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Comparative anti-inflammatory activities of *jatropha curcas*, *ocimum gratissimum* and *solanum scabrum* leaves

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ABSTRACT

The methanolic extracts of the leaves of Jatropha curcas Linn. (Euphorbiaceae), Ocimum gratissimum Linn. (Lamiaceae) and Solanum scabrum Mill. (Solanaceae) were investigated for possible anti-inflammatory activities. The plant extract, suspended in 0.5% carboxymethyl cellulose (CMC) was orally administered to formaldehyde-induced arthritic mice at a dose of 200mg/kg body weight for seven days. Control mice and standard mice, which were also injected with formaldehyde, were administered 0.5% CMC and 20mg/kg indomethacin respectively for seven days. Inflammation in the mice was assayed by monitoring paw oedema and by measuring the concentrations of serum alanine aminotransferase (ALT) as well as serum aspartate aminotransferase (AST). Paw oedema was substantially decreased by the oral administration of the methanolic extracts of J. curcas. O. gratissimum and S. scabrum also decreased the paw oedema but to a lesser extent. Both serum ALT and AST were substantially decreased by J. curcas and O. gratissimum but there was greater reduction of ALT than of AST by both plants extracts. S. scabrum also caused a significant decrease in serum ALT concentration but the serum AST value was only slightly decreased by this plant. These results strongly suggest the presence of strong anti-inflammatory activities in the methanolic extracts of J. curcas and O. gratissimum leaves while S. scabrum probably possesses a moderate anti-inflammatory activity.

Keywords: Anti-inflammation, serum alanine aminotransferase (ALT), serum aspartate aminotransferase (AST), paw oedema.

INTRODUCTION

The term inflammation describes a complexity of ailments which include redness, swelling, and feelings of heat, pain and sometimes loss of function of the affected part of the body. The most common inflammatory condition in humans is the joint inflammation, referred to as arthritis, which also presents in various forms such as rheumatoid arthritis and osteoarthritis [1]. There is a number of non-steroidal anti-inflammatory drugs (NSAIDs) used for the treatment of arthritis; these include aspirin, indomethacin, ibuprofen, meloxican and others. Many of the NSAIDs are effective in alleviating the pains of arthritis but they also have side effects such as gastrointestinal [2, 3], renal effects [4] and cardiovascular effects [5].

Many plant extracts are used ethno-medically to treat various ailments including arthritis [6, 7]. Investigations in many laboratories are aimed at finding the scientific basis for such ethnomedical treatment [8]. For the study of anti-inflammatory activity in plant extracts, such extracts are tested on animal models in which arthritis is artificially induced [9].

The plant *Ocimum gratissimum* Linn (Lamiaceae) is a herbaceous plant grown in the tropics and common in Southern Nigeria. It has various ethnomedical and culinary uses [10]. Scientific investigations have shown that Ocimum species possess antidiarrhoeal activity [11], antimicrobial activity [12] and hypoglycemic activity [13, 14]. It has also been shown that *O. gratissimum* contains and synthesizes prostaglandin-like compounds [15, 16], and contains smooth-muscles contracting activity [17]. Antioxidant activity has been demonstrated in *O. gratissimum* Linn and *Ocimum canum* Sims [18]. These findings have implications for some therapeutic potentials of the Ocimum species. The aqueous leaves extract of *O. gratissimum* has been shown to possess anti-inflammatory activity by its effects on rat paw oedema. In our work whose report is given here, we investigated possible anti-inflammatory activity in *O. gratissimum* using both paw oedema and enzyme activity assessment.

Jatropha Curcas Linn. (Euphorbiaceae) is a shrub grown in tropical areas. Virtually all parts of this plant are commonly used in traditional medicine in many parts of West Africa [19]. The seeds of *J. curcas* have been used as purgatives, antihelminthic, abortifacient and also for treating gout, paralysis and skin diseases. The seed's oil has been used as part of the concoction for the treatment of rheumatoid conditions, fever, jaundice and some have been used as a haemostatic agent [20]. The latex has been shown to possess both coagulant and anticoagulant activities [21]. A review of the medicinal uses of this plant has recently been published by Prasad and his colleagues [22]. We reasoned that *J. curcas* leaves might also contain anti inflammatory activity.

