



Uterotonic Properties of the Ethanol Extract of *Bryocarpus coccineus*

S. Amos, L. Binda, O.F. Kunle, C. Wambebe & K. Gamaniel

To cite this article: S. Amos, L. Binda, O.F. Kunle, C. Wambebe & K. Gamaniel (2002) Uterotonic Properties of the Ethanol Extract of *Bryocarpus coccineus*, *Pharmaceutical Biology*, 40:1, 33-38, DOI: [10.1076/phbi.40.1.33.5855](https://doi.org/10.1076/phbi.40.1.33.5855)

To link to this article: <https://doi.org/10.1076/phbi.40.1.33.5855>



Published online: 29 Sep 2008.



Submit your article to this journal [↗](#)



Article views: 179



View related articles [↗](#)



Citing articles: 1 View citing articles [↗](#)

Uterotonic Properties of the Ethanol Extract of *Brysocarpus coccineus*

S. Amos¹, L. Binda¹, O.F. Kunle², C. Wambebe¹ and K. Gamaniel¹

¹Department of Pharmacology and Toxicology, ²Department of Medicinal Plant Research and Traditional Medicine, National Institute for Pharmaceutical Research and Development, P.M.B. 21, Abuja-FCT, Nigeria

Abstract

The ethanol extract of the leaves of *Brysocarpus coccineus* Schum. & Thorn. (Connaraceae) was investigated for uterotonic activity in non-gravid rat uteri. Repeated treatment with the extract enhanced spontaneous uterine muscle contraction. A marked increase was observed on day 14, which was sustained up to day 28. Furthermore, the extract increased the contractile force of the isolated myometrial preparation in a concentration-dependent manner. This effect was not attenuated by atropine, cyproheptadine or piroxicam, but was blocked by salbutamol, suggesting the involvement of β -adrenergic receptors. This result indicates the presence of biologically active substances with comparable effects to oxytocin on the uterus. The intraperitoneal LD₅₀ in mice was found to be 547.7 ± 4.3 mg/kg. Preliminary phytochemical studies revealed the presence of carbohydrates, tannins, flavonoids and balsams.

Keywords: *Brysocarpus coccineus*, rat uterus, adrenergic receptors, uterotonic activity.

Introduction

Brysocarpus coccineus Schum. & Thorn. (Connaraceae) is a shrub that is distinctively decorative and of great medicinal value in West Africa. It is widely used in ethnomedicine for the treatment of ear ache. In Ghana, the roots and a decoction of the leaves are applied to mouth sores as well as sores on the skin. The root bark, with black pepper (*Piper guineense*), is used to cure wounds. The plant is also used in the treatment of jaundice, tumors and inflammation, to arrest bleeding, swelling, pile, venereal disease and as an antidote for poisoning. In Nigeria, a cold infusion of the bruised leaves is used for the treatment of gonorrhoea (Dalziel, 1937;

Irvine, 1961). The leaves have also been used traditionally in Northern Nigeria to induce labour and cause therapeutic abortion in Kogi State (Yakubu Habi – personal communication). From the available literature, no scientific report has been made on its oxytocic activity. In our effort to screen potential herbal medicines in Nigeria, we report in this paper the uterotonic activity of a 70% ethanol extract of the plant leaves. The oxytocic activity of the plant extract was evaluated using the isolated rat uterus and the effect on spontaneous contraction of the uteri following daily administration of the plant extract.

Materials and methods

Drugs

The drugs used in this present work were acetylcholine, atropine sulphate, oxytocin, ergometrine, indomethacin (Sigma Chemical Company, MO, USA), cyproheptadine (MSD, Lagos, Nigeria) and stilboestrol dipropionate (May and Baker, Dagenham, England).

Collection, identification and extraction

Leaves of *Brysocarpus coccineus* were collected from around Idu, Abuja, Nigeria. The plant was identified and authenticated by Mr. A. Oheari of the Department of Medicinal Plant Research and Traditional Medicine of the National Institute for Pharmaceutical Research and Development where a voucher specimen was deposited for future reference. The fresh leaves were washed clean with water, air-dried for 7 days and subsequently reduced to coarse powder using a mortar and pestle. The powder (83 g) was

Accepted: August 14, 2001

Address correspondence to: Samson Amos, Department of Pharmacology and Toxicology, National Institute for Pharmaceutical Research and Development, P.M.B. 21, Abuja-FCT, Nigeria. Tel: 234-9-5249089, Fax: 234-9-5231043, E-mail: samsonamos@yahoo.co.uk

extracted in a Soxhlet apparatus using 70% alcohol for a period of 8h. The extract was concentrated to dryness *in vacuo* under reduced pressure using a rotary evaporator to give a residue with a yield of 11.73% (w/w).

