

## Detection of Germination inhibitors in fruits of *Terminalia laxiflora* Engl. & Diels using biochemical assays

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**ABSTRACT:** *Terminalia laxiflora* is multipurpose tree in Sudan. But it has low germination percentage, which may affect its natural regeneration. There were evidence that the fruits have an inhibitory effect, so this study was carried out to determine which part of the fruit that affect germination and seedlings growth. Three parts of the fruit extraction were examine (Coat, Pulp and Wing) with tow concentration to each part (200 fruit/litre, 100 fruit/litre).the results showed that all extractions had no effect on germination percentage except fruit coat extract (200 fruit/litre), which reduced it significantly. The different extractions had no effect on root length and seem to elongate the shoot expect fruit pulp (200 fruit/litre), which no different from control. All extractions had no effect on first leave appearance, but they causing abnormal seedlings. The result suggest new treatments that could be applied on the fruit of this species like de winging or de pulping the fruit before sewing or soaking in acid or hot water after de winging, de pulping and de coating. These results can be of great value for the medicinal plant scientists and users to try *T. laxiflora* fruit extractions for controlling bacteria and fungi activities and it may be an addition benefit to the medicinal uses of this tree.

**KEYWORDS:** *Terminalia laxiflora*, Germination, Inhibitors, fruit extracts.

### 1 INTRODUCTION

Seeds of many tree species germinate readily when subjected to favourable conditions of moisture, oxygen and temperature, many other species posses some degree of dormancy. Where dormancy is strong, some form of seed pre-treatment is essential for artificial regeneration, in order to obtain reasonably high germination rate in a short time [10]. Dormancy may be of several different types and sometimes more than one type occurs in the same seed [4]. There were numerous publications of phytotoxic molecules being produced by plants, a phenomenon generally termed allelopathy, It has been known for centuries that walnut trees poison the soil for underlying vegetation [9], [8], [5] Some substances that are produced naturally by plants were found to inhibit seed germination. These inhibitors don't reduce seed viability or cause any defects in seedling after germination. These inhibitors were found sometimes in the seed coats or fruit pulp or endosperm or embryo. The presence of these inhibitors in plants are common, and some of the natural plant inhibitors that were determined include Coumarin, Parascorbic acid, Ferulic acid, ABA, Cyanide – releasing, Ammonia – releasing, Phenolic compounds, Alkaloids, Organic acids. But ABA is the most wide spread natural inhibitors. [11], [6]. *Terminalia* spp is common indigenous species in wood land and semi humid Savannah of the Sudan. It is useful multipurpose species with a high

potential of timber production, medicinal uses etc... From previous studies at the National Tree Seed Centre [7]) the poor germination of seeds is an obstacle for plantation of the species. Poor germination was found to be partly due to the combined dormancy chemical. With regards to other problems that faces the individuals of this family such like agricultural expansion, climate fluctuation and missus of these species may put them of the endangered trees in Sudan. So this work aimed is determined of inhibitory effects of the different parts of the *Terminalia laxiflora* fruit on seed germination and seedlings growth.

## **2 MATERIALS AND METHODS**

Fruits were collected from Elnour forest at Blue Nile State (2006) for extractions the fruit were prepared as followed:

1. Dewinged: The fruits wings were removed, crushed and grinded.
2. Decoated: The fruits coats were removed, crushed and grinded.
3. Depulped: The pulps were extracted, crushed and grinded.

Every part was extracted with methanol in the laboratory of the Aromatic and Medicine plants institute. Every extraction was diluted with distilled water for tow extractions 0.001 and 0.002.

The concentration 100 was equivalent to extracting 100 fruits as followed:

$$\text{Weight of one extracted unit} = \frac{\text{Weight of extracted substance} \times \text{Weight of one crushed unit}}{50 \text{ gram of each grinded substance (wing, pulp)}}$$

And 200 equivalent to 200 fruits.

*Acacia mellifera* seeds were sown in sand in petri- dishes and were irrigated with prepared extractions versus a control irrigated with tap water. 25 seeds in each Petri dishes for four replicate to each treatment were used (three seed extractions with two concentrations).

The germination percentage and first leaf emergence were calculated every 3day for 15 days. The length of shoot and root were measured. The abnormal seedlings (seedlings without root or without shoot, discolouration) were also recorded.

## **3 RESULTS AND DISCUSSION**

The results in (table 1) and (figures 1, 2,3,4,5,6,7) showed that the germination percentage of *Acacia mellifera* seeds was not affected when watered with *T. laxiflora* extractions of different fruit parts, except when *Acacia mellifera* seeds were watered with seed coat extraction with concentration 200 fruit/litre, which reduced the germination percentage significantly compared with other extractions. These extractions had the tendency to inhibit seeds germination when the extractions concentrations were raised.

