SOME PHARMACOLOGICAL EFFECTS OF DESMODIUM ADSCENDENS ON THE CONTRACTED TRACHEA IN VITRO AND IN VIVO AND SPONTANEOUSLY BEATING RIGHT ATRIUM OF THE GUINEA-PIG

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ABSTRACT

Pharmacological Studies have been carried out on a crude, aqueous extract of Desmodium adscendens, an anti-asthmatic traditional medical remedy in West Africa [1, 2]. In these studies, carbachol-induced contractile response on the guinea-pig tracheal chain, the isolated right atrium of the guinea-pig and antigen-induced bronchoconstriction in the conscious guinea-pig were used.

The extract caused a dose-dependent relaxation of the tracheal chain that did not involve ß-adrenoceptor activation. In a few preparations, the effect was biphasic. In the spontaneously beating guinea-pig atrium, the extract produced a depressant effect. On the conscious guinea-pig, the extract had a protective effect on antigen-induced broncho constriction. Compared with isoprenaline as a standard bronchodilator drug, however, an aqueous extract of the plant extract was found to be much less potent than the sympathomimetic.

Keywords: *Desmodium adscendens, isoprenaline, right atrium, tracheal chain, bronchoconstriction, relaxation.*

INTRODUCTION

Desmodium adscendens (Papilionaceae), an undergrowth shrub, is a common traditional medical remedy in West Africa for bronchial asthma [1, 2]. It has also been reported that the leaf decoction is drunk for constipation, the leaf infusion as a bath for convulsions and venereal sores and taken internally for ringworm in Liberia [1].

Since previous reports indicated the effectiveness of aqueous extracts of Desmodium adscendens in the clinical management of asthma at the Centre for Scientific Research into Plant Medicine (CSRPM) at Mampong Akwapim in Ghana [2] a number of phytochemical and pharmacological investigations of the plant have been carried out [3, 4].

The present studies have been carried out on a crude aqueous extract of the plant using the carbachol-induced contractile response of the guinea-pig tracheal chain preparation, and the antigen-induced bronchoconstriction in the conscious guinea-pig, to investigate possible anti-asthmatic properties of the plant; the effect was also observed on the isolated right atrium of the guinea-pig.

MATERIALS AND METHODS

PREPARATION OF THE PLANT EXTRACT

The leaves and stems of the plant from the CSRPM and the UST campus were sun-dried for a week. They were pulverized and 20g of the material were extracted continuously for 48 hours with 150ml distilled water in a Soxhlet extractor. The solutions were used freshly prepared or stored in a refrigerator at a temperature of 5-10°C. The concentration of the extract was expressed in terms of the dried weight of the pulverized plant material in solution which gave 133mg/ml.

PREPARATION OF THE TISSUE

i) Tracheal Chain:

Guinea-pigs of either sex weighing about 300g were used. They were killed by a blow on the head and the throat was cut as near the head as possible. The trachea was dissected and transferred to a dish containing Krebs' solution. A length of the trachea measuring about 12.5cm was opened by making a longitudinal cut through it at a point diametrically opposite the strip of smooth muscle. Opened rings of about 2mm in width were cut and sewn end to end, each preparation having five to six such rings. Each set of tracheal chain was mounted in Krebs' solution in an organ bath at 37°C and continuously aerated. Tension applied to the preparation was 0.2 - 0.5g. One end of the tracheal chain was attached to a fixed pin in the bath and the other to a frontal writing lever on a smoked drum. The tissue was allowed to equilibrate for ½ to one hour after which the drugs were investigated on it. This method was a modification of that used by Castillo and de Beer [5].

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In order to induce tone in the tissue, for the experiment, a suitable submaximal dose, $1.6 \times 10^{-5}\mu$ of carbachol was added. At a constant level of tone, $10^{-8}\mu$ isoprenaline was added cumulatively and in a geometric progression until complete relaxation was obtained.

After the tissue was washed and allowed to rest for 30 minutes, the experiment was repeated using the aqueous extract of D. adscendens in place of isoprenaline.

ii) Spontaneously beating right atrium:

Guinea-pigs of either sex were used. They were killed by a blow on the head and the throat was cut. The chest was opened and the heart removed as quickly as possible and placed in a Ringer Locke solution at room temperature. The right atrium was isolated, and mounted in a bath containing Ringer Locke solution at 30°C and aerated with oxygen. The tissue was left to equilibrate for ½ to one hour.

After normal rate and amplitude of the beats had been recorded for 5 minutes, increasing amounts of isoprenaline (Starting with $2.4 \times 10^{-8}\mu$) and 0.01ml the aqueous extract of Desmodium were investigated on it.

