarrosum* seedlings still remained alive even though sand burial depths reached 266% of the initial seedling height in the Horqin Sandy Land (Li et al. 2015). Thus, compared with other species, seedling of sandy elm sowed moderate resistance to sand burial. Harris and Davy (1987) and Perumal and Maun (2006) suggested that plant energy exhaustion and suppression of photosynthesis were implicated in the severely reduced intense radiation and high temperature to some extent. Seedlings of sandy elm have adapted to extreme conditions and previous research has confirmed that sandy elm had a higher transpiration rate and stomatal conductance with lower photosynthesis water-use efficiency and less sensitivity to high temperature and irradiance, compared with other native tree species such as *Malus baccata*, *Prunus padus* and *Pinus sylvestris*, especially in the midday (Park et al., 2012).

Effects of sand burial on seedling morphological traits

Sand burial modifies the environment of living plants, forming new microhabitat available for seedling (Disraeli, 1984; Sun et al., 2014). Plants would be expected to adjust their morphological performances and developments to maximize photosynthetic efficiency and sustain survival (Wang et al., 2014). Our results demonstrated that various seedling morphological traits (i.e., height; stem diameter; taproot length; total leaf area; dry biomass; partitioning of biomass to shoots and roots; RHGR and RMGR) were increased by partial butial, especially at T33. Thus seedling height of sandy elm was

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greater in the partially buried than in the unburied and completely buried treatments, which indicated that partial sand burial stimulated stem elongation. This might be explained by the fcat that the processes of growth and elongation, benefiting from improved water maintenance and nutrient uptake in these arid and semi-arid regions (Li et al., 2015). In Horqin Sandy Land, dry sandy layer was reported in the first 5 cm from soil surface level in semi-fixed dunes. This suggests that suitable sand burial depth could be beneficial in reducing soil temperature and keep moisture for root uptake, which are critical to seedlings survival and resource capturing (Niu et al., 2015). Our finding was also consistent with previous research reported from Disraeli (1984), who reported that partial burial stimulated growth of *Ammophila breviligulata* in coastal dunes of northeastern North America. Belcher (1977) also determind that seedling heights of *Rosa rugosa* was higher in the partial than in the unburied and completely continuous sand burial treatments in the desert.

Seedling height growth rate was a critical parameter to determine the speed of growth. The greater RHGR of seedlings in the partial (T33 and T67) than in the unburied and completely buryied treatments (T100) was an indication that partial burial did contribute to a greater seedling height after a 45-day-growth period via accelerating the speed of growth in height. Nevertheless, the greatest seedling growth in height in the T33 treatment came from its greatest RHGR in this than any other treatments. Liu et al. (2008) and Miao et al. (2012) also found that shallow soil burial depths could promote the relative growth rates in height on *Salix gordejevii*, *Artemisia wudanica* and *Artemisia halodendron*. However, there is not universally true as some species(e.g., *Artemisi gmelinii*) have decreased their growth rates as a result of sand burial (Liu et al., 2008). These findings confirmed that the phenotypic response to the degree of sand burial might be species-specific..

Compared with the unburied treatment, partial burial treatments fostered increments in stem diameter and taproot length. Sun

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et al. (2014a) also showed that seedling diameter and taproot length of *Suaeda salsa* were increased by a partial burial treatment in the coastal marches of the Yellow River estuary, China. Caldwell et al. (1998) found that increases in taproot length were conducive to a greater nutrient and water uptake from deeper soil depths. Total leaf area, however, was either similar or lower in the partial burial than in the unburied treatment. This was in agreement with the results of Liu and Guo (2005), who noted that increasing depth of sand burial decreased the total leaf area of *Caragana intermedia*.

Changes of biomass allocation and relative mass growth rate, which are involved in successful seedling recruitment, have been regarded as the results of variations in plant adaptive strategies in response to environmental changes, (Wu et al., 2013). Appropriate resource allocation is essential for plant establishment and growth (Bazzaz, 1997). Also, plants may shift resource allocation to minimize the effects of external environmental changes (Maun, 1997; Ni et al., 2015). Numerous findings, especially those on sandy environments, have reported that plants could withstand episodes sand burial by changing biomass allocation. Some species (e.g. *Artemisia ordosia, Elymusfarctus*) may transfer biomass from underground to leaf and stem organs (Brown, 1997; Li et al., 2010), while others (e.g.,*Caragana microphylla, Nitraria sphaerocarpa*) have either maintained or increased biomass allocation to roots (He et al., 2008; Sykes and Wilson, 1990). In our experiment, it was somewhat surprising that no differences were observed in the dry biomass of seedling stems among all sand burial treatments. However, seedlings showed an increased stem diameter in the T33 sand burial treatment; similar to results of Zhao et al.(2015), this was most likely because of fresh stem had a greater water content in the T33 than in the other treatments. Additionally, partial burial treatments produced greater dry biomass for leaves, roots and the whole seedlings in comparison with unburied and completely sand burial treatments. Previous studies also determined that 67% burial of seedlings of the

