

# STUDIES ON THE GERMINATION INHIBITORS IN THE FRUITS OF FOUR TROPICAL TREE SPECIES

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## ABSTRACT

Germination inhibitors in the aqueous extracts of the fruits of four tropical tree species were investigated. The germination inhibitors were characterised according to their Rf values in two solvents using the paper chromatography. Aqueous extracts of intact fruits of *Terminalia ivorensis*, *Terminalia superba*, *Tectona grandis* and *Gmelina arborea* contained germination inhibitors, which when applied to guinea corn grains inhibit their germination. Inhibitor with Rf values 0.1 – 0.2, 0.2 – 0.3, 0.5 – 0.7 and 0.8 – 0.9 were common in the four tree species. Inhibitors with Rf value of 0.4 were found in *T. ivorensis* and *T. superba*.

**KEY WORDS:** Germination inhibitors, Tropical trees, *Terminalia* species, *Tectona grandis*, *Gmelina arborea*.

## INTRODUCTION

*Terminalia ivorensis*, *Terminalia superba*, *Tectona grandis* and *Gmelina arborea* are four tropical tree species of economic importance. The *Terminalia* species belong to the family Combretaceae, a small family of trees, shrubs and climbers. *T. superba* is a distinctive tree which produces the well known timber locally called "Afara" (Yoruba). *T. ivorensis* produces the well known timber, "Idigbo". The two *Terminalia* species are high forest, especially of secondary regrowth (Keay et al., 1964).

*T. grandis* and *G. arborea*, belong to the family Verbanaceae. *T. grandis* produces durable heart-wood useful in construction works. *G. arborea* produces general utility timber for joinery, light construction and packaging. *T. grandis* has been found to be one of the best species for manufacture of match boxes and splints (Vickery and Vickery, 1979).

One common feature among these tree species is that the fruits' wall are heavily laden with germination-inhibiting substances which impose some degree of dormancy on the seeds contained in the fruits. This gives rise to poor germination performance in the nursery. These problems are alleviated by leaching and fermentation of the fruits in some cases (Agboola, 1991b). The presence of germination-inhibiting substances in the fruits of these species was corroborated by the methods used in breaking the dormancy of the seeds contained in

these fruits (Agboola, 1991b; Fasidi and Olofinboba, 1975). Moreover, on exposure, the mesocarp (pulp) of the fresh fruits especially those of *G. arborea* turned brown indicative of the presence of phenolic compound (Okoro, 1983). Phenolic compounds in general are found to be inhibitors of germination and because of their wide occurrence and distribution they have been regarded as natural germination inhibitors. Coumarin and its derivatives are also widely distributed in nature (Mayer and Roljakoff-Mayber, 1973).

Inhibitors of germination and growth have been isolated from pericarp, testa, endosperm and embryo (Fasidi et al., 1975; Khan, 1980). Many studies on the inhibitory effect of the contents of the pericarp and sap of fleshy fruits indicated that inhibitors might be involved in the control of germination. This study was aimed at detecting the germination-inhibiting substances in fruits of four tropical tree species chromatographically and characterising them on Rf basis. The knowledge from this investigation would add more to the understanding of the physiological basis of seed dormancy in tropical tree species.

## MATERIALS AND METHODS

### Extraction of Germination Inhibitors:

The aqueous inhibitor extracts from the fruit samples of the four tree species were prepared using modified method of Fasidi et al. (1979). Ten grams of

fruit sundried for 3 weeks were finely ground. In the case of *G. arborea* fresh drupes (from fruit-fall) with fruit-stones removed were used. Care was taken not to spill the fluid in the mesocarp as this part is known to contain growth-inhibiting substances (Okoro, 1983). The macerated fruits in the sterile water were placed on a shaker for 24 hours at room temperature ( $29 \pm 1^\circ\text{C}$ ) and the inhibitors were then extracted. The extracts were pooled, centrifuged and then evaporated to dryness in vacuo. The residue was dissolved in 4 ml. of sterile water for further analysis.

#### Chromatographic Separation:

One ml. of the concentrated inhibitor extracts from each of the 4 tree species was strip loaded on 10 x 30cm and 25 x 25cm No 1 whatman chromatography papers. The papers were developed for 13 hr by both methods of Fasidi *et. al.* (1979), Smith and Feinberg (1977). No locating reagent was used for developing the spots in the chromatogram as the separated constituents were coloured (Smith and Feinberg, 1977). The constituent inhibitors were identified on the basis of their Rf. Values after the solvent had moved up to 20cm from the origin. Graphical representations of the Rfs value and the solvent front distances of the inhibitors were made.

