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Study of Potentiality for Using Giant Sensitive Plant as Rootstock for Yard Long Bean Production

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ABSTRACT

The main problem limiting yard long bean production is the beans susceptibility to disease which leads to growth damage. The aim of this experiment was to prove the effects of different aged yard long bean scion grafted onto giant sensitive plant rootstock. The effects on yard long bean production, survival percentage, death percentage and graft union evaluation were studied. The experiment was arranged in a Completely Randomized Design, composed of five different scion ages: 15 Days after Planting (DAP), 20 DAP, 25 DAP, 30 DAP and 35 DAP. Each treatment consisted of four replications, with 15 plants in each replicate. The five scion ages of yard long bean were grafted by hand onto three-month-old giant sensitive rootstock. Grafting was evaluated by using the Tongue approach. The experiment was undertaken at the Department of Agricultural Technology, Faculty of Technology, Mahasarakham University, Mahasarakham, Thailand from March to July, 2011. The results showed that different ages of yard long bean scion had strong incompatibility with giant sensitive stock. The 25 DAP scion that were grafted onto the giant sensitive stock had the maximal survival percentage and the lowest death rate after grafting. No marked differences in graft union appearance were observed among those treated. However, the results of yard long bean scion grafted onto giant sensitive stock showed very few potential successes.

Key words: Grafting, scion, rootstock, giant sensitive plant, yard long bean

INTRODUCTION

Giant sensitive plant (*Mimosa pigra* L.), a bean plant, is native to tropical America. It is considered as an important invasive species of the genus *Mimosa*, in the family of Fabaceae. Giant sensitive plant is classified as a small shrub that can grow very well and spread throughout tropical areas, especially wet areas (Lonsdale *et al.*, 1995). When they are young, their stems are round, greenish and contain short stiff hairs along their stems (CABI, 2011). All year the growth of the giant sensitive plant proceeds quickly in many regions of Thailand. The plant will flower, when it gets old, usually between 4 and 12 months after germination (ARMCANZ and ANZECCFM, 2000). Hall *et al.* (2012) reported that the plant was first introduced into Thailand in 1947. At present, we see the giant sensitive plant abundantly widespread throughout Thailand, especially widely scattered along agricultural areas. As such, the giant sensitive plant is considered a serious weed and it leads the global invasive weed species in several countries, including Thailand (CABI, 2011). The growth characteristics of the giant sensitive plant reveals an upright growth habit 1-4 m tall that has many branched leaves (Hall *et al.*, 2004, 2012; CABI, 2011). From an agricultural perspective, the giant sensitive plants are the cause of many harmful effects including the loss of plant biodiversity and disturbance of cultivated plants (CABI, 2011). In contrast, the plant grows well and is capable of withstanding dry conditions by forming an adventitious root system from its aerial and submerged stems. In addition, it can tolerate and grow under infertile soils, or even in the alkaline soil found in the Northeast region in Thailand (Hall *et al.*, 2012). Hall *et al.* (2004) described the giant sensitive plants as having deep root systems because they can produce several adventitious roots from their stems. Lamp and Collet (1990) also reported that their roots can fix nitrogen from air to activate their growth. In addition, these plants are never damaged by natural pests. Thus, we can see these plants grow abundantly throughout all regions in Thailand. Yard long bean (*Vigna unguiculata* L.) is one of the economical vegetable legumes that are mostly cultivated in southeast Asia (Xu and Zong, 1993) including Thailand. It is arranged as an annual legume vegetable in family of Fabaceae (Papilionaceae). Its growth characteristics are as a vigorous climbing vine, to a height of 2-4 m, in bushy forms (Borget, 1992; Xu and Zong, 1993). In Thailand, it is popularly consumed as a young pod vegetable in several foods. Christine *et al.* (2007) cited that yard long beans are high sources of nutrition such as vitamins A and C. Yard long bean can be planted all year round and in every part of Thailand. However, the problem of pests occurs all through its growing life (Nokkoul *et al.*, 2011). Martin and Ruberte (1980) reported that yard long beans are sensitive to several plant diseases and pests, such as powdery mildew and aphids which affect plant growth (Bland and Knausenberger, 1985). During their growth, yard long beans also become feeble from root knot nematode infestation, resulting in reduced plant growth (Rhoden *et al.*, 1990). One method commonly used in vegetable production in order to control plant disease and pests is grafting. This method is often used on Solanaceace and Cucurbitaceae plants, such as cucumber, watermelon, tomato and pepper (King *et al.*, 2010). The grafting operation is an alternative plant production method that uses two close taxonomic plant species (Moncada *et al.*, 2013) composed of two sections; a rootstock and a scion which are linked together with a graft and grow as a single plant (Yetisýr *et al.*, 2003; Rouphael *et al.*, 2010). Tsaballa *et al.* (2013) also cited an important advantage of grafting is the protection against abiotic stresses such as high/low temperature, salinity (Colla *et al.*, 2010; King *et al.*, 2010) and drought or excessive water soil content (Schwarz *et al.*, 2010). Furthermore, grafting can increase scion vigor during the growing season (Lee *et al.*, 2010). This operation is rapidly spreading and expanding throughout the world (Lee *et al.*, 2010). Grafting, however, had never been applied in yard long bean scion onto giant sensitive rootstock and no published data is available on yard long bean/giant sensitive plant grafting. Therefore, the objective of this experiment was to evaluate the potential of grafting between yard long bean scion and giant sensitive rootstock.

