THE EFFECTS OF FERTILIZER AND ASPIRIN ON PROPAGATION OF *FICUS* SPECIES FROM SEED

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ABSTRACT

Ficus spp. (or fig trees) are keystone species in tropical forest and have been promoted as framework species for forest restoration in Northern Thailand. This study aimed to improve propagation of *Ficus* spp. seedlings from seeds. The effects of fertilizer, aspirin, individually and in combination, were determined. *Ficus* seeds were treated with 0.05mM aspirin solution and 2 fertilizer dosages, placed under the medium surface in germination baskets. Seed germination, seedling survival and seedling performance were monitored. Three months after sowing, seedlings were subjected to drought stress and assessed. Fertilizer and aspirin had no effect on seed germination, but fertilizer significantly accelerated seedling growth and increased seedling height and number of leaves (P≤0.05). Aspirin has a significant effect on drought stress by delaying and reducing the severity of drought injury of leaves. Both aspirin and fertilizer treatments are likely to improve the efficiency of propagation of *Ficus* seedlings in nurseries and produce drought -resistant planting stock in less time than conventional propagation methods.

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บทคัดย่อ

พืชสกุลมะเดื่อเป็นกลุ่มพืชที่ถือว่าเป็นสิ่งมีชีวิตที่มีความสำคัญในระบบนิเวศป่าเขตร้อน และเป็นพันธุ์ไม้ที่ได้รับการส่งเสริมในฐานะพันธ์ไม้โครงสร้างสำหรับการฟื้นฟูป่าในภาคเหนือ ของประเทศไทย การศึกษาครั้งนี้มีวัตถุประสงค์เพื่อปรับปรุงวิธีการขยายพันธุ์พืชสกุลมะเดื่อจาก เมล็ด ทดสอบผลของปุ๋ย แอสไพรินทั้งแบบแยกและรวมกันต่อการผลิตมะเดื่อ โดยการแช่เมล็ด มะเดื่อในสารละลายแอสไพรินความเข้มข้น 0.05 มิลลิโมลาร์ ก่อนเพาะในวัสดุปลูกที่มีปุ๋ย 2 ความ เข้มข้นในตะกร้าเพาะเมล็ด ทำการติดตามการงอกของเมล็ด การรอดชีวิตและการเติบโตของต้นกล้า เป็นเวลาสามเดือน จากนั้นชักนำให้เกิดความเครียดจากความแห้งแล้ง และประเมินผลกระทบที่มี ต่อต้นกล้า ผลการศึกษาพบว่าปุ๋ยและแอสไพรินไม่มีผลต่อการงอกของเมล็ด แต่ปุ๋ยมีผลเร่งการ เจริญเติบโตของต้นกล้า โดยการเพิ่มความสูงและจำนวนใบอย่างมีนัยสำคัญ (P<0.05) ส่วน แอสไพรินมีผลต่อการชะลอและลดอาการเครียดที่เกิดจากความแห้งแล้งต่อใบได้อย่างมีนัยสำคัญ ทั้งแอสไพรินและปุ๋ยมีส่วนช่วยให้การขยายพันธุ์พืชสกุลมะเดื่อในโรงเพาะพันธุ์มีประสิทธิภาพ มากขึ้น และสามารถผลิตกล้าไม้ที่มีความความด้านทานต่อความแห้งแล้งในระยะเวลาที่สั้นกว่าการ ขยายพันธุ์โดยวิธีปกติ

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CHAPTER 1

INTRODUCTION

1.1 Introduction

Ficus spp. or fig trees are distributed throughout subtropical and tropical America, Asia and Australia. There are 80-100 species in Thailand, at least 35 of which grow in northern Thailand. *Ficus* species are keystone species and have been promoted as framework tree species for restoring forest ecosystems in Northern Thailand. The characteristics that qualify *Ficus* as keystone and framework species include:

- Figs are food for many fruit-eating animals, such as primates, squirrels and bats. So, they can sustain populations of fruit-eating animals when other foods are scarce.
- 2. Figs can attract seed-dispersing animals and thus promote forest regeneration.
- 3. *Ficus* trees have mutualistic relationships with diverse groups of animals.
- 4. *Ficus* trees have a dense root system which can prevent soil erosion and enables the trees to survive and grow well under the harshest of conditions and rapidly grow back after burning.
- 5. *Ficus* spp. are important shade producers and thus suppress weed growth when planted out in degraded areas.

Many studies of *Ficus* spp. have been carried out in northern Thailand, over a long time, on their ecology, phenology, propagation and planting techniques. Kuaraksa and Elliott (2013) studied 6 *Ficus* species: *F. auriculata*, *F. fulva*, *F. hispida*, *F. oligoden*, *F. semicordata* and *F. variegata*. They recommended that *Ficus* spp. should be raised from seed in nurseries, before transplanting saplings into degraded forest. The appropriate medium for *Ficus* propagation is coarse sand: charcoalized rice husks (50:50) because this mix decreases damping-off disease. *Ficus* seeds are tiny. The length of the germination period varies among species.

FORRU (2006) showed that the median length of dormancy (MLD) of Ficus spp. varies from 25 to 67 days depending on species. After the seeds germinate, the seedlings remain very small for a considerable period, because the small seeds contain little endosperm to support the initial growth of the seedling. So, seedlings must be grown in germination trays for 5-10 months before pricking out into containers. Most seedlings grow large enough for out-planting for forest restoration projects from 18 to 22 months after germination (FORRU, 2006). Although, slow-release fertilizer is usually used on seedlings after picking out, seeding growth is still slow, compared with most other forest tree species. Drought is an important problem in forest restoration programs, since restoration sites are often hot dry, sunny open locations. Such conditions limit plant growth and survival. Moreover, fig trees are recommended for restoring forest to former mine sites where drought stress would be a severe problem since such sites have no soil or shade. Fig tree roots are capable of invading and breaking apart compacted soils and even rock on the most degrade of sites (Elliott et al., 2013). So, it is important to reduce drought stress in out-planted tree saplings used for forest restoration projects. Treatment with aspirin is one option to decrease drought stress in plants under water deficit conditions. For example, Khan et al. (2012) found that soaking wheat seeds in an aspirin solution was effective at ameliorating the negative impact of drought stress on wheat plants. Drought stress resistance was increased on bean and muskmelon plants when their seeds were soak in salicylic acid solution (Sadeghipour and Aghaei, 2012; Komaz et al., 2007).

Although, several propagation techniques for *Ficus* spp. have been developed, *Ficus* spp. still grow slowly and are affected by drought stress. Therefore, in this study, experiments were carried out to improve the propagation of *Ficus* spp. from seed by using fertilizer and aspirin. We examined the hypothesis that *Ficus* spp. have higher survival and faster growth if fertilizer is added to the germination medium and that aspirin will protect *Ficus* seedlings from drought stress.

1.2 Objectives

- 1. To develop an efficient nursery technique to improve propagation of *Ficus* spp. from seed, by using fertilizer and aspirin
- 2. To determine the effects of aspirin of young *Ficus* spp. seedlings on drought stress tolerance.

CHAPTER 2

LITERATURE REVIEW

2.1 Ficus species

Ficus is a genus in the plants family Moraceae. Although, commonly known as Fig trees, *Ficus* spp. also include some vines, woody climbers, shrubs, epiphytes, hemi-epiphytes, treelets as well as large forest trees. They are distributed mostly in tropical and subtropical America, Africa, Asia and Australasia. About 80-100 species have been recorded as growing in Thailand, of which 35 are found in the north. More species grow in evergreen forest than in deciduous forest types.

