Seed Germination and Seedling Growth of *Tithonia diversifolia* (Hemsl.) A. Gray, the Mexican Sunflower

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ABSTRACT

Studies on the seed germination and seedling growth of Tithonia diversifolia *(Hemsl.) A. Gray, the Mexican sunflower, was carried out. This involved the usage of some dormancy-releasing methods and the effect of some concentrations of three herbicide formulations on the young seedlings. Initial germination tests on fresh and stored seeds revealed a low percentage germination of less than 30%. The seeds of the weed exhibit dormancy. Subjecting the seeds to wet heat at 80 ºC and 100ºC and light treatment terminated dormancy both in the fresh and stored seeds. Light greatly enhanced the germination percentage of hot water – treated fresh seeds by about 70%. There was gradual increase in germination percentage with increase in storage period in dormancy-released seeds. The mean LAR (Leaf Area Ratio), NAR (Net Assimilation Rate) and RGR (Relative Growth Rate) are comparatively high in young seedlings. Concentrations of 0.5 – 2.0% of Gramoxone, Primextra and Galex are toxic to 1-month-old seedlings. The physiological basis of dormancy in the seeds of this noxious weed species as revealed through after-ripening and light requirements by the embryo are discussed.*

Key Words: - *Tithonia diversifolia*, Mexican sunflower, germination, weed, dormancy, herbicide, seedlings.

Introduction

Tithonia diversifolia (Hemsl.) A. Gray, Asteraceae (Compositae), commonly called Marigold, wild sunflower or Mexican sunflower, is a perennial noxious weed of field crops, waste land and road sides. The plant is about 2.5 m in height at full maturity and reproduce from seeds and from vegetative regrowth of basal stem when cut. It is native to Mexico and the Central Americas where it has been introduced into West Africa as an ornamental plant (Akobundu and Agyakwa 1987).

Tithonia diversifolia is now prominent and fast growing in Nigeria, inhabiting the rainbelt of the southern part of Nigeria. The weed also occurs in the wet part of the Guinean Savanna especially along the fringes of the rain belt (Latitude 6-9ºN). These areas (Fig. 1) have an annual rainfall of more than 1000 – 1250 mm with an optimum temperature of 28-33ºC (Akobundu and Agyakwa 1987). The weed grows along the fringes of highways and abandoned farmlands very close to highways. The heavy deposits of seeds from the previous year's population which have remained dormant

in the dry season (November-March) germinate at the onset of the early rain in April-May. The seedlings grow fast and attain maturity during the second rainfall peak of July-August when they flower and seed heavily (Akobundu 1987). The plant dries up before January of the following year yielding a lot of litter, which is often destroyed through bush burning in December-January (Agboola, 1998).