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shrub *Caragana intermedia* determined a greater biomass allocation to leaves and stems than to roots, compared with values in the unburied control (Xu et al., 2013). Meanwhile, there was a trend for an increasing aboveground (leaf + stem), and a decreasing belowground allocation with increasing burial depth for sandy elm seedlings. Nearly 50% of the total seedling biomass corresponded to leaves. This indicated sufficient leaves are necessary on sandy elm to sustain photosynthesis and maintain evapotranspiration after exposure to high temperature and intense irradiance environments during the growing season (Dulamsuren et al., 2009; Li et al., 2003; Park et al., 2012). The relatively investment in root was slight decreased, which indicated that, on one hand, greater soil moisture availability weakened the dependence on root function, and on the other hand, the plant's need to divert biomass aboveground for light interception and net assimilation rate (Maun, 1994; Sun et al., 2010; Wang et al., 2014) .

Relative mass growth rates measure the mean efficiency rate for producing new biomass (Walck et al., 1999). Dalling and Hubbell (2002) showed that seedling growth rate was a better determinant of successful seedling establishment than biomass. In our experiment, relative mass growth rates were higher in the partial than in the other treatments, indicating that moderate sand burials were beneficial for a rapid biomass increase. However, all mass relative growth rates were small compared with those of other plant species (e.g.,*Artemisia wudanica*, *Solidago shortii* and *Solidago. altissima*) in the same area. This suggests that the relative lower biomass accumulation on sandy elm seedlings during the first growing season places these seedlings at a disadvantage when considering soil resource competition and coexistence with other species (Brown, 1997; Liu et al., 2014; Wu et al., 2013). Reduced dry matter accumulation could also have contributed to increased mortality. Although there were striking effects on biomass accumulation and allocation reflecting the plasticity of various

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morphological traits. Partial sand burial treatments did not change the survivorship, and these treatments facilitate individual seedling growth and population regeneration through phenotylic plasticity or various morphological traits. Our study, however, was conducted in pots. We have to recognize it has limitations in comparison to field studies. Under natural rangeland conditions, some factors (e.g., abrasion of plant tissues by sand grains; grazing by herbivores and granivores) might reduce or eliminate some of the positive effects of the partial burial treatments (Dulamsuren et al., 2009; Jeffreyt et al., 2009). Furthermore, we found evidence of allometry of each plant proportion with increasing seedling age in our experiment. Therefore, more comprehensive studies on physiological and biochemical mechanisms involved on sandy elm seedling survivorship and performance under field, sand burial conditions at different growth stage in future research.

Conclusion

Sand burial affected seedling survivorship, growth and biomass allocation of *Ulmus pumila* var. *sabulosa* through phenotypic plasticity of morphological traits. Seedlings of sandy elm showed adaptive responses to moderate sand burial, consistent with its evolution in the sandy environment. Partial sand burial treatment did not influence seedling survivorship, but complete sand burial significantly increased mortality. Compared with the unburied treatment, seedling height, relative height growth rates, taproot length, total biomass and relative mass growth rates were stimulated by partial burial with sand. At the same time, percentage biomass allocation of seedlings was changed, diverting more biomass to aboveground organs (e.g., leaf and stem) to sustain normal photosynthesis and evapotranspiration. Complete sand burial after seedling emergence, however, inhibited their growth, and even resulted in seedling death. Consequently, burial depths should be controlled by making enclosures and increasing vegetation coverage to facilitate regeneration or re-establishment of sandy elm. The observed

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variation in all parameters has defined the tolerance of *Ulmus pumila* var. *sabulosa* to sandy environments and its capacity to acclimate them. Hence our research provides a theoretical support for recruitment in sandy sparse elm woodlands.

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Acknowledgements

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Table legends

Table. 1 Dry biomass of leaves, stems and roots for seedlings of Ulmus pumila var.sabolusa exposed to various sand burial treatments during a 45-day-growth period. These treatments included sand burial of seedlings to a depth equivalent to 33 (T33), 67 (T67) or 100% (T100) of the mean seedling height at the initiation of the study. Each histogram is the mean ±1 S.E. of n=15. Different letters in a row among seed burial depths are significantly different at P<0.05.

Table. 2 Percentage biomass allocation to leaves, stems and roots on seedlings of *Ulmus pumila* var.*sabolusa* expose to various sand burial treatments during a 45-day-growth period. These treatments included sand burial of seedlings to a depth equivalent to 33 (T33), 67 (T67) or 100% (T100) of the mean seedling height at the initiation of the study. Each histogram is the mean ±1 S.E. of n=15. Different letters above histograms among seed burial depths are significantly different at P<0.05.

Figure legends

Fig. 1 The geographic location of study area in Horqin Sandy Land, China.

Fig.2 Seedling height and relative growth rate for height (RHGR) of *Ulmus pumila* var.*sabolusa* exposed to various sand burial treatments during a 45-day- growth period. These treatments included sand burial of seedlings to a depth equivalent to 33 (T33), 67 (T67) or 100% (T100) of the mean seedling height at the initiation of the study (see the Material and Methods Section for further details). Each histogram is the mean ±1 S.E. of n=15. Different letters above histograms among seed burial depths are significantly different at P<0.05.