#### Effectiveness of aqueous inhibitor extracts:

Guinea corn grains were divided into lots. Fifty surface-sterilized grains were plated for germination in Petri dishes with filter paper and moistened with 5ml. of aqueous inhibitor extract. This was done for the extracts from the four tree species, distilled water served as the control. Mean percentage germination from the five replicates was determined.

### RESULT

The inhibitors in the aqueous extracts from the fruits of four tropical tree species based on the Rf values from paper chromatography in two different types of solvent are as shown in Figures 1A-D and 2A-D. It was observed that inhibitors with Rf. values of 0.1-0.2, 0.2-0.3, 0.5-0.7 and 0.8-0.9 were common to all the four tree species investigated in the two solvents (n-Butanol, Glacial-Acetic Acid - (60:15:25 v/v) and Isopropanol:

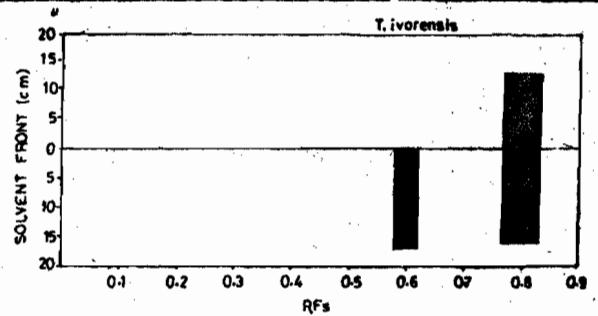


Figure 1A: Germination inhibitors in the fruits of *T. ivorensis* through Rf values in ascending and descending order in Butanol-Glacial acetic acid-water (60:15:15v/v).

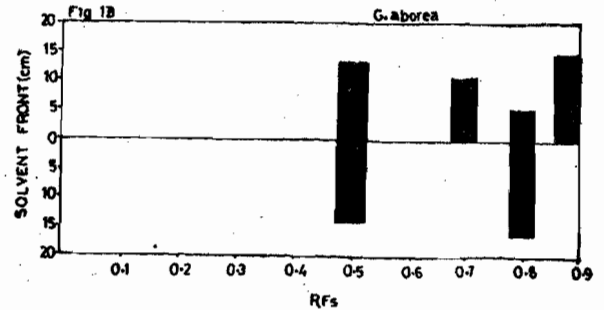


Figure 1B: Germination inhibitors in the fruits of *G. arborea* through Rf values in ascending and descending order in Butanol-Glacial acetic acid-water (60:12:15v/v)

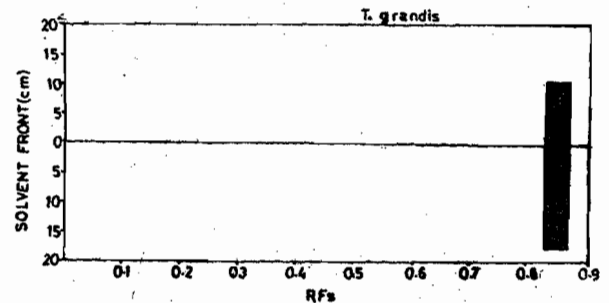


Figure 1C: Germination inhibitors in the fruits of *T. grandis* through Rf values in ascending and descending order in Butanol-Glacial acetic-water (60:15:15v/v)

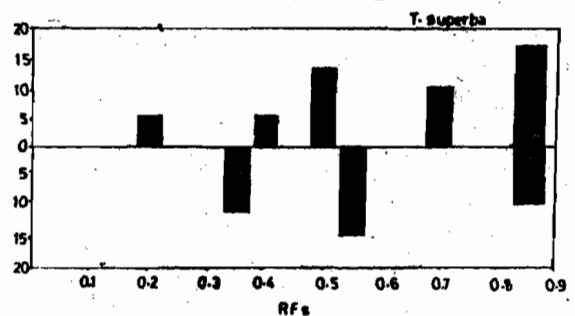


Figure 1D: Germination inhibitors in the fruits of *T. superba* through Rf values in ascending and descending order in Butanol-Glacial acetic acid-water (60:15:15v/v)

Ammonium hydroxide water - (10:1:1 v/v) both in the ascending and descending chromatography techniques (Figs. 1A-D and 2A-D). Inhibitors with 0.0 Rf were

only found in Isopropanol -  $\text{NH}_4\text{OH}$  -  $\text{H}_2\text{O}$  (10:1:1 v/v) in ascending chromatographic technique (Figs 2A-D). It was also observed that some inhibitors were specific to a particular species. For example, inhibitors with Rf values 0.4 were found in the two *Terminalia* species in the two solvents used (Figs. 1D and 2C).

Aqueous extracts from the fruits of all the four tree species significantly reduced germination of guinea corn grains. It was observed, that percentage germination ranged between 70 and 90% in water and 12-42% in inhibitor extracts.

