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ANALGESIC AND ANTI-INFLAMMATORY ACTIVITIES OF ASPARAGUS AFRICANUS ROOT EXTRACT

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Abstract

The methanolic extract of the roots of *Asparagus africanus* Lam (Liliaceae) which contains mainly saponins and carbohydrate showed significant analgesic and anti-inflammatory activities (P<0.05) in the tail-flick/hot-plate test and egg albumen-induced rat paw oedema tests that were comparable to the test drugs (morphine 20mg/kg and indomethacin 50mg/kg respectively). These results indicate that the extract possesses analgesic and anti-inflammatory properties.

Key words: Asparagus africanus, roots, analgesic and anti-inflammatory activities

Introduction

Asparagus africanus Lam (Liliaceae) is an erect armed herb that grows up to 5ft high. The plant is widely distributed in tropical Africa. In Nigeria, the plants are known as "Shekan bera" in Hausa and "aluki" in Yoruba (Dalziel, 1956). In traditional medicine, the plant is used for the treatment of headache, backache, stomach pain and as an aid in child birth (Msonthi and Magombo, 1983). The plant is also used for heamaturia, heamorrhoids (Desta, 1993), malaria, lishmaniansis, bilharziasis, syphilis and gonorrhoea (Oketch-Rabah et al., 1992). The root extract is applied externally for the relief of pain, rheumatism and chronic gout (Watt and Breyer-Brandurijk, 1962). It is also used as a diuretic, for sore throat and otitis (Oliver, 1960). Three steroidal saponins have been isolated from the roots of *A. africanus* (Debella et al., 1999).

This study was carried out to ascertain the traditional uses of the roots of the plant as analgesic and antiinflammatory agents.

Materials and Methods Plant Materials

The root of *Asparagus africanus* was collected in the month of November from a bush near Samaru, Zaria. The plant was authenticated at the Herbarium of the Department of Biological Sciences, Ahmadu Bello University, Zaria where a reference specimen (No. 900129) has been deposited. The roots were air-dried for several weeks and then powdered using mortar and pestle.

Extraction

The powdered roots (300g) were defatted using petroleum ether (60-80°C) exhaustively in a soxhlet extractor apparatus for 24 hrs. The marc was then air-dried and extracted exhaustively with methanol for 2 days. The

methanolic extract (ME) was concentrated in a rotary evaporator at reduced pressure to attain a dark brown residue (68.5 g).

Phytochemical tests

The extract (ME) was subjected to phytochemical tests to detect the various types of chemical constituents present using standard procedures (Evans, 1996).

Pharmacological tests Animals

Adult Swiss albino mice (20-25g) and rats (200-250g) of both sexes were used for the tests. The animals were obtained from the animal house of the Department of Pharmacology and Clinical Pharmacy, Ahmadu Bello University, Zaria. They were housed in steel cages under standard conditions, fed with standard pellets and water *ad libitum*. Procedures for animal handling were consistent with international guidelines.

Acute toxicity test

The acute toxicity (LD_{50}) of the extract (ME) was determined in mice by the method of Lorke (1983) using the oral route.

Analgesic activity Tail-flick test

Mice (20-25g) of both sexes were fasted overnight before the study. A tail-flick analgesic meter Ugo Basile was used to measured response latencies as described by Santos et al. (1995). five groups of six animals each were pretreated with 0.2ml normal saliva intraperitoneally to group one as negative control, morphine 20mg/kg subcutaneously to group two as positive control while the remaining three groups received 250, 500 and 1000mg/kg of ME orally respectively. Measurement of responses were taken at time zero, 30, 60, 90, and 120 mins after treatment of the animals with drugs and extract by application of pressure from the analgesiometer onto their tail (1cm from the tip of the tail).

Hot-plate test

Mice (20-25g) of both sexes were fasted overnight before the study. Hot-pate was used to measure response latencies according to the methods of Turner, (1965) and Guzman et al. (2001) with minor modifications. In this study, the hot-plate was maintained at $55 \pm 1^{\circ}$ C and the animals were individually placed on the heated surface. The time in seconds between placement and shaking, paw licking and jumping off the plate was recorded as response latency. Five groups of six animals each were pre-treated with 0.2ml normal saline intraperitoneally to group one as negative control, morphine 20mg/kg intraperitoneally to group two as positive control while the remaining three groups received 250, 500 and 1000mg/kg of ME orally respectively. Measurements were taken at zero, 30, 60, 90 and 120 mins after the treatment of animals.