Solanum Scabrum Mill. (Solanaceae) is a green vegetable popularly eaten by West Africans. The leaves and fresh shoots are eaten as a cooked vegetable [23, 24]. *Solanum nigrum*, a name which encompasses a few solanum species, including *S. scabrum* [23], has been used traditionally to treat various ailments including inflammation, fever, wound and liver disease [25]. It was of interests to substantiate scientifically, the possible anti-inflammatory activity of *S. scabrum* leaves.

MATERIALS AND METHODS

Plant materials

J. curcas leaves were collected from hedges in Ilishan town, Ogun state. *O. gratissimum* leaves were collected from the farmland in Babcock University. *S. Scabrum* cuttings were collected from Akinyele Area, Ibadan, Oyo State. All the plants were authenticated by Prof. E. B. Esan, an Ethno-botanist at Babcock University.

Preparation of plant extract

Leaves of each plant were air dried at room temperature and then extracted with 80% methanol. The methanolic extracts were then evaporated to dryness at 40^{o}C using a rotary evaporator.

Animals

Male albino mice were purchased from the Nigerian Institute of Medical Research (NIMR), Yaba, Lagos. The animals were acclimatized for about a week before the treatments. They were kept in cages in the laboratory and fed with livestock feed obtained from NIMR and with water *ad libitum.*

Induction of arthritis in mice and the effects of the plants leaf methanolic extract

Arthritis was induced in mice with formaldehyde according to the method used by [9]. The mice, each with an average weight of 30g, were divided into four groups namely: test, standard, control and normal groups. Arthritis was induced in all mice, except those in the normal group, by injecting 0.2ml of 2% (v/v) formaldehyde on the first and third day into the left hind foot paw, just beneath the plantar aponeurosis. The "normal" mice were injected with 0.02ml of normal saline solution. Arthritis in the mice was assayed by two methods. The first method involved an assay for paw oedema by measuring paw thickness before and at intervals after injection of formaldehyde. The other method involved the assay of two serum enzyme activities: serum alanine aminotranferse (ALT) and aspartate aminotransferase (AST).

The test animals were orally administered once a day, for seven days, with 200mg/kg plant methanolic extract suspended in 0.5%carboxymethyl cellulose (CMC). The standard groups of animals were each administered with 20mg/kg indomethacin (indocid) suspended in 0.5% CMC. Control mice were administered with 0.5% CMC alone. The normal groups of animals were not induced with arthritis and were not subjected to any special oral treatments. All the special feeding treatments were in addition to their normal feeding regime. These experiments were replicated.

Assay of serum AST and ALT

At the end of the seven days treatment period, the mice were anaesthetized with chloroform and their blood obtained through cardiac puncture. The blood was allowed to clot and the serum separated by centrifugation in a refrigerated centrifuge. The sera from all the four mice in each group were pooled. Serum AST and ALT were assayed using RANDOX kits. The assays were carried out following the instruction manuals in the assay kits.

RESULTS

Effects of *O. gratissimum*, *J. curcas* and *S. scabrum* leaf methanolic extracts on mice paw oedema

Injection of formaldehyde into the paw of the mice caused paw oedema as shown by a substantial increase in paw thickness (Figure 1). The figure compares the paw thickness in mice injected with formaldehyde with the control mice injected with normal saline.