2.1.1 The characteristics of Ficus tree species

1) Fruit

Figs are often referred as the fruits of *Ficus* tree, but in fact they are "syconia" the invaginated peduncles (or stalk) of flower spikes, such that the flowers become enclosed and surrounded by the peducular walls (Cook and west, 2005).



Figure 1 Fig diagram. (Source: Adapted from Storey, 1975)

Figs are the most characteristic feature of mature *Ficus* trees. They are often borne on directly from the tree trunks or large branches (cauliflory). The flowers within figs are pollinated by fig wasp (FORRU, 2006; Cook and West, 2005).



Figure 2 Figs: (a) F. benjamina, (b) F. racemosa and (c) F. semicordata

2) Root system

Ficus trees have strong, highly penetrative, fibrous root systems that extend out up to three times the diameter of canopy (Condit, 1974) and are capable of growing into crevices and highly compacted substrates, with little or no soil lateral roots are typically shallow, but their taproots penetrate more deeply. Their extensive root systems make fig trees tolerant of poor soils, moderate salinity and drought (Golombek and Ludders, 1990).

3) Latex, leaf and bark

Fig trees contain a white milky exudate in all tissues (Moshe, 2008). The barks of fig trees are usually gray or brown. Leaves are single and large, leaf arrangement and shape are variable (FORRU, 2006).

2.1.2 The importance of *Ficus* tree species in forest

Ficus spp. are well known as keystone species in tropical forest ecosystems because they can maintain populations of frugivorous animals during seasons of food scarcity because within each *Ficus* species figs are always produced by some individuals throughout the years (Thorntron *et al.*, 1996). Abalaka (2008) showed that 49 bird's species visit the *Ficus* trees to feed on figs. Moreover, mammals and reptiles are also regular visitors to fig trees. Almost all animal species observed visiting fig trees did so when figs were ripe.

2.1.3 Study species

1) Ficus racemosa

Ficus racemosa is a large deciduous tree. The figs are red when ripe, borne in large clusters, on short leafless branches that emerge from the trunks or main branches (Paarakn, 2008). This specie grows in the lowland, at elevation 300 - 350 m. above sea level in Northern Thailand. This is a very narrow elevation band for forest restoration programs; seeds are usually collect on February. Median length of dormancy (MLD) is about 20 - 27 days (FORRU, 2006).



Figure 3 F. racemosa (a) Whole tree, (b) Leaves and (c) Figs

2) Ficus semicordata

Ficus semicordata is a deciduous tree with wide – spreading branches, brown hairs on leafy twigs, leaves and syconia. The leafless fig-bearing branched develop at the base or trunk and often become stolon like, trailing across the forest floor. The figs are red-brown at maturity. This species can found from 350 to 1,555 m. elevation in northern Thailand. For forest restoration programs, seeds are usually collected from December to March. The MLD is about 52 days (FORRU, 2006).



Figure 4 F. semicordata

2.2. Forest restoration

2.2.1 What is forest restoration?

Forest restoration attempts to re-establish former levels of ecosystem biomass, structure, biodiversity and ecological functioning by reviving natural forest regeneration mechanisms and encouraging growth of natural regenerant (i.e. surviving seedlings, saplings or coppicing tree stumps of remnant trees, if present) and/or by planting tree species, known to play an essential role in the functioning of the original forest ecosystem (FORRU, 2006).

2.2.2 The Framework species method

Framework species method involves planting the minimum number of key tree species (about 20 - 30 species) capable of catalyzing full recovery. It concentrates on re-establishing natural seed dispersal mechanisms to achieve

rapid tree species recruitment, biodiversity recovery in restoration sites. For this method to work a remnant of the target forest type must survive near to the forest restoration site (as a seed source). The trees planted should be typical or characteristic of target forest, but also share the following characteristics:

- High survival when planted out in deforested sites
- Rapid growth
- Dense, spreading crowns that shade out herbaceous weeds
- Flowering, fruiting, or the provision of other resources, at a young age, which attract seed-dispersing wildlife
- Resilience after burning and
- Seedling should be easy to propagate in nursery

2.2.3 Forest restoration and *Ficus* tree species

1) The importance of *Ficus* species in forest restoration programs

Fig trees are considered to be a framework tree species for forest restoration in northern Thailand. Elliott *et al.* (1998) recommended that about 20 % of planted seedling should be *Ficus* species in the framework species method. Two main characteristic make *Ficus* spp. ideal as framework tree species. First, the figs are essential food for a wide range of seed dispersing animals, including birds, bats, primates, civets, squirrels, bears, deer and pigs. They help to maintain healthy populations of seed dispersers, which are vital for recover of tree species richness in regenerating forest. Secondly, their dense root system enables them to survive under harshest condition and grow back rapidly after burning. Moreover, their roots can tap into soil moisture deep underground, which allows most species to retain their leaves throughout the dry season and helps to prevent soil erosion.

2) Nursery techniques for propagating *Ficus* species

Raising trees in nurseries is important for forest restoration program. Elliott and Kuaraksa (2008) suggested that for restoration projects in northern Thailand, saplings grown in nurseries, must be ready for planting at the beginning of rainy season (May – June). Seed germination should be carried out in modular trays. After seeds germinate, seedlings should be picked out into polybags, 9 x 2.5 inches. The most suitable potting mix is 50% forest soil, 25% peanut husk and 25 % coconut husk. Fertilizer should be applied every 3 months, 10 grains of Osmocot (slow release fertilizer) is the most effective fertilizer treatment for most species. Damping off of young seedling in germination tray is sometimes controlled with fungicides (Captan or Thiram) (FORRU, 2006).

FORRU (2006) reported that *Ficus* seedling must be grown in a nursery for 5- 10 months before picking-out. After that, saplings of most species grow rapidly, but they are not large enough for planting out until 2^{nd} planting season after seed collection (i.e. about 18 - 22 months after germination). So, a long standing down time is required. This consumes water, fertilizer and the time of nursery staff in sapling care. Figs appear on different individuals of the same species nearly all year-round, with different fruiting peaks among the species. Figs are cut from the trees only when they are fully ripe.

For seed germination, Kuaraksa (2012) recommended a germination medium of sand and charred rice husk (1:1) in the nursery, although using this medium with fungicides had a negative effect on seed germination. Four months after seed germination, seedlings were still tiny only 1 to 2 cm tall. The mean overall success of all species is fairly low. Saplings can be planted into deforested sites when 20 cm tall (Kuaraksa and Elliott, 2013).

2.3 Aspirin and drought stress in plant

Aspirin or acetyl salicylic acid (ASA) is a derivative of salicylic acid (SA). This substance is found as a phenolic acid and as a conjugated form in plants. Bandurska (2013) reviewed that under non-stressful conditions, SA is found naturally at very low concentration in tobacco, corn, tomato, bean, barley and rice plants tissues (several ng to several mg per g of fresh weight) but its concentration increases in

plants exposed to a water deficit. Water deficit increases the activity of enzymes involved in SA synthesis which consequently increases SA levels in plants.

