Characteristics and Germination of *Xanthoceras sorbifolia* Bunge Seeds Originated from Inner Mongolia and Liaoning, China

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ABSTRACT : Seeds of *Xanthoceras sorbifolia* Bunge were collected from two plantations and two superior trees in Inner Mongolia; and one plantation and one superior tree in Liaoning, China in late August, 2011. Yellowhorn or goldenhorn is an important tree species, from the aspects of source of edible oil and biodiesel and pioneering capacity of degraded and desert land. Characteristics investigated were seed length, width, and weight; weight and volume of 1,000 seeds; and weight and volume of one-liter seeds. The seeds of Qingsonglingxiang No. 1, growing alone in an open space, showed the highest values in seed length (16.08 mm), width (14.48 mm) and weight (1.40 g), while those of Tree No. 160 in Ar Khorqin Banner were the lowest ones; that is, 11.48mm for length, 11.81 mm for width, and 0.73 g for weight, respectively. Traits of seeds varied quite much between trees and among areas; for example, Tree No. 38 and No. 160 produced quite different seeds in several traits, although they are adjacent to each other in the same farm. Weight of 1,000 seeds varied from 718.0 g to 1,010.1 g and volume from 0.76 L to 1.52 L. Weight of one-liter seeds were 522.3 g to 688.2 g, while the number of seeds were 603 to 935. Seeds which were soaked in the water at 4°C for 2 days showed the highest germination rate (89%) in a 30-day test, which was about 10% to 40% higher than those of non-treatment and dipping treatment at 36°C followed by keeping under room temperature for 2 days. 81% of seeds in the wet sand at room temperature germinated, while 23% of seeds deprived of seed coat germinated. It is necessary to understand seed traits to select superior clones or provenances for the increased, unfluctuating production of seed.

Keywords : Xanthoceras sorbifolia, Seed characteristics, Germination, Clone, Provenance

INTRODUCTION

Xanthoceras sorbifolium Bunge, the sole species in the genus *Xanthoceras*, is a flowering plant in the family Sapindaceae. It is known as yellowhorn, shiny leaf yellowhorn, goldenhorn, or Chinese flowering chestnut in English and native to northern China in the provinces of Gansu, Hebei, Henan, Liaoning, Inner Mongolia, Ningxia, Shaanxi, and Shandong. It grows up to 8 m, but often looks like a shrub. The leaves are arranged alternately, 1 $2\sim30$ cm long, and are pinnate, with $9\sim17$ leaflets, the leaflets $3\sim6$ cm long, with a sharply serrated margin. The flowers are $2\sim3$ cm in diameter, with five white petals, and are produced in erect panicles $10\sim20$ cm long in mid spring. The fruit is an oval leathery capsule $5\sim6$ cm diameter, which splits into three or four sections at maturity to release the $6\sim18$ seeds. The seeds are black,

around 1.5 cm in diameter, resembling a small chesnut seed (http://en.wikipedia.org/wiki/Xanthoceras#cite_note-ubcagp-1).

The flowers are beautiful in shape and color. It has been paid much attention to because seeds have a lot of unsaturated fatty acids and used traditionally as a source of Chinese medicine to cure high blood pressure and to increase anti-inflammatory capacity (Cui et al., 1987). As it is known that more than 50% to 70% oil consists in crushed seeds, it is recently expected as one of highly valuable biodiesel plants for the future (Mou et al. 2008). Thus it was planted in large scale in 1970's in Inner Mongolia, Liaoning and Gansu, and recently also tested and planted in Heilongjiang, Liaoning and Jilin.

As it was mentioned as 'a tree of thousand flowers but one fruit', it bears so many flowers but produces few fruits, and thus varies so much from tree to tree in seed production (Ding and Ao, 2008). To increase seed productivity it is highly recommended that we should look into the variation in fecundity at first and select the superior tree or family. The trait of a plant is the result of genetic and environmental interaction. Thus there should be a genetic test to understand this interaction. To reach such a goal, selection and/or hybridization will be followed.

There are some reports on the analysis of components and physiologically active substances of seeds (Li et al., 2010; Zhang et al., 2010). One report suggested that the treatment of seeds with brassinolide was effective to improve the resistance to dryness (Kai and Feng, 2011). Development of random amplified polymorphic DNA marker was tried to study variation among populations (Guan et al., 2010). However, few researches are found on the selection of superior tree or family in seed productivity. In addition, as much attention has not been paid to the traits of seeds, it is necessary to obtain information on seed characteristics first and then also to explore the resource status including distribution, cultivation, growth status, etc. (Mou et al., 2008). In this study the traits of seeds were investigated and compared among trees and provenances in addition to the germination capacity. Primary data obtained from these analyses will provide basic information for further research on the selection of superior trees and provenances.