MATERIALS AND METHODS

The experiment was set up as a Completely Randomised Design (CRD) including five levels of scion age as follows: T_1 = scion aged at 15 Days after Planting (DAP), T_2 = scion aged at 20 DAP, T_3 = scion aged at 25 DAP, T_4 = scion aged at 30 DAP and T_5 = scion aged at 35 DAP, with four replications. Each replication consisted of 15 plants. The experiment was done at the greenhouse of the Department of Agricultural Technology, Faculty of Technology, Mahasarakham University, Mahasarakham, Thailand. It was carried out between March to July, 2011 at the Experimental Farm of the Department of Agricultural Technology, Faculty of Technology, Mahasarakham University. Seeds of giant sensitive plant were sown three months before the sowing date of the yard long bean seeds. Seeds of giant sensitive plant were cultivated in 10-L plastic pots filled with soil: Husk: Manure at ratio of 2:1:1 and placed in a glasshouse. There was one plant in each pot.

Three months after planting, the seedlings were used as rootstock to investigate the effect of different scion ages grafted onto giant sensitive stock when their branches grew at semi-mature stage. Seeds of yard long bean var. 'Kheawdok no. 5' were obtained from the East West Seed Co. Ltd. Yard long bean seeds were cultivated for use as scion in pot size 8×12 inch with planting media of soil: Husk: Manure at a ratio of 2:1:1. When the yard bean seedlings grew to each selected treatment (15 DAP, 20 DAP, 25 DAP, 30 DAP and 35 DAP), they were brought to be grafted onto the giant sensitive stock using the procedure of Tongue grafting on 1 June. The grafting was done in a glasshouse in the morning. The stems of the yard long bean scions were cut with a razor blade below the third leaf from the apex. While the rootstocks were also cut transversely with a razor blade above soil level 10 cm. Prepared scion pieces were placed on the top portion of the giant sensitive stocks. After grafting, grafted yard long bean shoots were covered with a transparent plastic bag and a paper bag to maintain a humid environment and protect them from direct sunlight. The glasshouse temperature ranged from 30 to 35°C and 83% mean relative humidity. The recorded data consisted of survival percentage, death percentage and graft union. The level of the graft union was assessed as a percentage by visual means and scores were given ranging from 0 to 100% as follow 0= no connective tissue appearance, (1) Very slight connective tissue appearance around 1-20%, (2) Slight connective tissue appearance around 21-40%, (3) Moderately connective tissue appearance around 41-60%, (4) Rather abundant connective tissue appearance around 61-80% and (5) Abundant connective tissue appearance around 81-100%. Data were recorded at seven-day intervals after grafting. All collected data were statistically analyzed by ANOVA using the SPSS Computer Program, Version 6 (SPSS, 1999).

RESULTS

After grafting with different scion ages of yard long bean (15, 20, 25, 30 and 35 DAP) onto giant sensitive rootstock, the data were recorded as follows.

Survival percentage: After grafting, scions of all treatments still showed fresh in the first week. During the second week, scion aged 15, 20 and 25 DAP started to die. There was no significant difference of plant survival among treatments on week 2 and week 3. However, on week 3, the survival percentage of all scion ages became lower than week 2, survival means all treatments were between 58.75-68.75%. On week 4, the results showed that all ages of yard long bean scion decreased their survival percentage to a low level, especially scion aged at 15 DAP and 35 DAP. They showed the least survival percentage of 0.00 and 0.25%, respectively. The highest survival value was from scion aged 25 DAP (9.25%) (Table 1).

