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Studies on the Effects of *Brachystegia eurycoma* (Harms) on Termites and Fungi

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Studies were conducted on the proximate, mineral and phytochemical constituent of *Brachystegia eurycoma* (Harms) wood extracts. The termicidal and antifungal properties of aqueous and ethanol extract were determined. The survival of *Amitermes evuncifer* Silvestri on wood shavings of *B. eurycoma* for seven days was also investigated. The growth of *Fomes heterobasidium, Polyporus coridus* and *Daedalea daedaleopsis* was inhibited by the extracts. The test for the termicidal effect of *B.eurycoma* revealed that ethanol extract was more active. Survival rate of termites on *B.eurycoma* was 0% after the end of a seven day no-choice food experiment, while the consumption rate of the wood was 0.3%. Phytochemical studies of the wood sample revealed the presence of oxalate (12.83 ± 0.08mg/100g), hydrocyanic acid (22.62± ± 0.08 mg / 100g), tannin (8.10 ±0.89mg/100g) and phytic acid (3.01± 0.00mg/100g). Magnesium had the highest value of 875.0± 0.01 ppm while lead had the lowest value of 1.67± 0.01 ppm. Cobalt was not detected.

Key words: *Brachystegia eurycoma*, fungi, termite, survival.

All sorts of wood from standing timber, logs of felled timber, stored and seasoned timber and the wood finally installed in the house are subjected to the attack of many pathogens that cause incalculable damage every year. The natural ability of wood to resist the attack of decaying organism such as bacteria, fungi and insects is known as durability (Nakayama *et al*., 2000; Peralta *et al*., 2004). Plants produce secondary metabolites acting as defense mechanism that can be used in pest control (Coelho *et al*., 2006). The presence of potentially toxic compounds particularly common in the heartwood like tannin and various other phenolic compounds like terpenes, stilbenes, flavonoids, tropolones, glycosides, sugars and fatty acids confer durability on woods (Nakayama *et al*., 2000; Chen *et al*., 2004; Neya *et al*., 2004; Peralta *et al*., 2004; Taylor *et al*., 2006). These insecticidal substances can affect the feeding behavior and growth regulators, disrupting the endocrinology balance of the insects (Coelho *et al., 2006*). The development of new termicides from plant origin can be an alternative for the control of termites. Natural resistive wood and wood extractives have great promise for prevention of termite attacks. Environmental concern about the use of persistent chlorinated hydrocarbons as termicides has resulted in studies on the development of products that are not hazardous to the environment from wood origin (Abdolahi *et al*., 2004; Nakata *et al*., 2005).

Brachystegia eurycoma belongs to the family Leguminosae and subfamily Fabaceae. It grows along the stream banks in the high forest

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zone of tropical Africa. In Nigeria, it is found widely distributed along Ogun river bank, Ibadan–Ife road and Imo river bank (Omotayo, 1999; Amah *et al*., 2001). In a preliminary study, wood-eating termites *Amitermes evuncifer* survived poorly when exposed to *B. eurycoma* wood chips. Previous workers (Nagnan and Clement, 1990; Roger *et al*., 2006) had reported that fungal decay process makes wood more susceptible to termite infestation. While durability and susceptibility of wood to termite infestation has been attributed to the presence of chemical extractives. This research was initiated to study the extractive components of the wood and the effect of the aqueous and ethanol extracts on the survival of a wood-eating termite, *Amitermis evuncifer* and some wood decay fungi. We have also studied the survival of the termites on the *Brachystegia eurycoma.*

MATERIAL AND METHODS

The *Brachystegia eurycoma* wood chips and bark were collected from Bashiri Sawmill in Ado-Ekiti, Nigeria. Termite samples were collected from decayed planks on the campus of University of Ado-Ekiti, Nigeria. The study was conducted during the rainy season (April to November).

Proximate analysis

This was carried out on the wood sample for ash, moisture, crude fiber and fat using the method described by Association of Official Analytical Chemists (AOAC, 1990). Nitrogen was determined by the micro-Kjeldahl method reported by Pearson (1976). The percentage nitrogen was converted to crude protein by multiplying with 6.25. The carbohydrate was calculated by difference.