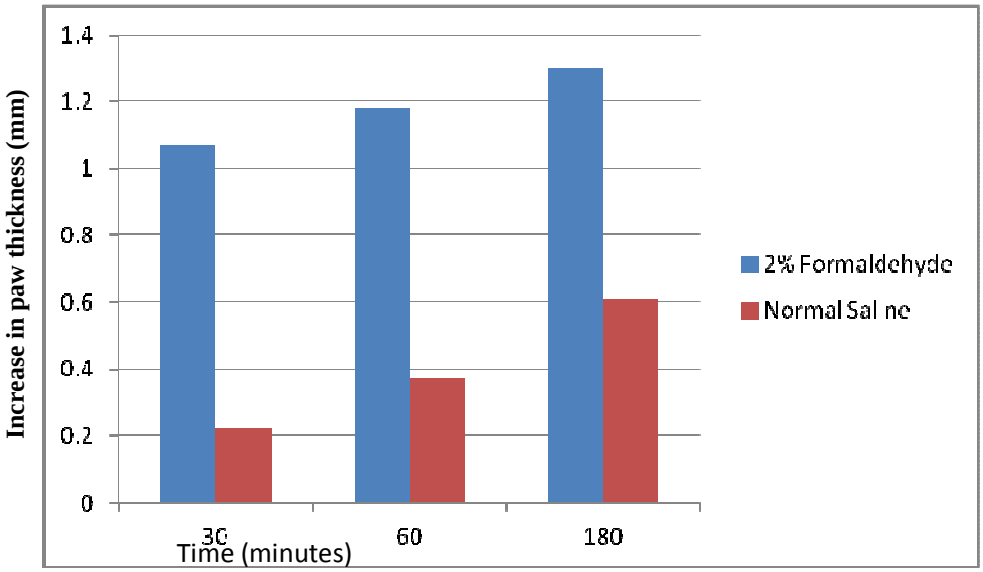

Figure 1: Effects of 2% formaldehyde and Normal saline on the left hind paw thickness in mice

J. curcas leaf methanolic (200mg/kg) extract reduced paw thickness in formaldehyde induced arthritic mice to about 10% by 6hrs and 2.5% by 24 hours (Figure 2A). Similarly, oral administration of *O. gratissimum* leaf methanolic extract to formaldehyde induced arthritic mice reduced the paw oedema to about 61% within 24 hours (Figure 2B); while *S. scabrum* leaf methanolic extract decreased the paw thickness to about 70% by 24 hours (Figure 2C). Furthermore, 24 hours after the administration of the leaves extracts the effects of the methanolic extracts were comparable to the effect of indomethacin administered to the mice at a concentration of 20mg/kg as seen in figures 2A, 2B and 2C.