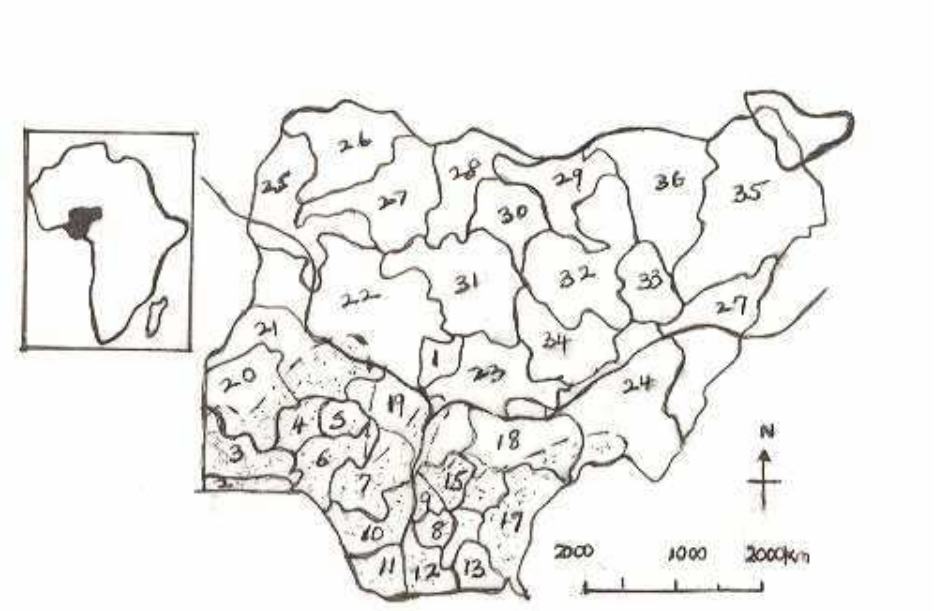

Fig.1.Map showing areas (shaded) of *Tithonia* infestation in Nigeria. States with infestation include: (2) Lagos, (3) Ogun, (4) Osun, (5) Ekiti, (6) Ondo, (7) Edo, (8) Imo, (9) Anambra, (10) Delta, (11) Bayelsa, (12) Rivers, (13) Akwa-Ibom, (14) Abia, (15)Enugu, (16) Ebonyi, (17) Cross River, (18) Benue, (19) Kogi, (20) Oyo, (21) Kwara.

In very recent times, *T. diversifolia* has now made an incursion into the farmland thus becoming a nuisance to farmers. The weed produces large biomass which farmers do not fully utilize as green manure. In the highlands of Kenya, the plant is one of those used as traditional hedges to demarcate both external and internal boundaries of farms and compounds. These hedges also protect soil and crops as well as producing fodder, green manure and mulch (Nyasim *et al.* 1997).

An infusion made from *Tithonia* leaves and buds is used as a medicine for constipation, stomach pains, indigestion, sore throat and liver pains. *Tithonia diversifolia* ash or solution of fermented extracts have been used to control termite infestation in agroforestry programmes (Spore 1998).

The vigour and luxuriance of the growth of *T. diversifolia* has stimulated research on the aspects of its biology and the problem posed by this weed in the environment (Smith and Anisu 1997). This study focuses on dormancy and germination of viable seeds, growth performance and chemical control of young seedlings. This is with a view to contributing to the growing knowledge on the ecology and physiology of this weed for effective control.

Materials and Methods

Survey of Tithonia *infestation in Nigeria*

A general survey of Nigeria was made for the spread of *Tithonia* infestation between June and August, 1999. These months fall within the period of heavy rainfall in the Southern areas and the beginning of rainfall in the Northern areas of the country. The journey took three routes each of which started from Lagos in the southwest coastal zone. The survey was made by traveling on land on six different occasions i.e. twice per route. The first route was along a vertical axis from Lagos in the southwest through Ilorin, Mokwa, Kotangora, Zuru to Sokoto in the northwest. The second was a diagonal route through, Ife, Akure, Okene, Lokoja, Abuja, Akwanga, Jos, Bauchi to Maiduguri in the northeast, while the third was in the southeastern direction through Benin, Onitsha, Owerri to Calabar.

Seed collection

Seed collection was made in January 2000 from dead and dried stands of *T. diversifolia* plants on an abandoned farmland separating the fence of the University of Agriculture Abeokuta, Nigeria from the major road. Because of the closeness of the stands, a devise for the collection of the seeds was made. This involved clearing the space in front of the first row of stands with cutlass and spreading a white cotton sheet (2 m x 3 m) on it. About two to three stands were brought together and shaken in such a way that the black 'seeds' which are actually the achenes (dry indehiscent one-seeded fruits characteristic of the Compositae) dropped on the white sheet. The seeds were then gathered together by folding the sheet and the dried leaves and branches that dropped with them were removed. The 'seeds' were collected and sun-dried again for two to three days and later stored in specimen bottles with cork lids at room temperature (28-30^{o}C).

Initial germination tests

Germination tests were carried out on freshly harvested seeds using some randomly selected ones according to the methods of Etejere and Ajibola, (1980); Agboola (1996; 1998). One hundred seeds were surface sterilized with 0.1% Mecuric chloride solution for 30 seconds and rinsed in several changes of distilled water. The seeds were then placed on moistened filter papers placed on 9 cm Petri dishes. The set up was maintained at 28-30^{o}C under a light intensity of 2000 lux as given by four 2 m Phillip flourescent tubes. Some sets of Petri dishes were wrapped in aluminium foil and incubated in the dark. Each germination test described below in the dormancy studies had five replicates. Germination counts were made with the emergence of the radicle to a length of 2 cm after 8 days incubation. The mean values for the percentage germination were calculated.

