

ARTIFICIAL FLOWERING IN *TRIPLOCHITON SCLEROXYLON*

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Received April 2010

DJAGBLETEY GD, OFORI DA & COBBINAH JR. 2011. Artificial flowering in *Triplochiton scleroxylon*. *Triplochiton scleroxylon* is an important indigenous tropical hardwood species in West Africa. Due to its erratic flowering cycles, seeds are not readily available on a year to year basis, posing considerable difficulty in plantation establishment. To ensure abundance and continuous supply of planting stock, various treatments to induce flowering on grafts and mature trees were tested. These included spraying of urea solution on leaves, bark ringing and soil drenching with 2,4-dichlorophenoxyacetic acid solution. The studies were carried out in two agroecological zones in Ghana, namely, the dry and moist semi-deciduous ecozones. In the dry semi-deciduous ecozone, fruiting occurred from November–December 2005 in girdled (80%) and ungirdled (30%) trees whereas in the moist semi-deciduous ecozone there was less fruiting, i.e. girdled (40%) and ungirdled (0%) trees in the same period. In all plants (30), the region within 10 cm above and below the girdles were swollen four months after girdling. Soluble carbohydrate concentrations in the bark before flowering were higher above the girdle (8.5%) than below (6.0%). Leaf shedding and bud development were greatest in treatments with 50, 37.5 and 25% urea compared with 12.5 and 0% urea. The results suggest that *T. scleroxylon* can be artificially induced to flower.

Keywords: Girdling, urea spray, defoliation, fruiting, soluble carbohydrates

DJAGBLETEY GD, OFORI DA & COBBINAH JR. 2011. Pembungaan *Triplochiton scleroxylon* secara tiruan. *Triplochiton scleroxylon* ialah spesies kayu keras tropika yang asli dan utama di Afrika Barat. Kitaran bunga *T. scleroxylon* yang tidak menentu mengakibatkan kuantiti biji berubah-ubah dari setahun ke setahun dan ini menyulitkan penubuhan ladang spesies tersebut. Untuk memastikan sumber stok penanaman yang banyak dan berterusan, beberapa rawatan untuk mengaruh pembungaan pada cantuman dan pokok matang diuji. Rawatan termasuk penyemburan daun dengan larutan urea, penggelangan kulit dan pelencunan tanah dengan larutan asid 2,4-diklorofenoksiasetik. Kajian dijalankan di dua zon ekologi tani di Ghana iaitu ekozon daun separa luruh kering dan ekozon daun separa luruh lembap. Di ekozon daun separa luruh kering, pembuahan berlaku dari bulan November–Disember 2005 pada pokok yang digelang (80%) dan pokok yang tidak digelang (30%). Pada tempoh yang sama, ekozon daun separa luruh lembap kurang berbuah iaitu pokok yang digelang (40%) dan pokok yang tidak digelang (0%). Empat bulan selepas penggelangan, kawasan 10 cm di atas dan di bawah gelang didapati bengkak pada semua pokok (30). Sebelum pembungaan, kepekatan karbohidrat terlarut dalam kulit adalah lebih tinggi di atas gelang (8.5%) berbanding bahagian bawah gelang (6.0%). Keguguran daun dan perkembangan tunas paling banyak dalam rawatan 50% = 37.5% = 25% urea berbanding rawatan 12.5% = 0%. Keputusan mencadangkan yang *T. scleroxylon* boleh diaruh untuk berbunga secara tiruan.

INTRODUCTION

Triplochiton scleroxylon (trade name obeche) is a very important tropical hardwood timber species in the West and Central African humid forests. Its distribution ranges from Guinea in West Africa to Democratic Republic of Congo in Central Africa. The wood is used for interior joinery, panelling, moulds, furniture, profile boards and sculptures. Despite its great economic importance and the associated high exploitation, regeneration has only been

achieved through a small number of plantations (Longman & Leakey 1995, Mayaka et al. 1995, Adu-Anning & Anglaere 1999). Typically the establishment of large-scale plantations of *T. scleroxylon* has been hindered by irregular seed production. Flowering of *T. scleroxylon* is naturally erratic, occurring at four- to five-year intervals (Taylor 1960, Jones 1976, Oni 1990).

