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Scientific Investigation of Antitrypanosomal Activity of *Crateva Adansonii* DC Leaves Extracts

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ABSTRACT: *Crateva adansonii* DC is used in traditional medicines in the West of Africa. The crude hexane (**CAN-1**) and ethyl acetate (**CAN-2**) extracts were evaluated for their *in vitro* bioactivity against African trypanosome *Trypanosoma brucei brucei* (S427) blood stream forms. The crude extracts showed moderate anti-trypanosomal activity (MIC 12.5µg/ml). We recommend its use either alone or in combination with other natural/semi-synthetic/synthetic drugs for the treatment of Human African Trypanosomiasis.

KEYWORDS: *Crateva adansonii* DC; Anti-trypanosomal activity; *T. brucei*; Medicinal Plants; Tsetse Fly; Sleeping Sickness; Human African Trypanosomiasis.

INTRODUCTION

Herbal products are gaining progressively attention due to less toxicity and high efficacy against free radical mediated diseases. At present, approximately 25% of drugs in modern pharmacopoeia were derived from plants (phytomedicines) and many others were synthetic analogues built on the prototype compounds isolated from plants [1-4].

Crateva adansonii DC, also known as *Crateva religiosa* or sacred garlic pear, belongs to family Capparaceae is in high demand, especially its leaves for the treatment of ear infections. The bark is widely used for stomach troubles and held to have tonic properties. In Senegal the roots figure in several treatments for syphyilis, jaundice and yellow fevers[5]. Ayodeji and coworkers claimed the antimicrobial properties of its leaves[6]. Organic extract(dichloromethane & methanol, 1:1) of *C. adanosonii* DC seeds had been evaluated for their bioactivity against brine shrimp found to have very high activity. Two phytoconstituents had also been isolated and identified as oleanolic acid(**Figure 1**) and 4-epi-hederagenin(**Figure 1**). When done the comparative analysis, oleanolic acid have LC_{50} 2.51 µg/ml as compared to the standard cycloheximide(LC_{50} is 40 µg/ml)[7].

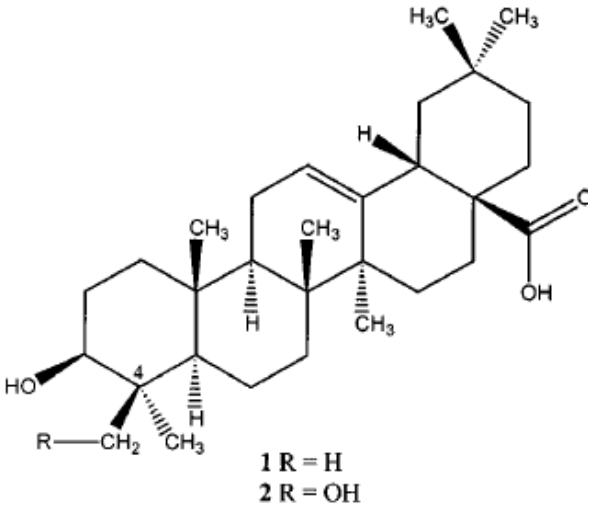

Figure 1 Structure of Oleanolic Acid(1) & 4-epi-hederagenin(2)

Unlike other communicable diseases that receive a high level of attention from health systems, a group of parasitic and infectious diseases has been characterized by historically low investment by the pharmaceutical industry[8,9]. Human African trypanosomiasis, also known as sleeping sickness, is a vector borne disease where the vector is tsetse fly. Sleeping sickness threatens millions of people in 36 countries in sub-Saharan Africa[10]. Till date, there is no information related to the anti-trypanosomal activity of this handsome tree, *C. adansonii* DC.

The aim of the present study was to investigate the anti-trypanosomal activity of *Crateva adansonii* in order to evaluate its medicinal value and to point to an easily accessible source of natural anti-trypanosomals that could be used as possible replacement of existing allopathic medicines.

