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BRIEF COMMUNICATION

Local Anesthetic Effects of Cocaine and Several Extracts of the Coca Leaf (*E. coca*)[1]

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BEDFORD, J. A., C. E. TURNER AND H. N. ELSOHLY. *Local anesthetic effects of cocaine and several extracts of the coca leaf* (E. coca). PHARMACOL BIOCHEM BEHAV 20(5) 819–821, 1984.—Cocaine and a number of different fractions of a crude ethanol extract of the coca leaf *(E. coca)* were subjected to a local anesthetic screen using rat tail withdrawal from electric shock. Following an intradermal injection of 0.1 ml of a 2.0% (w.v) solution of cocaine HCl, an immediate response was observed. Two of the coca fractions also produced some local anesthesia. An alkaloidal fraction, containing an equivalent amount of cocaine, produced a maximum effect that was approximately 20% less than that observed with cocaine. The only other fraction producing any effect, a water soluble cocaine-free fraction, showed a maximum response that was approximately 30% of that observed with cocaine.

Local anesthesia Coca Cocaine Rat

FOR the past several years our laboratory has been engaged in the process of determining the biological effects of several different fractions of an ethanol extract of the coca leaf. Since cocaine (the major plant alkaloid) has been and continues to be one of the primary drugs of abuse, our efforts to date have been primarily concerned with the behavioral pharmacology of the fractions as they relate to the known effects of cocaine. We have demonstrated substantial anorexic activity in both cocaine and non-cocaine containing fractions [2]. We have also reported [3] on a water soluble cocaine free fraction that, like cocaine, reduced food consumption but unlike cocaine does not appear to have any central nervous system stimulant effects. Finally, we recently reported [4] on a water soluble cocaine free fraction that alters food reinforced bar pressing but does not generalize to cocaine in a discrimination procedure.

Aside from its prominent effects on the central nervous system, cocaine also possess potent local anesthetic and vasoconstrictor activity. Although originally used extensively as a local anesthetic in ophthalmic surgery, cocaine has been replaced by several synthetic local anesthetics (e.g., procaine, lidocaine and others) because it was found that repeated use may be followed by the toxic effects of clouding, pitting and ulceration of the cornea [1]. The potent vasoconstrictor activity and the local anesthetic activity of cocaine are the only medical uses of the drug today. It is routinely used in surgical procedures involving the nose, throat, larynx and lower respiratory passages because blood loss from these highly vascularized areas can be considerable. In addition, excess bleeding may obscure the operative field.

Because local anesthesia is one of the more prominent effects of cocaine, we felt it appropriate to screen the several fractions studied thus far for their local anesthetic activity. In addition, by comparing the cocaine containing fractions to cocaine alone, we may obtain preliminary information concerning plant constituents that alter the effect of cocaine.

METHOD

Subjects

The subjects (n=56) were male, Wistar rats (Harlan Industries) weighing between 200–250 g. Subjects had ad lib access to water and chow. Ambient temperature was maintained at 20±2°C and the light/dark cycle was 12 hr on/12 hr off. Subjects were housed individually in galvanized steel suspension cages.

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[2]Leave of absence.

Apparatus

The local anesthetic apparatus used has been described elsewhere [5]. Briefly, the subjects are restrained in a standard tubular Plexiglas rat restrainer (Fisher Scientific) with the tail protruding from one end. The subject's tail is connected to a strain gauge by a piece of No. 3 surgical silk. The strain gauge is then set to close a relay when the rat exerts a force of 100 g. Shock is supplied to the rats tail from a constant current shock generator (BRS-LVE) by stainless steel needle electrodes placed intradermally (depth of penetration 2 mm) 3 mm apart.

Solution Preparation

Solutions were prepared on the morning of use. The analytical procedure used to obtain the coca extracts is outlined in Fig. 1 and described in detail below. Cocaine content of the solutions was determined by a method developed in this laboratory [6].

