Short Communication

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CYTOTOXIC ACTIVITY OF SELECTED NIGERIAN PLANTS

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Abstract

Cancer is one of the most prominent human diseases which has stimulated scientific and commercial interest in the discovery of new anticancer agents from natural sources. The current study investigates the cytotoxic activity of ethanolic extracts of sixteen Nigerian plants used locally for the treatment of cancer using the MTT assay on the HeLa cell line. *Sapium ellipticum* leaves showed activity comparable to the reference compound Cisplatin and greater cytotoxic activity than *Combretum paniculatum*, *Celosia trigyna, Drymaria cordata, Cyathula achyranthoides* and *Cyathula prostata*. *Justica extensa, Pupalia lappacea, Hedranthera barteri* leaves, *Alternanthera sessilis, Ethulia conyzoides* leaves, *Combretum zenkeri* root, *Sapium ellipticum* stembark and *Lannea nigritana* stembark showed very low activity while *Combretum molle, Adenanthera parvoniana* and *Lannea acida* showed no activity. The results justify the use of *Sapium*, *Combretum*, *Celosia*, *Drymaria* and *Cyathula* in traditional treatment of cancer.

Keywords: Medicinal plants; Cytotoxicity; Cancer; *Sapium*; *Combretum*

Introduction

Plants have formed the basis for the treatment of diseases in traditional medicine systems for many years, and continue to play a major role in the primary health care of about 80% of the world's inhabitants (Farnsworth et al., 1985; Sofowora, 1984; Koduru et al, 2007a). Research interest has focused on various plants that possess anticancer properties and this has led to the discovery and development of efficacious anticancer agents such as vinblastine and vincristine from *Catharanthus roseus,* and taxol from *Taxus brevifolia*. (Noble, 1990; Wani et al, 1971). Although the use of ethnomedicines is widespread in Africa, many of these plants are yet to be investigated for their anticancer activity.

This paper reports the cytotoxic activity of the ethanolic extracts of sixteen plants against HeLa cervix adenocarcinoma cells. *Sapium ellipticum, Combretum paniculatum, Celosia trigyna, Pupalia lappacea, Justica extensa, Hedranthera barteri, Alternanthera sessilis, Ethulia conyzoides, Lannea nigritana, L. acida, Combretum zenkeri, C. molle, Adenanthera parvoniana, Cyathula achyranthoides, Drymaria cordata* and *Cyathula prostata* were selected based on their frequency in recipes for the management of cancer from an ethnobotanical survey of traditional medical practitioners in Western Nigeria.

Materials and Methods

Plant material

All the tested plants collected from the Olokemeji Forest Reserve and from the Campus of Obafemi Awolowo University, Ile-Ife in Nigeria in July 2006 were authenticated by comparison with corresponding herbarium specimens at the Forestry Research Institute, Ibadan, Nigeria (FRIN) where voucher specimens were

also deposited. The plants were air dried for two days followed by drying in a hot air oven at 40°C, ground to powder and stored in amber coloured bottles. 100 g each of powdered plant material was macerated with 80% ethanol at room temperature respectively. Extracts were filtered and concentrated to dryness *in vacuo* at room temperature. The respective plant parts used are as stated in Table 1.

Cytotoxicity assay

The cytotoxic effect of plant extracts on HeLa (cervix adenocarcinoma) cell line was determined using a modification (Koduru et al., 2007b) of the MTT assay (Mossman, 1983). Briefly, cells were seeded into 96-well culture plates (Nunc) at 6 000 cells/well in RPMI1640:10% fetal bovine serum (FBS) and left for 24 hours. Plant extracts or cisplatin (positive control) were added and the cells incubated for a further 48 hrs after which the medium was replaced with 200 µl MTT (Sigma) (0.5 mg/ml in RPMI 1640:10% FBS). After a further 4 hr incubation at 37°C, the MTT was removed and the purple formazan product dissolved in DMSO and absorbance measured at 540 nm on a multiwell scanning spectrophotometer (Multiscan MS, Labsystems). All incubation steps were carried out in a 37°C humidified incubator with 5% CO_2.

Results and Discussion

The cytotoxicity results are as shown in Figure 1 with extract numbers corresponding to those in Table 1. Cisplatin at 10 and 100 µM caused 49.25 ± 3.33% and 88.19 ± 0.60% (SEM, n=4) inhibition, respectively. In consideration of the cytotoxicity, the extracts could be classified into four categories. Firstly, potentially cytotoxic are *Sapium ellipticum* leaves, *Combretum paniculatum* leaves, *Celosia trigyna*, *Drymaria cordata* and *Cyathula prostata* showing over 50% activity at 500 µg/ml. Secondly, moderate cytotoxic are *Ethulia conyzoides* leaves, *Hedranthera barteri* leaves and *Cyathula achyranthoides* showing between 40-50% activity at 500 µg/ml. Thirdly, low cytotoxic activities are *Pupalia lappacea*, *Justica extensa*, *Sapium ellipticum* stembark, *Alternanthera sessilis*, *Lannea nigritana* stem-bark and *Combretum zenkeri* root showing less than 40% activity at 500 µg/ml. Lastly, non-toxic are *Combretum molle, Adenanthera parvoniana* fruit and *Lannea acida* stem bark with no activity against the HeLa cell line.

The plants in the first three categories justify their inclusion in traditional recipes for the treatment of cancer. The crude extracts in the first two categories could be taken for further bioassay guided experiments. However, *Sapium* leaves possessed the highest activity indicating its potential for biopharmaceutical use.