Anti-inflammatory activity

Anti-inflammatory activity was carried out using the rat paw oedema test according to the method of Winter et al. (1962) and Guzman et a., (2001) with minor modifications. Rats (200-250g) of both sexes were divided into five groups of six animals each. 0.2ml of fresh undiluted egg albumen was injected into the right hind paw of the rats after pre-treatment with 0.2ml of normal saline intraperitoneally to group one as negative control, indomethacin 50mg/kg intraperitoneally to group two as positive control while the remaining three groups received 250, 500 and 1000mg/kig of ME orally respectively.

Paw volumes were measured using Ugo Basile 7140 phethysometer immediately after injection of egg albumen and at 30, 60, 90 and 120 mins after treatment. Percentage oedema inhibition was calculated according to the formula of Barbera et a., (1992).

 $\frac{MV_t}{MV_i} x \quad 100 - 100 = (A)$

= % Oedema inhibition

Where MV_i = mean paw volume at start of experiment

 MV_t = mean paw volume at time 30, 60, 90 and 120 minutes

(A) = % Oedema increase

Statistical analysis

Results were expressed as mean \pm standard error of mean (SEM). Student's *t*-test was used to analyze the data. P<0.05 was considered to be statistical significant.

Result and Discussion

The extraction process yielded 12.83% of the methaolic extract. Phytochemical tests showed the presence of carbohydrates and saponins mainly with small quantity of flavonoids and tannins.

The methanolic extract of the roots of *Asparagus africanus* did not cause any mortality unto a dose of 5000mg/kg orally and was thus considered to be none toxic (Dubois and Geiling, 1959).

Table 1: Effect of ME on response latencies in the tail-flick test

Treatment	Doses	Mean response latencies + SEM					
groups	mg/kg	0	30	60	90	120	
Control	_	4.17 <u>+</u> 0.93	5.58 <u>+</u> 0.80	5.42 <u>+</u> 1.28	6.33 <u>+</u> 1.20	6.25 <u>+</u> 1.29	
Morphine	20	5.17 <u>+</u> 1.99	18.83 <u>+</u> 4. 45*	15.17 <u>+</u> 3.92*	14.25 <u>+</u> 3.25*	11.25 <u>+</u> 3.25*	
ME	250	4.58 ± 0.86	6.75 <u>+</u> 1.84	11.17 <u>+</u> 2.21*	14.17 <u>+</u> 3.53*	10.33 <u>+</u> 3.44*	
ME	500	5.17 ± 0.82	9.67 <u>+</u> 2.80*	15.08 <u>+</u> 4.71*	12.83 <u>+</u> 3.14*	12.08 <u>+</u> 2.35*	
ME	1000	4.42 ± 0.74	$11.25 \pm 0.08*$	15.33 <u>+</u> 5.28*	14.83 <u>+</u> 3.61*	13.25 <u>+</u> 3.43*	

Table 2: Effect of ME on Response Times in the Hot-Plate Test

Treatment	Doses	Mean response latencies <u>+</u> SEM						
groups	mg/kg	0	30	60	90	120		
Control	-	5.33 <u>+</u> 1.03	5.17 <u>+</u> 0.75	5.33 <u>+</u> 2.07	5.5 <u>+</u> 2.35	5.5 <u>+</u> 2.35		
Morphine	20	6.33 <u>+</u> 1.91	34.33 <u>+</u> 4.13*	30.5 <u>+</u> 4.46*	16.17 ± 2.04*	10.5 ± 0.84 *		
ME	250	6.0 <u>+</u> 1.26	9.17 <u>+</u> 2.79*	14.33 ± 2.5*	22.67 ± 8.04*	12.17 <u>+</u> 3.87*		
ME	500	5.67 <u>+</u> 1.21	8.83 <u>+</u> 1.6*	17.5 <u>+</u> 4.37*	28.83 ± 8.73*	17.17 <u>+</u> 7.63*		
ME	1000	6.33 ± 1.21	$19.5 \pm 2.74*$	$24.1 \pm 4.05*$	$30.5 \pm \overline{3.83}$ *	$21.33 \pm 5.35*$		