Phytochemical screening

Standard screening for various constituents, alkaloids, flavonoids, glycosides, etc., were carried out on the extract

following the methods described by Trease and Evans (1983).

Acute toxicity studies (LD₅₀)

The intraperitoneal (i.p.) acute toxicity (LD₅₀) study of the extract was carried out in Swiss albino mice as described by Miller and Tainter (1944). Briefly, the method involved administration of 5 different doses of the extract to 5 groups

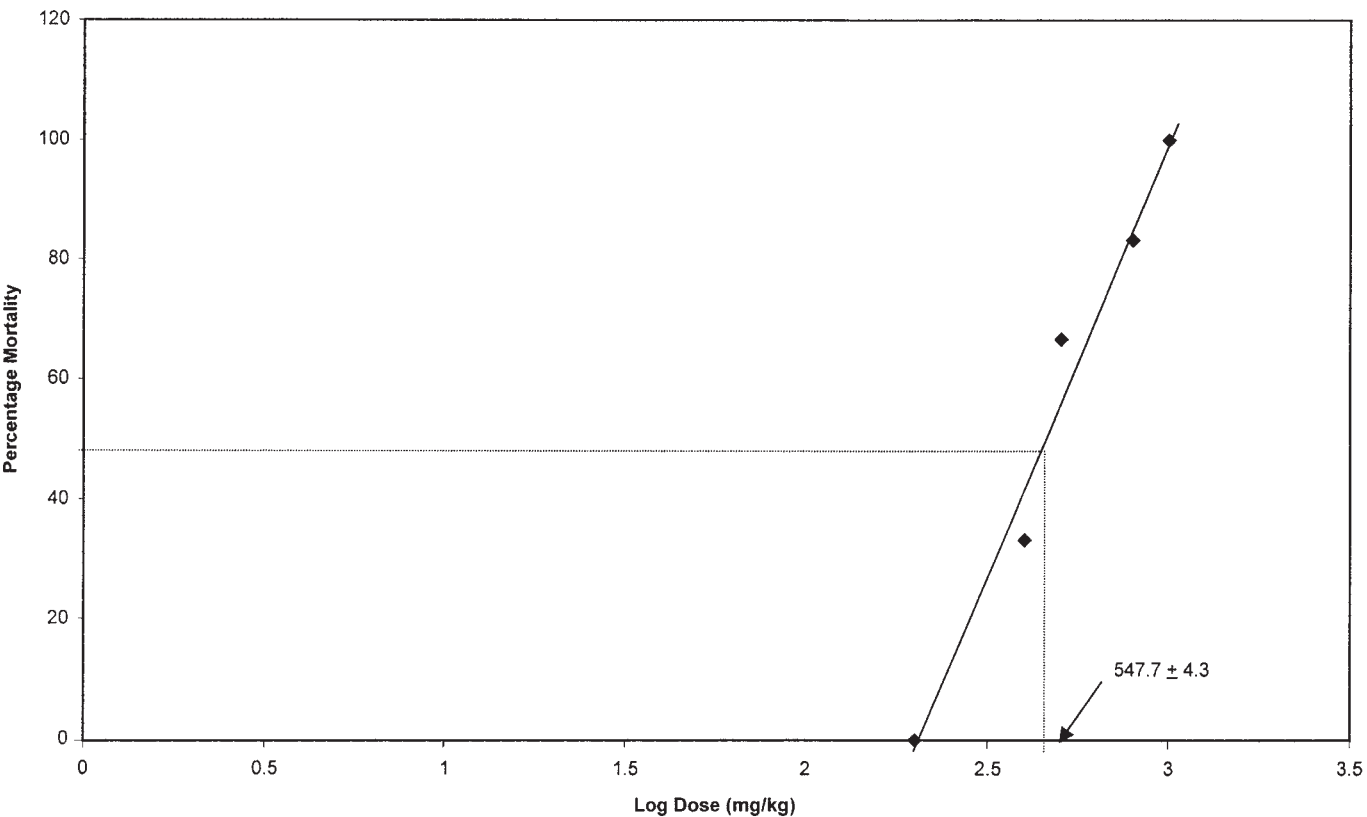


Figure 1. Graph showing the percentage mortality against log dose of the extract of *B. coccineus*.

Table 1. Phytochemical Screening of *Brysocarpus coccineus* leaf Extrart.

