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# INHIBITION OF MICROSOMAL LIPID PEROXIDATION AND PROTEIN OXIDATION BY EXTRACTS FROM PLANTS USED IN BAMUN FOLK MEDICINE (CAMEROON) AGAINST HEPATITIS

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## Abstract

The antioxidant activities of 53 medicinal plants used in Bamun Folk Medicine for the management of jaundice and hepatitis were investigated. The studies were done using rat hepatic microsomes for lipid peroxidation and bovine serum albumin (BSA) for carbonyl group formation. Silymarine was used as reference compound. Fifteen different extracts were effective at a dose of 200µg/ml in both experiments. Specifically, 25 extracts inhibited lipid peroxidation initiated non-enzymatically by ascorbic acid while 18 inhibited peroxidation as determined by reduced Nicotinamide Adenine Dinucleotide Phosphate (NADPH). The inhibitory concentration 50 ($IC_{50}$) of 23 different plant extracts was lower than 200µg/ml in the microsomal lipid peroxidation inhibition study. Fifteen of the 23 extracts were active in preventing protein oxidation by inhibiting the formation of the carbonyl group on BSA with an $IC_{50}$ value less than 200µg/ ml. The results suggest that the antioxidant activity of the extracts, may be due to their ability to scavenge free radicals involved in microsomal lipid peroxidation or in protein oxidation. These biochemical processes are involved in the aetiology of toxic hepatitis.

**Key words**: lipid peroxidation, protein oxidation, medicinal plants, Bamun, toxic hepatitis.

## Introduction

Lipid peroxidation and protein oxidation are reported to be involved in the aetiology of several human diseases such as atherosclerosis, ischemia-reperfusion injury, ageing, and liver-related diseases (Dean et al., 1997; Aruoma, 1998). In paracetamol- and $CCl_4$-induced hepatitis particularly, the most widely used animal models for the study of the hepatocurative or preventive effect of many medicinal plants (Lin et al., 1995; Shenoy et al., 2001; James et al., 2003), lipid peroxidation and protein oxidation play the main role in the development of the disease (Recknagel, 1983; Fleurentin and Joyeux, 1990; Vuletich and Osawa, 1998; Michael *et al.*, 1999). Thus, the inhibition of these oxidation phenomena may be important for the alleviation of the resulting diseases.

In the Bamun folk medicine, quite a number of plants are reported to be used for the treatment of hepatitis and other liver related-diseases (Mongbet, 1975, Moundipa et al., 2001). However, for a good number, no report is available to prove their therapeutic activity. Since toxic hepatitis is often associated with the oxidative destruction of lipids and proteins, the plants used by the Bamun in order to alleviate liver-related diseases may contain compounds which protect lipids and proteins from oxidation since such compounds have been suggested as prophylactic agents (Aruoma, 1997).

Therefore, the present work was aimed at identifying among these plants, those that are potentially active in the protection of biomolecules of the liver and other organs against oxidation. Thus, the inhibitory effect of their respective extracts on the course of lipid peroxidation induced non-enzymatically or enzymatically in rat liver microsomes has been assessed. We also assayed the inhibitory action of these extracts on the hydroxyl-mediated oxidation of bovine serum albumin (BSA).

## Materials and Methods

**Chemicals**: All reagents used in this study were purchased from Sigma Chemicals Company (St. Louis, MO, USA) and Prolabo (Paris, France).

**Plant extracts**: Fifty-four plants were used in this study, selected according to a previous survey carried out in the Bamun region (Moundipa et al., 2002). The parts used were either the bark, the leaves, tubers or rhizomes. The air-dried and powdered parts (50g) of each plant were extracted by maceration with a mixture of methanol-methylene chloride (200ml, v/v) for 24 hrs with constant shaking. The plant extracts were evaporated to dryness under vacuum, the yield computed and the residue kept at –40°C for pharmacological studies.

### Screening of lipid peroxidation and protein inhibitory activities

### Lipid peroxidation assay

Male Wistar rats weighing 180 – 200g were sacrificed by cerebral dislocation after overnight fasting. The liver was removed and homogenised in ice cold 150mM KCl solution. Liver microsomes were isolated by the calcium aggregation procedure as described by Garle and Fry (1989). Protein concentration in the microsomal suspension was assayed by the Bradford method (Bradford, 1976) using BSA as standard. The resulting suspension was diluted to 10mg of microsomal protein/ml in buffer (25mM Tris-HCl, 115mM KCl, pH 7.5), and stored at –40°C. Experiments were carried out according to the method described by Ulf et al., (1989). Silymarine and plant extract concentrations were tested at 10, 100 and 200µg/ml. Lipid peroxidation was initiated non-enzymatically using ascorbate or enzymatically by NADPH (only for plant extracts for which $IC_{50}$ was less than 200µg/ml). The reaction mixture consisted of microsomes (0.4mg protein/ml), plant extract and 0.5mM ascorbate or 0.3mM NADPH in 25mM Tris-HCl buffer, pH 7.5 containing 115mM KCl. The reaction was initiated by the addition of 1.5µM $Fe^{2+}$ (in the form of $(NH_4)_2Fe(SO_4)_2$) complexed with 1mM ADP. After the incubation period (15 min, 37°C), the reaction was stopped by the addition of thiobarbituric acid reagent. The samples were then assayed for thiobarbituric acid-reactive substances (TBA-RS) as described by Wills (1987). Lipid peroxidation was expressed as the change in absorbance of TBA-RS at 530nm. The amount of TBA-RS which existed in the mixture before the peroxidation reaction was substracted from the value obtained.