Plant death: The effects of grafting on plant death using different ages of yard long bean scion grafted onto rootstocks are given in Table 2. No significant difference among treatments was observed on plant death in week 1, 2 or 3. The scion death at the early stage of week 2 and 3 showed no significant difference (0.00-3.75 and 31.25-41.25%, respectively). A highly significant difference ($p = 0.01$) between treatments was observed in week 4. The giant sensitive rootstocks grafted with scion at 15 DAP and 35 DAP showed the highest death percentage. The lowest value of plant death (90.75%) was recorded on the 25 DAP yard long bean scion. Thus, a negative effect of grafting was obtained when 'Kheawdok no. 5' yard long bean was grafted onto giant sensitive rootstock. It can be concluded that scions of yard long bean have a low success rate when grafted onto giant sensitive rootstock.

Table 1: Survival percentage of different yard long bean scion after grafting onto giant sensitive stock

	Survival percentage of scion (weeks)			
Scion age	1	2	3	4
15 DAP	100	96.25	58.75	0.00[c]
20 DAP	100	96.25	63.75	3.50[b]
25 DAP	100	97.50	66.25	9.25[a]
30 DAP	100	100.00	67.50	1.25[bc]
35 DAP	100	100.00	68.75	0.25[c]
F-test	-	ns	ns	**
CV (%)	-	3.84	3.41	15.67
LSD	-	1.88	10.51	0.80

Letters within columns indicate least significant differences (LSD) at p** = 0.01, ns: Non significant

Table 2: Plant death percentage of different yard long bean scion ages grafted onto giant sensitive rootstock

	Death percentage of scion (weeks)			
Scion age	1	2	3	4
15 DAP	0.00	3.75	41.25	100.00[a]
20 DAP	0.00	2.50	31.25	96.50[b]
25 DAP	0.00	0.00	32.50	90.75[c]
30 DAP	0.00	3.75	33.75	98.75[ab]
35 DAP	0.00	0.00	36.25	99.75[a]
F-test	-	ns	ns	**
C.V. (%)	-	18.81	19.51	1.66
LSD	-	1.88	3.41	0.80

Letters within columns indicate least significant differences (LSD) at P ** = 0.01, ns: Non significant

Table 3: Evaluating of connective score at site grafting between yard long bean scion and giant sensitive rootstock

	Score of connective tissue between scion and rootstock (weeks)			
Scion age	1	2	3	4
15 DAP	5.00	4.50	3.250	0.63
20 DAP	5.00	5.00	3.880	0.88
25 DAP	5.00	5.00	4.000	1.00
30 DAP	5.00	5.00	4.500	0.75
35 DAP	5.00	4.50	4.000	0.50
F-test	-	ns	ns	ns
C.V. (%)	-	7.60	12.42	5.67
LSD	-	0.18	0.310	0.22

ns: Non significant

Graft union at wounding site: The effects of different scion ages grafted onto giant sensitive rootstock on the graft union appearance by visual means at site grafting are presented in Table 3. The results showed that scores of all graft union between scion and rootstock decreased and all treatments showed similar connective scores of two portions at site grafts, with the low mean values of 4.50-5.00, 3.25-4.50 and 0.50-1.00 after grafting on week 2, 3 and 4, respectively. The most visible tissue injury in grafted plants was a drying of the scion after grafting. These symptoms lead to plant death. Thus, a negative effect of grafting was shown when different yard long bean ages were used as scion grafted onto giant sensitive rootstock.

DISCUSSION AND CONCLUSION

For plant survival, when comparing the different scion ages grafted onto the giant sensitive plant, it can be seen that plant survival decreased significantly. The survival percentage of yard long bean scion was significantly affected by the scion age (Table 1). The grafting success of the yard long bean scion was reduced by grafting on giant sensitive plant rootstock. The poor success rate was similar for all scion ages. At week 4, only the yard long bean scion at 25 DAP grafted onto giant sensitive plant rootstocks showed maximal percentage of plant survival after grafting (9.25%). The results showed that after grafting, all yard long bean scion started to die in the second week. It is possible that the scion survival rate after grafting was significantly affected by the genotypic characteristics between scion and rootstock. Thus, the effect of grafting between yard long bean scion and giant sensitive plant was regarded as unsuccessful when using the yard long bean scion grafted onto giant sensitive plant. This is in agreement with the previous data of Santa-Cruz *et al*. (2002) who found that grafting may be possible due to the dependency of the different genotypes between scion and rootstock plants. These results do not corresponded to Rodriquez and Bosland (2010) who reported the opposite results; they found a high percentage (100%) of successful grafting obtained with *Capsicum* onto tomato rootstocks while Lee (1994) reported that watermelons can be used as scion grafted onto gourd. These responses indicated that not only scion-rootstock species but also compatibility can affect the scion survival (Marsic and Osvald, 2004). However, no published data is available concerning the effects of grafting of yard long bean scion onto giant sensitive rootstock.