Tests	Observation	Inference
Sample + Molisch reagent + concentrated Sulphuric acid	Purple ring at interface	Carbohydrate present
Sample + Fehling’s Solution A & B + heat	Brick red precipitate	Reducing sugars present
Sample + Ferric Chloride	Green colour	Tannins present
Sample + lead sub acetate	Precipitate formed	Tannins present
Sample + glacial acetic acid + ferric chloride, poured over concentrated sulphuric acid	No blue layer	Negative for digitalis glycoside
Sample + water + vigorous shaking	No froth formed	Negative for saponins
Sample + lead acetate	Yellow precipitate	Flavonoid present
Sample + amyl alcohol	Yellow colour	Flavonoid present
Sample + alcoholic ferric chloride	Dark green colour	Balsam present
Sample + Dragendorff’s reagent	No precipitate	Negative for alkaloids
Sample + Picric Acid	No colour change	Negative for alkaloids

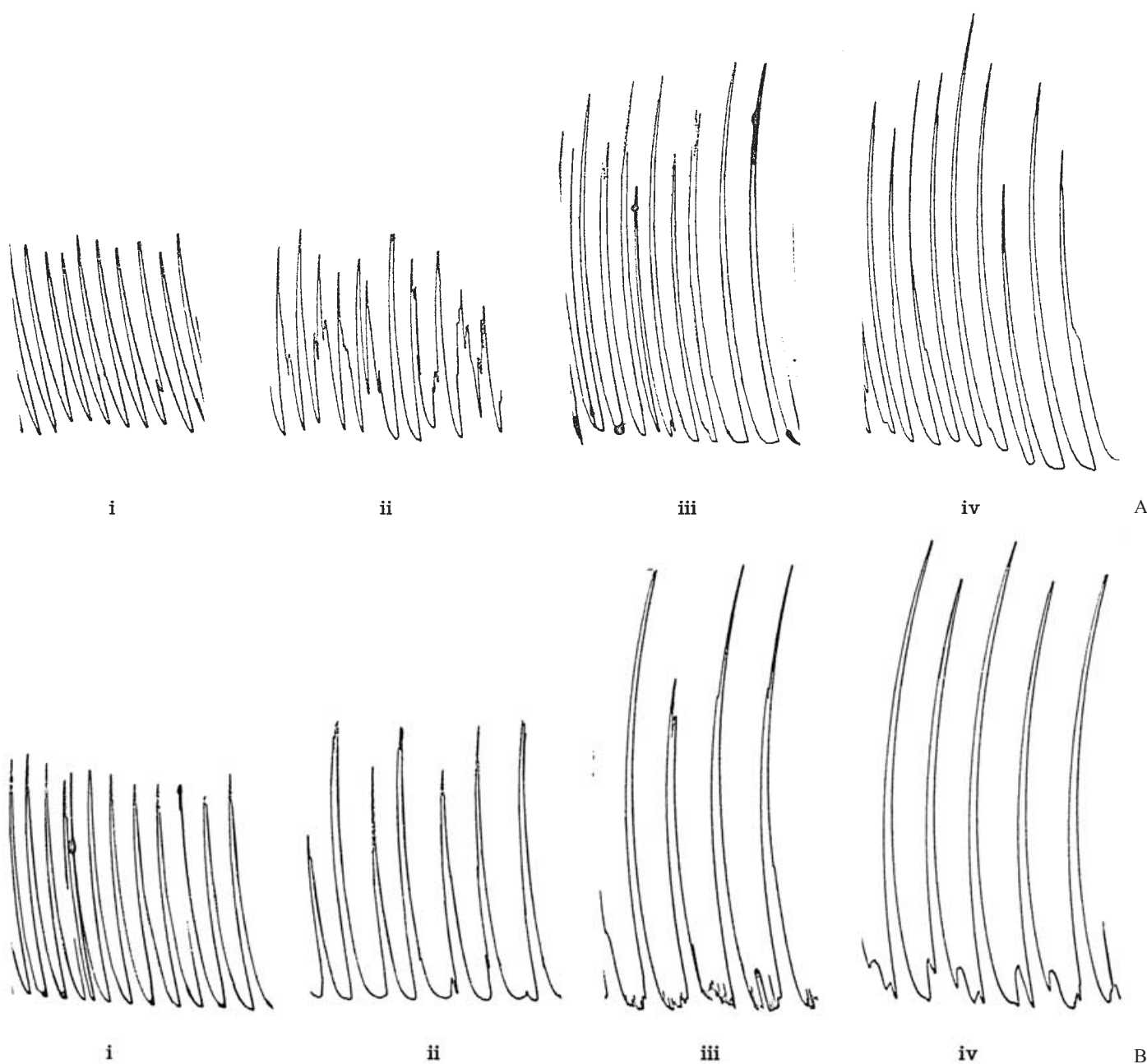


Figure 2. Typical tracings of the effect of ethanolic extract of *B. coccineus* (A: 25 mg/kg) and (B: 50 mg/kg) pretreatments (i) for day 0 (control); (ii) for 7 days; (iii) for 14 days and (iv) for 28 days on uterine contractions *in vivo*.

of mice (6 mice/group). The mortality in each group was recorded within 24 h. The (LD_{50}) was estimated from the graph of percentage (%) mortality (converted to probit) against log-dose of the extract probit 5 being 50%.