MATERIALS & METHODS

Collection of Material

Fresh leaves of *C. adansonii* were harvested from Nsukka, Enugu State of Nigeria in August 2011, sun-dried and ground to powder. The plant was authenticated by the Department of Botany of the University of Nigeria, Nsukka where a voucher specimen No 1152 was deposited in the University herbarium.

Preparation of Samples

Finely grounded leaves (501.08g) were extracted successively with hexane and ethyl acetate in a soxhlet extractor to obtain 11.83g(**CAN-1**) and 14.20g(**CAN-2**) of crude extracts respectively.

Antitrypanosmal Activity

A modification of the microplate Alamar blue assay to determine drug sensitivity of African trypanosomes *in vitro* according to Raj & coworkers, 1997 was used[11]. Stock solutions of plant extracts were prepared as 10mg/ml in DMSO and serially diluted in HMI-9 medium, the negative control for MIC determination. 10µM Suramin was the positive control.

Trypanosomes blood forms used were *Trypanosoma brucei brucei* S427 and the concentrations of trypanosomes were 2 - 3 x10 ^{4} trypanosomes/ml. Incubation was at 37ºC, 5% CO_2 with a humidified atmosphere for 48 hours for three replicates made. Alamar blue was used and fluorescence determined on a Wallac Victor microplate reader in fluorescence mode (Excitation 530nm; Emission 590nm).

RESULTS & DISCUSSION

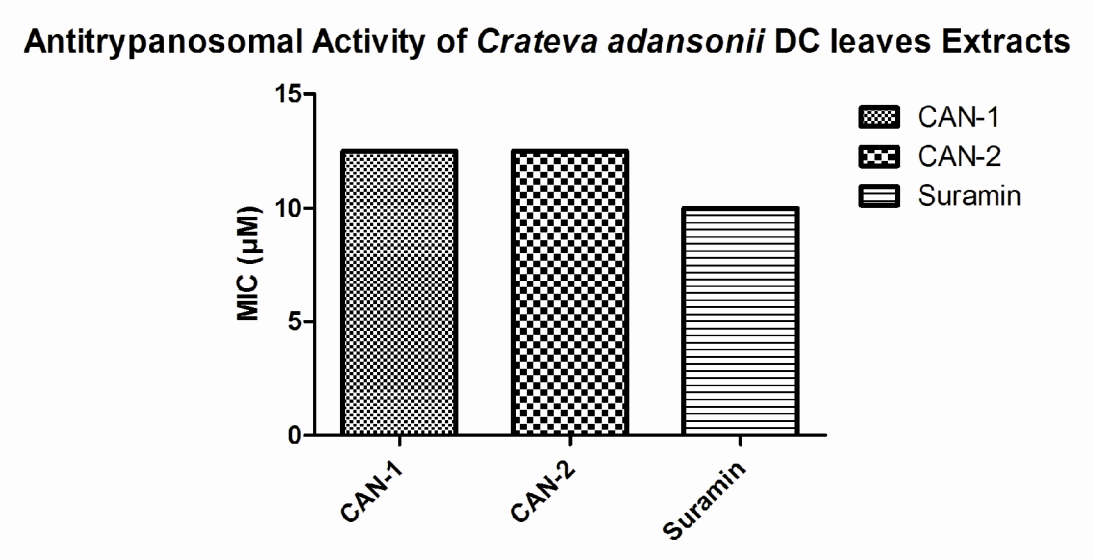

Figure 2

Hexane extract (**CAN-1**) and ethyl acetate extract(**CAN-2**) of *C. adansonii* DC leaves were evaluated in vitro against African trypanosome blood forms *Trypanosoma brucei brucei* S427(**Figure 2**). Both **CAN-1** and **CAN-2** have an MIC value of 12.5 µg/ml, comparable to that of standard Suramin. Oleanolic acid reported to have anti-trypanosomal activity[12], could be attributable phytoconstituent for the activity of **CAN-1** and **CAN-2** against *T. brucei brucei* blood forms. This study leads us to conclude that *C. adansonii* DC can certainly replace the modern medicines for the treatment of trypanosomiasis. Further isolation and characterization of the responsible constituents from these extracts of *C. adansonii* DC is desirable.

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