Dormancy studies

Seed lots of 100 each were soaked in concentrated sulphuric acid for 1 and 5 min. The acid was poured away and the seeds rinsed in several changes of distilled water before placing in Petri dishes for germination. Some seed lots were also immersed in hot water between 80^{o}C -100^{o}C for 30 seconds and one minute before cooling them in cold water. A total of 100 seeds from each temperature regime were prepared for germination in the laboratory and incubated both in the light and dark. Untreated seeds served as the control. Mean values for the percentage germination were calculated from germination counts from five replicates. The treatment means were

compared using the Analysis of Variance (ANOVA) at $P \geq 0.05$ and the Least Significance Difference (LSD) test.

Effect of storage period on seed germination

Pretreated seeds were sampled monthly from the specimen bottles where they have been stored for germination both in the light and dark for twelve months (January to December, 2000). The specimen bottles were corked after each sampling.

Growth Analysis

One to three-month old seedlings of *T. diversifolia* were used for the growth analysis in the green house. Dry weights of leaves and whole seedlings were made monthly for 3 months from June – August according to the methods of Beadle, (1982) and Agboola, (1996). This involved drying the whole plant in the electric oven at 60°C for 3 days until a constant weight was obtained. The leaf areas were also measured. The mean values of these parameters from five replicates were used to calculate 3 components of growth analysis including the Net Assimilation Rate (NAR), Leaf Area Ratio (LAR) and Relative Growth Rate (RGR).

RGR is calculated from the measured values of the dry weights of plants (W_2 & W_1) at different time (t_2 & t_1) using the formula:

$$RGR = \frac{\log_e W_2 - \log_e W_1}{t_2 - t_1}$$

The NAR was calculated from the measured values of leaf area (A_2 & A_1), dry weights of values of plant (W_2 & W_1) and time (t2 & t1) applying the formula.
W_1 and W_2 are initial and final dry weight measure at 1 and 3 months respectively.
t_1 and t_2 are the initial and final period of growth at the 1st and 3rd month when the measurements were taken.

$$NAR = \frac{W_2 - W_1}{(A_2 - A_1)} \bullet \frac{\log_e A_2 - \log_e A_1}{t_2 - t_1}$$

The calculation of LAR was done from the formula:

$$LAR: \frac{(LA_1)}{(LW_1)} \bullet \frac{(LA_2)}{(LW_2)}$$

Using the measured values of leaf area (LA_1 & LA_2) and dry weights of leaves (LW_1 & LW_2) the mean values from the calculations of NAR, LAR and RGR were used for the graphical interpretation of these growth components over three months.

Effect of herbicides on seeds and young seedlings

Six concentrations (0.1, 0.3, 0.6, 0.9, 1 and 2% solution) each of three commercially formulated herbicide solutions were prepared. The herbicides used include Galex, Gramoxone and Primextra. Hot water pre-treated seed lots of 100 each were prepared for germination in Petri dishes as earlier described. The various herbicide concentrations were used as soaking solutions while water served as the control. Five replicates of each set-up were made. Germination counts were made at two-day interval for ten days. Mean percentage germination were recorded for each treatment.

Five seedlings of *T. diversifolia* were raised in heat-sterilized loamy soils contained in black polythene bags for one to three months. Seedlings in each bag were thinned down to one per bag. Seedlings were divided into four lots for three herbicide higher concentrations of 0.5, 1.0 and 2.0% and water. The various herbicide concentration were tested on the young seedlings by spraying of the foliage leaves using sprayer and allowing to dry up. Plants were left in the open for observation. The experimental design was a Complete Randomized Block Design with three age groups x three herbicides x seven concentrations for the seed treatment and three age groups x three herbicides x four concentrations for the seedlings. Data was also subjected to Analysis of Variance ($P \geq 0.05$) to determine the significant differences between the control and herbicide treatments.