Experimental protocol

Daily doses of 25 and 50 mg/kg of the leaf extract were given orally to 2 groups of 10 female virgin Wistar rats weighing 100–120 g. A third group was given isotonic saline (0.9%

NaCl). The rats were killed on day 0, 7, 14 and 28 by decapitation. The effect of repeated treatments on the spontaneous contractions of the uteri in De Jalon's solution aerated with air, maintained at a temperature of $37 \pm 1^\circ\text{C}$ were recorded isometrically on a Ugo Basile Unirecorder 7050. A basal tension of 1 g was applied throughout the period of the experiment.

In another experiment, female Wistar rats weighing 150–180 g were pretreated with stilbestrol, 1 mg/kg subcutaneously, 24 h before the experiment. The rats were killed by

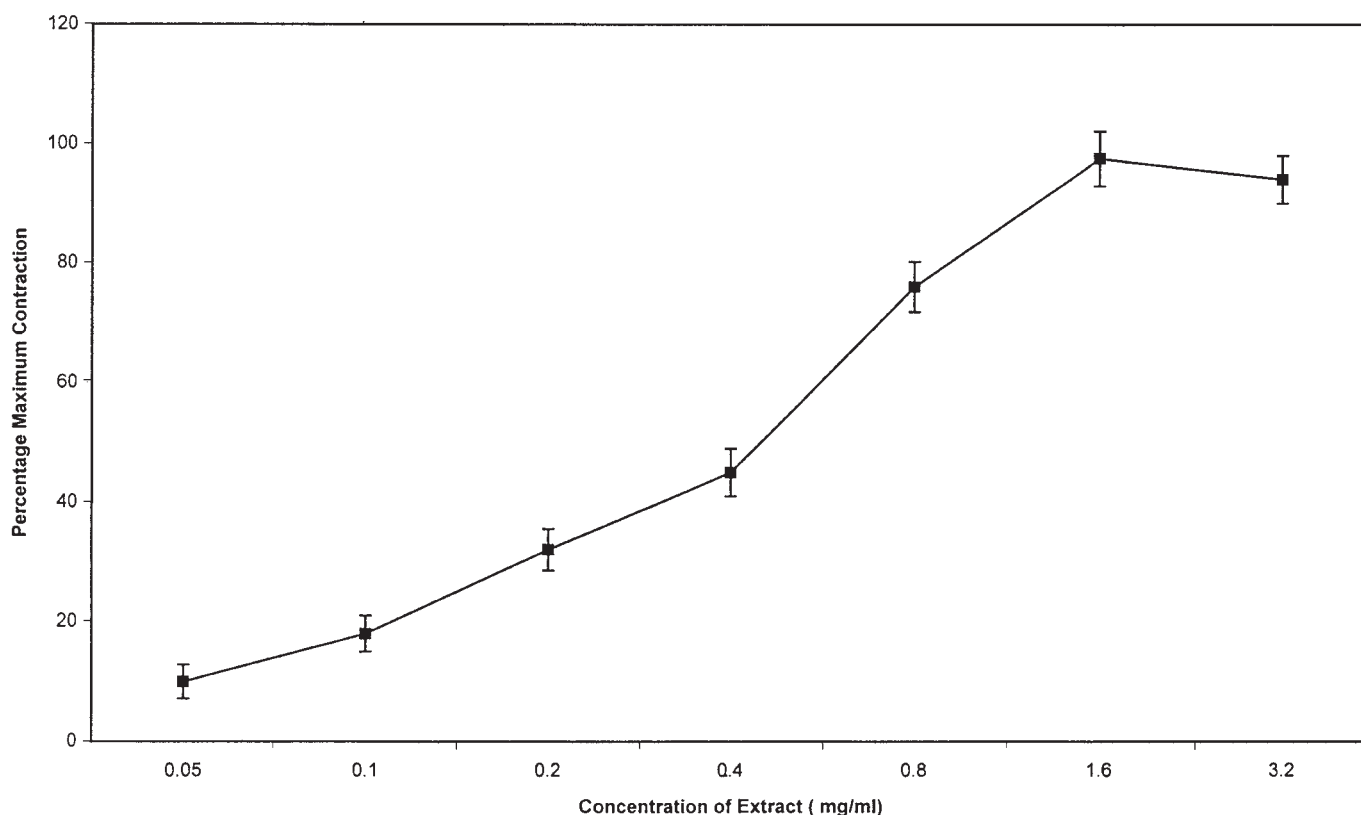


Figure 3. Concentration-dependent contraction of *B. coccineus* (0.05–3.2 mg/ml) on the isolated rat uterus.

cervical dislocation and exsanguinated. The uterus was isolated and the uterine horns dissected free of adhering tissues. A length of about 2 cm was obtained and mounted in a 20 ml organ bath containing De