The effect of the plant extract on the atrium in the presence of isoprenaline was also investigated.

iii) Antigen-induced bronchoconstriction in the guinea-pig:

Twelve guinea-pigs of either sex weighing between 250 and 350g were used. They were sensitized by a single intraperitoneal injection of 1mg crystallized crude egg albumen (as antigen) contained in 1ml normal saline (0.9% sodium chloride).

After 28 days of sensitization, the guinea-pigs were randomized into three groups of 4 per group.

The first group served as control and were each given an intraperitoneal injection of 1ml/kg saline.

The second group received an intraperitoneal injection of 0.3mg/kg isoprenaline and the third group were each given an intraperitoneal injection of the extract of Desmodium adscendens in a dose equivalent to 46mg/kg of the powdered drug.

Fifteen minutes after injection of the drugs, all the animals were challenged by exposure to the antigen aerosol (i.e. 1% w/v crystallized egg albumen in normal saline) in an airtight perspex gas chamber.

Histamine-induced bronchospasm:

During the period of exposure to the aerosol of the antigen solution, the animals were closely observed for signs of respiratory distress or dyspnoea such as rippling and twitching of the body and eventual collapse. The experimental method used for this work was based on the work of Carney *et al* [6]. The times taken for the appearance of the first signs of respiratory distress as well as for complete collapse of the animal were noted in each case.

The degree of hypersensitivity and drug protection was a function of the time of exposure required to induce:

(a) recognizable signs of the onset of respiratory distress manifested as a rippling spasm of the body or as a cough, this being the preconvulsion time (PCT) of Herxheimer [7].

(b) The prostration of the animal or collapse time (CT).

DOSES OF DRUGS

Doses of isoprenaline and the plant extract used in this experiment were based on the therapeutic doses used in man. The average dose of isoprenaline was taken as 20mg and that of the extract as 3g based on information from the CSRPM.

Taking the average weight of the adult as 65kg, this works out as 0.3mg/kg and 46mg/kg for isoprenaline and the extract respectively. These values were extrapolated to the guinea-pig.

RESULTS

Tracheal chain

Whereas isoprenaline produced a relaxation of the tissue in all cases, the plant product produced relaxation in about 80% of the experiments and a biphasic effect, that is, relaxation followed by contraction, in the remaining cases. The relaxant actions of both isoprenaline and the extract were dose-dependent but the latter was not inhibited by propranolol. Fig.1 shows the dose-response effects of isoprenaline on the tissue and Fig.2 the dose-response effects of the extract on it.

The lower tracing in Fig.2 shows the biphasic effect of the plant extract. Fig.3 shows the log dose-response curves for isoprenaline and the plant extract on the tissue with the ED_{50s} for both Isoprenaline and Desmodium being 8.9×10^{-5}mg and 7.5mg respectively with corresponding slopes of 1.4 and 2.2. The ED_{50} dose ratio of desmodium to isoprenaline was 8.4×10^{4}

Spontaneously beating atrium:

Isoprenaline had a stimulant action on the spontaneously beating atrium but the extract depressed it. Both the plant extract and propranolol inhibited the stimulant action of isoprenaline on the tissue.

Inhibition of bronchospasm in the conscious guinea-pig

The results are shown in Fig.4. There was a significant difference between the time of onset of respiratory crises in the saline pretreated (i.e. control) and the

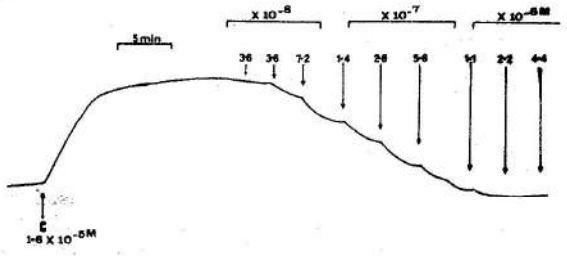

Fig. 1 Cumulative Dose-response Effects of Isoprenaline on the Guinea-pig Tracheal Chain. Carbachol was added. Carbachol was added at C.

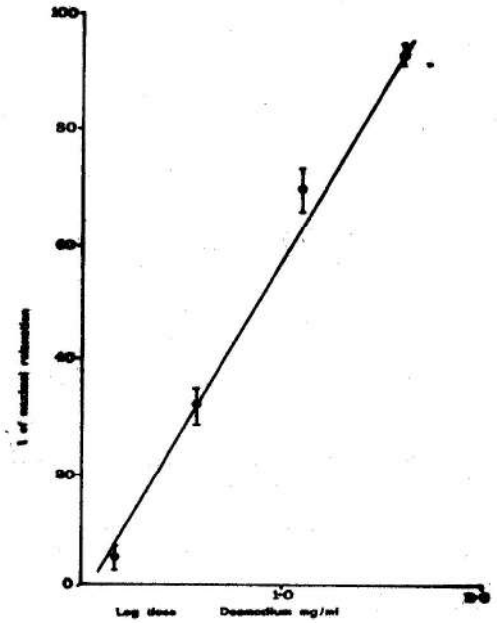

Fig. Dose-response Curve for Desmodium Extract on relaxation of Carbachol-Induced Tracheal Chain Preparation. n = 4.

extract and the isoprenaline-pretreated groups ($p < 0.05$) Similarly, there was a significant difference between the time of onset of respiratory crises between the extract- and isoprenaline-pretreated groups ($p < 0.05$).