Coca leaves *(Erythroxylon coca)* obtained from the Tingo Maria area of Peru were powdered using a Wiley mill and exhaustively extracted by percolation with 95% ethanol. Evaporation of the solvent resulted in a crude ethanol extract containing all alkaloidal constituents of coca. The crude ethanol extract was then partitioned between water and chloroform resulting in two fractions (water fraction, A, and chloroform fraction, B). All water soluble compounds (e.g., water soluble alkaloids, quaternary ammonium compounds, sugars, cyclitols, glyosides, etc.) were contained in fraction A while fraction B contained the water insoluble constituents (e.g., alkaloids, terpenes, sterols, fatty acids, etc.). Fraction A was then washed repeatedly with chloroform to ensure that all cocaine had been removed. After solvent evaporation, fraction A was then partioned between butanol and water in order to separate some of the slightly polar organic constituents from the other water soluble constitutents. Following fractionation and evaporation of solvents the fraction residues were then prepared for injection in the following manner: Fraction B (cocaine content of the residue=5.5%) was suspended in distilled water, using 2 drops of Tween 60 and Arlacel 83 per ml of water as suspending agents, such that the suspension contained 2% cocaine (w/v). Fractions C and D were prepared as 2% (w/v) solutions in distilled water. Cocaine was prepared both as a 2% (w/v) solution in distilled water and as a 2% (w/v) suspension using Tween 60 and Arlacel 83 as described above for fraction B. Injection volume was held constant at 0.1 ml and was given intradermally.

Procedure

Following placement in the restrainer and attachment of the electrodes, groups of 8 subjects each were given several minutes to adapt to the restraint system. Next, five baseline measures were conducted at one minute intervals. Each measurement consisted of gradually increasing the shock intensity (0.1 mA/sec) until a response (100 g tail jerk) turned the shock off. Responses were recorded in terms of % of maximum (2 mA). Following baseline determination, groups of 8 subjects were injected with either a vehicle (distilled water, or a suspension of 2 drops of Tween 60 and Arlacel 83 per ml of water), cocaine in either vehicle, or one of the fractions of the coca leaf. Five minutes after injection local anesthetic readings were taken for four hours at the following times post injection: 5, 10, 15, 30, 45, 60, 75, 90, 105, 120, 135, 150, 165, 180, 210, 240 min.

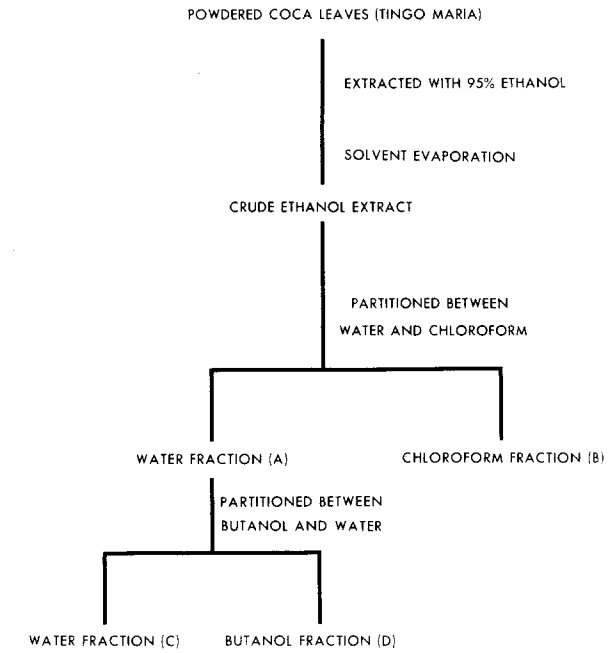

FIG. 1. Analytical procedure employed to separate coca fractions studied. Tingo Maria refers to the area in Peru where the leaves were obtained.

RESULTS AND DISCUSSION

Gas chromatographic analysis [6] of the various fraction residues revealed the following cocaine contents. Original ethanol extract, 0.6%, Fraction A, 0.0%; Fraction B, 5.5%; Fraction C, 0.0%; Fraction D, 0.0%.

The results of the anesthetic screening are presented in Figs. 2a–2f. As can be seen neither of the vehicles (Figs. 2A and 2B) produced any systematic change in the response to shock. There was a tendency to show a very slight gradual increase over the entire four hour testing period. None of the readings, however, differed significantly from the baseline readings. Fraction C, likewise, did not produce any anesthetic effect and therefore the data from this fraction were not included in the figure. In a recent paper [3] we tested this fraction (C) for its effects on food consumption and found only a slight anorexic effect at 480 mg/kg and no effect on locomotor activity.

Figures 2C and 2D present the results obtained when cocaine was tested as a solution in distilled water (Fig. 2C) and when suspended in distilled water with Tween and Arlacel (Fig. 2D). Although both treatments produced near maximum response, the cocaine in distilled water appeared to produce greater local anesthesia than the cocaine in the Tween/Arlacel suspension during the last 2 hours of testing. Upon analysis, these differences (Figs. 2C vs. 2D) were not found to be statistically significant.