Jalon's solution of the following composition (mM): NaCl 154, KCl 5.63, CaCl₂ 0.41, NaHCO₃ 1.7, MgCl₂ 1.4 and D-glucose 5.5, maintained at $37 \pm 1^\circ\text{C}$ and aerated with air. A tension of 1 g was applied throughout the experiment. The inhibitory effects of atropine ($5 \times 10^{-9}\text{M}$), piroxicam ($1 \times 10^{-3}\text{M}$) cyproheptadine ($1 \times 10^{-8}\text{M}$) and salbutamol ($2 \times 10^{-8}\text{M}$) on the extract-induced contraction of the rat uterus were evaluated. Furthermore, the effect of cyproheptadine ($1 \times 10^{-8}\text{M}$) on ergometrine ($1.5 \times 10^{-5} - 1.2 \times 10^{-4}\text{M}$) and 5-HT ($7.5 \times 10^{-4} - 5.6 \times 10^{-4}\text{M}$) induced responses were investigated. The responses were recorded isometrically on an Ugo Basile Unirecorder 7050 through an isometric transducer 7004 after 1 h equilibration period.

Statistical analysis

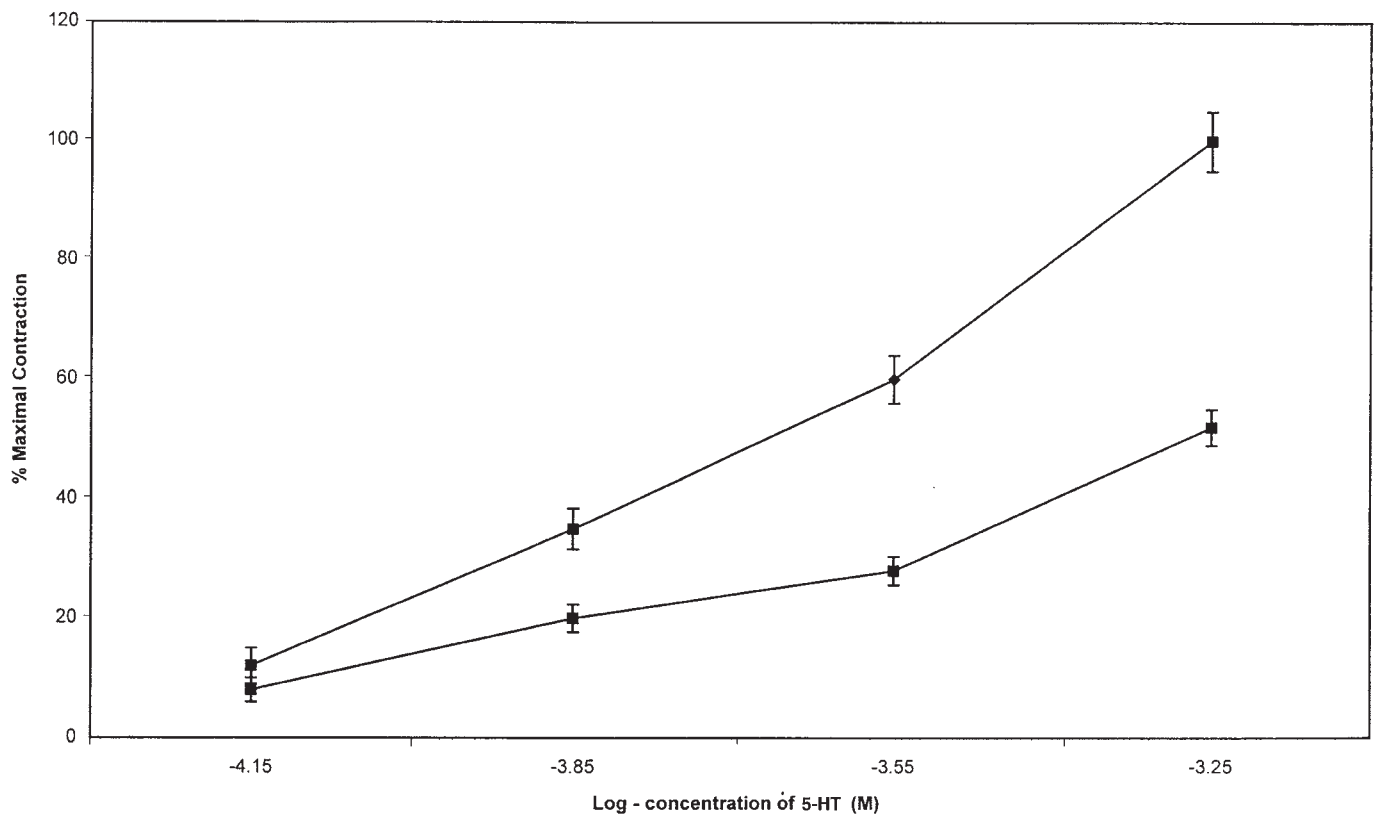
Data obtained were expressed as mean \pm SEM. The results obtained were analyzed statistically using the Student's *t*-test. The level of significance was $p < 0.05$.