### BSA oxidation assay

BSA was oxidised by a Fenton-type reaction (Martinez et al., 2001). The reaction was carried out in 2ml polypropylene tubes with lids. Plant extracts were added to the medium and, after incubation and protein precipitation by TCA, the mixture was centrifuged (3000g, 4°C, 5 min) and the pellet used for protein carbonyl content determination. This was assayed as a 2,4-dinitrophenylhydrazine (DNPH) derivative by of the method described by Martinez et al., (2001) with some modifications. After extraction and a second precipitation of the precipitate, the protein pellets were dissolved in 1ml of 6M urea and centrifuged (3000g, 4°C, 5 min). The different spectra of the DNPH derivatives were obtained at 372nm.

### Phytochemical studies

Groups of phytochemical compounds (flavonoids, polyphenols, leucoanthocyanins, alkaloids, tannins, triterpens and sterols, anthranoids) were tested for their presence in each extract using commonly accepted phytochemical methods (Bruneton, 1999).

### Calculations

Different $IC_{50}$ values were estimated using the EPA probit analyses, on computer program version 1.3 used by C. Stephen of the Duluth USEPA, Research Laboratory.

## Results

### Lipid peroxidation and protein inhibitory activities of extracts
### Inhibition of microsomal lipid peroxidation

The respective inhibition percentages (IP) obtained for each extract are shown in Table 1. These values varied considerably for the different plant extracts. For each extract, this variation also depended on the mode of initiation of peroxidation. Based on the IP, in the non-enzymatical microsomal lipid peroxidation system, at a concentration of 200µg/ml, plant extracts with values equal to or above 50 were selected for further experiments with the Fe(II)-NADPH system. Twenty-five extracts were thus selected and tested in the system where the reaction was sustained by NADPH. Of these, only 18 extracts were active with an IP value above 50 at 200µg/ml (Table 1). Table 3 presents the $IC_{50}$ values of different plant extracts according to the mode of initiation the lipid peroxidation reaction.

### Inhibition of BSA oxidation

The IP values of hydroxyl-mediated oxidation of BSA are presented in Table 2 and the $IC_{50}$ in Table 3. The values of the former varied between different extracts. Only 26 different plant extracts were active above 50 at 200µg/ml.

### Groups of compounds in different plant extracts

The phytochemical studies of plant extracts active in inhibiting microsomal lipid peroxidation or/and protein oxidation revealed the presence of flavonoids, polyphenols, alkaloids, among other classes of compounds as shown in Table 4.

## Discussion

In many traditional practices, there are medicines used for the treatment of liver-related diseases (Fleurentin and Joyeux, 1990). These medicines are generally based on medicinal plants and their systematical screenings often permit leads to the identification of the effective plants (Joyeux et al., 1990, Lin et al., 1995). Extracts of plants under study were tested for their microsomal lipid peroxidation and protein oxidation inhibitory activities. On the whole, the active extracts inhibited both biochemical processes in a dose-dependent manner. Similar results were obtained by Czinnera et al. (2001) on the action of *Helichrysi flos* regarding the inhibition of microsomal lipid peroxidation.

In the present study, plant extracts inhibiting both oxidation phenomena with an $IC_{50}$ less than 200µg/ml were considered as possessing a high protein and lipid oxidation inhibitory potential. In this respect, *Mangifera indica*, *Enantia chlorantha*, *Voacanga africana*, *Aspilia africana*, *Senna alata*, *Piliostigma thonningii* (bark), *Piliostigma thonningii* (leaves), *Kalonchoe crenata*, *Alchornea laxiflora*, *Crotalaria lachnophora*, *Erythrina senegalensis*, *Khaya grandifoliola*, *Entada africana*, *Melinis minutiflora* and *Curcuma longa* (Table 2) were found to be active. Among these active plant species, some of them, namely *E. chlorantha* (Virtanen et al., 1993), *E. africana* (Sanogo et al., 1998) and *C. longa* (Pulla and Lokesh, 1994; Sreejayan and Rao, 1994; Ruby et al., 1995), have been reported to be active against experimentally induced hepatitis. *M. indica* on its part, has been shown to be very effective against lipid and protein oxidation *in vitro* and injury associated to hepatic ischemia reperfusion (Martinez et al., 2001; Sanchez *et al*., 2000). Concerning *S. alata*, the choleretic effect of its extract on rats was demonstrated by Assane et al. (1993).