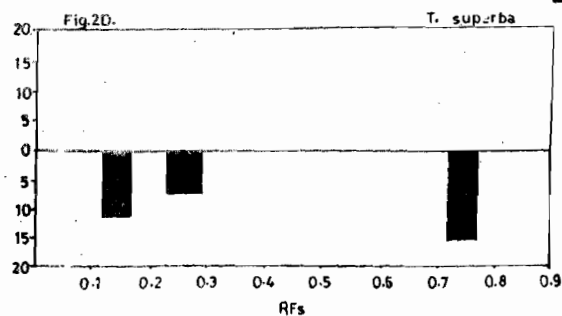


Figure 2A: Germination inhibitors in the fruits of *G. arborea* through Rf values in ascending and descending order in Isopropanol -  $\text{NH}_4\text{OH}$ - $\text{H}_2\text{O}$  (10:1:1 v/v)

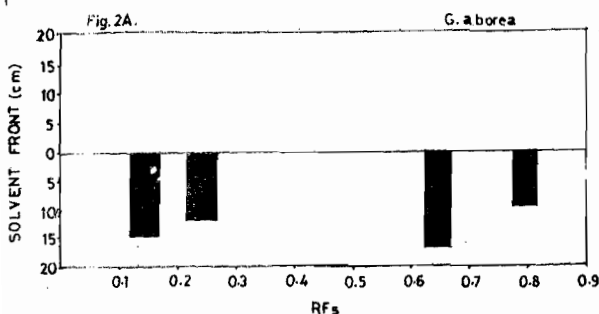


Figure 2B: Germination inhibitors in the fruit of *T. grandis* through Rf values in ascending and descending order in Isopropanol -  $\text{NH}_4\text{OH}$ - $\text{H}_2\text{O}$  (10:1:1 v/v)

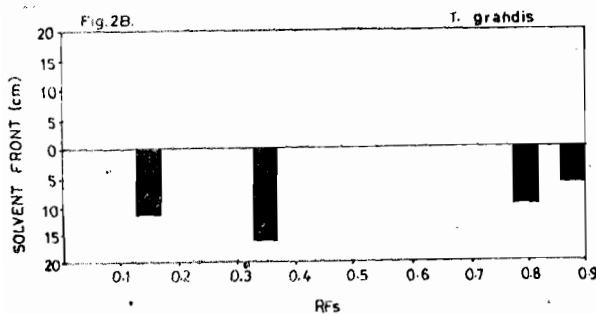


Figure 2C: Germination inhibitors in the fruits of *T. ivorensis* through Rf values in ascending and descending order in Isopropanol -  $\text{NH}_4\text{OH}$ - $\text{H}_2\text{O}$  (10:1:1 v/v)

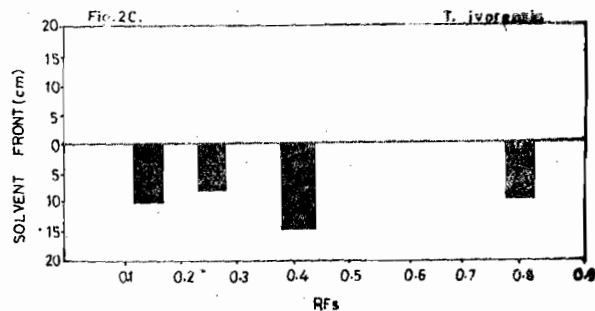


Figure 2D: Germination inhibitors in the fruit of *T. superba* through Rf values in ascending and descending order in Isopropanol -  $\text{NH}_4\text{OH}$ - $\text{H}_2\text{O}$  (10:1:1 v/v)

## DISCUSSION

Aqueous extracts from fruits of *T. ivorensis*, *T. superba*, *T. grandis* and *G. arborea* reduced germination capacity of guinea corn grains. The aqueous extracts also contained some inhibitors which were associated with definite Rf values. In a related study Fairlamb and Davidson (1976) demonstrated that chemical inhibitors may also be involved in the poor germination of seeds of *T. ivorensis* and *T. grandis*. Brookman-Amisah (1976) using ether ethyl acetate extracts of fruits of *T. ivorensis* found that the inhibitory effects may be due partially to combined effects of 3, 4-dihydroxybenzoic acid, coumarin, ascorbic acid and salicylic acid. These findings also compared closely with those of Fasidi and Olofinboba (1975) and Fasidi et al. (1979). These same workers found that unwashed seeds and fruits of *Chlorophora excelsa* (*Milicia excelsa*) contained considerable level of inhibitor which showed intense activity at definite Rf values. Similarly seeds of *Rosa* species have been found to be notoriously dormant and a powerful inhibitor is known to be present in the epicarp (Black, 1972). The action of this tissue is so potent that even isolated embryos of lettuce seeds in the vicinity of pieces of the *Rosa* epicarp fail to grow.

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