Table 3: Effects of ME on Egg Albumen – Induced Rat Paw Oedema Test

Treatment group	ment group Doses Mean Paw Volume <u>+</u> SEM (Percentage Oedema Inhibition)					1)
	mg/kg	0	30	60	90	120
Control	-	0.88 <u>+</u> 0.34	1.44 <u>+</u> 0.34	1.71 <u>+</u> 0.32	2.16 <u>+</u> 0.44	2.24 <u>+</u> 0.49
Indomethacin	20	1.01 <u>+</u> 0.20	1.49±0.33(25.33)*	1.70 <u>+</u> 0.37(27.57) *	1.97 <u>+</u> 0.36(34.65)*	2.06 <u>+</u> 0.22(32.73) *
ME	250	0.97 <u>+</u> 0.16	1.51 <u>+</u> 0.28 (12.52)	1.68 <u>+</u> 0.23(22.39)	2.02 <u>+</u> 0.13(25.58)*	2.18 <u>+</u> 0.17(19.46) *
ME	500	0.93 <u>+</u> 0.13	1.42 <u>+</u> 0.24 (17.21)	1.56 <u>+</u> 0.18(24.76)	1.68 <u>+</u> 0.18(44.55)*	1.99 <u>+</u> 0.22(26.25)
ME	1000	0.89 <u>+</u> 0.21	1.31 <u>+</u> 0.4 (25.85)*	1.43 <u>+</u> 0.46(35.68)	1.69 <u>+</u> 0.62(38.20)*	1.99 <u>+</u> 0.67(20.03)

^{* =} Statistical significant (P<0.05) compared to control

The mathanolic extract of the roots of A. africanus showed significant dose-dependent analgesic activity in the tail-flick and hot-plates tests (Tables 1 and 2). Analgesic activity of the extract increases with time while that of the test drug (morphine) decreases with time. The extract has shown central and peripheral analgesic activities since the results showed significant (p<0.05) analgesic activity of the extract at all the doses used in the tests. Agents such as histamine, serotonin and bradykinin are natural pain substances liberated by injured tissues which stimulate the nociceptive mechanism to elicit pain (Collier, 1964). Analgesic agents are known to diminish the perception of pain by depressing the nociceptive mechanism. The extract may possibly have acted as an analgesic agent by antagonizing the activities of the natural pain substances that stimulate the nociceptive mechanism.

The results of the anti-inflammatory effects of the methanolic extract of the roots of analgesic *A. africanus* on egg albumen-induced oedema in rats' hind paws are presented in Table 3. The extract at a dose of 500mg/kg showed the highest percentage inhibition of oedema (44-55%) at 90 mins which was comparable to that of indomethacin 50mg/kg (34.65%) at the same time. Saponins isolated from about 50 plants have been shown to possess anti-inflammatory activities against several experimental models of inflammation in mice and rats (Lacaille-Dubois and Wagner, 1996).

The initial phase of oedema which develops in rats paw after the injection of oedemagenic agents which is known to last for 90 minutes is due to the release of histamine and serotonin. The last phase of the inflammation which last for 5 hrs is due to the release of prostaglandins (DiRosa et a., 1971). Egg albumen is an oedemagenic agent which produces inflammatory reactions through the release of serotonin and histamine. The resulting oedema has been shown to be inhibited by antihistaminic agents (Winter, 1966). The activity of the extract was observed to be more pronounced in the first phase of the rat paw oedema (90 mins) thus suggesting that the extract may possess antihistaminic activity.

Conclusion

Based on the results of the present study it can be concluded that the methanolic extracts of the roots of *Asparagus africanus* has potential dose-dependent analgesic and anti-inflammatory activities. Hence this study has confirmed the use of the plant in traditional medicine as a pain reliever in the treatment of headache, backache, as an aid in child birth and in the treatment of inflammatory diseases such as haemorrhoids, rheumatism and chronic gout.

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