METERIALS AND METHODS

Seed collection

Seeds were collected late August, 2011 from four origins in Chifeng, Inner Mongolia and two in Choyang, Liaoning, in China. The information on all locations is given in Table 1. Tree No. 38 and No. 160 were planted in a seed orchard in 1980. Tree No. 38 produced 1.5 kg of seeds in 2010 and 5.5 kg in 2011; and Tree No. 160 2.0 kg and 2.5 kg, respectively. Qingsonglingxiang No. 1, around 30 years old, grew alone in an open space in a small village, and 3.0 kg of seeds in 2010 and 2.5 kg in 2011(Li Fengri, private communication). Other seedlots were 30 to 40 years old, but no documentation was available.

Seed measurements

The seeds were stored at room temperature until measurement. Ten seeds were selected for each seedlot at random to measure length, width and weight. In addition, seeds were classified into 3 groups, that is, large, medium, and small; and then 10 seeds were measured in each class for the traits above. The moisture content was measured with HB43-S Moisture Measurer, Mettler Toledo, Switzerland.

Weight and volume of 1,000 seeds are helpful for understanding seed traits of each seedlot. However, many

Name	Location	Latitude	Longitude	Altitude
Jingjilinchang	Ongniud Banner, Chifeng, Inner Mongolia	N42°57'50.7"	E119°00'31.7"	633 m
Lindonglinchang	Baarin Left Banner, Chifeng, Inner Mongolia	N43°58'17.1"	E119°22'45.3"	485 m
Tree No. 38	Ar Khorqin Banner, Chifeng, Inner Mongolia	N44°13'55.3"	E119°56'20.4"	451 m
Tree No. 160	Ar Khorqin Banner, Chifeng, Inner Mongolia	N44°13'55.3"	E119°56'20.4"	451 m
Jianping Agri. Res. Station	Jianping County, Chaoyang, Liaoning	N41°21'34.0"	E119°34'06.5"	499 m
Qingsonglingxiang No. 1	Jianping County, Chaoyang, Liaoning	N41°46'15.7"	E119°56'51.7"	547 m

Table 1. Location of trees and plantations for seed collection.

sources could not supply 1,000 seeds and one-liter seeds. Weight and volume of 100 seeds were measured five times per seedlot and the data on 1,000 seeds were estimated through this information. Weight and number of one-liter seeds were estimated based on the data of 200ml seeds measured five times.

Seed germination

Seeds were placed on two sheets of filter paper in a petri dish, watered everyday to prevent dryness and investigated for germination 30 days after placement. The incubator was kept at $24\pm1^{\circ}$ C without light. Four treatments were adopted for 48 hours before trials of seeds: that is, keeping in wet, mixed sand (seed:sand= 1:1 v/v) at room temperature(20°C to 25°C), in water at 4°C, in water at 36°C at first and then under room temperature after

Table 2. Several seed traits of each seedlot.

cooling, or in wet, mixed sand (seed:sand= 1:1 v/v) at room temperature after deprived of seed coat. Ten seeds were tried twice for each treatment except for the test of peeled seeds, in which 30 seeds were used per trial. Water content was measured as above mentioned before placing seeds for trials.

Statistical analysis

Data were analyzed for differences using Duncan's multiple range test of SPSS (ver.18).

RESULTS AND DISCUSSION

The seeds of Qingsonglingxiang No. 1, growing alone in an open space, showed the highest values in seed length (16.08 mm), width (14.48 mm) and weight (1.40

Seedlot Name	Length (mm)*	Width (mm)	Fresh weight (g)	Water content (%)	
Jingjilinchang	14.33°±0.35	13.65 ^c ±0.27	$1.08^{b} \pm 0.06$	25.20 ^a ±3.46	
Lindonglinchang	$14.08^{bc} \pm 0.33$	13.38 ^{bc} ±0.36	$1.04^{b}\pm 0.07$	$26.40^{a}\pm2.70$	
Tree No. 38	13.28 ^b ±0.23	$13.08^{bc} \pm 0.24$	$0.93^{b} \pm 0.05$	24.13 ^a ±3.18	
Tree No. 160	$11.48^{a} \pm 0.23$	11.81 ^a ±0.23	0.73 ^a ±0.04	23.53 ^a ±3.26	
Jianping Agri. Res. Station	$13.80^{bc} \pm 0.27$	12.61 ^b ±0.29	$0.75^{a}\pm0.04$	26.67 ^a ±2.27	
Qingsonglingxiang No. 1	$16.08^{d} \pm 0.21$	$14.48^{d} \pm 0.20$	$1.40^{\circ} \pm 0.06$	37.53 ^b ±4.43	
Average	13.64±0.76	13.00±0.46	0.95±0.13	27.97±2.66	

*The same letter means that there is no significant difference at 5% level by Duncan's multiple range test.