2.3.1 The application of SA or ASA on plant under drought condition.

Many studies have shown that exogenous application of SA, though roots, seed soaking and foliar spray decreases drought stress of plants, under water deficit. For example, Senaratna *et al.* (2000) showed that soaking seeds of bean and tomato plants, with 0.1 or 0.5 mM of SA or ASA maintain a high degree of cell turgidity after subject to drought stress, whilst non-treated control plants lose turgor and wilt. Komaz *et al.* (2007) found that SA can decrease drought stress on muskmelon and the application of SA, by seed soaking or foliar spray, has similar effects on drought stress tolerance of muskmelon. Drought stress resistance in bean and cucumber plants is increased by soaking seeds in SA solution (Sadeghipour and Aghaei, 2012). Soaking wheat seeds in 0.03-0.05 mM ameliorates the negative impact of drought stress on wheat (Khan *et al.*, 2012).

2.3.2 The action of SA on plant under drought condition

Bandurska (2013) who reviewed the effects of SA on plants under drought stress reported that exogenous application of SA to leaves, roots or seeds activate protective mechanisms that enhance resistance to water deficit. Exogenous application of SA improves water status under water deficit condition by the following mechanisms:

- Reduced stomatal conductance
- Reduced damage of cell membranes
- Increased proline production, which increases the capacity of roots to absorb of water from soil and reduces tissue dehydration

The result of all the above mechanisms not only induces drought resistance but also improves crop yields (Fig 5).



Figure 5 The role of SA in the enhancement of plant resistance to water deficit.

CHAPTER 3

MATERIAL AND METHODS

3.1 Seed collection and extraction

Mature, fully ripe figs of two *Ficus* spp: *F. racemosa* and *F. semicordata* were collect from trees of each species in September, 2016 from Chiang Mai University and Doi Suthep-Pui National Park. Figs were opened and tiny, light brown, fruits (achenes), each containing a single seed, were scraped out. The achenes were dropped in water and sieved through a mosquito net. The viable seeds passed through the mosquito net and sank. Seeds were spread out on paper and left to dry for 1-2 days, as recommended by The Forest Restoration Research Unit (2006).

3.2 Application of fertilizer and aspirin

3.2.1 Application of fertilizer

Slow release fertilizer (Osmocote 13-13-13) was placed on about 1 cm depth of germination medium in baskets and covered with additional germination medium (as show in 3.3) also about 1 cm deep. The amount of fertilizer was varied according to each treatment (see in 3.3).

3.2.2 Application of aspirin.

After seed extraction and drying, seeds were soaked in 15 ml of 0.05 mM aerated aspirin solution, on a double layer of filter paper, in covered transparent polystyrene boxes. The boxes were kept in darkness for 24 hours, after which the seeds were washed and dried (Korkmaz et al., 2007).

3.3 Seed germination and growth

A 1:1 mixture of coarse sand and charcoalized rice husk was used for the germination medium (Kuaraksa, 2002) and placed in baskets ($25 \times 17 \times 7.5 \text{ cm}$) to a depth of about 4.5 cm. deep. The effects of fertilizer and aspirin on germination were tested. There were three fertilizer treatments: i) no fertilizer, ii) low dose of fertilizer:

1.5 g per basket, and iii) high dose of fertilizer: 3.0 g per basket. The two aspirin treatments were i) no aspirin and ii) soaking seeds in the aspirin solution, as described in 3.2.2. Combination treatments were therefore i) low dose fertilizer +/- aspirin and ii) high dose fertilizer +/- aspirin as listed in Table 1. The experimental design was a randomized complete block design (RCBD) with 3 replicates of each of 6 treatments. One hundred seeds were sown for each replicate. After seed sowing, the germination baskets were watered everyday by hand, using a fine spray bottle (Kuaraksa, 2012).

Treatments	Description
T1	coarse sand + charcoalized rice husk + no fertilizer + no aspirin (Control)
T2	coarse sand + charcoalized rice husk + aspirin
Т3	<pre>coarse sand + charcoalized rice husk + low dose fertilizer + no aspirin</pre>
T4	coarse sand + charcoalized rice husk + high dose fertilizer + no aspirin
T5	<pre>coarse sand + charcoalized rice husk + low dose fertilizer + aspirin</pre>
T6	<pre>coarse sand + charcoalized rice husk + high dose fertilizer + aspirin</pre>

 Table 1 Experiment design on seed germination trials.

Seed germination was defined as the emergence of cotyledons. For each germinated seed, a toothpick was put into the germination medium near the seedling. The total number of toothpicks therefore represented the total number of seeds germinated. However, as seedlings died and disappeared, the toothpicks were replaced with white plastic sticks. The numbers of white plastic sticks indicated number of seed that germinated and died. Seedling survival was therefore the total of number of toothpicks as a percentage of total cumulative seed germination (= toothpicks + white sticks).

Seed germination was monitored daily. Seedling survival was monitored weekly. Two months after planting, seedling height, number of leaves and number of nodes were recorded every 2 weeks. The experiment ended after 13 weeks, when root and shoot length, fresh and dry weights were measured.

For seed germination, the median length of dormancy (MLD) was calculated as the length of time between sowing and germination of half the seed which eventually germinated (Elliot *et al.* 2013).

Seedling survival percentage was defined by the number of seedling survival divided by the number of seed germination, then converted to percentage.

Seedling survival percentage= $\frac{\text{Seedling survival}}{\text{Seed germination}} \times 100$

Relative growth rate (RGR) of the young seedlings was calculated using height increase data and the equation below:

$$RGR = \frac{(lnH1-lnH2) \times 36500}{No. days between measurements}$$

H1 is height at first measurement; H2 is

Shoot: root ratio were calculate form

Shoot:root ratio= $\frac{\text{Dry weight of shoot}}{\text{Dry weight of root}}$

3.4 Drought stress tolerance

Three months after seeds had germinated; seedlings were subject to drought stress by withholding water. Damage to the seedlings was indicated as the necrotic leaf area (see 3.5.2) and seedling survival 1, 3, 5 and 7 days after withholding water. Drought injury was classified by 5 injury level:

1 = none: no visible damage

2 = slight: small necrotic areas on shoots (< 5 % of leaves area)

3 = moderate: well defined necrotic areas on shoot (< 25 % of leaves area)

4 = severe: extensive necrotic areas on shoots (> 25 % of leaves area but plant still alive)

5 = killed: plant appears dead.

Plants were assessed by drought injury level (1, 2, 3, 4 or 5). Then the average value for each replicate of each treatment was calculated (Korkmaz, 2002).

No. of days to reach to injury value 2 was defined by the length of time after withholding water until small necrotic area was appeared on leaves (injury level 2). Moreover, mean injury rating values among treatments were compared at 7 days after withholding water.

3.5 Data analysis

Data were subject to analysis of variance (ANOVA) using SPSS 17.0. The dependent variables were seed germination percentage, seedling survival percentage, MLD, seedling height, the number of leaves, fresh weight, dry weight, RGR, No. of days to reach to injury value 2 and Injury rating values at 7 days after withholding water. The independent factors were treatment and block (or replication). When ANOVA showed significant treatment effect, Tukey's HSD (Honest Significant Difference) test was applied to compare the mean at $P \le 0.05$.

CHAPTER 4 RESULTS

4.1 Seed germination and Seedling survival

The treatments applied had no significant effect on both germination and dormancy of *F. semicordata* and *F. racemosa* seeds and seedling survival within 3 months of seed sowing.