The remaining two fractions (B and D) did demonstrate local anesthetic activity. Fraction B (Fig. 2E) was prepared such that the suspension tested contained 2% cocaine (w/v). This fraction produced a substantial local anesthetic effect; however, a comparison between Fraction B and the cocaine suspension (Fig. 2D) revealed that the maximum effect produced by this fraction was considerably less. Although the times of onset of the effects were essentially the same, the cocaine suspension produced statistically significantly greater local anesthetic effects from the 30–240 min post in-

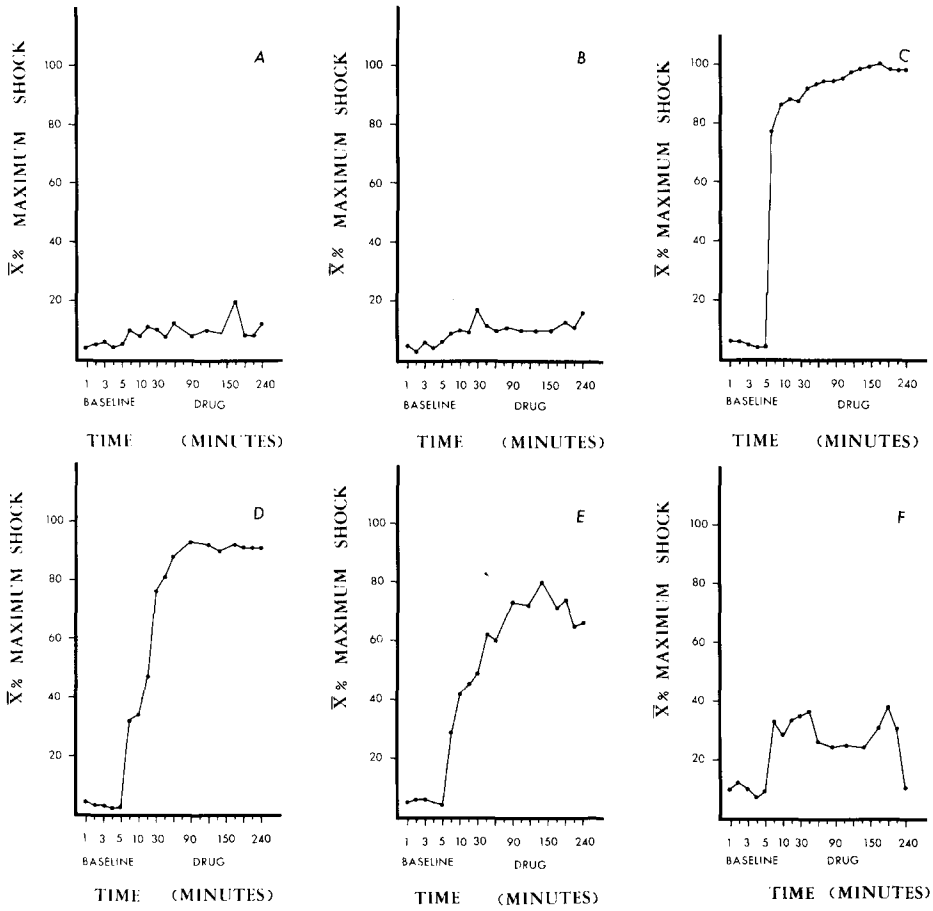

FIG. 2. Percent maximum (2 mA) shock intensity achieved before response as a function of time post injection. 2A-distilled water, 2B-Tween/Arlacel suspension, 2C-Cocaine in distilled water, 2D-Cocaine in the Tween/Arlacel Suspension, 2E-Cloroform fraction B, 2F-Butanol fraction D (see Fig. 1).

jection reading. Since both of these suspensions contained identical amounts of cocaine (2% w/v), it appears that one or more of the other alkaloidal constituents contained in fraction B partially blocked the effect of the cocaine contained in the fraction.

The only other fraction demonstrating a local anesthetic effect (fraction D, Fig. 2F) engendered a response that was 25–30% of that produced by cocaine. In addition the response had returned to baseline as of the final 4 hr reading which was not the case with cocaine or the cocaine containing fraction (B, Fig. 2E).

SUMMARY

The foregoing data support our previous contention [2] that the coca leaf contains constituents other than cocaine that are biologically active. Fraction D (Fig. 2F), although engendering activity considerably less than with cocaine or the cocaine containing fraction, nonetheless produced significant local anesthetic effects.

Finally, the significant difference observed between fraction B and the cocaine suspension clearly indicated that the coca leaf contains constituents that alter the activity of cocaine.

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