Table 3. Length, width and fresh weight of each seedlot when divided into three categories.

No.* Length (mm)**		Width (mm)			Fresh weight (g)				
INU.	Small	Medium	Large	Small	Medium	Large	Small	Medium	Large
1	$12.60^{b} \pm 0.46$	$14.26^{bc} \pm 0.27$	$16.10^{\circ}\pm0.48$	$12.58^{cd} \pm 0.40$	$13.20^{b} \pm 0.25$	$15.17^{b}\pm0.28$	$0.76^{c}\pm0.76$	$0.98^{b} \pm 0.03$	$1.49^{\circ}\pm0.05$
2	$10.43^{a}\pm 0.33$	$14.87^{b}\pm0.55$	$15.10^{b} \pm 0.30$	$11.32^{b}\pm0.36$	$13.70^{b} \pm 0.30$	$15.10^{b} \pm 0.54$	$0.63^{b}\pm 0.07$	$1.10^{c}\pm0.05$	1.39 ^c ±0.06
3	$11.93^{b}\pm 0.31$	$13.64^{b} \pm 0.19$	$14.26^{b} \pm 0.25$	$11.77^{bc} \pm 0.10$	$13.24^{b}\pm 0.26$	$14.23^{b}\pm0.37$	$0.62^{b}\pm 0.03$	$0.98^{b} \pm 0.03$	1.19 ^b ±0.02
4	$10.44^{a}\pm 0.30$	$11.41^{a}\pm0.24$	$12.58^{a} \pm 0.31$	$10.51^{a}\pm0.29$	$11.81^{a}\pm0.12$	$13.12^{a}\pm0.20$	$0.47^{a}\pm0.04$	$0.74^{a}\pm0.01$	$0.97^{a}\pm0.03$
5	$12.56^{b} \pm 0.36$	$13.90^{b} \pm 0.32$	$14.94^{b}\pm0.43$	$11.83^{bc} \pm 0.25$	$11.71^{a}\pm0.43$	$14.29^{b} \pm 0.34$	$0.55^{ab}\!\!\pm\!\!0.03$	$0.66^{a} \pm 0.03$	1.03 ^a ±0.03
6	$14.88^{c}\pm0.22$	$16.51^{d} \pm 0.25$	$16.85^{c} \pm 0.23$	$13,28^{d}\pm0.24$	$14.94^{c}\pm0.14$	$15.22^{b}\pm0.27$	$1.01^{d} \pm 1.01$	$1.52^{d}\pm 0.04$	$1.65^{d} \pm 0.05$
Avg.	12.14±0.68	14.10±0.68	14.97±0.61	12.22±0.50	13.64±0.50	14.68±0.35	0.75±0.0.09	1.14±0.13	1.39±0.11

^{*}1: Jinjilinchang, 2: Lindonglinchang, 3: Tree No. 38, 4: Tree No. 160, 5: Jianping Agri. Res. Station, and 6: Qingsonglingxiang No. 1.

**The same letter means that there is no significant difference at 5% level by Duncan's multiple range test.

g), while those of Tree No. 160 in Arhorqin county were the lowest ones; that is, 11.48 mm for length, 11.81 mm for width, and 0.73 g for weight, respectively (Table 2). Water content ranged from 37.53% (Qingsonglingxiang No. 1) to 23.53% (Tree No. 160). Dryness may affect the size of seed due to shrinkage, if we look at many traits carefully. Tree No. 160 containing the least water among seedlots showed the least figures in each trait. Traits of seeds varied quite much between trees and among provenances; for example, Tree No. 38 and No. 160 produced quite different seeds in several traits, although they are adjacent to each other in the same farm.

Qingsonglingxiang No. 1 also showed the highest values for several traits in the classified seed size(Table 3). It was the same trend as that with the traits for each seedlot in Table 2.

Weight of 1,000 seeds varied from 718.0 g (Jianping Agri. Res. Station) to 1,010.1 g (Qingsonglingxiang No. 1) and volume from 0.76 L (Lindonglinchang) to 1.52 L (Qingsonglingxiang No. 1) (Table 4). Weight of 1-liter seeds were 522.3 g (Jianping Agri. Res. Station) to 688.2 g (Qingsonglingxiang No. 1), while the number of seeds were 603(Tree No. 38) to 935(Tree No. 160).