The length of shoot of *Acacia mellifera* was significantly affected with *T. laxiflora* extractions and unexpectedly it seems to elongate the shoot length compared with the control and 200 fruits pulp extraction. This result may be explained by the effect of extractions on root pattern (table 1). Such pattern was recorded [8] when *Glycine max* subjected to ABA treatment and water – deficit, water – water deficit always reduces protein synthesis. Reference [1] reported wheat cultivars root growth affected with some plant residues rather than shoot. May be the seedlings have a certain mechanism to escape the effect of inhibitors in the extracts by this elongation. The length of root of *Acacia mellifera* seedlings was significantly affected when it was irrigated with *T. laxiflora* fruits pulp extraction compared with other treatments including the control (table 1). *T. laxiflora* fruits pulp extractions reduce the root length of the seedlings which indicates that it had some substances which inhibit or delays root growth. This finding agrees with [2] who stated that aqueous extracts of *Solanum lycocarpus* significantly reduced root growth and inhibited root hair and lateral root. Reference [3] also reported an inhibitory effect of siam weed on growth of root and stem differentiation in Sesame seedlings.

*T. laxiflora* seeds extract significantly doesn't affect the first leaf appearing although there was a tendency to delay the first leaf appearing compared with the control (table 2). The results showed the inhibitory effect of different extractions on creating abnormal seedling especially the extraction of fruits wings (table 3).

Table 1. Effect of Terminalia laxiflora fruits extract on A. mellefera seeds germination and seedlings performance

Treatments	Mean of germination %	Mean of shoot length/ cm	Mean of root length/ cm
Control	45.6 a	3.1 b	2.3 a
Fruits pulp 200 fruit/litre	41.4 a	3.9 b	1.9 b
Fruits pulp 0.001	50.1 a	5.2 a	2.7 a
Fruits wing 200 fruit/litre	42.6 a	4.8 a	3.4 a
Fruits wing 0.001	47.3 a	4.7 a	2.5 a
Fruits coat 200 fruit/litre	35.8 b	5.4 a	2.5 a
Fruits coat 0.001	41.4 a	6.1 a	3.5 a
P ≥	0.04	0.006	0.02
SE ±	4.9	0.05	0.4
CV=	22	38	38

Table 2. Effect of Terminalia laxiflora fruits extracts on A. mellefera seedlings first leaf appearance

Treatments	Mean No of first leaf appearing after 3 days	Mean No of first leaf appearing after 6 days	Mean No of first leaf appearing after 9 days	Mean No of first leaf appearing after 12 days/No
Control	9.1 a	20.1 a	31.7 a	39.7 a
Fruits pulp 200 fruit/litre	10.2 a	15.1 a	20.5 a	29.9 a
Fruits pulp 100 fruit/litre	0 a	17.5 a	27.3 a	42.5 a
Fruits wing 200 fruit/litre	8.2 a	14.5 a	16.1 a	33.5 a
Fruits wing 100 fruit/litre	16.2 a	17.2 a	22.1 a	30.7 a
Fruits coat 200 fruit/litre	4.1 a	13 a	14.7 a	32.7 a
Fruits coat 100 fruit/litre	6.9 a	16.2 a	24.9 a	33 a
	p≤0.93	p≤0.45	p≤0.6	p≤0.8
	SE±2.5	SE±5.1	SE±6.9	SE±6.7
	CV=104	CV=128	CV=59	CV=36

Table 3. Effect of Terminalia laxiflora fruit extracts on seedling abnormality Acacia mellifera

Treatments	Mean/ No of seedling up normality	Rank
Fruits pulp 100 fruit/litre	6.8	ab
Fruits pulp 200 fruit/litre	8.1	ab
Fruits Wings 100 fruit/litre	16.5	a
Fruits Wings 200 fruit/litre	5.1	ab
Fruits Coats 100 fruit/litre	4.1	b
Fruits Coats 200 fruit/litre	0	b
Control	2.9	b

P≤ 0.002 SE± 4.2



**Fig. 1.** Effect of *T. laxiflora* fruit s coats extractions (200 fruit/litre concentration) on *A. mellifera* seedlings



**Fig. 2.** Effect of *T. laxiflora* fruit s pulps extractions (100 fruit/litre concentration) on *A. mellifera* seedlings



**Fig. 3.** Effect of *T. laxiflora* fruit pulps extractions (200 fruit/litre concentration) on *A. mellifera* seedlings



**Fig. 4.** Effect of *T. laxiflora* fruit s coats extractions (100 fruit/ litre concentration) on *A. mellifera* seedlings



**Fig. 5.** Effect of *T. laxiflora* fruit s wings extractions (100 fruit/ litre concentration) on *A. mellifera* seedlings



**Fig. 6.** Effect of *T. laxiflora* fruit s wings extractions (200 fruit/litre concentration) on *A. mellifera* seedlings



**Fig. 7.** *A. mellifera* seedlings watered with tap water (control)

#### 4 CONCLUSION AND RECOMMENDATIONS

It is clear from these results that the wings of *T. laxiflora* had chemical substances that affected the normal formation of the seedlings, and when concentration was raised, the number of abnormal seedlings increased (abnormality seedling appears as seedlings have shoot without root, the shoot was reddish). These substances may play an important role for this species in its natural habitat by eliminating the competition of other species that live in the same ecological place especially in the first stage of the seedlings development.

These results suggest new treatments that could be applied on the fruit of this species like de winging or de pulping the fruit before sewing or soaking in acid or hot water after de winging, de pulping and de coating. These results can be of great value for the medicinal plant scientists and users to try *T. laxiflora* fruit extractions for controlling bacteria and fungi activities and it may be an addition benefit to the medicinal uses of this tree.

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