The protection of the hydroxyl-mediated oxidation of BSA takes place essentially by reducing the $H_2O_2$ concentration, a fundamental component in Fenton-type reaction, by chelating iron or by scavenging the hydroxyl radical formed on the immediate side of the target protein during oxidation (Kingu and Wei, 1997). This may suggest that these plant extracts are able to scavenge hydroxyl radical or chelate iron. The inhibitory effect against the free radical-mediated degradation of BSA and the microsomal lipid peroxidation by plant extracts mentioned above may also be attributed to flavonoids and polyphenols as many of these phytoconstituents are known to be antioxidants (Faurè et al., 1990; Markus, 1996; Middleton et al., 2000). The presence of these two families of compounds was revealed in all the above cited plant extracts. This is in accordance with phytochemical screening done by Noguchi et al. (1994) and Wandji et al., (1994) respectively

**Table 1:** Inhibition percentages of microsomal lipid peroxidation of different plants extracts and initiation modes.

| **Species** | **Family** | **Inhibition percentage** | | | | | |
|---|---|---|---|---|---|---|---|
| | | Fe(II)-Ascorbate[1] | | | Fe(II)- NADPH[1] | | |
| | | Concentrations of plant extracts (µg/mL) | | | Concentrations of plant extracts (µg/mL) | | |
| | | 10 | 100 | 200 | 10 | 100 | 200 |
| Control silymarine | | 61.86 ± 2.61 | 86.81 ± 1.45 | 99.29 ± 3.23 | 62.15 ± 1.65 | 78.79 ± 3.54 | 99.40 ± 2.65 |
| *Eremomastas speciosa (hochst.) Cufod* | Acanthaceae | 5.58 ± 1.23 | 15.01 ± 1.23 | 46.16 ± 0.35 | | | |
| *Draceana deisteliana Engl.* | Agavaceae | -2.63 ± 0.48 | -1.72 ± 1.45 | -0.46 ± 0.00 | | | |
| *Mangifera indica Lin.* | Anacardiaceae | 5.46 ± 0.49 | 66.75 ± 0.33 | 75.35 ± 1.32 | 15.42 ± 1.17 | 60.56 ± 0.00 | 77.5 ± 0.40 |
| *Annona senegalensis Pers.* | Anonaceae | -3.76 ± 4.96 | 14.29 ± 0.35 | 16.17 ± 0.88 | | | |
| *Enantia chlorantha Oliv.* | Anonaceae | 33.25 ± 0.71 | 42.06 ± 0.52 | 53.97 ± 0.52 | 12.19±2.34 | 28.87±0.98 | 53.87±0.00 |
| *Voacanga africana Stapf* | Apocynaceae | 57.82 ± 1.05 | 100.00 ± 0.00 | 100.00±0.00 | 53.04±1.18 | 92.13±2.14 | 100.00±0.00 |
| *Xanthosoma sagittifolium L. Schott* | Araceae | 1.37 ± 053 | 25.31±0.71 | 48.01 0.88 | | | |
| *Polyscias fulva (Hiern.) Harms.* | Araliaceae | -0.02 ± 0.00 | -0.04±0.01 | -0.01±0.27 | | | |
| *Ageratum conyzoides Lin.* | Asteraceae | -2.79 ± 1.00 | -2.56±3.00 | -9.31±3.29 | | | |
| *Aspilia africana (Pers.) C.D. Adams* | Asteraceae | 18.26 ± 0.37 | 35.72±0.37 | 52.91±0.37 | 32.95±0.54 | 51.53±0.18 | 68.45±0.72 |
| *Bidens pilosa Lin.* | Asteraceae | 14.19± 0.33 | 39.82±2.91 | 50.92±1.38 | 4.86±1.37 | 33.06±0.39 | 52.92±0.59 |
| *Chrysanthellum americanum (Lin.) Vatke* | Asteraceae | 2.91 ± 0.00 | 22.89±0.19 | 17.20±1.12 | | | |
| *Dichrocephala integrifolia (Lin.F ) O.Ktze* | Asteraceae | 1.24 ± 0.35 | 2.61±0.87 | 25.43±0.88 | | | |
| *Emilia coccinia (Sims.) G. Don* | Asteraceae | -7.15 ± 6.20 | -5.77±3.19 | -1.51±1.77 | | | |
| *Sonchus oleraceus Lin.* | Asteraceae | -0.75 ± 0.35 | -5.71±0.35 | 31.02±0.35 | | | |
| *Spilanthes filicaulis (Sch. et Th.) C.D. Adams* | Asteraceae | 9.18 ± 0.35 | 18.99±1.23 | 58.93±0.17 | 0.55±0.00 | 3.18±0.20 | 12.29±0.98 |
| *Vernonia amygdalina Del.* | Asteraceae | 0.93 ± 0.00 | 26.98±0.33 | 33.49±1.97 | | | |
| *Dacryodes edulis (G.Don) H.Lam* | Burseraceae | -0.26 ± 0.00 | -6.75±1.68 | -8.20±0.00 | | | |
| *Carica papaya Lin.* | Caricaceae | -3.38 ± 1.15 | -4.07±3.78 | -2.68±1.80 | | | |
| *Senna alata (Lin.) Link* | Cesalpilaceae | 34.53 ± 0.94 | 71.30±0.56 | 88.50±0.56 | 46.44±0.18 | 92.37±0.00 | 100.00±0.00 |
| *Piliostigma thonningii (Sch.) M. Red.* (L) | Cesalpilaceae | 32.84 ± 0.16 | 58.70±0.16 | 74.26±0.81 | 35.14±0.59 | 61.81±0.59 | 68.47±1.37 |
| *Piliostigma thonningii (Sch.) M. Red.* (B) | Cesalpilaceae | 37.53 ± 0.33 | 64.99±3.56 | 78.03±0.33 | 40.56±1.58 | 60.70±0.59 | 67.50±0.00 |