DISCUSSION

The guinea-pig tracheal chain preparation is one of the models for studying the effects of drugs on the trachea [5]. Since bronchoconstriction is the principal causative factor of asthma, tone was induced in the tracheal

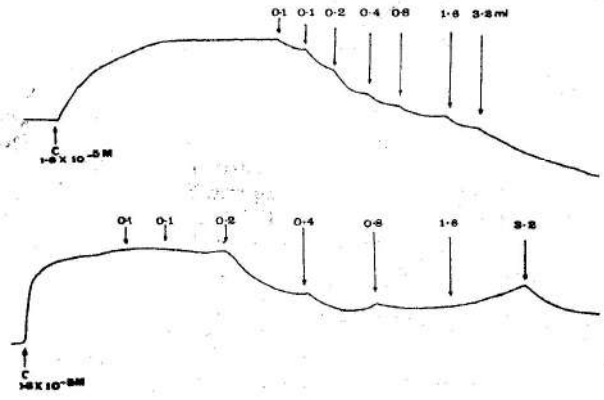

Fig. 2 Cummulative Dose-response Effects of Desmodium extract on the Guinea-pig Tracheal Chain. the Lower tracing shows the biphasic effect of the extract. Carbachol was added at C.

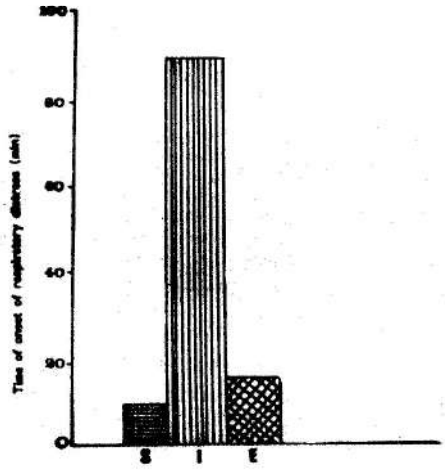

Fig. 4 Histogram of Protective Effects of Isoprenaline and Desmodium Extract on Antigen-Induced Bronchoconstriction in the Guinea-pig. $p < 0.05$.

S = Normal Saline, I = Isopenaline, E = Desmodium

preparation to simulate the bronchoconstrictor component of an asthmatic attack. A herbal preparation which is used in the treatment of bronchial asthma would be expected to inhibit at least one or some of the aetiological factors associated with the disease. Carbachol as a spasmogen was chosen because it produced a constant effect that is stable. Isoprenaline is an established bronchodilator and a standard one for the evaluation of bronchodilator activity.

Whereas isoprenaline has been shown to cause gradual relaxation of the preparation throughout, the plant extract, on the other hand, caused a gradual relaxation in about 80% of the preparations and a biphasic effect, i.e. relaxation followed by contraction, in about 20% of the cases studied.

The isoprenaline effect was inhibited by a non-selective blocking agent propranolol which did not affect the action of the plant extract on the trachea. The fact that the tracheal action of the extract was not antagonised by propranolol indicates that its action on the tissue was not mediated by β adrenoceptor activation.

The biphasic action of the extract is consistent with the observation of Addy [3] who investigated the inhibitory effects of the aqueous and ethanolic extracts of D. adscendens on induced anaphylaxis on the isolated guinea-pig ileum. Ampofo [8], in a trial of the plant extract on 12 asthmatic patients reported 75% success.

The pharmacological actions of the drug can be attributable to its constituents which included glycosides, saponins and alkaloids [4].

The anti-anaphylactic action of Desmodium extract has been reported by several workers including Addy [3] and Addy & Awumey [9]. Asante-Poku, Sakakibara & Addy [4] reported the isolation of five alkaloids from the stem-leaves of the plant. Addy & Burka [10] using flash chromatographic methods reported the isolation of certain active fractions from the plant extract and suggested that the pharmacological actions of the extract might be due to the modulation of arachidonic acid metabolism, possibly by affecting both the cyclo-oxygenase and lipoxygenase pathways with the subsequent production of prostaglandins, thromboxanes and leukotrienes.

The cardiodepressant action on the atrium might be due to the accumulation of glycosides in the heart muscle as previously reported from chronic toxicity studies in the rat [11].

On the whole, the results of our preliminary pharmacological investigations of D. adscendens justify its continued use in the management of bronchial asthma.

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