Results and discussion

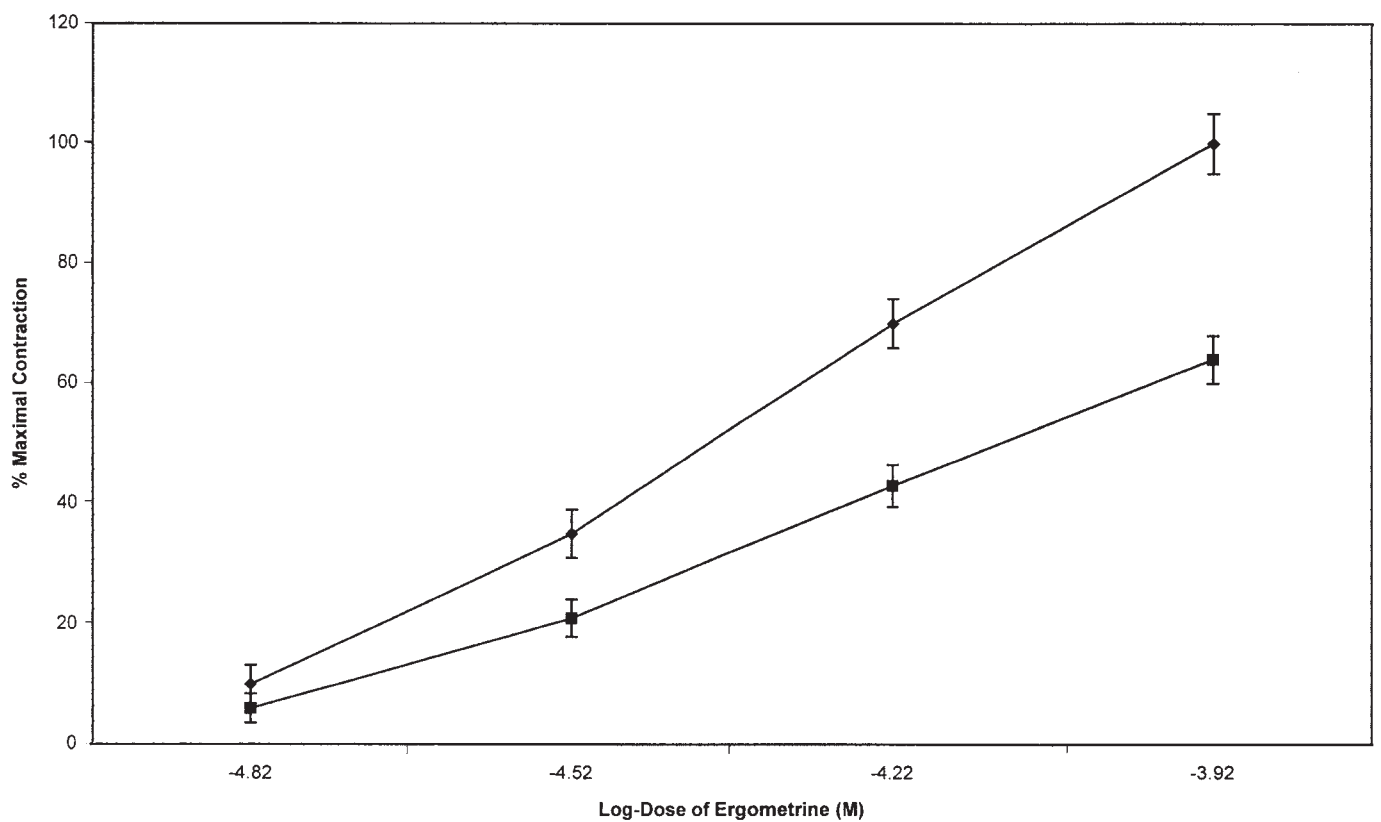
The intraperitoneal LD₅₀ of the extract was found to be 547.7 \pm 4.3 mg/kg (Fig. 1). Preliminary phytochemical screening