**Table 1** (continued): Inhibition percentages of microsomal lipid peroxidation of different plants extracts and initiation modes.

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| *Terminalia glaucescens Planch.ex benth.* | Combretaceae | 2.10 ± 0.66 | 7.32±0.49 | 47.68±1.65 | | | |
| *Ipomea batatas (Lin.) Lam* | Convolvulaceae | 2.10 ± 0.33 | 5.12±1.65 | 14.30±0.49 | | | |
| *Kalonchoe crenata (Andr.) Haw.* | Crasulaceae | 17.18 ± 0.76 | 36.31±0.00 | 73.70±0.50 | 11.88±0.39 | 44.48±0.39 | 59.81±1.76 |
| *Alchornea laxiflora (benth.) Pax& K.H* | Euphorbiaceae | 58.07 ± 9.91 | 84.39±0.75 | 95.90±0.57 | 40.84±0.39 | 65.42±1.77 | 79.17±1.57 |
| *Manihot esculenta Crantz* | Euphorbiaceae | -1.15 ± 0.64 | -0.46±0.00 | -4.80±0.00 | | | |
| *Crotalaria lachnophora Hochst.ex A.R.* | Fabaceae | 22.54 ± 0.76 | 84.26±0.50 | 97.41±0.37 | 38.26±1.37 | 68.37±1.75 | 74.45±0.98 |
| *Erythrina senegalensis D.C* | Fabaceae | 39.32 ± 3.79 | 75.20±2.12 | 94.25±0.71 | 35.91±0.78 | 61.33±3.91 | 75.69±0.40 |
| *Harungana madagascariensis Lam.* | Hypericaceae | 13.60 ± 0.50 | 71.38±1.01 | 81.75±0.00 | 31.08±0.59 | 67.96±0.39 | 76.25±0.39 |
| *Gladiolus dalenii Van Geel* | Iridaceae | -0.36 ± 0.25 | -3.94±0.25 | -8.23±1.27 | | | |
| *Occimum Gratissimum Lin.* | Labieae | 14.99 ± 0.16 | 55.38±1.62 | 68.01±0.49 | 1.11±0.00 | 23.89±1.57 | 43.75±1.37 |
| *Persea americana Mill. (L)* | Lauraceae | -6.02 ± 1.77 | 2.63±3.01 | 24.31±0.00 | | | |
| *Persea americana Mill. (B)* | Lauraceae | 29.52 ± 0.65 | 29.87±0.16 | 44.85±4.52 | | | |
| *Anthocleista schweinfurthii Gil.* | Loganiaceae | -0.01 ± 0.01 | -0.07±0.01 | -0.05±0.01 | | | |
| *Gosypium barbadense (Mac fedyen) J.B.H.* | Malvaceae | 8.24 ± 1.60 | 53.32±1.29 | 56.98±0.33 | 0.56±0.39 | 7.78±0.78 | 40.14±0.98 |
| *Khaya grandifoliola D.C.* | Meliaceae | 1.51 ± 0.49 | 60.94±0.66 | 78.91±1.08 | 12.50±0.00 | 53.33±0.00 | 59.87±0.98 |
| *Entada africana (Guill. et Pers.)* | Mimosaceae | 25.17 ± 0.00 | 50.34±0.00 | 82.73±0.49 | 38.48±0.59 | 74.03±1.37 | 100.00±0.00 |
| *Ficus exasperata Vahl.* | Moraceae | -1.40 ± 0.00 | -6.98±3.29 | -6.98±0.00 | | | |
| *Ficus sp.* | Moraceae | -0.01± 0.00 | -0.04±0.00 | -0.07±0.00 | | | |
| *Musa sapientum Lin.* | Musaceae | -2.15 ± 2.78 | -6.09±0.76 | -9.33±1.39 | | | |
| *Eucalyptus sp.* | Myrtaceae | 50.93± 0.19 | 78.18±0.94 | 76.19±0.00 | 29.77±1.44 | 65.90±0.00 | 85.63±0.18 |
| *Psidium guayava Lin.* | Myrtaceae | 2.21 ± 0.49 | 14.54±0.83 | 50.47±2.64 | 7.08±1.77 | 24.31±0.98 | 32.92±1.38 |
| *Olax subscorpioideae Oliv.* | Olacaceae | 18.26 ± 0.37 | 42.86±0.00 | 44.71±0.00 | | | |
| *Cymbopogon citratus ( D.C.) Stapf* | Poaceae | -1.25 ± 1.10 | -5.71±0.35 | -9.66±0.38 | | | |
| *Melinis minutiflora P. Bearw* | Poaceae | 32.67 ± 1.68 | 86.11±0.93 | 58.47±0.37 | 10.56±0.54 | 61.32±0.72 | 71.33±0.90 |
| *Coffea arabica Lin.* | Rubiaceae | 39.69 ± 1.12 | 25.00±1.32 | 10.05±0.00 | | | |
| *Coffea robusta lin.* | Rubiaceae | 8.27 ± 0.35 | 19.43±7.62 | 41.23±1.60 | | | |
| *Nauclea latifolia Sm.* | Rubiaceae | 25.97 ± 1.13 | 33.41±0.33 | 43.02±1.61 | | | |
| *Citrus aurantifolia Swingle* | Rutaceae | 1.61 ± 0.18 | 26.31±0.71 | 54.59±1.40 | 11.74±058 | 20.31±0.59 | 30.80±4.10 |
| *Citrus sinensis* L. ( Osbeck ) | Rutaceae | 15.01± 1.23 | 54.71±0.52 | 100.00±0.00 | 12.57±0.59 | 24.72±0.98 | 46.83±2.54 |
| *Solanum acaleastrum* Dunal | Solanaceae | 11.00 ± 1.00 | 40.23±7.62 | 28.57±0.00 | | | |
| *Trema orientalis* Lour. | Ulmaceae | -1.28 ± 1.15 | -2.91±0.81 | -5.23±4.77 | | | |
| *Costus afer* Ker .Gawl | Zingiberaceae | 7.51 ± 0.00 | 25.31±0.38 | 68.16±2.02 | 0.83±0.00 | 2.62±0.98 | 11.61±3.51 |
| *Curcuma longa* Lin. | Zingiberaceae | 53.26 ± 1.95 | 77.44±0.35 | 90.36±0.18 | 91.60±0.00 | 100.00±0.00 | 100.00±0.00 |