RESULTS

The results on the initial germination tests and dormancy studies on seeds of *T. diversifolia* are shown in Figs. 2, 3, 4 and 5. Initial germination of fresh untreated seeds, both in light and dark showed that the seeds do not germinate easily. Less than 30% germination was observed after 5 days of sowing of untreated seeds (Fig. 2). It was however observed that germination of the few seeds started 24h after sowing. There was 10-16% and 20-25% germination in seeds exposed to the light and dark respectively (Fig. 2). Seeds treated with concentrated sulphuric acid for 1 to 5 min. and sown in light showed a germination percentage of 20 to 32 after 5 to 7 days of

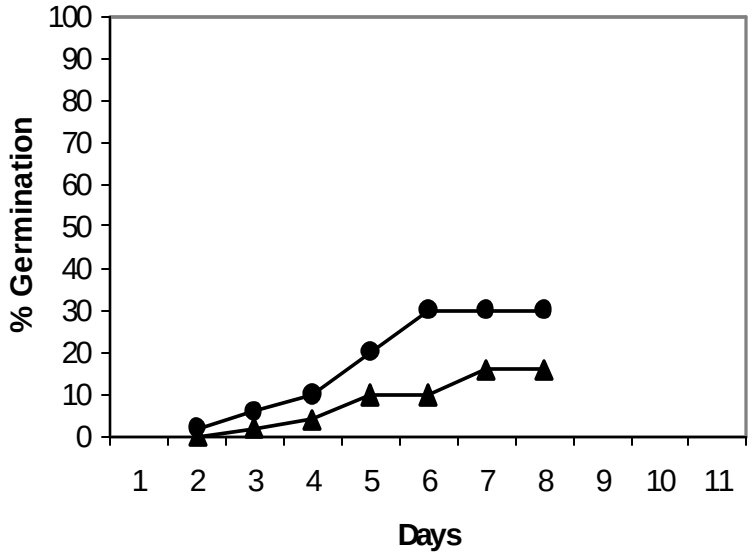

Fig. 2: Germination of fresh *Tithonia diversifolia* seeds after 10 days incubation under light and dark. Each point is the mean of five replicates

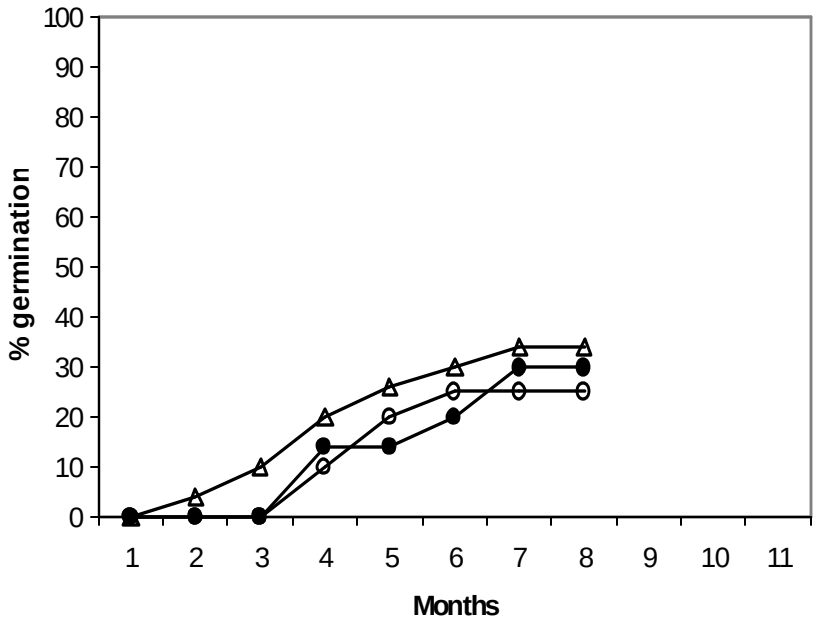

Fig. 3: Germination of *Tithonia diversifolia* seeds treated with concentrated sulphuric acid and sown in the light.