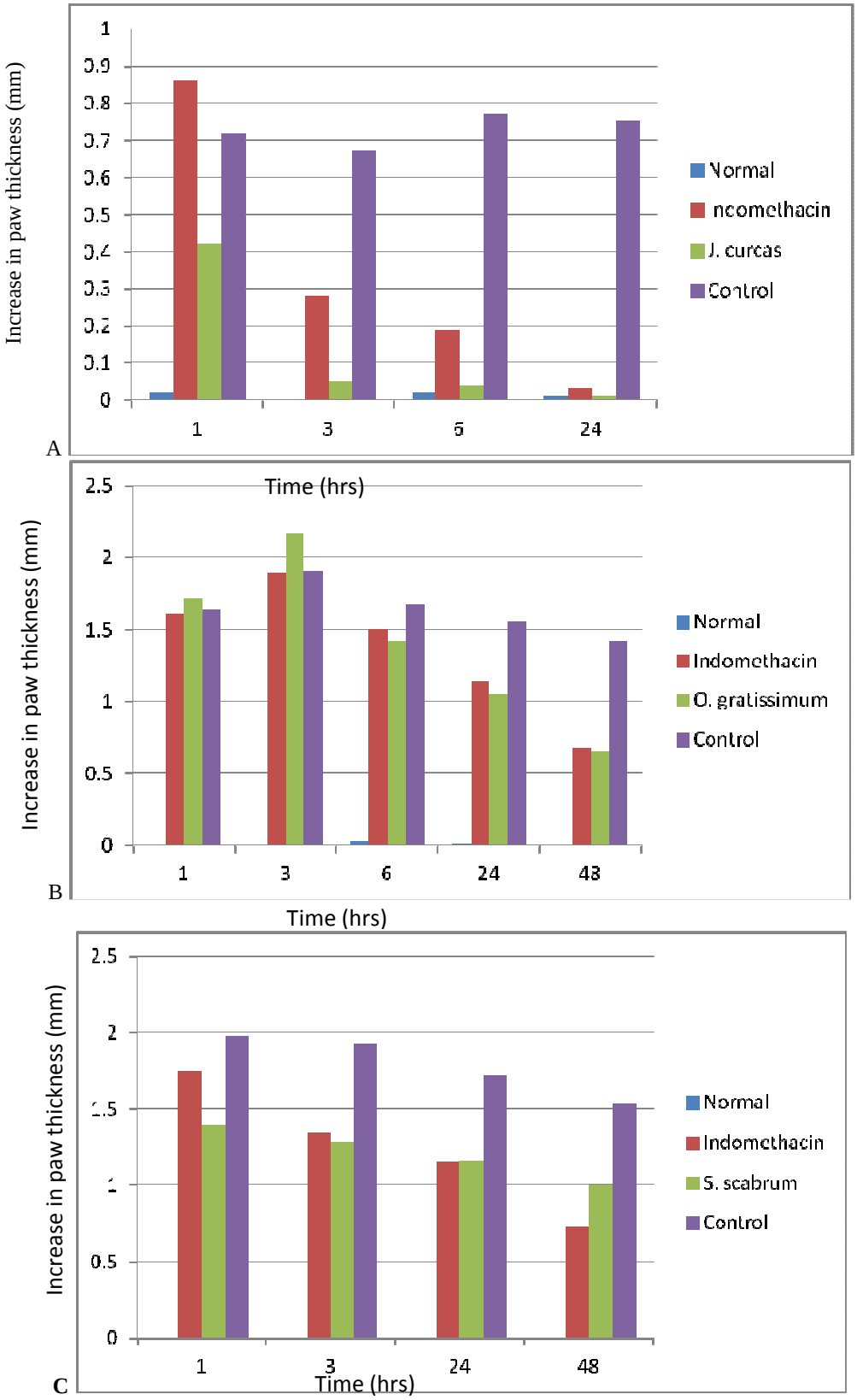

Figure 2: Effects of *J. curcas* (A), *O. gratissimum* (B) and *S. scabrum* (C) methanolic extracts and indomethacin on paw thickness in formaldehyde-induced inflammation in mice