Mean seed germination percentage of *F. racemasa* was significantly higher than that of *F. semicordata* (P \leq 0.05). Median length of dormancy (MLD) of *F. racemosa* was significantly shorter than that of *F. semicordata* (P \leq 0.05) but seedling survival percentages of both species (surviving seedlings as a per cent of the number of seeds that germinated) were not significantly different (P \leq 0.05).



Figure 6 Seed germination and dormancy of *F. semicordata* and *F. racemosa*. (Control treatment): (A) Mean seed germination, mean number of seedlings which died (white box) and mean number of surviving seedlings (after 3 months) (blackbox), (B) MLD

4.1.1 Ficus semicordata

Mean seed germination percentage (across all three replicates) of *F.semicordata* varied from 35 to 45% (across treatments) 3 month after seed sowing(Fig 7). Some seedlings died soon after seed germination. Mean survival, measured as

the number of seedlings that survived as a per cent of the number of seeds that germinated varied from 44 to 77%, across treatments (Fig 8), but differences among treatments were not significant (P \leq 0.05).

MLD varied from 16 to 18 days (across treatments), but the differences among treatments were insignificant (P \leq 0.05).



Figure 7 *F. semicordata*: Mean seed germination (total bar height), mean number of seedlings which died (white box) and mean number of surviving seedlings (after 3 months) (black box).



Figure 8 Per cent seedling survival of *F. semicordata* at 3 months after sowing (as a per cent of the seeds that germinated).

4.1.2 Ficus racemosa

Mean seed germination percentage (across all three replicates) of *F. racemosa* varied from 95 to 100% (across treatments) 3 months after seed sowing (Fig 9). Some seedlings died shortly after seed germination.



Figure 9 *F. racemosa*; Mean seed germination (total bar height), mean number of seedlings which died (white box) and mean number of surviving seedlings (after 3 months) (black box).



Figure 10 Per cent seedling survival of *F. racemosa* at 3 months after sowing (as a per cent of the seeds that germinated).

Mean survival, measured as the number of seedlings that survived as a per cent of the number of seeds that germinated varied from 72 to 91% (Fig 10), across treatments, again but differences among treatments were not significant (P \leq 0.05).

The median length of dormancy (MLD) for all treatments was 10 -11 days, with no significant differences among treatment means ($P \le 0.05$).

Due to low seedling survival of *F. semicordata*, seedling performance and drought injury were accessed only for *F. racemosa* seedlings.

4.2 Seedling performance

Fertilizer alone had significantly increased seedling height, fresh weight and dry weight, compared with the control, but aspirin had no significant effects. Both fertilizer and aspirin had no clearly significant effects on relative growth rate (RGR).

Three months after sowing, seedlings of *F. racemosa* with fertilizer had significantly more leaves than those without (Table 28, P \leq 0.05). Seedlings which did not receive fertilizer had 2-4 leaves, whilst those that received fertilizer had 2-6 leaves, with the vast majority having 3-6 leaves (Fig 11).



Figure 11 The number of seedlings, which show the percentage form total seedling in each treatment, separated by length of the number of leaves.

For the seedling height, almost seedlings in non-fertilizer treatments are 0.8 - 1.0 cm height, whereas seedlings in fertilizer treatment are 1.1 - 1.3 cm height (Fig 12). The average height across all treatments varied form 0.9 - 1.2 cm at 3 months after sowing. It has a significantly different between fertilizer and non-fertilizer treatment but no significant among low dose and high dose fertilizer. The treatments with aspirin did not effect on seedling height (P ≤ 0.05) (Fig 13).



Figure 12 The number of seedlings, which showed the percentage form total seedling in each treatment, separated by length of the seedling height.



Figure 13 Mean heights at 13 weeks after sowing.

Mean relative growth rates (RGR) which calculated from High of all treatments varied from 325.07 to 364.11. RGR of High dose fertilizer treatment was higher than that of all other treatments but the difference was insignificant from low dose fertilizer and aspirin + high dose fertilizer treatments (Fig 14).



Figure 14 Mean relative growth rate

Because seedlings were very tiny, fresh and dry weights were measured for groups of five seedlings and root and shoot weight was not measure individually in this case. Fertilizer significantly increased both fresh and dry weights ($P \le 0.05$). The higher dose of fertilizer resulted in higher mean seedling weights but the increase was not significant compared with the lower dose of fertilizer ($P \le 0.05$). Aspirin had no effect on seedling dry and fresh weight. Mean fresh weights varied from 7.62 to 9.27 mg among non-fertilizer treatments and 28.17 to 33.38 mg among fertilizer treatment. Mean dry weights varied from 1.32 to 1.51 mg among non-fertilizer treatments and 3.29 to 4.53 mg, among fertilizer treatments.



Figure 15 Mean fresh (A) and dry weight (B) (per 5 seedlings)

4.3 Drought Stress

Aspirin had a significantly increased drought stress tolerant (P \leq 0.05). When seedlings were subjected to drought stress, necrotic spots appeared on the leaves, which were used an index of stress injury (Fig 18). With the control, the necrotic areas appeared and expanded faster than with the other treatments. The aspirin treated seedlings developed stress injury significant slower than non-aspirin treatments. Mean No. of day to reach to the injury rating value "2" (first necrotic area was appeared) of treatments varied from 4.98 to 5.48 days, whilst those of non-aspirin treatments varied from 4.33 to 4.76 days (Fig 16 and Fig 17).

The average injury rating of seedlings with aspirin treatments was significantly lower, compared with the control, but was not significantly different compared with the fertilizer treatments.

High dose fertilizer + aspirin conferred the greatest drought tolerance. Seven days after withholding water, the mean injury rating value of all treatment varied from 2.53 to 3.25 across treatments (Fig 19).



Figure 16 Mean injury rating value



Figure 17 Mean No. of days to reach to the injury rating value 2



Figure 18 Drought stress injury on leaves of F. racemosa seedling



Figure 19 Mean injury rating value at 7 days after withholding water

CHAPTER 5

DISSCUSSION

5.1 Seed germination and seedling survival

Seed germination percentage of *F. racemosa* was significantly higher than that of *F. semicordata*. During the experiment, I observed that some *F. semicordata* seeds were carried away from the germination tray by ants that may have contributed to the decrease in germination percentage, whereas *F. racemosa* seeds were not attractive to ants. *F. semicordata* seeds were bigger than those of *F. racemosa*, because their seeds had a thick mucilaginous seed coat that may have attracted the ants. This result is supported by Kuaraksa (2012), who reported that the low germination rate of some *Ficus* species maybe due to a thick mucilaginous coat which is absent in other species. He also speculated that the mucilaginous coat is necessary to attract ants.

The treatments applied had no significant effect on seed germination and MLD. In previous studies, various authors reported that the environment factor affecting *Ficus* spp. seed germination were medium moisture, soil pH, and sometimes light (Kuaraksa, 2012; Musa, 2005). Fertilizer is not a direct factor that affects *Ficus* seed germination. This result agrees with Kuaraksa (2012), who found that using fertilizer in the germination medium does not affect seed germination. Hayat *et al.* (2003) reported that aspirin can either inhibit seed germination or increase it, depending on the concentration applied and the plant species. No previous reports on the effects of aspirin on germination of *Ficus* spp. have been published. I found that aspirin did not affect seed germination of *F. semicordata* and *F. racemosa*.