gave positive reactions to carbohydrates, tannins, flavonoids and balsams (Table 1). Daily doses of 25 and 50 mg/kg of *B. coccineus* was found to enhance spontaneous uterine and muscle contraction of virgin female rats causing a graded increase in the force of contraction with respect to duration of treatment (Fig. 2). The extract produced an initial enhancement up to day 14 of treatment and this was sustained up to day 28.

On the isolated uterine preparations, the extract evoked a concentration-dependent increase in the amplitude of contractions (Fig. 3). This effect was similar to the findings of Uguru et al. (1998) on the effects of acetylcholine, oxytocin, ergometrine and 5-HT on the rat uterus. Atropine ($5 \times 10^{-9}\text{M}$), a known muscarinic blocker (Bolton, 1979), did not attenuate the contractions induced by the extract, but completely blocked the effects of acetylcholine, suggesting the lack of involvement of cholinergic receptors in the observed effects. Cyproheptadine ($1 \times 10^{-8}\text{M}$), a 5-HT blocker (Hashimoto et al., 1977; Hartig, 1989), blocked the effect of 5-HT and ergometrine (Figs. 4A,B) but produced no effect on the extract induced contraction of the rat uterus, indicating that 5-HT receptors might not be involved in the observed effect. Piroxicam ($1 \times 10^{-3}\text{M}$) is one of the most widely used non-steroidal anti-inflammatory drugs which produce their actions primarily through the inhibition of prostaglandin biosynthesis (Vane, 1971; Gamani et al., 1988). The drug did not attenuate extract induced contraction of the uterus.



A



B

Figure 4. Inhibitory effects of cyproheptadine (\blacksquare , 1×10^{-8} M) on (A) 5-HT (\blacklozenge , 7.5×10^{-4} – 5.6×10^{-4} M) and (B) ergometrine (\blacktriangle , 1.5×10^{-5} – 1.2×10^{-4} M).