[1]**-Initiation mode of lipid peroxidation: Fe (II)-Ascorbate (non-enzymatical lipid peroxidation), Fe (II)-NADPH (enzymatical lipid peroxidation), Data are given as mean ± SD of two experiments; L = Leaves; B = Stem bark**

**Table 2:** Inhibition percentages of carbonyl-group formation of different plants extracts

| **Species** | **Family** | **Inhibition Percentage** | | |
|---|---|---|---|---|
| | | Fe(III)-EDTA / $H_2O_2$ / Ascorbate[1] Concentration of plant extracts (µg/ml) | | |
| | | 10 | 100 | 200 |
| Control silymarine | | 51.19 ± 2.34 | 84.18 ± 4.54 | 99.40 ± 3.56 |
| *Eremomastas speciosa* (hochst.) Cufod | Acanthaceae | 9.52±0.00 | 31.79±0.00 | 45.03±0.00 |
| *Draceana deisteliana* Engl | Agavaceae | 5.07±0.35 | 16.55±0.76 | 38.27±1.30 |
| *Mangifera indica* Lin. | Anacardiaceae | 69.47±0.27 | 89.44±0.15 | 99.21±0.00 |
| *Annona senegalensis* Pers. | Anonaceae | 44.49±1.63 | 68.59±3.23 | 85.17±0.82 |
| *Enantia chlorantha* Oliv. | Anonaceae | 20.67±1.44 | 44.95±0.57 | 61.62±0.72 |
| *Voacanga africana* Stapf | Apocynaceae | 24.80±1.29 | 36.34±0.71 | 76.75±0.59 |
| *Xanthosoma sagittifolium* L. Schott | Araceae | -0.81±0.30 | 14.50±1.56 | 29.58±0.30 |
| *Polyscias fulva* (Hiern.) Harms. | Araliaceae | 5.00±0.45 | 27.40±1.48 | 42.89±0.31 |
| *Ageratum conyzoides* Lin. | Asteraceae | 34.13±0.15 | 35.24±0.33 | 48.71±0.42 |
| *Aspilia africana* (Pers.) C.D. Adams | Asteraceae | 59.39±3.24 | 81.63±1.00 | 86.33±0.81 |
| *Bidens pilosa* Lin. | Asteraceae | 5.81±0.26 | 15.68±0.12 | 33.24±1.83 |
| *Chrysanthellum americanum* (Lin.) Vatke | Asteraceae | 61.09±1.90 | 88.02±0.00 | 99.58±0.59 |
| *Dichrocephala integrifolia* (Lin.F ) O.Ktze | Asteraceae | 2.74±0.45 | 19.44±0.27 | 42.92±0.00 |
| *Emilia coccinia* (Sims.) G. Don | Asteraceae | 14.01±2.59 | 29.59±3.23 | 35.22±1.46 |
| *Sonchus oleraceus* Lin. | Asteraceae | 4.46±1.44 | 4.46±0.00 | 22.16±2.14 |
| *Spilanthes filicaulis* (Sch. et Th.) CD. Adams | Asteraceae | 5.27±0.30 | 12.56±1.41 | 42.41±0.72 |
| *Vernonia amygdalina* Del. | Asteraceae | 27.83±1.44 | 58.77±1.47 | 75.27±1.47 |
| *Dacryodes edulis* (G.Don.) H.Lam | Burseraceae | 9.10± 0.00 | 26.68±3.76 | 47.20±1.10 |
| *Carica papaya* Lin. | Caricaceae | 32.89±1.32 | 71.66±0.70 | 87.20±0.60 |
| *Senna alata* (Lin.) Link | Cesalpilaceae | 70.05±1.16 | 84.38±0.30 | 98.94±0.48 |
| *Piliostigma thonningii* (Sch.) M. Red. (L) | Cesalpilaceae | 61.38±0.68 | 71.36±0.69 | 91.81±0.26 |
| *Piliostigma thonningii* (Sch.) M. Red. (B) | Cesalpilaceae | 34.94±0.40 | 68.03±1.90 | 95.33±1.70 |
| *Terminalia glaucescens* Planch.ex benth. | Combretaceae | 22.41±0.00 | 59.33±0.70 | 74.58±1.11 |
| *Ipomea batatas* (Lin.) Lam | Convolvulaceae | 35.51±0.99 | 40.14±0.00 | 50.97±1.40 |
| *Kalonchoe crenata* (Andr.) Haw. | Crasulaceae | 45.84±0.15 | 62.13±0.00 | 69.93±0.45 |
| *Alchornea laxiflora* (benth.) Pax & K.H. | Euphorbiaceae | 58.40±0.40 | 85.61±0.40 | 95.60±0.59 |
| *Manihot esculenta* Crantz | Euphorbiaceae | 2.41±0.57 | 10.69±0.12 | 29.45±0.15 |
| *Crotalaria lachnophora* Hochst.ex A.R | Fabaceae | 0.43±0.60 | 21.36±1.07 | 54.32±0.40 |
| *Erythrina senegalensis* D.C. | Fabaceae | 54.48±0.18 | 95.43±1.36 | 98.53±0.00 |
| *Harungana madagascariensis* Lam. | Hypericaceae | 7.18±0.42 | 17.43±0.30 | 47.48±0.12 |
| *Gladiolus dalenii* Van Geel | Iridaceae | -0.19±0.27 | 3.55±0.15 | 7.29± 0.57 |
| *Occimum Gratissimum* Lin. | Labieae | 11.99±0.30 | 25.44±0.71 | 49.42±0.71 |
| *Persea americana* Mill. (L) | Lauraceae | 3.47±1.29 | 17.81±0.33 | 44.05±0.00 |
| *Persea americana* Mill. (B) | Lauraceae | 0.00±0.00 | 16.98±1.42 | 35.54±1.43 |
| *Anthocleista schweinfurthii* Gil. | Loganiaceae | 37.99±0.30 | 51.96±0.00 | 57.57±0.44 |
| *Gosypium barbadense* (Mac fedyen) J.B.H. | Malvaceae | 11.96±1.17 | 19.47±0.16 | 40.52±1.01 |
| *Khaya grandifoliola* D.C. | Meliaceae | 19.69±1.62 | 68.56±2.21 | 83.09±0.28 |