Seedling survival was also unaffected by the treatments applied. This contrasts with Kuaraksa (2012) who found that using fertilizer in a germination medium reduces *Ficus* spp. seedling survival. However, the concentrations of fertilizer used were different. Kuaraksa used higher concentration than my experiment. Kuaraksa reported that *Ficus* seedling response negatively when received over fertilizer.

5.2 Seedling performance

Fertilizer significantly increased seedling height and fresh and dry weights of F. racemose in compared with the control in my experiments. Clearly the young seedlings responded positively to the extra N, P and K provided by the Osmocote fertilizer in the germination medium. These three elements play an important role in photosynthesis and support the formation of oils, sugars, and starches that promote seedling growth. Non-fertilizer treatments had access to less nutrients and therefore grew more slowly. This result agrees with Henley et al. (1999), who found that moderate application of 5-10-5 or 5-10-10 NPK fertilizer is sufficient for the growth of Ficus spp. as measured by height and fresh and dry seedling weights. In my experiments, seedlings that received the high dose fertilizer grew better than those which received the lower dose, although the difference was not statistically different. This result could have two explanations. Firstly, the difference between the low and high dose fertilizer treatments might have been too small to elicit a significant effect. Secondly, nutrient uptake of the tiny seedlings may have been limited by their small root systems and may have already reached maximum uptake capacity with the lower fertilizer dose. In such a case, adding more nutrients than their maximum uptake capacity would have no additional effect. In this experiment, the high dose fertilizer treatment may have been in excess of the seedlings maximum uptake capacity or the seedlings' requirements.

Mean relative growth rate (RGR) of seedlings receiving the fertilizer treatments was higher than for seedlings with non-fertilizer treatments. The high dose fertilizer treatment significantly increased RGR compared with the non-fertilizer treatments. However the low dose fertilizer treatment did not significantly increase RGR compared with the non- fertilizer treatments, because the variability of the results was high. This may have been due to the very small sizes of the seedlings being monitored and the short period (1 month) over which RGR was calculated and extrapolated to a per year value.

Aspirin treatments had no significant effect on seedling height, fresh weight, dry weight and RGR because under normal conditions, aspirin has no effect on plant growth. Mostly, aspirin affects plants under stressful conditions (Hayat *et al.* 2013).

5.3 Drought stress

All plants, subjected to drought stress developed stress injuries, but the level of drought stress injuries of the seedlings treated with aspirin was lower than that of seedlings without aspirin. Aspirin decreased drought injury by promoting plant protection mechanism that decreases the necrotic area on leaves. This result is support by a lot of research, which reported that aspirin decreases the negative impacts of drought stress. In this experiment drought stress injury declined when seeds were treated with 0.05 mM of aspirin solution. This result agrees with those of Khan et al. (2012) who reported that soaking wheat seeds in 0.03-0.05 mM aspirin solution ameliorated the negative impact of drought stress. In contrast, Senaratna et al. (2000) reported that soaking bean (Phaseolus vulgaris L.) and tomato (Lycopersicon esculentum L.) seeds in 0.05 mM or 1.0 mM of aspirin solution were not effective in ameliorating drought injury. The optimal concentration, which effectively decreases drought injury, is 0.1 and 0.5 mM of aspirin solution according to Korkmaz et al. (2007) who worked on Muskmelon (Cucumis melo L.). However, plant species in each those experiments were different and that may account for differences in effect. Moreover, my experiment was carried out with only one concentration of aspirin solution (0.05 mM). So, my experiment found that Ficus spp. seeds that soaked with 0.05 aspirin solution have higher tolerance than seeds which were not soaked with aspirin. However, 0.05 mM of aspirin solution was not reported as the most effective concentration for Ficus spp. seed because this experiment have studied on only one concentration of aspirin.

After 7 days, seedlings of all treatments wilted and died, because the seedlings had been subjected to drought conditions longer than those that can be ameliorated with aspirin. Therefore, although, aspirin decreases drought stress injury, but it cannot increase seedling survival over a long time of water deficit condition. However, Senaratna (2000) found that if seedlings of bean (*Phaseolus vulgaris* L.) and tomato (*Lycopersicon esculentum* L.) were watered again, after withholding water for 7 days, those treated with aspirin recovered, whereas control seedlings died.

The experiment reported here was limited because it involved only two quantities of fertilizer and one concentration of aspirin. Moreover, this work was carried out over a very short time, so, root and shoot dry weights were very tiny and difficult to measure accurately. However, the results showed conclusively that fertilizer accelerated *Ficus* spp. seedling growth and aspirin decreased drought injury, at least in the short term. More work is needed to test a wider range of fertilizer and aspirin concentrations and combinations.

Water deficit can be an important factor that limits seedling growth and survival, particularly in the first dry season after planting, since deforested sites are open, sunny and hot. Therefore, experiments with aspirin should be expanded to cover other framework species. The concentration of aspirin should be varied more to determine optimal concentrations that may maximize survival of planted tree saplings through the first dry season. For *Ficus* spp., it seems clear that addition of fertilizer to the germination medium (not usually recommended for most native forest tree species) does generate a significant growth spurt, which could reduce the time to pricking out and thus accelerate seedling production in nurseries. However, more work needs to be done to determine the optimal fertilizer treatments for a wider range of *Ficus* spp.

CHAPTER 6

CONCLUSIONS

For the propagation of *Ficus* species, seeds should be protected from ants. Osmocote 13-13-13 (slow release fertilizer) accelerates seedling growth of *F. racemosa* and *F. semicordata* and its use is therefore recommended to reduce seedling production time. Soaking seeds in 0.05 mM of aspirin solution decreases drought stress of very young *F. racemosa* seedlings for about 7 days. The effect of different concentrations of aspirin on large seedlings of a wider variety of framework tree species is recommended.

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APPENDICES

APPENDIX A

PICTURE ABOUT THE EXPERIMENTS



Figure 20 Ficus racemosa seed (left) and Ficus semicordata seed (right)



Figure 21 Seed extraction; (a) put the mosquito net on the top of bottle, (b) Figs were put on mosquito net, (c) and (d) sieved through strainer, (e) spread out on paper and left to dry, (f) dried seeds



Figure 22 Seed pretreatment or seed priming (a) seeds were put on filter layer which in polystylene box, (b) close the box and keep it in darkness for 24 hrs.



Figure 23 Germination medium and seed sowing (a) Germination medium: charcoalized rice husk and coarse sand, (b) put the medium in basket which has newspaper on the bottom, (c) Fertilizer were put on the medium and cover with the medium 1 cm. deep, (d) Seeds were put on the top of medium, non-covering.



Figure 24 Randomized complete block design (RCBD) with 3 replicates of each of 6 treatments



Figure 25 Seed germination monitoring (a) tooth stick indicates that seed was germinated, (b) white stick indicated that seedling died after germinated.