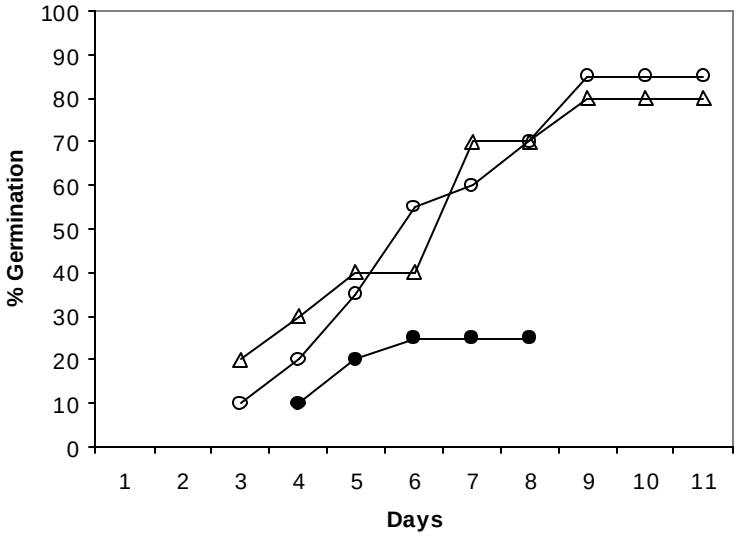

Fig. 4: Germination of *Tithonia diversifolia* seeds treated with hot water and sown under light. Each point is the mean of five replicates.

planting (Fig. 3). Sulphuric acid treatment therefore did not terminate the dormancy in the seed of this weed species effectively. Boiling water treatment for 30 to 60 seconds at 80 ^{o}C to 100^{o}C gave 80 to 85% compared to 25% observed in the control (Fig. 4). This gave a high significant effect at $P \geq 0.05$.

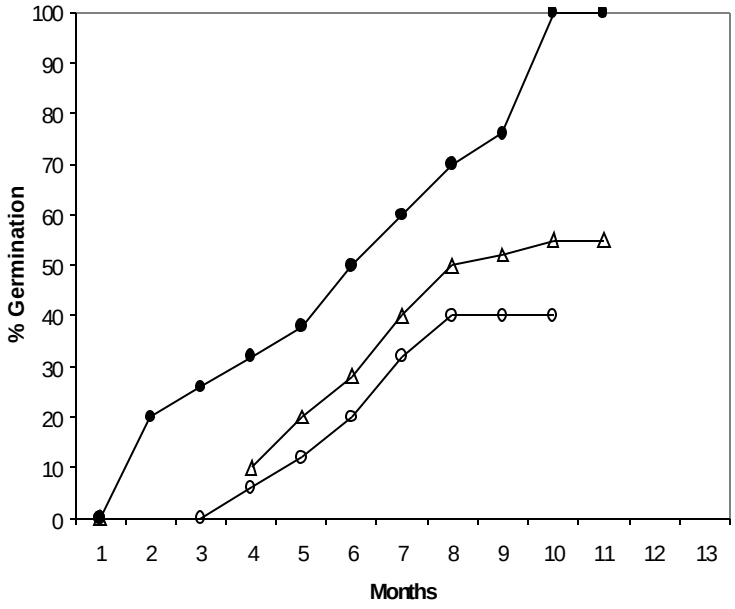

Fig. 5: Germination of hot water pretreated, and stored seeds of *Tithonia diversifolia* sown under light and dark regimes control - non-stored.

Germination percentage of 80 to 100% was observed in pre-treated seeds of *T. diversifolia* stored for 8 to 12 months and light-sown compared to 52 to 55% in the dark-grown seeds. Light had a significant effect ($P \geq 0.05$) on the germination of pre-treated fresh seeds of *T. diversifolia*. (Fig. 5). However, dark grown seeds had better germination than light grown seeds in those seeds stored for 12 months (Fig. 5). There was a gradual increase in germination percentage with increase in storage period in pre-treated seeds (Fig. 5). The results on the growth analysis on 1 to 3 month-old seedlings of *T. diversifolia* are shown in Figs. 7, 8 and 9. The mean RGR was 0.19-0.34 gg^{-1} $month^{-1}$, mean LAR 243.68 - 420.24 cm^2g^{-1} and mean NAR 0.52 - 1.37 x 10^{-4} gcm^{2-1} $month^{-1}$. It was generally observed that these mean values of LAR, NAR and RGR are higher in the seedlings especially at early phase of growth.