Mineral analysis

Fe, Cu, Zn, Mg, Pb, Mn, Ca, Co, Na and K were analyzed by the AOAC (1990) method. One gram of the wood sample was dry-ashed at 550°C in a murfle furnace. The ash obtained was dissolved in 10% hydrochloric acid (HCl), filtered and made up to standard volume with deionised water. The minerals were determined by atomic absorption spectrophotometer using a Buck model 200-A. The phytochemical contents were determined by the method of the Association of Official Analytical Chemist (1990) for hydrocyanic acid, oxalate and saponin; phytic acid (Maga, 1980)

Preparation of extract

Eighty grams each of grounded wood samples of *B. eurycoma* was suspended in 250ml of ethanol and deionised water respectively in a conical flask for 48h. The aqueous suspension was filtered and the filtrate allowed evaporating to dryness at room temperature (27±2°C).

Test for antifungal activity

The test fungi *Fomes heterobasidium, Polyporus coridis* and *Daedalea daedaleopsis* were isolated from decayed wood and grown on Malt Extract Agar (MEA).Using the agar punch-hole method (Stokes,1975), the antifungal activity of the different extract concentration was determined in MEA. Plates were incubated at 27± 2°C for 5days.

Test for antitermic activity

Externally undifferentiated termites (workers) beyond the third instars were selected from colonies of *Amitermes evuncifer* on a decayed wood. Extract of *B. eurycoma* were applied to absorbent paper pads and allowed to dry. One milliliter aliquots of different concentration of test material was applied to each pad and placed in the bottom of a disposable Petri plate. Twenty termites were then placed on the pad and the conditions of the termites were monitored for 6h.

Termite survival test

Worker termites were maintained on 10g chips of *Brachystegia eurycoma* placed on a sand substrate in a plastic container (12 cm ×11 cm). The container was filled with 500g of sand and moistened with 50 ml of water to maintain a relative humidity near saturation (Carter *et al*., 1978, Duryea *et al*., 1999). Forty termites were added to each container and were maintained at 27 ± 2°C while their conditions were checked daily for 2 weeks.

RESULTS

The proximate composition of *B. eurycoma* wood is shown in Table 1. The carbohydrate content was high (46.45±0.0g/100g), while the protein content (2.57± 0.9g/100g) and the nitrogen content (0.379±0.0g/100g) were relatively low. The crude fat and fiber content were 8.16± 0.9g/100g and 16.40± 0.1g/100g respectively. The mineral element composition in Table 2 showed that magnesium, potassium, zinc and manganese were the most abundant elements present in *B. eurycoma* with values recorded were 875.0±0.01, 666.6± 0.01, 609.2±0.01 and 562.3± 0.01(ppm) respectively. Lead was the least abundant element (1.67±0.01), while cobalt was not detected in *B. eurycoma*. Table 3 presents the phytochemical constituents of *B. eurycoma* wood. While it is rich in hydrocyanic acid (22.62± 0.90mg/100g), oxalate (12.83± 0.08mg/100g) and tannin (8.10± 0.89mg/100g), saponin was not present. However the value of phytic acid (3.01± 0.00) was low. The result on the effect of *B. eurycoma* aqueous and ethanol extract on the growth of some wood decay fungi is shown in Table 4. Both the aqueous and ethanol extract of *B. eurycoma* inhibited the growth of the test fungi.

The result of the survival test of termites exposed to absorbent paper pads treated with 1ml aliquots of either aqueous or ethanol extracts are shown in Fig. 1 and 2 respectively. Termites survived well on paper pads treated with the aqueous extract (Fig. 1). However, all the termites died in less than 96min when exposed to paper pads treated with 0.4g/ml of the ethanol extract (Fig. 2).