It is necessary to understand seed traits to select a superior clone or provenance in the increased, successful production of seed. The variation in seed traits within a seedlot was not evaluated in this study and thus further studies are required to continue selection for seed production. In Table 5, 39% to 43% of water content showed more than 80% of germination rate. It seems to be likely that seed dormancy is not deep and physiological in yellowhorn, although seed coat is oily and a little bit thick like chestnut, which are not hard or stiff as walnut. This is also inferred from the results in the only two-day treatments at room temperature in wet sand or cold condition in water. Thus there might be no serious problems in the germination itself of seeds for the commercial production of seedlings in this species. Naturally germination of seeds is heavily related with moisture content, but further studies for clarification are necessary for many seeds originated from different provenances and/or families.

When moisture was measured, water content of seeds soaked in hot water was 42%, the highest one among treatments, but dipping in the cold water resulted in the highest germination rate, close to 90%, which must be more effective as this condition is very much similar to the natural situations in the mountain or field. This difference may be caused by some damages inside the seed coat due to high temperature (at $36^{\circ}C$ at first and then at room temperature after cooling) even though hot water facilitated the absorption of water into the seed. Little difference in germination between treatments in room and high temperature may indicate that difference of more or less $10^{\circ}C$ is not critical to make large differences, because the seeds were soaked in the high

 Table 4. Weight and volume of 1,000 seeds, estimated from the data based on 100 seeds; and weight and number of 1-L seeds, estimated from the data based on 200-ml seeds.*

Name	Weight of 1,000 seeds (g)	Volume of 1,000 seeds (L)	Weight of 1-L seeds (g)	Number of 1-L seeds
lingiilinghang	805.00	0.96	597.00	714
Jingjilinchang	735.00	0.76	615.90	830
Lindonglinchang	950.50	1.05	570.60	603
Tree No. 38	727.60	0.80	673.00	935
Tree No. 160 Jianping Agri. Res. Station	718.00	0.86	522.30	695
Qingsonglingxiang No. 1	1,010.10	1.52	688.20	689
Average	824.37±51.46	0.99±0.11	611.17±25.51	743.33±48.33

*Means are shown without standard deviation and cannot be compared because every mean is estimated from the measurements of 100 seeds and 200ml seeds, respectively.

temperature just at the beginning of the treatment. If they were kept at 36° for 2 days, the result may be quite different from those in Table 5.

Untreated seeds showed 27% of moisture content, which is far lower than those of treated, and 51% of germination rate, which is also 30% to 40% lower than those of treated. It might be inferred that about 40% of moisture is necessary to obtain about 80% of germination rate. Moisture is critical for germination and thus priming method was adopted to improve seed germination (Finch-Savage et al., 2004; Murungu et al., 2004). Interestingly, 35% of moisture in tomato and 37% in Chinese aster were most effective in germination and showed the germination rate close to 100% (Badek *et al.*, 2006). These values in moisture content are much similar to that of Qingsonglingxiang No. 1 (Table 2), which resulted in the highest germination rate (data not shown).

Absorption of water is the first, prerequisite step by protoplasma and organelles of cells in germination and results in starting many kinds of metabolic activities in a cell (Ching, 1972). Such metabolic activities include increased vitality of enzymes, synthesis and digestion of organic substances, increment of osmotic pressure, transportation of nutritional materials, etc (Hartman et al., 1997). Such activities lead to the breakage of dormancy. In this study on germination using the current year's seeds of yellowhorn tree, it was found that sinking in water only can improve germination. However, the long period of storage in unfavorable environment will increase the dormancy intensity and require pretreatment of seeds in order to increase germination rate. Removal of seed coats lessened the moisture content and showed low germination rate. If any mechanical damage happens in the seed coat in this species, low rate of germination is expected due to loss of water, although mechanical damage is necessary to the seeds of some species.

Seeds of *Xanthoceras sorbifolia*, which were collected from Inner Mongolia and Liaoning, China, showed much variation in seed traits and high germination rate after treated with hot or room-temperature water and after cold stratification in sand. In consideration of fluctuating seed productivity, it's necessary to investigate seed traits and production for several years to select superior clones or provenances.

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Table 5. Dry weight, water contents and germination rate of each treatment.

Treatments	Dry weight(g)	Moisture content(%)	Germination rate(%)
Control	$0.78^{\circ}\pm0.02$	27.31 ^a ±0.03	51
Room temp.(20-25°C)	$0.78^{\circ}\pm0.03$	38.94 ^c ±2.03	81
Low temp.(4℃)	$0.70^{b} \pm 0.02$	42.00 ^c ±2.06	89
High temp. (36℃)	$0.72^{bc} \pm 0.02$	42.80 ^c ±1.56	79
Removal of seed coat	$0.49^{a} \pm 0.02$	26.71 ^b ±1.38	23
Average	0.69±0.05	31.55±6.71	64.60±12.22

*The same letter means that there is no significant difference at 5% level by Duncan's multiple range test.

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