Observations on the effect of some concentrations (0.1, 0.3, 0.6, 0.9, 1 and 2%) of three herbicide formulations on the germination of pretreated seeds are as shown in Fig. 6. Seeds in 0.1 and 0.3 - 2.0% Galex gave 25% and 2 to 4% germination respectively while those in 0.1 - 2% Primextra gave 0-10% germination (Fig. 6). The effect of the herbicides most especially Gramoxone, on 1-month old seedlings caused their rapid death within 4 days of application. Galex and Primextra at 0.5% concentration was only effective after 3 days of application as compared to the other whose effect was noticed within 24h of application (Table 1). Statistical differences among the three herbicides showed that 0.5 to 2.0% Gramoxone, 1 to 2% Galex and Primextra had high significant effect ($P \geq 0.05$) on the weed seedlings

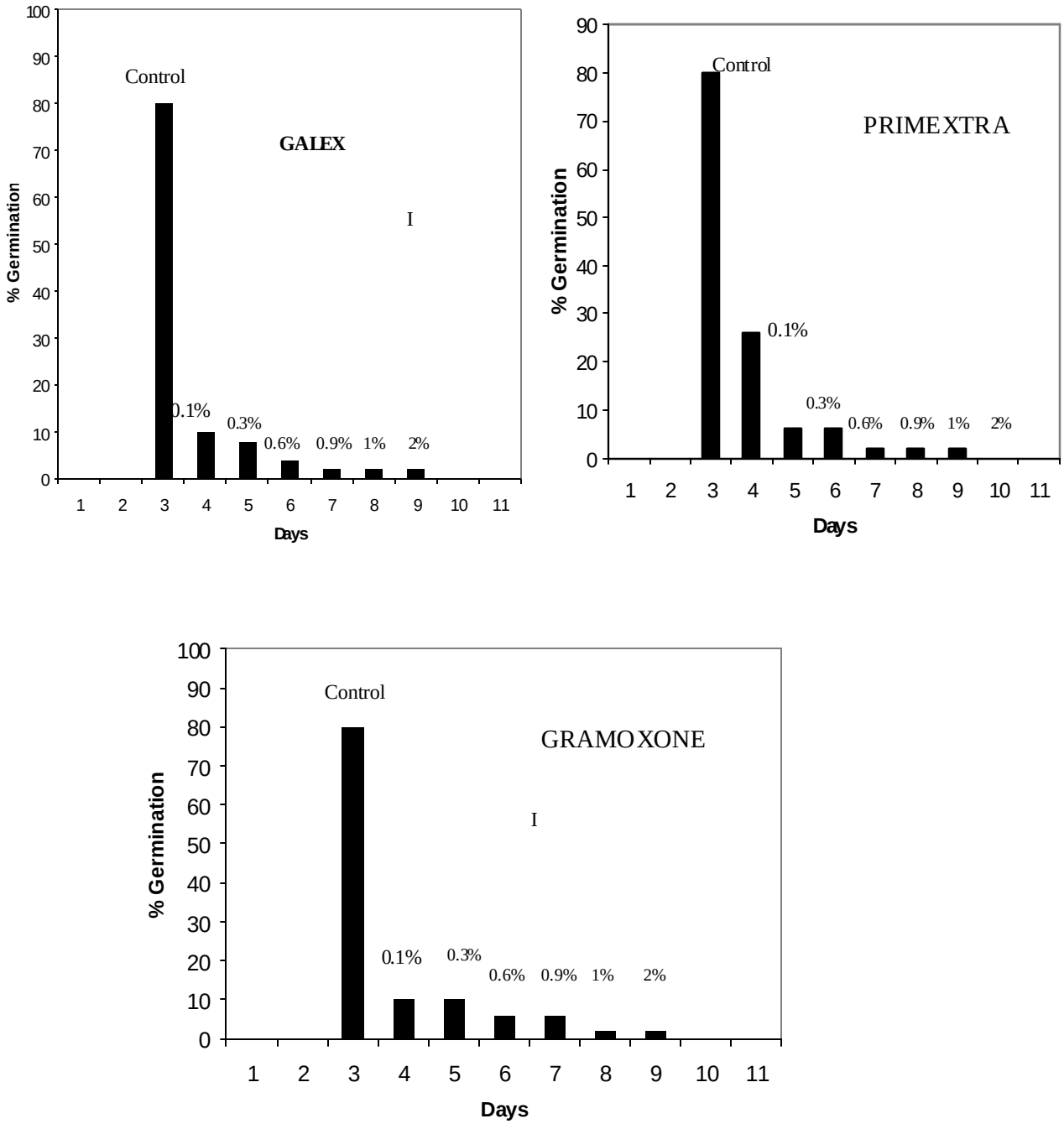

Fig 6: Effect of different concentrations of three herbicides on the germination of pretreated seeds of *Tithonia diversifolia* after four days of application.