With the rapid interest in private sector forest plantation in Ghana, it has become necessary to

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implement measures to guarantee sustainable production of this timber species. Plantation development calls for the supply of abundant planting stock. In the absence of good seeds, clonal techniques have been used. Vegetative propagation techniques using leafy stem cuttings have been developed (Ladipo et al. 1991, Nketiah 1994, Nketiah & Ofori 1996, Nketiah et al. 1998, Leakey 2004). However, demands for *T. scleroxylon* by large-scale commercial plantation developers cannot be met through this technique. Mass propagation through tissue culture (Ofori & Cobbinah 2007) can be used to supplement propagation by cuttings but this technique is expensive and requires both specialised laboratories and highly skilled technicians.

Research to induce flowering regularly is one option towards overcoming the difficulties arising from irregular seed production. In conifers, application of gibberellic acid is an effective flower induction technique (Longman 1982), but in tropical trees there have been few successes (Longman 1985).

Despite the irregularity in flowering and fruiting in *T. scleroxylon*, precocious flowering in successive years has been reported in 26- and 82-month-old cuttings of *T. scleroxylon* grown in a tropicalised glasshouse in Penicuik, Scotland (Leakey et al. 1981). This illustrates that the species can be manipulated to flower and produce seeds under artificial conditions even in young plants. In this case, it is thought that root dormancy is induced by low root temperatures, while shoots are active in warm air (30 °C), perhaps mimicking drought stress during the short dry season.

The objective of this study was to identify suitable methods of flower induction in *T. scleroxylon* to enhance seed production, regeneration and sustainable production of the species.

MATERIALS AND METHODS

Study sites

The nursery experiment was conducted at the Forestry Research Institute of Ghana (FORIG) nursery. FORIG is located in Fumesua which is in the moist semi-deciduous (MSD) forest zone of Ghana. It is located at 6° 44' N, 1° 30' W and at 280 m above sea level. Mean annual rainfall during the study period was 1500 mm. The area experiences a major rainy season from April till

June, and a minor rainy season, from September till early November. The minor rainy season is followed by the dry season, which ends in mid-March.

The field experiments were conducted in South Formangso Forest Reserve (SFFR) in the MSD forest zone of Ghana and Afram Head-Waters Forest Reserve (AHWFR) in the dry semi-deciduous forest zone (DSD) of Ghana. SFFR is located at 6° 35' N, 1° 57' W with mean annual rainfall between 1500 and 1750 mm. AHWFR is located at 7° 10' N, 1° 40' W with mean annual rainfall between 1250 and 1500 mm (Hall & Swaine 1981, Hawthorne & Abu-Juam 1995, Wagner et al. 2008).

Bark ringing/girdling

Scions were collected from five large mature trees (AB1, AB2, AB3, AB4 and AB5) located in AHWFR and grafted onto seedling rootstocks. At one year after grafting, 15 grafts (one graft per clone per treatment = 5 × 3 treatments) were subjected to three treatments as follows. (1) Five grafts were subjected to bark ringing on a vigorous growing branch, (2) five grafts were subjected to bark ringing at 5 cm above soil, (3) five grafts were left intact as control. The experiment was set up at FORIG, Fumesua, Kumasi, Ghana in randomised complete blocks with five replications (one plant per treatment per plot). Bark ringing started in December 2004 and was repeated in June 2005.

In the field studies, 30 mature trees growing within 1 km radius were selected at each zone. Fifteen trees at each zone were girdled at 1.3 m above ground and were compared with the other 15 un-girdled trees. Girdling operations were performed in June and September 2004 in the MSD and DSD forest zones respectively. The girdles were re-opened in June 2005 and June 2006. Samples of bark (3 cm²) were taken from below and above the girdle just before flowering (November 2005) and after fruiting (June 2006) for analysis of soluble carbohydrate content using the Anthrone method (Trevelyan et al. 1952). The number of girdled and un-girdled trees that flowered and fruited were recorded.

Defoliation using urea sprays

This experiment was set up in February 2005 with 25 grafts (five grafts each of AB1, AB2, AB3, AB4 and AB5) growing in 10 litre plastic

pots in a nursery at FORIG. The grafts were arranged in randomised complete blocks with five replications with each treatment represented by one graft in a block. The treatments were 12.5, 25, 37.5, 50 and 0% (water only) urea sprayed on leaves. Treatments were repeated every 6 months for 24 months. The rate of leaf fall and bud development were recorded.