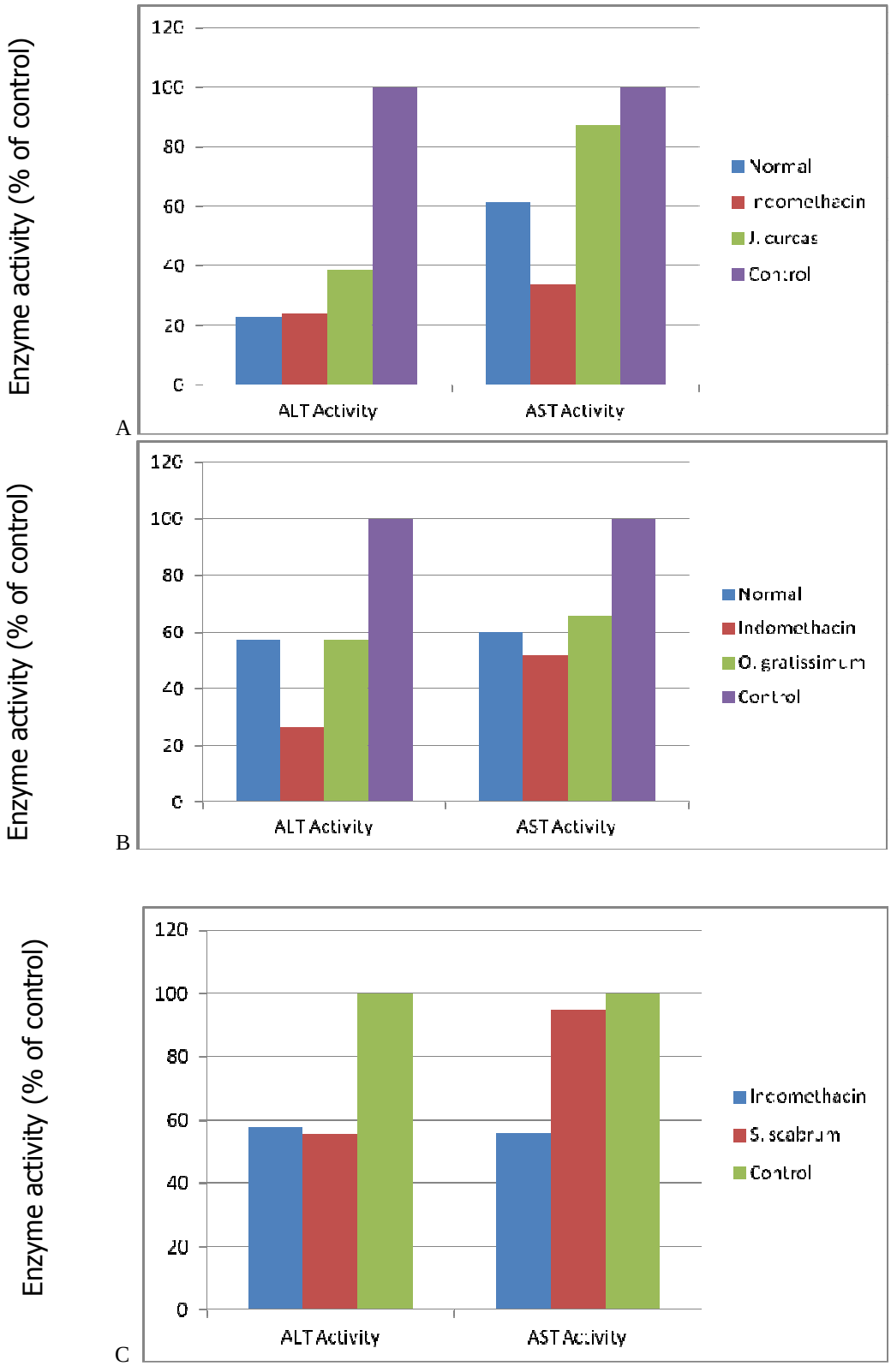

Figure 3: Effects of *J. curcas* (A), *O. gratissimum* (B) and *S. scabrum* (C) methanolic extracts and indomethacin on alanine aminotransferase (ALT) and aspartate aminotransferase (AST) in formaldehyde-induced inflammation in mice

Effects of *O. gratissimum*, *J. curcas* and *S. scabrum* leaf methanolic extracts on serum ALT and AST of formaldehyde induced arthritic mice

Figure 3 shows a typical set of results obtained when serum ALT and AST activities were determined in the formaldehyde-induced arthritic mice. Mice which were injected with formaldehyde but which received no plant extract were the controls and their enzymatic activities were equated to a hundred percent. Normal mice were neither injected with formaldehyde nor treated with the plant extract. Their enzyme activities therefore constituted the basal serum ALT and AST activities in the mice. It can be seen from figure 3 that the enzymatic activities in the control mice are much higher than the basal activities showing that the induced arthritis in the mice led to increased serum ALT and AST. When the *J. curcas* extract was administered to the formaldehyde-induced arthritic mice, serum ALT activity of the mice was substantially reduced to 31±6% of the control, while AST activity was moderately reduced to 78±8% of the control (Figure 3A).

Oral administration of *O. gratissimum* leaf methanolic extract to formaldehyde-induced arthritic mice also caused a moderate reduction in ALT and AST activities compared to the control (Figure 3b). The ALT activity was reduced to 56±6% of the control, while AST was reduced to 64±4% of the control.

Administration of *S. scabrum* leaf methanolic extract also moderately reduced serum ALT concentration (55±2% of control), but the serum AST concentration was only slightly reduced (94±2% of control) (Figure 3C).

DISCUSSION

The results showed that injection of formaldehyde into the mice paw caused paw edema as shown by increased paw thickness (Figure 1 & 2) and increase in the serum concentrations of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) (Figure 3), comparing the 'normal' and 'control' groups. These increases were reversed by oral administration of *O. gratissimum, J. curcas* and *S. scabrum* methanolic leaf extracts, although to different extent.