Table 2. Mineral composition of *Brachystegia eurycoma*

Minerals	Level (mg/100g)
Sodium	384± 0.01
Potassium	666.6± 0.01
Copper	124.6± 0.01
Zinc	609.2± 0.01
Iron	8.3± 0.02
Manganese	562.3± 0.01
Magnesium	875.0± 0.01
Calcium	122.0± 0.02
Lead	1.67± 0.0
Cobalt	Not detected

Mean± SD of triplicate determinations

Table 1. Proximate composition of *Brachystegia eurycoma* wood

Components	Percentage
Moisture	15.14±0.4
Protein	37 ±0.9
Crude fat	8.16±0.9
Crude protein	16.40±0.1
Ash content	1.48±0.0
Carbohydrate (by difference)	46.45±0.0
Nitrogen	0.38±0.0

Mean± SD of triplicate determinations

Table 3. Phytochemical constituent of *Brachystegia eurycoma*

Component	Level (mg/ml)
Oxalate	12.83± 0.08
Tannin	8.10± 0.89
Hydrocyanic acid	22.62± 0.09
Phytic acid	3.01± 0.00
Crude saponin	Not detected

Mean± SD of triplicate determinations.

Table 4. Effect of aqueous and ethanol extract of *B. eurycoma* on the growth of some wood decay fungi

Extract concentration extract	Diameter of zone of inhibition in mm(mg/ml) Aqueous (Ethanol)*		
	F. heterobasidium	*P. coridis*	*D. daedaleopsis*
0.1	0.2(0.3)	0.2(0.0)	0.3(0.4)
0.2	3.0(3.0)	2.5(2.0)	2.0(2.0)
0.3	3.5(3.5)	3.0(2.5)	3.0(4.0)
0.4	5.5(5.0)	5.0(5.0)	7.0(8.0)

*Diameter of zone of inhibition in ethanol extract in parentheses

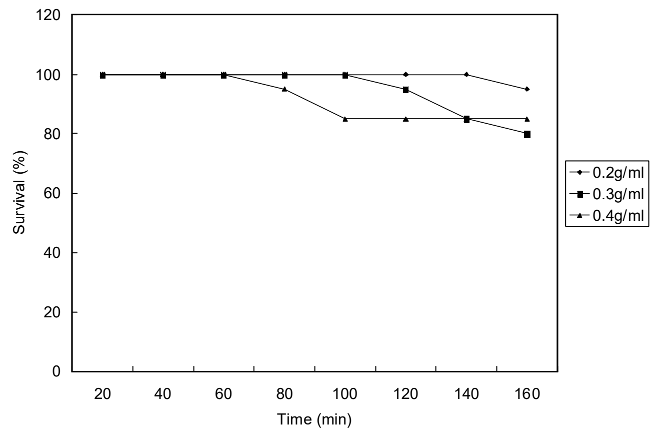


Fig. 1. Survival of *A. evuncifer* on paper pads containing aqueous extract of *B. eurycoma*

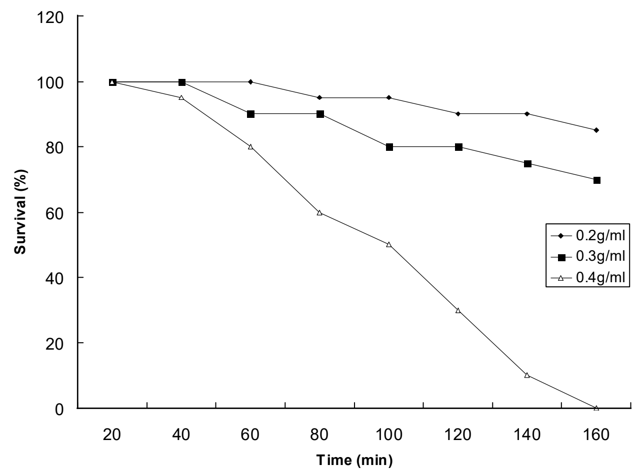
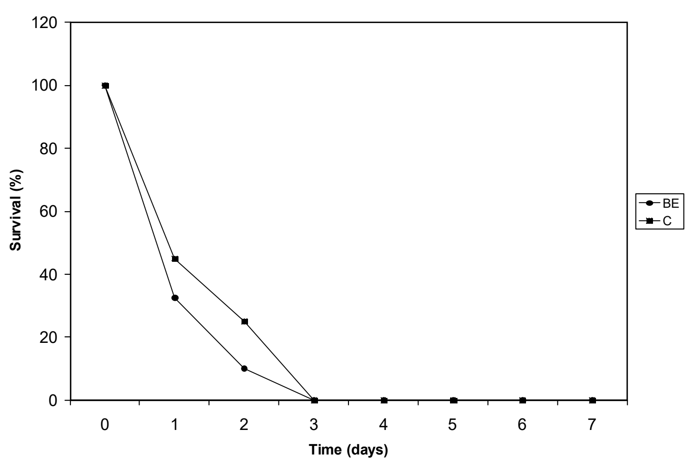