Table 1: List of plants used in the cytotoxicity test against HeLa cells.

Extract number	Name of Plant	Plant part used
1a	*Sapium ellipticum* (Krauss.) Pax. (Euphorbiaceae)	Leaves
1b	*Sapium ellipticum* (Krauss.) Pax. (Euphorbiaceae)	Stembark
2	*Combretum paniculatum* Vent. (Combretaceae)	Leaves
3	*Celosia trigyna* L. (Amaranthaceae)	Whole plant
4	*Pupalia lappacea* (L.) A. Juss (Amaranthaceae)	Whole plant
5	*Justica extensa* T. Anders (Acanthaceae)	Whole plant
6	*Hedranthera barteri* (Hook. f.) Pichon (Apocynaceae)	Leaves
7	*Alternanthera sessilis* L. DC. (Amaranthaceae)	Whole plant
8	*Ethulia conyzoides* Linn. F. (Asteraceae)	Leaves
9	*Lannea nigritana* (Scott Elliot) Keay SB. (Anacardiaceae)	Stembark
10	*Combretum zenkeri* Engl. & Diels (Combretaceae)	Root
11	*Combretum molle* R.Br (Combretaceae)	Leaves
12	*Adenanthera parvoniana* L.(Mimosaceae)	Fruits
13	*Lannea acida* A. Rich (Anacardiaceae)	Stembark
14	*Cyathula achyranthoides* (Kunth.) Moq. (Amaranthaceae)	Whole plant
15	*Drymaria cordata* (Linn.) Willd. (Caryophyllaceae)	Whole plant
16	*Cyathula prostata* (Linn.) Blume (Amaranthaceae)	Whole plant

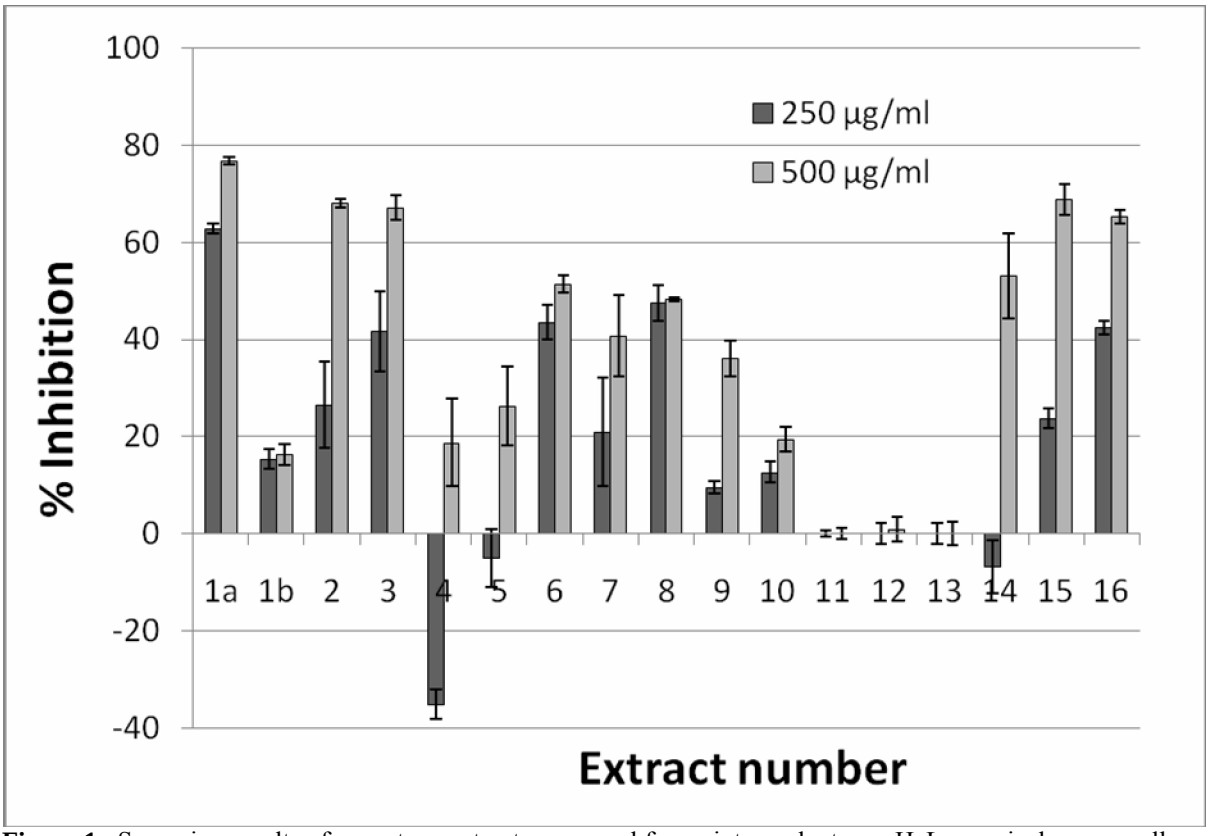

Figure 1. Screening results of seventeen extracts, prepared from sixteen plants, on HeLa cervical cancer cells. Results represent the mean ± standard error of the mean of quadruplicate determinations. Cisplatin as positive control at 10 and 100 µM caused 49.25 ± 3.33% and 88.19 ± 0.60% (SEM, n=4) inhibition, respectively.

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