Table 1. Effect of some herbicide concentrations on young seedlings of *T. diversifolia*. (Values presented are means of five replicates).

DAYS	GRAMOXONE			Herbicide concentrations (%). GALEX			PRIMEXTRA		
0	0.5	1.0	2.0	0.5	1.0	2.0	0.5	1.0	2.0
				% of seedlings alive					
1	*0.0	*0.0	*0.0	90.0 ± 4.2	*0.0	*0.0	100	*0.0	*0.0
2	*0.0	*0.0	*0.0	82.0±3.4	*0.0	*0.0	64.0±3.6	*0.0	*0.0
3	*0.0	*0.0	*0.0	60.5 ± 2.1	*0.0	*0.0	*0.0	*0.0	*0.0
4	*0.0	*0.0	*0.0	*0.0	*0.0	*0.0	*0.0	*0.0	*0.0

*Significant at ($P \geq 0.05$) LSD = 2.46

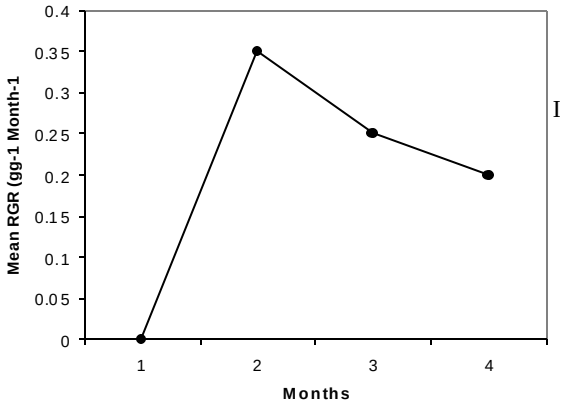

Fig. 7. The Relative Growth Rate (RGR) of 1-3 month-old seedlings of *Tithonia diversifolia* each point is the mean of five replicates.

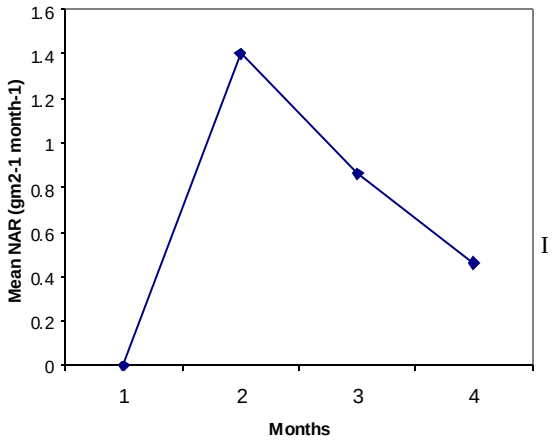

Fig 8: The Net Assimilation Rate (NAR) of 1 – 3 month-old seedlings of *Tithonia diversifolia*. Each point is the mean of five replicates

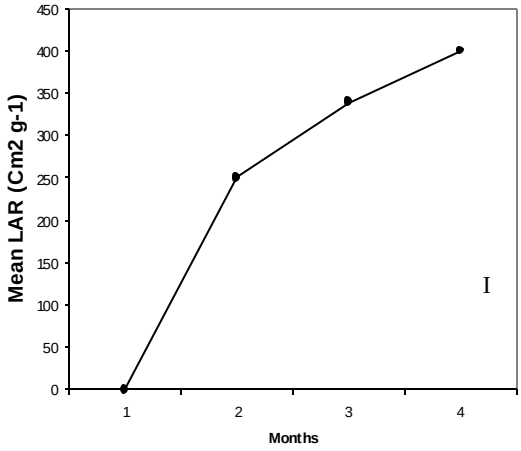

Fig. 9: The Leaf Area Ratio (LAR) of 1-3 month-old seedlings of *Tithonia diversifolia*. Each point is the mean of five replicates.