Fig. 2. Survival of *A. evuncifer* on paper pads containing ethanol extract of *B. eurycoma*



BE = *Brachystegia eurycoma* C = No wood feeding

Fig. 3. Survival of *A.evuncifer* on *B. eurycoma* shaving during aseven day no-choice food experiment

The survival of *Amitermis evuncifer* on *Brachystegia eurycoma* was 0% at the end of the seven days no-choice food experiment, while termites without food also died within 48 h (Fig. 3). Termites consumed about 0.3% of the wood.

DISCUSSION

The study showed that the aqueous and ethanol extract of *Brachystegia eurycoma* has inhibitory effect on the growth of wood decay fungi like *Polyporus coridis*, *Daedalea daedaleopsis* and *Fomes heterobasidium*. Adekunle (2000) had earlier reported the antifungal activity of the plant against *Aspergillus flavus, A. fumigatus, A. niger, Candida albicans* and *Mucor mucedo*. Taylor *et al*. (2006) had also reported the inhibitory effect of *Thuga plicata* and *Chamaecyparis nootkatensis* on termites and fungi. The ethanol extract appeared to be more effective on termites than the aqueous extract. The rate of death was faster and it could be that the antitermic components are extracted maximally with ethanol.

The non-survival of termites on wood chips and extractives might be due to the repellent or toxic activity of *B. eurycoma*. While termites consumed about 0.3% of *B. eurycoma,* termites without food starved and also died within 48 h. Death of termites fed with *B. eurycoma* may also be due to starvation Crude water extract of *Larix leptolepsis* wood had also been reported to show strong feeding deterrent activities against the subterranean termites *Coptotermes formosanus* (Chen *et al*., 2004). The Hardwood has been reported to be repellent to termites (Moore, 1979; Nagnan and Clement, 1990). It has been suggested that the inhibition might be due to some intrinsic factors present in the wood. According to Illston *et al*. (1981), extractives in form of hormones, resins and fatty acids account for 3± 2% of soft wood and 5± 3% of hardwood. These naturally occurring chemical components confer durability on woods (Peralta *et al*., 2004; Neya *et al*., 2004). Many workers have attributed the presence of these extractives to the protection of a variety of wood against termite attack, thereby contributing immensely to the economic importance of woods. (Rigol *et al*., 2004; Gonzalez-villa, 2006). The inhibition of the growth of wood decay fungi; *P. coridis*, *D. daedaleopsis* and *Fomes heterobasidium* by the aqueous and ethanol extract of *B. eurycoma* may have a relationship with the inability of the termites to survive on paper pads treated with extracts of *B. eurycoma*. It has been reported that termites are attracted to the odors of wood decaying fungi (Nagnan and Clement, 1990; Roger *et al*., 2006). There may be a relationship between susceptibility of a wood to fungal and termite attack, because wood infestation by termites often follows fungal attack. According to Roger *et al*. (2006), fungal decay process makes the wood easier for termite to penetrate. In addition to this, fungi are known to provide a source of nitrogen in the termite diet.

CONCLUSION

Both the aqueous and ethanol extracts inhibited the growth of wood decay fungi, while the ethanol extract of *B. eurycoma* was more active on *Amitermes evuncifer*. The survival of termites on *B. eurycoma* was 0% (within 48 h). This study has confirmed that *B. eurycoma* promises to be a good source of extractive that could be used in the alternative control of wood decay fungi and termites.

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