Soil drenching with growth retardant

The experiment was set up in March 2005 with 25 grafts (five grafts each of AB1, AB2, AB3, AB4 and AB5). The grafts were grown in 10 litre plastic pots filled with sandy loam soil and maintained in a green house at FORIG. Temperatures within the green house ranged from 24 to 29 °C. The design was five grafts per treatment (one ramet per clone) arranged in randomised complete blocks with five replications. The treatments were water (control), 0.25, 0.5, 0.75 and 1.0 g 2,4-dichlorophenoxyacetic acid (2,4-D Amin 720 SL) applied to potted grafts. The treatments were applied after the grafts had not been watered for two days and were repeated every four weeks.

Data analysis

For the effect of girdling on flowering and fruiting, the numbers of trees that flowered and fruited at each site were presented in percentages. The differences in percentage

soluble carbohydrate above and below girdles were analysed using *t*-test. Data on soil drenching could not be analysed statistically since only one graft flowered. Data on rate of defoliation and bud development (percentages) were arc-sine transformed and then subjected to analysis of variance (ANOVA).

RESULTS

Bark ringing

Bark-ringed grafts in the nursery did not flower, but flowering occurred in mature trees in the field with minor differences in leaf shedding. In both ecozones, the number of girdled trees that completely shed their leaves was higher than that of the ungirdled trees (Figure 1). Leaf flushing after leaf shedding coincided with flowering. In the DSD ecozone, fruiting occurred from November–December 2005 in girdled (80%) and ungirdled (30%) trees (Table 1). In the MSD ecozone, there was less fruiting on the girdled (40%) and ungirdled (0%) trees in the same period (Table 1). Regions just above and below the girdles were swollen at four months after girdling. Analysis of soluble carbohydrates in the bark before flowering showed significantly ($p \leq 0.05$) higher concentration of carbohydrates above the girdle (8.5%) than below (6.0%) (Table 2). Soluble carbohydrate concentrations in the bark after fruiting on the other hand showed no significant ($p \geq 0.05$) difference (Table 2).

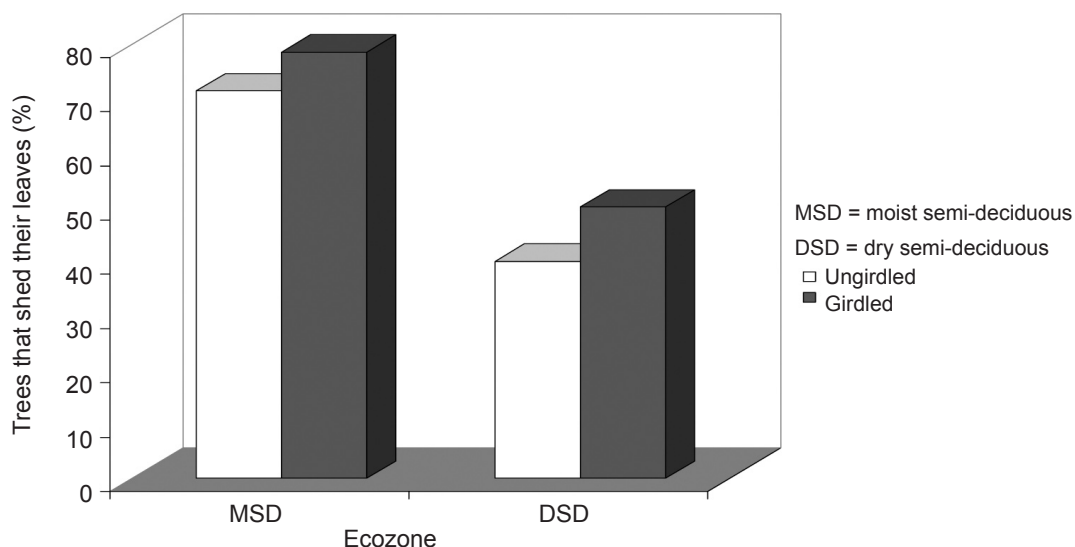


Figure 1 Percentages of trees in each treatment that completely shed their leaves in the two ecological zones

Urea application and defoliation

Urea sprayed on leaves significantly promoted leaf shedding. The rates of leaf shedding for 25, 37.5 and 50% urea were significantly higher ($p \leq 0.05$) than those for control and 12.5%. The control had the lowest leaf shedding and remained fresh throughout (Figure 2). Bud development was also influenced by urea applicatio. Plants that received urea spray on their leaves had significantly ($p \leq 0.05$) greater rate of bud development than the control (Figure 3).