Figure 26 Seedling monitoring (a) Measuring height and counting no. of leave, (b) Measuring fresh and dry weight (1 sample contain 5 seedlings)



Figure 27 Seedlings in each treatment at 3 month after sowing

APPENDIX B

SEED GERMINATION DATA

Table 2 Number of seed germination of F. semicordata	

Donligation -	No. of seed germination								
Kepilcation -	T 1	T2	T3	T4	T5	T6			
R 1	23	40	31	34	47	27			
R2	27	18	22	25	11	46			
R3	54	52	49	46	50	62			
Average	35	37	34	35	36	45			
SD	16.86	17.24	13.75	10.54	21.70	17.52			
Statistic result	a	a	a	a	a	a			

 Table 3 Number of seedling died of F. semicordata

Donligation -	No. of seedling died								
Kephcation -	T1	T2	T3	T4	T5	T6			
R 1	7	23	24	14	16	27			
R2	10	10	11	14	10	22			
R3	1	3	8	31	5	12			
Average	6	12	14	20	10	20			
SD	4.58	10.15	8.50	9.81	5.51	7.64			

Table 4 Number of seedling survival of F. semicordata

Replication	No. of seedling survival									
-	T1 T2 T3 T4 T5 T6									
R 1	16	17	7	20	31	0				
R2	17	8	11	11	1	24				
R3	53	49	41	15	45	50				
Average	29	25	20	15	26	25				
SD	21.08	21.55	18.58	4.51	22.48	25.01				

Doulisation	Seedling survival (%)								
Replication	T1	T2	T3	T4	T5	T6			
R1	70	43	23	59	66	0			
R2	63	44	50	44	9	52			
R3	98	94	84	33	90	81			
Average	77	60	52	45	55	44			
SD	18.70	29.32	30.60	13.14	41.55	40.90			
Statistic result	a	a	a	a	a	a			

Table 5 Seedling survival percentages (measured form seed germinated) of F.semicordata

 Table 6 The median length of dormancy (MLD) of F. semicordata

Donligation	MLD (days)								
Replication	T1	T2	T3	T4	T5	T6			
R1	16	17	17	17	17	17			
R2	17	16	20	18	17	17			
R3	17	16	18	19	17	19			
Average	17	16	18	18	17	18			
SD	0.58	0.58	1.53	1.00	0.00	1.15			
Statistic result	a	а	a	a	a	a			

Table 7 Number of seed germination of F. racemosa

Doplication	No. of seed germination								
Kephcation	T1	T2	T3	T4	T5	T6			
R1	100	97	100	95	100	97			
R2	99	100	95	100	99	100			
R3	94	96	89	96	100	99			
Average	98	98	95	97	100	99			
SD	3.21	2.08	5.51	2.65	0.58	1.53			
Statistic result	a	a	a	a	a	a			

Dauliaatian			No. of se	edling died		
Replication	T1	T2	T3	T4	T5	T6 20 10 37 22 13 65
R1	12	8	27	15	24	20
R2	4	24	6	40	11	10
R3	10	22	22	27	18	37
Average	9	18	18	27	18	22
SD	4.16	8.72	10.97	12.50	6.51	13.65

Table 8 Number of seedling died of F. racemosa

Table 9 Number of seedling survival of F. racemosa

Donligation	No. of seedling survival							
Kephcation	T1	T2	T3	T4	T5	T6		
R1	88	89	73	80	76	77		
R2	95	76	89	60	88	90		
R3	84	74	67	69	82	62		
Average	89	80	76	70	82	76		
SD	5.57	8.14	11.37	10.02	6.00	14.01		

Table 10 Seedling survival percentage (measured form seed germinated) of F.racemosa

	Seedling survival (%)									
Replication	T1	T2	T3	T4	T5	T6 79 90 63 77 13 80				
R 1	88	92	73	84	76	79				
R2	96	76	94	60	89	90				
R3	89	77	75	72	82	63				
Average	91	82	81	72	82	77				
SD	4.26	8.80	11.34	12.11	6.45	13.80				

Doulisation	MLD (days)								
Replication	T1	T2	T3	T4	T5	T6			
R1	11	9	11	11	9	9			
R2	11	11	10	11	10	10			
R3	11	11	10	10	10	10			
Average	11	10	10	11	10	10			
SD	0.00	1.15	0.58	0.58	0.58	0.58			
Statistic result	a	a	a	a	a	a			

Table 11 The median length of dormancy (MLD) of F. racemosa

APPENDIX C

SEEDLING GROWTH DATA

No of loovo	Average number of Seedling (%)							
NU. OI leave	T1	T2	T3	T4	T5	T6		
1-2	36	38	0	1	2	1		
3-4	64	62	47	57	51	38		
5-6	0	0	53	42	47	61		
7-8	0	0	0	0	0	0		

Table 12 The number of leave (F. racemosa)

 Table 13 Seedling height at 13 weeks after sowing

Donligation		Height at 13 weeks(cm)							
Replication	T1	T2	T3	T4	T5	T6			
R1	0.91	0.91	1.25	1.29	1.24	1.24			
R2	0.76	0.82	1.19	1.28	1.2	1.22			
R3	1.04	1.01	1.17	1.19	1.16	1.2			
Average	0.90	0.91	1.20	1.25	1.20	1.22			
SD	0.14	0.10	0.04	0.06	0.04	0.02			
Statistic	0	0	h	h	h	h			
result	d	d	U	U	U	U			

Table 14 Seedling height

Treatment _	Height(cm)						
Treatment -	9 weeks	11 weeks	13 weeks				
T1	0.72 ± 0.13	0.85 ± 0.15	0.90 ± 0.14				
T2	0.73 ± 0.14	0.90 ± 0.12	0.91 ± 0.10				
T3	0.95 ± 0.03	1.16 ± 0.05	1.18 ± 0.08				
T4	0.95 ± 0.01	1.15 ± 0.04	1.25 ± 0.06				
Т5	0.95 ± 0.02	1.12 ± 0.04	1.19 ± 0.03				
T6	0.96 ± 0.01	1.13 ± 0.02	1.22 ± 0.02				

Donligation	Relative growth rate								
Replication	T1	T2	T3	T4	T5	T6			
R 1	368.30	356.88	330.72	379.88	320.12	343.57			
R2	301.77	344.78	346.59	400.23	345.18	333.75			
R3	245.15	177.60	266.94	312.22	258.38	311.78			
Average	305.07	293.09	314.75	364.11	307.89	329.70			
SD	61.64	100.20	42.16	46.07	44.67	16.28			
Statistic	а	a	ab	ah	а	h			
result	u	u	ub	uU	u	0			

 Table 15 Relative growth rate

Table 16 Fresh weight

Doplication		Weight (mg)							
Replication	T1	T2	T3	T4	T5	T6			
R1	7.90	10.17	27.25	42.02	24.47	23.50			
R2	8.33	8.42	41.13	30.08	39.08	54.43			
R3	6.65	9.20	16.13	16.75	19.70	22.23			
AVG	7.62	9.27	28.17	29.62	27.75	33.38			
SD	0.87	0.88	12.53	12.64	10.09	18.23			
Statistic	9	0	h	h	h	h			
result	a	d	U	U	U	U			

Table 17 Dry weight

Donligation			Dry w	veight(mg)		
Kephcation	T1	T2	T3	T4	T5	T6
R1	1.45	1.83	5.80	7.43	3.87	7.10
R2	1.10	1.17	4.08	3.30	4.28	4.73
R3	1.40	1.51	1.28	1.47	1.72	1.75
AVG	1.32	1.51	3.72	4.07	3.29	4.53
SD	0.19	0.33	2.28	3.05	1.37	2.68
Statistic result	а	а	а	а	а	а