Increase in the serum concentration of ALT is indicative of damage to some tissues, especially the liver, as the enzyme is largely found in the liver. Such damage causes the release of the enzyme from the damaged cells into the serum. Similarly, AST is an intracellular enzyme found in a variety of tissues, which include the liver, heart, muscle, kidney and brain. Damage to any of these tissues causes the release of the enzyme into the serum. Thus, an increase in the serum level of either ALT or AST is indicative of tissue damage [26, 27]. The increase in the activities of ALT and AST in formaldehyde-induced arthritic mice is therefore not surprising, as inflammation is known to cause tissue damage. The reduction of these enzymes activities to normal or below normal value in the arthritic mice administered with the plant methanolic leaf extracts strongly indicates the presence of anti-inflammatory components in these extracts. Comparing the results of the three plants investigated, when ALT activity and paw oedema are considered, *J. curcas* apparently has the highest anti-inflammatory activity, while *O. gratissimum* and *S. scabrum* exhibited comparable moderate activities. However, the serum AST concentration in *O. gratissimum* treated mice was lower than in those treated with *J. curcas*; treatment with *S. scabrum* resulted in only a slight decrease in serum AST activity.

It may be noted that ALT has been shown to be a more specific and sensitive indicator of liver injury than AST in some animals [27]. Thus the ALT results obtained with these three plant extracts may be reflecting their relative hepatoprotective activities while the AST results may be reflecting the relative effects of these extracts on extrahepatic tissues. Overall, from these results, it can be concluded that *J. curcas* leaf methanolic extract exhibited the highest anti-inflammatory activity, followed by *O. gratissimum* while *S. scabrum* has the least anti-inflammatory activity.

It has been reported that reactive oxygen species (ROS) such as superoxide anion and hydroxyl radicals participate in inflammatory processes in tissues. Therefore, chemicals which have scavenging activities towards these radicals have potentials for the treatment of inflammatory diseases. It may be noted the *O. gratissimum* leaf methanolic extract possesses strong antioxidant activity [18]. Work in our laboratory also showed that *J. curcas* and *S. scabrum* extracts contain strong antioxidant activity.

Evidence suggests that prostaglandins are involved in the processes of inflammation. Cyclooxygenase-1 (COX-1) and cyclooxygenase-2 (COX-2) are enzymes involved in prostaglandin and thromboxane synthesis (from

arachidonic acid) in different tissues [28]. The classic NSAIDS act as non-selective inhibitors of COX-1 and COX-2, thus alleviating inflammation [29]. It is possible that the anti-inflammatory component of *O. gratissimum* leaves may also act by inhibiting prostaglandin synthesis in the arthritic mice. Such anti-inflammatory component may be a regulatory factor in prostaglandin synthesis as the possible presence of prostaglandin synthetase in *O. gratissimum* leaves has been reported [16].

Although *S. scabrum*, at the administered concentration (200mg/kg) seemed to exhibit only a moderate anti-inflammatory activity, since it is a vegetable which can be consumed in large quantity, it is possible that such large quantity may lead to higher anti-inflammatory activity.

CONCLUSION

O. gratissimum, *J. curcas* and *S. scabrum* leaf methanolic extracts all have some anti-inflammatory activities but to different extent. *J. curcas* apparently exhibited the highest anti-inflammatory activity, while *S. scabrum* exhibited the least. However, since *S. scabrum* is normally a vegetable eaten as part of human diet, its moderate anti-inflammatory activity may nevertheless be therapeutically important.