**Table 2** (continued): Inhibition percentages of carbonyl-group formation of different plants extracts

| | | | | |
|---|---|---|---|---|
| *Entada africana* (Guill. et Pers.) | Mimosaceae | 50.00±0.71 | 71.25±0.83 | 79.55±0.42 |
| *Ficus exasperata* Vahl. | Moraceae | 17.22±1.01 | 28.84±0.00 | 39.56±0.58 |
| *Ficus sp.* | Moraceae | 27.60±1.20 | 34.71±0.16 | 51.39±0.60 |
| *Musa sapientum* Lin. | Musaceae | 6.99±0.45 | 25.42±0.42 | 34.13±0.42 |
| *Eucalyptus sp.* | Myrtaceae | 58.48±0.30 | 29.37±2.93 | 12.95±0.82 |
| *Psidium guayava* Lin. | Myrtaceae | 17.61±1.32 | 54.64±0.29 | 83.72±1.17 |
| *Olax subscorpioideae* Oliv. | Olacaceae | 13.33±0.33 | 31.66±0.30 | 39.12±2.76 |
| *Cymbopogon citratus* ( D.C.) Stapf | poaceae | 19.12±0.72 | 26.82±1.56 | 40.08±0.30 |
| *Melinis minutiflora* P. Bearw | Poaceae | 5.60±0.40 | 64.00±1.90 | 96.92±2.46 |
| *Coffea arabica* Lin. | Rubiaceae | 11.03±0.35 | 18.03±0.17 | 36.45±0.00 |
| *Coffea robusta* Lin. | Rubiaceae | 29.37±0.00 | 45.98±0.48 | 45.80±0.00 |
| *Nauclea latifolia* Sm. | Rubiaceae | 48.30±0.57 | 51.89±0.00 | 70.96±1.54 |
| *Citrus aurantifolia* Swingle | Rutaceae | 14.90±1.00 | 38.22±3.38 | 47.77±0.30 |
| *Citrus sinensis* L. ( Osbeck ) | Rutaceae | 20.65±1.44 | 29.86±1.00 | 45.35±1.44 |
| *Solanum acaleastrum* Dunal | Solanaceae | 2.19±0.51 | 12.07±1.12 | 25.23±0.34 |
| *Trema orientalis* Lour. | Ulmaceae | 13.80±0.88 | 21.63±1.44 | 36.49±0.89 |
| *Costus afer* Ker .Gawl | Zingiberaceae | 36.00±1.36 | 49.38±0.47 | 57.87±0.44 |
| *Curcuma longa* Lin. | Zingiberaceae | 95.31±0.52 | 99.57 ± 0.00 | 100.00 ± 0.00 |

[1]-Initiation mode of BSA oxidation, Data are given as mean ± SD of two experiments; L = Leaves; B = Stem bark

**Table 3**: Computed $IC_{50}$ (µg/mL) of microsomal lipid oxidation and protein oxidation by some plant extracts.

| **Species** | **Family** | Microsomal lipid peroxidation | | Protein oxidation |
|---|---|---|---|---|
| | | Non-enzymatical | Enzymatical | |
| Control silymarine | | 5.5 ± 1.98 | 22.70 ± 3.34 | 10.43 ± 2.39 |
| *Mangifera indica* | Anacardiaceae | 69.84 ± 0.70 | 51.70 ± 2.83 | 3.33±0.22 |
| *Annona senegalensis* | Annonaceae | NC | NC | 16.21±1.05 |