APPENDAIX D DROUGHT STRESS INJURY DATA

Trootmont	Days after withholding water							
Treatment	1 day	3 days	5 days	7 days				
T1	1.17 ± 0.03	1.52 ± 0.14	2.00 ± 0.35	3.25 ± 0.58				
T2	1.08 ± 0.10	1.35 ± 0.22	1.87 ± 0.20	2.73 ± 0.06				
T3	1.00 ± 0.00	1.50 ± 0.15	1.98 ± 0.29	3.13±0.38				
T4	1.03 ± 0.06	1.35 ± 0.10	1.80 ± 0.30	2.85 ± 0.23				
Т5	1.00 ± 0.00	1.30 ± 0.05	1.82 ± 0.25	2.80 ± 0.22				
T6	1.00 ± 0.00	1.18 ± 0.08	1.60 ± 0.22	2.53±0.13				

 Table 18 Injury rating value

Table 19 No. of days to reach to injury rating value 2

Donligation			No. of da	ys (days)		
Replication	T1	T2	T3	T4	T5	T6
R1	4.60	4.87	4.2	4.6	4.67	5.6
R2	3.60	4.13	4.27	4.27	5	5.2
R3	5.60	5.93	4.53	5.4	5.6	5.67
Average	4.60	4.98	4.33	4.76	5.09	5.49
SD	1.00	0.90	0.17	0.58	0.47	0.25
Statistic result	а	a	а	ab	bc	b

 Table 20 Injury rating value at 7 days after withholding water

Donligation	Injury rating value(at 7 days)						
Replication	T1	T2	T3	T4	T5	T6	
R 1	3.85	2.7	3.5	3.05	2.9	2.55	
R2	3.2	2.8	3.15	2.9	2.95	2.65	
R3	2.7	2.7	2.75	2.6	2.55	2.4	
Average	3.25	2.73	3.13	2.85	2.80	2.53	
SD	0.58	0.06	0.38	0.23	0.22	0.13	
Statistic result	а	bc	ab	ab	bc	с	

APPENDIX E

STATISTIC TABLE

Statistic table of seed germination and seedling survival

1.F. semicordata

Table 21 ANOVA of 1	F. semicordata's seed	germination	(Data in table 2))
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Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2538.833	7	362.690	3.695	0.031
Intercept	24420.500	1	24420.500	248.766	0.000
Treatment	234.500	5	46.900	0 .478	0.785
Replication/block	2304.333	2	1152.167	11.737	0.002
Error	981.667	10	98.167		
Total	27941.000	18			
Corrected Total	3520.500	17			

Table 22 Turkey's HSD of replication/block for F. semicordata's seed germination(Data in table 2)

Replication/	N	Subset for alpha = 0.05		
block	11	1	2	
2	6	24.8333 <mark>a</mark>		
1	6	33.6667 <mark>a</mark>		
3	6		52.0000 <mark>b</mark>	
Sig.		0.3130	1.0000	

Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	7466.006	7	1066.572	1.739	0.206
Intercept	55675.620	1	55675.620	90.767	0.000
treatment	2187.357	5	437.471	0.713	0.628
Replication/block	5278.650	2	2639.325	4.303	0.045
Error	6133.896	10	613.390		
Total	69275.523	18			
Corrected Total	13599.902	17			

 Table 23 ANOVA of F. semicordata's seedling survival percentage (Data in table 5)

Table 24 ANOVA of F. semicordata's MLD (Data in table 6)

Source of	Sum of				
variances	Squares	df	Mean Square	F	Sig.
Corrected Model	11.667	7	1.667	2.000	0.155
Intercept	5408.000	1	5408.000	6489.600	0.000
Treatment	9.333	5	1.867	2.240	0.130
Replication	2.333	2	1.167	1.400	0.291
Error	8.333	10	0.833		U.
Total	5428.000	18			i I
Corrected Total	20.000	17			

2. F. racemosa

 Table 25 ANOVA of F. racemosa's seed germination (Data in table 7)

Source of	Sum of				
variances	Squares	df	Mean Square	F	Sig.
Corrected Model	76.556	7	10.937	1.441	0.290
Intercept	171307.56	1	171307.56	22573.470	0.000
Treatment	43.111	5	8.622	1.136	0.402
Rep	33.444	2	16.722	2.204	0.161
Error	75.889	10	7.589		
Total	171460.00	18			
Corrected Total	152.444	17			

Source of	Sum of	df Mean Square		F	Sig.
variances	Squares				
Corrected Model	786.118	7	112.303	1.108	0.427
Intercept	117628.67	1	117628.67	1160.793	0.000
Treatment	594.131	5	118.826	1.173	0.387
Replication/block	191.988	2	95.994	0.947	0.420
Error	1013.347	10	101.335		
Total	119428.13	18			
Corrected Total	1799.466	17			

Table 26 ANOVA of F. racemosa's seedling survival percentage (Data in table10)

 Table 27 ANOVA of F. racemosa's MLD (Data in table 11)

Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5.056 ^a	7	0.722	1.585	0.245
Intercept	1901.389	1	1901.389	4173.780	0.000
Treatment	4.278	5	0.856	1.878	0.185
Replication/block	0.778	2	0.389	0.854	0.455
Error	4.556	10	0.456		
Total	1911.000	18			
Corrected Total	9.611	17			

Statistic table for seedling growth of F. racemosa

1. No. of leave

Table 28 ANOVA of Number of leave

Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	1304.631	7	186.376	406.530	0.000
Intercept	21132.684	1	21132.684	46095.436	0.000
Treatment	1101.400	5	220.280	480.483	0.000
Replication/block	200.954	2	100.477	219.164	0.000
Error	646.880	1411	0.458		
Total	23022.000	1419			
Corrected Total	1951.511	1418			

 Table 29 Turkey's HSD of treatment of Number of leave

Tractmont	N	Subset for alpha = 0.05				
Treatment	reatment IN		2	3	4	
T1	267	2.6667 <mark>a</mark>				
T2	239	2.7029 <mark>a</mark>				
T4	206		4.2913 <mark>b</mark>			
T5	249		4.4096 <mark>b</mark>	4.4096 <mark>c</mark>		
T3	229			4.5240 <mark>c</mark>		
T6	229				4.7686 <mark>d</mark>	
Sig.		0.992	0.406	0.446	1.000	

Table 30 Turkey's HSD of replication/block for Number of leave

Replication/	Ν	Subset for $alpha = 0.05$			
block		1	2	3	
3.00	439	3.3098 <mark>a</mark>			
2.00	499		3.9739 <mark>b</mark>		
1.00	481			4.2245 <mark>c</mark>	
Sig.		1.000	1.000	1.000	

2. Seedling height

Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	32.844	7	4.692	116.984	0.000
Intercept	1708.624	1	1708.624	42600.628	0.000
Treatment	31.747	5	6.349	158.309	0.000
Replication	1.154	2	0.577	14.392	0.000
Error	55.389	1381	0.040		
Total	1788.560	1389			
Corrected Total	88.233	1388			