Soil drenching with growth retardant

All grafts that received soil drenching with 2, 4-dichlorophenoxyacetic acid showed some signs of stress within the first week of application but later recovered. Only one graft flowered (Figure 4).

DISCUSSION

Flowering only occurred in treated trees in the field and was associated with leaf shedding. Bark-ringing of grafts in the nursery did not cause flowering. Leaf shedding occurred in both ecological zones and was greatest in girdled trees. The higher incidence of flowering in the DSD forest zone could be attributed to the more severe earlier and longer drought. This is in conformity with the findings of Knudsen et al. (2001), who reported on increased flowering in *Anaccacia xanthorrhiza* during a period of low rainfall. Precocious flowering in the glasshouse environment may be explained by root dormancy, a phenomenon likely to occur during a period of drought (Leakey et al. 1981). Thus, the results of the present study support the hypothesis that flower induction in *T. scleroxylon* is triggered by water stress.

Table 1 Proportion of *T. scleroxylon* trees that fruited in response to bark ringing

Zone	% Girdled tree	% Ungirdled tree
Dry semi-deciduous forest	80	30
Moist semi-deciduous forest	40	0

Table 2 Carbohydrate content in bark before flowering and after fruiting

Bark location	% Soluble carbohydrate	
	Before flowering	After fruiting
Above girdle	8.49 ± 0.77 a	2.62 ± 0.46 a
Below girdle	6.04 ± 0.21 b	2.92 ± 0.74 a

Means in the same columns followed by different letters are significantly different.

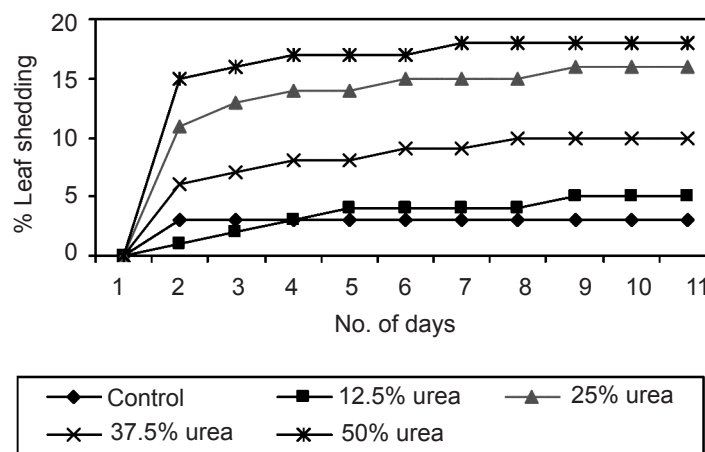


Figure 2 Rate of leaf shedding after urea spray application

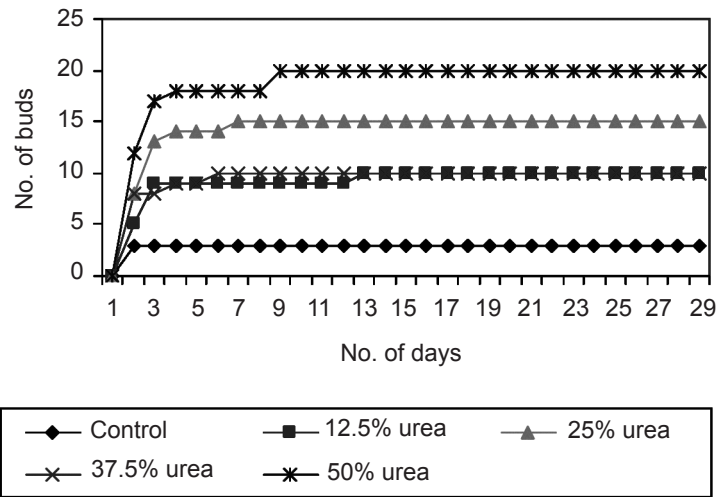


Figure 3 Rate of bud development after urea application



Figure 4 Graft of *Triplochiton scleroxylon* that flowered at six months after soil drenching with 0.5 g 2,4-dichlorophenoxyacetic acid

The swelling above girdle was attributed to accumulation of assimilate reserves as illustrated by the higher concentration of soluble carbohydrates above the girdle (8.5%) compared with below the girdle (6.0%) prior to flowering (Table 2). Carbohydrate accumulation is probably due to interruption of phloem transportation by the girdle as reported by several studies showing that girdling impedes the flow of sugars to the roots and promotes the development of reproductive organs (Li et al. 2003, Onguso et al. 2004, Rivas et al. 2007, Hossain & Boyce 2009).