REFERENCES

[1] ED Jr Harris; GS Firestein, In: GS Firestein; RC Budd; ED Jr Harris; IB McInnes; S Ruddy; JS Sergent (Eds.), Kelley's Textbook of Rheumatology, 8th ed., vol. 2, (Saunders/Elsevier, Philadelphia, **2009**), pp. 1087-1118.
[2] LA Garcia-Rodriguez; H Jick, *Lancet*, **1994**, 343(8900): 769-772.
[3] S Hernandez-Diaz; LA Garcia-Rodriguez, *American Journal of Medicine*, **2001**, 110(Suppl. 3A): 20S-27S.
[4] AL Linton, *Canadian Medical Association Journal*, **1984**, 131: 189-191.
[5] IL Meek; MAFJ van de Laar; HE Vonkeman, *Pharmaceuticals*, **2010**, 3: 2146-2162.
[6] A Sofowora, In: Medicinal plants and traditional medicine in Africa. Wiley, Chichester, West Sussex and New York, **1982**, 256 pp.
[7] BN Shah; BS Nayak; AK Seth; SS Jalapure; KN Patel; MA Patel; AD Mishra, *Pharmacognosy Magazine*, **2006**, 2: 77-86.
[8] BN Shah; AK Seth; KM Maheswari, *Research Journal of Medicinal Plants*, **2011**, 5: 101-115.
[9] EM Choi; JK Hwang, *Fitoterapia*, **2004**, 75: 557-565.
[10] KS Prabhu; R Lobo; AA Shirwaikar; A Shawaikar, *The Open Complementary Medicine Journal*, **2009**, 1: 1-15.
[11] MO Ilori; AO Sheteolu; EA Omonigbehin; AA Adeneye, *Journal of Diarrhoeal Disease Research*, **1996**, 14: 283-285.
[12] AM Janssen; JJC Scheffer; L Ntezurubanza; A Baerhem-Suendsen, *Journal of Ethnopharmacology*, **1989**, 26: 57-63.
[13] JC Aguiyi; CI Obi; SS Geng; AC Igweh, *Fitoterapia*, **2000**, 71(4): 444-446.
[14] OO Orafidiya; AO Oyedele; AO Shittu; AA Elujoba, *Phytomedicine*, **2004**, 11(2-3): 249-254.
[15] FD Onajobi; JB Ajayi; VN Nwakolo, *Nigerian Journal of Biochemistry*, **1986**, 3: 65-73.
[16] AJ Famurewa. Ph D thesis, Obafemi Awolowo University (Ile-Ife, Nigeria, **2003**).
[17] FD Onajobi, *Journal of Ethnopharmacology*, **1986**, 18: 3-11.
[18] OG Tade; T Adejorin; AT Sennuga; AC Akinmoladun; AJ Famurewa; EM Obuotor; FD Onajobi, *Ife Journal of Science*, **2008**, 10(1): 11-18.
[19] JA Duke. In: CRC Handbook of Medicinal Herbs. CRC Press, Boca Raton, FL, **1988**, pp. 253–254.
[20] HD Neuwinger, In: African Ethnobotany: Poisons and Drugs, Chapman & Hall, New York, **1996**, pp. 500–509.
[21] O Osoniyi; FD Onajobi, *Journal of Ethnopharmacology*, **2003**, 89: 101-105.
[22] DMR Prasad; A Izam; MdMR Khan, *Journal of Medicinal Plant Research*, **2012**, 6(14): 2691-2699.
[23] RR Schippers, UK Natural Resources Institute/ ACP-EU Technical center for Agricultural and Rural Co-operation, Chatham, UK, **2000**, pp. 147-192.
[24] DA Fontem; RR Schippers, In: GJH Grubben; OA Denton (Eds.), Plant Resources of Tropical Africa (PROTA), vol. 2, (PROTA Foundation, Wageningen, Nertherlands, **2004**), pp.493-498.
[25] R Jain; A Sharma; S Gupta; IP Sahety; R Gabrani, *Alternative Medicine Review*, **2011**, 16(1): 78-85.
[26] DE Amacher, *Regulation Toxicology Pharmacology*, **1998**, 27(2): 119-30.
[27] L Boone; D Meyer; P Cusick; D Ennulat; A Provencher Bolliger; N Everds; V Meador; G Elliot; D Honar; D Bounous; H Jordan, *Veterinary Clinical Pathology*, **2005**, 34(3): 162-188.

[28] LJ Marnett; WR Scott; CG Douglas; SK Amit; AL Cheryl, *Journal of Biochemical Chemistry*, **1999**, 274: 22903-22906.
[29] G Dannhardt; W Kiefer, *European Journal of Medical Chemistry*, **2001**, 36:109-126.