| *Enantia chlorantha* | Apocynaceae | 197.16 ± 3.85 | NC | 108.28±1.00 |
| *Voacanga africana* | Apocynaceae | < 10 | < 10 | 79.91±0.90 |
| *Aspilia Africana* | Asteraceae | NC | 53.91 ± 2.26 | 4.69±1.90 |
| *Bidens pilosa* | Asteraceae | 194.00 ± 9.07 | 190.91 ± 071 | NC |
| *Chrysanthellum americanum* | Asteraceae | NC | NC | 6.24±0.37 |
| *Spilanthes filicaulis* | Asteraceae | 239.58 ± 8.05 | NC | NC |
| *Vernonia amygdalina* | Asteraceae | NC | NC | 45.31±1.88 |
| *Carica papaya* | Caricaceae | NC | NC | 25.44±1.73 |
| *Senna alata* | Cesalpiniaceae | 23.86 ± 1.03 | 11.57 ± 0.26 | 2.83±0.39 |
| *Piliostigma thonningii (bark)* | Cesalpiniaceae | 26.13 ± 2.66 | 28.46 ± 4.04 | 23.18±0.93 |
| *Piliostigma thonningii (leaves)* | Cesalpiniaceae | 31.39 ± 9.45 | 37.14 ± 0.55 | 4.27±0.37 |
| *Terminalia glaucescens* | Combretaceae | NC | NC | 53.72±0.03 |
| *Kalonchoe crenata* | Crasulaceae | 97.96 ± 0.97 | 125.25 ± 6.99 | 17.37±0.00 |
| *Alchornea laxiflora* | Euphorbiaceae | 6.95 ± 4.31 | 21.64 ± 1.15 | 6.43 ±0.18 |
| *Crotalaria lachnophora* | Fabaceae | 25.62 ± 0.50 | 24.79 ± 2.93 | 189.92 ±7.76 |
| *Erythrina senegalensis* | Fabaceae | 33.11 ± 3.78 | 31.75 ± 3.65 | 8.57±0.23 |
| *Harungana madagascariensis* | Hypericaceae | 48.35 ± 1.37 | 33.28 ± 0.78 | NC |
| *Occimum gratissimum* | Labieae | 77.75 ± 1.44 | NC | NC |
| *Anthocleista schweinfurthii* | Loganiaceae | NC | NC | 67.09±2.72 |
| *Gossypium Barbadense* | Malvaceae | 114.80 ± 3.87 | NC | NC |
| *Khaya grandifoliola* | Meliaceae | 81.70 ± 3.30 | 102.04 ± 2.52 | 42.04±0.16 |
| *Entada Africana* | Mimosaceae | 50.67 ± 0.46 | 18.33 ± 0.76 | 9.85±0.66 |
| *Eucalyptus sp.* | Myrtaceae | 8.14 ± 0.06 | 31.33 ± 1.45 | NC |
| *Psidium guyava* | Myrtaceae | NC | NC | 53.20±2.55 |
| *Melinis minutiflora* | Poaceae | 27.42 ± 297 | 71.29 ± 2.59 | 54.93±3.25 |
| *Nauclea latifolia* | Rubiaceae | NC | NC | 17.62±1.41 |
| *Citrus aurantifolia* | Rutaceae | 190.48 ± 6.24 | NC | NC |
| *Citrus sinensis* | Rutaceae | 45.01 ± 0.49 | NC | NC |
| *Costus afer* | Zingiberaceae | 151.33 ± 6.42 | NC | 80.43±0.76 |
| *Curcuma longa* | Zingiberaceae | 8.39 ± 1.25 | < 10 | < 10 |