With respect to plant death, the results showed that the high percentage of plant death found in all yard long bean scions were quite similar to be the high values at short time after grafting. Plant death was also significantly affected by scion age. Recorded data in terms of survival percentage and death percentage after grafting indicate that yard long bean scions are rather restricted to their compatibility with the giant sensitive stock. The results revealed that the increase in plant death was in harmony with the reduction of plant survival after grafting. It is interesting to point out the trend of low survival in this experiment also indicated a severe negative effect of grafting between yard long bean scion and giant sensitive rootstock. These negative effects may be due to the influence of incompatibility between scion and rootstocks. In addition, Temperini *et al*. (2013) found that wounding stress may significantly reduce the survival percentage of scion after grafting onto rootstock. When a graft takes unsuccessfully, the two separate pieces of the two plants appear to die after grafting. The reasons may be due to lack of a vascular bridge connection between the scion and the rootstock (Harada, 2010). The negative influence of some rootstocks in increasing scion death revealed that the capacity for using giant sensitive plant as rootstock to improve the production of yard long bean is very low. The negative influence brought about a hastening of scion death after the longer grafting period evolved (Webster and Schmidt, 1996). It is therefore necessary to investigate the effect of different yard long bean scion ages on the morphological and physiological parameters of yard long bean scion related to giant sensitive rootstocks.

For graft union, the two plant pieces binding at the grafting site, as measured by visual means, were insignificantly affected by the different scion ages. The score evaluation for graft union between different ages of scion grafted on giant sensitive rootstock showed similar values at each data recording session. However, the recorded connective scores of all treatments gradually decreased after grafting. It may be possible that after grafting, all scions appeared to be wilting beginning in the second week. This was probably due to the fact that yard long bean scion seems to exhibit a higher incompatibility when grafted onto giant sensitive rootstock. The incompatibility is more clearly shown at four weeks after grafting and indicates the failure of giant sensitive plant as rootstock for yard long bean production. This is evident even though both yard long bean and giant sensitive plant are closely related in the same family of Fabaceae (Leguminosae), with sub-family Mimosoidea. These results may reveal an incompatibility in plant types between yard long bean scion and giant sensitive rootstock. Incompatibility characteristics between two plant types could reduce the survival percentage of the plant. Thus, all different ages of yard long bean scions grafted onto the giant sensitive rootstock could not grow and eventually died. Another possible explanation for disadvantageous effects of the graft union may be the involvement of root–shoot interactions in the formation of graft union between scion and rootstock which was thought to partly explain the increased death rate of scions after grafting. These results are also consistent with the findings of Davis *et al.* (2008) who indicated that the type of rootstock could alter the growth attributes of scion. Thus, the results from this experiment indicate that genotype characteristics of both selected scion and rootstock should have a close taxonomy relationship to improve scion growth. These results are also in line with the results of Lee *et al.* (2010) who found that scion-rootstock interaction graft incompatibility leads the disadvantageous effects of grafting in vegetable production. A similar finding in a previous report by Schwarz *et al.* (2010) reported that compatibility characteristics are regulated by plant type specifics, in which many genes are involved while Davis *et al.* (2008) reported on the capabilities in forming graft union between watermelons onto bottle gourd and melon onto wax gourd rootstocks (Lee and Oda, 2003). Sriwichan (2011) also reported on *Lycopersican esculentum* grafted onto different Solanaceae plants. He found that *Lycopersican esculentum* grafted onto *Solanum Torvum* Swartz showed the best graft union appearance when compared with *Solanum torvum*, *Solanum trilobatum*, *Solanum stramonifolium* and *Solanum xanthocarpum* Schrad and Wendl. However, this experiment does not focus on the physiological and biochemical aspects of the rootstock-scion interaction for both yard long bean and giant sensitive plant. Furthermore, giant sensitive plant is known for its poor resemblance relationship in morphological characteristics with yard long bean scion. At present, the mechanism of scion-rootstock compatibility has not been intensively investigated (Lee, 1994).

In conclusion, shoots of yard long bean cv. 'Kheawdok No. 5' of five different ages were grafted onto giant sensitive rootstocks. The results showed that after grafting, the plants had the rather low survival rate and a high percentage of deaths. In addition, all different yard long bean scion ages grafted onto giant sensitive rootstock had the similar low score evaluation of graft union. The deleterious effects of using yard long bean scion grafted onto giant sensitive rootstocks were noted and likely contributed to the poor survival rate after grafting.

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