Carbohydrate concentrations above and below girdled bark, however, showed no significant difference after fruiting (Table 2). The lack of difference may be due to the need for

high energy soluble substances for flower and fruit development. So the accumulated carbohydrate or sugar above the ring may have been utilised in flower production. Girdling resulted in carbohydrate (soluble sugar and starch) accumulation in leaves and shoot bark above the girdle of citrus trees during the fruitless year, but during heavy fruiting, the trees did not accumulate carbohydrates above the girdle due to the high demand for carbohydrates by the developing fruit (Li et al. 2003). An increase in sugar level and other structural changes during transition to flower was reported in *Sinapise alba* (Bernier et al. 1993). In *Dryobalanops aromatica* (Dipterocarpaceae), total non-structural carbohydrate in branches decreased significantly

during the flowering period, suggesting that stored assimilates in branches were used for flowering (Ichie et al. 2005). Many other authors (Jose 1997, Li & Xiao 2000, Rivas et al. 2007, Hossain & Boyce 2009) working with different tree species have also found that girdling is effective in flower induction. The blockage of assimilate reserves above the girdle is used for flowering. In *T. scleroxylon*, an experiment using a modified deep-freezer cabinet to chill the root systems of plants tested the effects of different root:shoot temperatures. Soluble carbohydrates increasingly accumulated in leaves as root temperature was lowered (28, 20, 16 to 8 °C, with constant shoot temperature of 28 °C), but was highest in stems and roots, when the roots were in 16 °C (Longman et al. 1990). In the experiment, flowering occurred in all treatments, but the number of flowers was greatest when roots were at 16 °C.

In *T. scleroxylon*, flowering occurs when the trees are leafless during the dry season. Activities that reduce vegetative growth also tend to stimulate flowering and fruiting. Accordingly, urea solution is applied on the leaves to stimulate leaf shedding and to induce flower and fruit development. Urea application indeed leads to leaf shedding which in-turn promotes bud development. This is in accordance with the findings of Syamswida and Owen (1997) who reported that flower induction results from leaf bud transformation into flower buds. High flower production was recorded 30 days after induced defoliation (Parra-Tabla et al. 2004). Foliar application with 0.5 g paclobutrazol + 0.4 g of ethephon per litre promoted flowering in unproductive litchi trees of cultivar Tai So in Mauritius (Ramburn 2000). Furthermore, Fumie et al. (2007) and Inoue (1990) induced flowering in Satsuma mandarin by defoliating potted trees. In *T. scleroxylon*, however, forced leaf shedding in potted trees did not induce flowering. This could be due to the fact that the grafts were too small, structurally not well developed and have restricted assimilate reserves. Another possible reason for the failure to flower could be attributed to the decline in carbohydrate reserves in small grafts. Bennett et al. (2005) recorded decline in carbohydrate reserves in chardonnay grapevines after induced defoliation, resulting in significant decrease in flower development. Restricted carbohydrate reserve accumulation as a consequence of defoliation may have negative

impact on subsequent grapevine flowering and productivity.

In contrast to *Tabebuia pallida* (Longman et al. 1990), only one graft of *T. scleroxylon* flowered after application of 2,4-D Amin 720 SL solution at different concentrations. Ramburn (2000) also observed erratic fruiting with application of growth retardant although application of growth retardant has been effective as the flowering agent in mango, pineapple and durian fruit crop production in South-East Asia (Coronel 2001).

The present study suggests that girdling is the most effective physiological treatment for flower induction in *T. scleroxylon*. However further studies are required to examine the interactions between physiological and environmental factors for regular and effective flower induction and fruiting.

ACKNOWLEDGEMENT

We thank the African Forest Research Networks (AFORNET) for funding the *Triplochiton* project (Grant No. 3/3003).

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