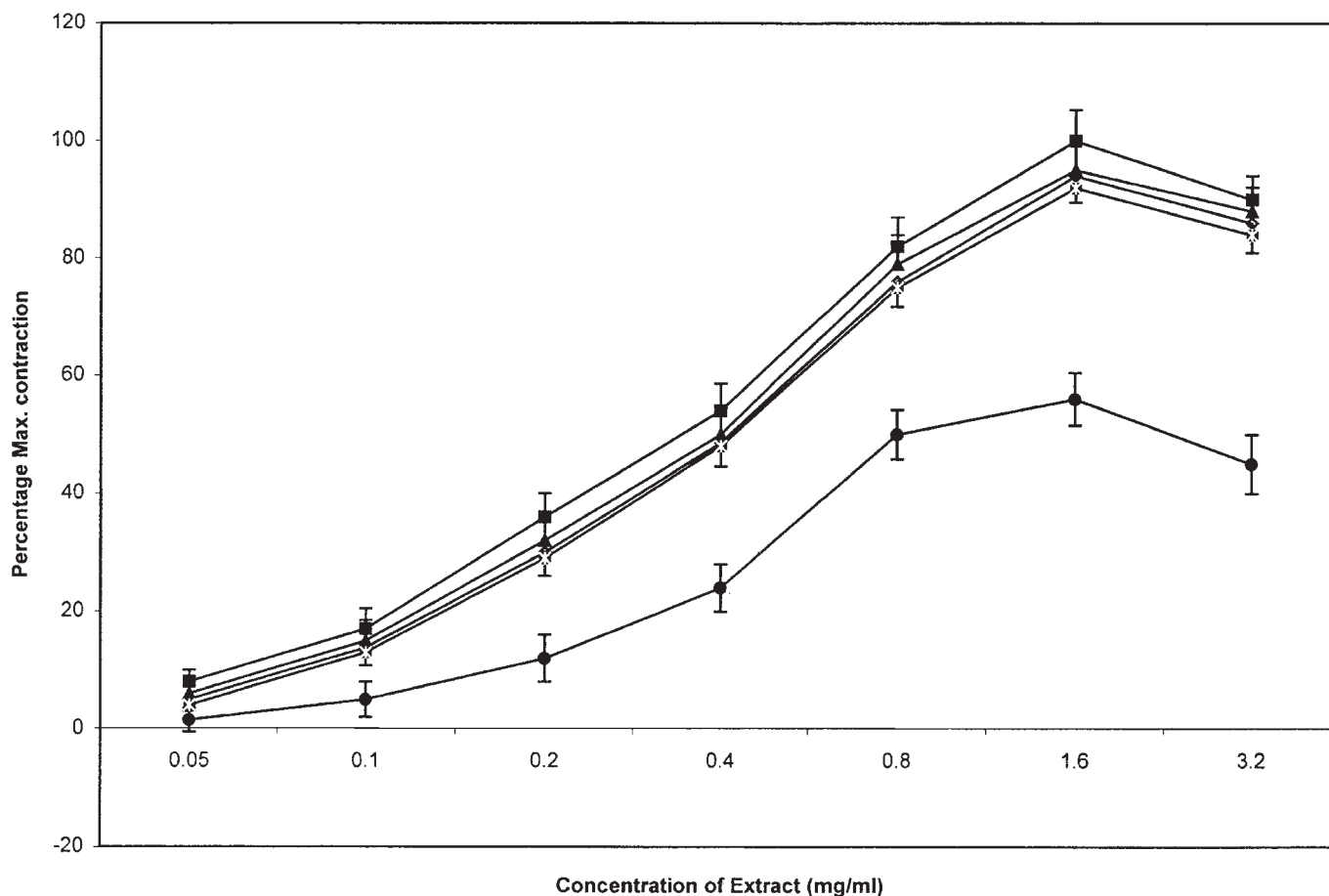


Figure 5. Contraction response curves to ethanolic extract of *B. coccineus* alone (■) and in the presence of (▲) atropine 5×10^{-9} M, (◇) piroxicam 1×10^{-3} M, (✕) cyproheptadine 1×10^{-8} M and (●) salbutamol 2×10^{-8} M.

The response observed with both oxytocin and the extract were abolished by the β_2 agonist salbutamol (Fig. 5). These effects might have been achieved via physiological antagonism (Rang et al., 1995). The extract and salbutamol may therefore physiologically antagonize the effects of each other similar to oxytocin. Work is ongoing in our laboratory to isolate and characterize the active compound with possible application in humans.

Acknowledgement

The authors gratefully acknowledge the technical assistance of David Akumka, Hauwa Abdullahi and Charles Balogun for typing the manuscript.

References

- Bolton TB (1979): Cholinergic mechanisms in smooth muscles. *Brit Med Bull* 35: 275–283.
- Dalziel JM (1937): *The Useful Plants of West Tropical Africa*. The Crown Agents for the Colonies, p. 343.
- Gamaniel K, Wuorela H, Metsa-Ketela T, Vapaatalo H (1988): Influence of piroxicam on the contractile action of prostaglandin E_1 on the isolated rat uterus. *Meth & Find Exptl Clin Pharmacol* 10: 493–496.
- Hartig PR (1989): Molecular biology of 5-HT receptors. *Trends Pharmacol Sci* 10: 64–69.
- Hashimoto H, Hyashi M, Nakahara Y, Niwaguchi T, Ishii H (1977): Actions of D-lysergic acid diethylamide (LSD) and its derivatives on 5-HT receptors in the isolated uterine smooth muscles on the rat. *Eur J Pharmacol* 45: 341–348.
- Irvine FR (1961): *Woody Plants of Ghana*. Oxford, Oxford University Press, pp. 568–570.
- Miller LC, Tainter ML (1944): Estimation of LD_{50} and its errors by means of log probit graph paper. *Proceed Soc Exper Biol & Med* 57: 261–264.
- Rang HP, Dale MM, Ritter JM (1995): *Pharmacology*. Churchill Livingstone, USA.
- Trease GE, Evans MC (1983): *Textbook of Pharmacognosy*. Bailliere, Tindall, London, pp. 343–383, 12th Edition.
- Uguru MO, Okwuasaba FK, Ekwenchi EE, Uguru, VE (1998): Uterotonic properties of the methanolic extract of *Monochroma ciliatum*. *Ethnopharmacol* 62: 203–208.
- Vane JR (1971): Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. *Nat New Biol* 231: 232–235.