Discussion

The general survey of Nigeria for *T. diversifolia* infestation showed that almost one third of the country has been inhabited by this weed starting from the southwestern coast. These areas enjoy moderate to heavy annual rainfall with a bimodal peak in June and September. The survey was carried out during this wet period, hence the luxuriant growth observed in the infested areas.

Initial germination test showed a low germination percentage in seeds of *T. diversifolia*. The seeds exhibit dormancy and required pre-treatment for best germination result. Etejere and Ajibola, (1990) and Agboola, (1998) also observed such situation in seeds of *Rotboella cochinchinensis, Calapogonium mucunoides, Cassia hirsuta* and *Cassia obtusifolia,* some of which are serious weeds of wasteland corn, cowpea and sorghum in Nigeria. Seed pre-treatment such as hot water abolished dormancy in their seeds. The pre-treatments that terminated dormancy in seeds of *T. diversifolia* in this study (i.e combination of heat, light and storage) showed that the seeds suffer from physiological dormancy. This is also true of seeds of prickly sida, *Sida spinosa* (Baskin and Baskin, 1984) and centro, *Centrosema pubescens* (Omokanye and Onifade 1993). Seeds from many weed species are subject to dormancy of many kinds causing delayed germination of variable duration. This is a strategy for the survival of weedy species (Kolk 1979). Heat treatment in the case of *T. diversifolia* seeds had probably helped to cause some metabolic changes within the dormant seeds. The ability of the embryo to germinate appears only when seeds have undergone warm stratification (Nikolaeva 1980; Esenewo and Adebona 1990).

Light significantly enhanced the germination of pre-treated fresh seeds of *T. diversifolia*. Seeds usually germinate to a lightly higher percentage in light than in the dark (Li *et. al.* 1999). Photoblastic seeds will germinate better in the open than under the forest canopy (Olatoye 1965). In the open, solar radiation comes unhindered. Moreover associated with this increase, is the air and soil temperature. This best

explains the luxuriant growth of the seedlings of *T. diversifolia* in the open by the road sides especially along the highways at the onset of the rainy season (April - May) in Nigeria.

Percentage germination increased with increase in storage period in pre-treated seeds than in the non-treated ones. The seeds of *T. diversifolia* probably require some after-ripening period which allows for embryo maturation. According to Li *et al.*, (1999), maturation seems to be a pre-requisite for seeds to respond and germinate.

The mean LAR, NAR and RGR are high in the young seedlings of *T. diversifolia*. This shows that the weed species is a fast growing type. Smith and Anisu, (1997) observed a rapid early vegetative growth especially in plant height and leaf production of *T. diversifolia* between 6 - 12 weeks. The growth rate of plants have been found to be dependent on the effectiveness of the leaf area. This is not far-fetched as the leaf is the major assimilatory surface (Hunt 1978; Black 1972).

Gramoxone, Primextra and Galex at 0.5 to 2.0% and 0.3 to 2% concentration were effective in suppressing the germination and growth of seeds and seedlings respectively. Many herbicide formulations have been tried on some noxious weeds including *Chromolaena odoratum* (*Eupatorium odoratum*), and there have been appreciable success with low concentration of the herbicides (Etejere and Ajibola 1980). Herbicides have been known to distrupt the enzyme systems thus affecting the entire physiology (including respiration, chlorophyl formation and photosynthesis) of the living system (Akobundu 1987). A good approach to the problem of eradication of this noxious weed is to find useful application such as in Kenya where *T. diversifolia* is used as green manure and for medicinal purposes (Nyasim *et. al.* 1997). It is suggested that for eradication, the seedlings should be attacked at one month stage. By this time all viable seeds that must have been naturally heat-treated by sunlight and bush fire during the dry season would have germinated.

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