Values are mean ± SD of two experiments

NC: Values not computed because of the low inhibition percentages obtained with the highest dose of extract during the test (< 50%)

**Table 4 :** Phytochemical composition of some selected active plant extracts

| Classes of compounds<br>Families &Species | Flavonoids | Triterpens | Sterols | Alcaloids | Polyphenols | Tannins | Anthranoids | Leucoanthocyans |
|---|---|---|---|---|---|---|---|---|
| **Anacardiaceae** | | | | | | | | |
| *Mangifera indica* | + | - | - | - | + | - | - | + |
| **Annonaceae** | | | | | | | | |
| *Annona senegalensis*(leaves) | - | - | + | - | + | + | - | - |
| *Enantia chlorantha* | - | - | - | + | + | + | - | - |
| **Apocynaceae** | | | | | | | | |
| *Voacanga Africana* | - | - | - | + | + | + | - | - |
| **Asteraceae** | | | | | | | | |
| *Aspilia Africana* | - | - | + | - | + | + | - | - |
| *Chrysanthellum americanum* | + | - | + | - | + | + | - | - |
| *Vernonia amygdalina* | + | - | + | - | + | - | - | - |
| **Caricaceae** | | | | | | | | |
| *Carica papaya* | - | - | + | - | + | - | - | - |
| **Cesalpiniaceae** | | | | | | | | |
| *Senna alata* | + | - | + | - | + | + | - | + |
| *Piliostigma thonningii*(bark) | + | - | + | - | + | + | - | + |
| *Piliostigma thonningii*(leaves) | + | - | + | - | + | + | - | + |
| **Combretaceae** | | | | | | | | |
| *Terminalia glaucescens* | + | - | - | - | + | + | - | + |
| **Crasulaceae** | | | | | | | | |
| *Kalonchoe crenata* | - | - | + | - | + | - | - | - |
| **Euphorbiaceae** | | | | | | | | |
| *Alchornea laxiflora* | + | - | + | - | + | - | - | - |
| **Fabaceae** | | | | | | | | |
| *Crotalaria lachnophora* | - | - | + | - | + | - | - | - |
| *Erythrina senegalensis* | + | - | - | - | + | - | + | - |

**Table 4 (continued): Phytochemical composition of some selected active plant extracts**

| Classes of compounds<br>Families &Species | Flavonoids | Triterpens | Sterols | Alcaloids | Polyphenols | Tannins | Anthranoids | Leucoanthocyans |
|---|---|---|---|---|---|---|---|---|
| **Hypericaceae** | | | | | | | | |
| *Harungana madagascariensis* | + | - | - | - | + | + | - | + |
| **Loganaceae** | | | | | | | | |
| *Anthocleista shweinfurthii* | + | - | - | - | + | + | - | - |
| **Meliaceae** | | | | | | | | |
| *Khaya grandifoliola* | + | - | - | - | + | + | - | + |
| **Mimosaceae** | | | | | | | | |
| *Entada Africana* | + | - | - | - | + | + | - | + |
| **Myrtaceae** | | | | | | | | |
| *Psidium guayava* | + | - | + | - | + | - | - | - |
| *Eucalyptus sp.* | + | - | + | - | + | - | - | - |
| **Poaceae** | | | | | | | | |
| *Melinis minutiflora* | + | - | - | - | + | + | - | - |
| **Rubiaceae** | | | | | | | | |
| *Nauclea latifolia* | + | - | - | - | + | + | - | + |
| **Zingiberaceae** | | | | | | | | |
| *Curcuma longa* | + | + | - | - | + | + | + | + |

(+) positive test for the class of compounds

(-) Negative test for the class of compounds

on *Curcuma longa* and *Erythrina senegalensis*. However, in extracts from *Enantia chlorantha* and *Voacanga africana* which also inhibited both studied biochemical phenomena the presence of alkaloids was also demonstrated.

Since protein degradation and lipid peroxidation seem to occur by distinct mechanisms (Davies and Goldberg, 1986), it may be suggested that the above 15 plant extracts have strong lipid and protein oxidation inhibitory potency. Therefore, these plant species may be a good source of medicines against diseases in which lipids and proteins oxidation are involved such as toxic hepatitis. Further *in vitro* and *in vivo* studies on some of these plant extracts are in progress.

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