 Table 31 ANOVA of seedling height at 13 weeks after sowing

 Table 32 Turkey's HSD of treatment for seedling height at 13 weeks after sowing

Trootmont	N	Subset for $alpha = 0.05$			
Treatment	11	1	2		
T1	263	0.8962 <mark>a</mark>			
T2	227	0.9110 <mark>a</mark>			
T5	244		1.1975 <mark>b</mark>		
T3	227		1.2044 <mark>b</mark>		
T6	225		1.2218 <mark>b</mark>		
T4	203		1.2502 <mark>b</mark>		
Sig.		0.969	0.055		

 Table 33 Turkey's HSD of replication/block for seedling height at 13 weeks after sowing

Replication/	N	Subset for $alpha = 0.05$				
block	IN	1	2	3		
3	439	3.3098 <mark>a</mark>				
2	499		3.9739 <mark>b</mark>			
1	481			4.2245 <mark>c</mark>		
Sig.		1.000	1.000	1.000		

3. Relative growth rate (RGR)

Table 34 ANOVA of RGR

Source of	Sum of	df	Moon Squara	Б	Sig
variances	Squares	ui	Mean Square	Γ	Sig.
Corrected	2.826E+06	7	403700.109	12.222	0.000
Model					
Intercept	1.390E+08	1	1.390E+08	4206.849	0.000
Treatment	607936.989	5	121587.398	3.681	0.003
Replication	2.259E+06	2	1.130E+06	34.194	0.000
Error	4.555E+07	1379	33031.828		
Total	1.895E+08	1387			
Corrected Total	4.838E+07	1386			

Table 35 Turkey's HSD of treatment for RGR

Treatment	N	Subset for al	pha = 0.05
Treatment	1	1	2
T2	227	299.2833 <mark>a</mark>	
T1	262	303.9458 <mark>a</mark>	
T5	244	308.5594 <mark>a</mark>	
T3	227	321.6273 <mark>a</mark>	321.6273 <mark>b</mark>
T6	224	325.8616 <mark>a</mark>	325.8616 <mark>b</mark>
T4	203		362.2980 <mark>b</mark>
Sig.		0.620	0.157

Table 36 Turkey's HSD of replication/block for RGR

Replication/	N	Sut	oset
block	1	1	2
3	426	258.9646 <mark>a</mark>	
2	486		343.4782 <mark>b</mark>
1	475		347.7036 <mark>b</mark>
Sig.		1.000	0.934

4. Fresh and dry weight

Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	10248.991	7	1464.142	16.728	0.000
Intercept	36887.807	1	36887.807	421.454	0.000
Treatment	7500.544	5	1500.109	17.139	0.000
replication	2748.447	2	1374.223	15.701	0.000
Error	5601.612	64	87.525		
Total	52738.410	72			
Corrected Total	15850.603	71			

 Table 37 ANOVA of fresh weight (data in table 16)

Table 38 Turkey's HSD of treatment for fresh weight

Treatment	Ν	Subset for $alpha = 0.05$	
		1	2
1	12	7.625000 <mark>a</mark>	
2	12	9.266667 <mark>a</mark>	
5	12		27.750000b
3	12		28.166667 <mark>b</mark>
4	12		29.616667 <mark>b</mark>
6	12		33.383333 <mark>b</mark>
Sig.		0.998	0.681

 Table 39 Turkey's HSD of replication/block for fresh weight

Replication/	NI	Subset for $alpha = 0.05$			
block	IN	1	2	3	
3	24	15.108333 <mark>a</mark>			
1	24		22.554167 <mark>b</mark>		
2	24			30.241667 <mark>c</mark>	
Sig.		1.000	1.000	1.000	

Source of	Sum of	df	Mean Square	F	Sig.
variances	Squares		1		0
Corrected Model	221.074	7	31.582	18.450	0.000
Intercept	678.961	1	678.961	396.637	0.000
Treatment	109.088	5	21.818	12.745	0.000
replication	111.986	2	55.993	32.710	0.000
Error	109.555	64	1.712		
Total	1009.590	72			
Corrected Total	330.629	71			

 Table 40 ANOVA of dry weight (data in table 17)

Table 41 Turkey's HSD of treatment for dry weight

Treatment	N	Subset for a	llpha = 0.05
		1	2
T1	12	1.316667 <mark>a</mark>	
T2	12	1.508333 <mark>a</mark>	
T5	12		3.291667 <mark>b</mark>
Т3	12		3.716667 <mark>b</mark>
T4	12		4.066667 <mark>b</mark>
Т6	12		4.525000 b
Sig.		0.999	0.206

Table 42 Turkey's HSD of replication/block for dry weight

Replication/		Subs	et for alpha =	= 0.05
block	Ν	1	2	3
3	24	1.525000 <mark>a</mark>		
2	24		3.108333 <mark>b</mark>	
1	24			4.579167 <mark>c</mark>
Sig.		1.000	1.000	1.000

5. Statistic table of drought injury rating value

Table 43 ANOVA of No. of days to reach to the injury rating value 2 (data in table19)

Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	110.594	7	15.799	6.233	0.000
Intercept	8275.211	1	8275.211	3264.842	0.000
Trement	49.356	5	9.871	3.894	0.002
Replication	61.239	2	30.619	12.080	0.000
Error	892.194	352	2.535		
Total	9278.000	360			

Table 44 Turkey's HSD of treatments of No. of days to reach to the injury ratingvalue 2

Treatment	N	Subset for a	llpha = 0.05
		1	2
T1	60	4.3500 <mark>a</mark>	
T2	60	4.5000 <mark>a</mark>	
T5	60	4.6667 <mark>a</mark>	
T3	60	4.8000 <mark>a</mark>	4.8000 <mark>b</mark>
T4	60	4.9500 <mark>a</mark>	4.9500 <mark>b</mark>
T6	60		5.5000 <mark>b</mark>
Sig.		0.309	0.156

Table 45 Turkey's HSD of replication/block for No. of days to reach to the injuryrating value 2 (data in table 19)

Replication/	Ν	Subset for $alpha = 0.05$	
DIOCK		1	2
3	120	2.6167 <mark>a</mark>	
2	120		2.9417 <mark>b</mark>
1	120		3.0917 <mark>b</mark>
Sig.		1.000	0.287

Source of variances	Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	35.150	7	5.021	8.500	0.000
Intercept	2992.900	1	2992.900	5066.126	0.000
Trement	21.000	5	4.200	7.109	0.000
Replication	14.150	2	7.075	11.976	0.000
Error	207.950	352	0.591		
Total	3236.000	360			
Corrected Total	243.100	359			

Table 46 ANOVA of drought injury rating value at 7 days after withholding water(data in table 20)

Table 47 Turkey's HSD of treatment for drought injury rating value at 7 days after

 withholding water

Treatment	Ν	Subset			
	- 1	1	2	3	
T6	60	2.5333 <mark>a</mark>			
T2	60	2.7333 <mark>a</mark>	2.7333 <mark>b</mark>		
T5	60	2.8000 <mark>a</mark>	2.8000b		
T4	60	2.8500 <mark>a</mark>	2.8500b	2.8500c	
T3	60		3.1333 <mark>b</mark>	3.1333c	
T1	60			3.2500c	
Sig.		0.215	0.052	0.052	

Table 48 Turkey's HSD of replication/block for drought injury rating value at 7 days

 after withholding water

Replication/	Ν	Subset for alpha = 0.05		
block		1	2	
3	120	2.6167 <mark>a</mark>		
2	120		2.9417 <mark>b</mark>	
1	120		3.0917 <mark>b</mark>	
Sig.		1.000	0.287	

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