Fig.3. Stem diameter, taproot length and total leaf area on seedlings of *Ulmus pumila* var. *sabolusa* exposed to various sand burial treatments during a 45-day-growth period. These treatments included sand burial of seedlings to a depth equivalent to 33 (T33), 67 (T67)

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or 100% (T100) of the mean seedling height at the initiation of the study. Each histogram is the mean ±1 S.E. of n=15. Different letters above histograms among seed burial depths are significantly different at $P<0.05$.

Fig.4 Relative mass growth rates (RMGR) on seedlings of *Ulmus pumila* var. *sabolusa* exposed to various sand burial treatments during a 45-day-growth period. These treatments included sand burial of seedlings to a depth equivalent to 33 (T33), 67 (T67) or 100% (T100) of the mean seedling height at the initiation of the study. Each histogram is the mean ±1 S.E. of n=15. Different letters above histograms among seed burial depths are significantly different at $P<0.05$.

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Fig.4 Seedling dry biomass and percentage biomass allocation to leaves, stems and roots on seedlings of *Ulmus pumila* var.*sabulosa* exposed to various sand burial treatments during a 45-day-growth period. These treatments included sand burial of seedlings to a depth equivalent to 33 (T33), 67 (T67) or 100% (T100) of the mean seedling height at the initiation of the study. Each histogram is the mean ±1 S.E. of n=15. .

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Table 1

Treatment	Stem mg	Root mg	Leaf mg	Total mg
T0	66.62 ±3.49a	91.7 ±7.51b	160.63 ±9.15b	318.95 ±14.85b
T33	69.52 ±7.76a	104.59 ±9.89c	195.55 ±11.54c	369.65 ±17.27c
T67	68.69 ±4.05a	101.64 ±6.87c	200.17 ±14.73c	372.5 ±15.74c
T100	65.51 ±3.52a	69.61 ±9.84a	137.55 ±12.43a	272.67 ±16.42a

Treatment	Stem %	Root %	Leaf %
T0	20.9 ±1.4b	28.8 ±1.6b	50.3 ±1.4b
T33	18.8 ±1.4b.	28.3 ±1.4b	52.9 ±1.6c
T67	18.5 ±1.3a	27.3 ±1.7ab	54.2 ±2.4c
T100	24.1 ±2.4c	25.5 ±2.4c	50.4 ±2.4c

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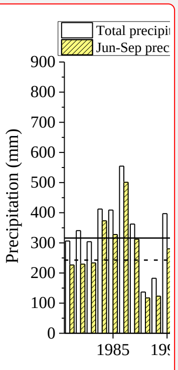

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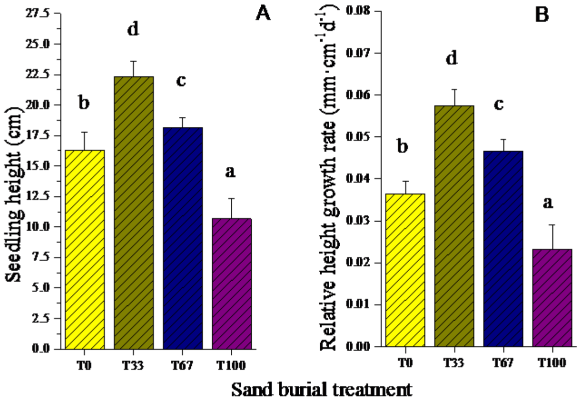

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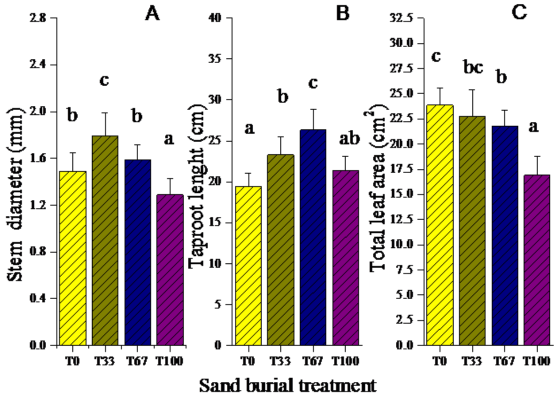

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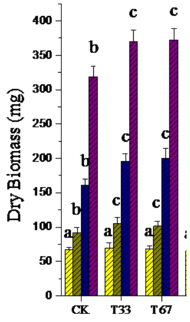

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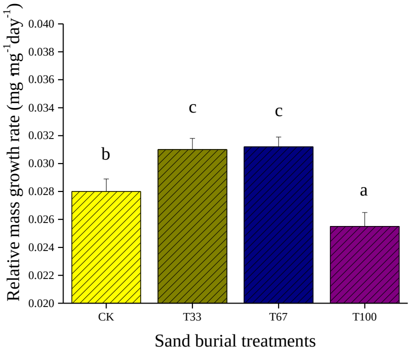

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Figure 4

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