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The Anorexic and Actometric Effects of Cocaine and Two Coca Extracts[1,2]

JOHN A. BEDFORD,[3] D. KAREN LOVELL, CARLTON E. TURNER, MAHMOUD A. ELSOHLY AND MARVIN C. WILSON

Research Institute of Pharmaceutical Sciences, and Department of Pharmacology, School of Pharmacy University of Mississippi, University, MS 38677

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BEDFORD, J. A., D. K. LOVELL, C. E. TURNER, M. A. ELSOHLY AND M. C. WILSON. *The anorexic and actometric effects of cocaine and two coca extracts.* PHARMAC. BIOCHEM. BEHAV. 13(3) 403–408, 1980.—The effects of cocaine and two extracts of the coca leaf were compared using locomotor activity and limited access food consumption paradigms. The three treatments were tested using both IP and PO routes of administration. The extracts were prepared by first extracting the powdered leaves with 95% ethanol, evaporating the ethanol and then partitioning the residue between water and chloroform. The doses of the extracts studied were 60, 120, 240, and 480 mg/kg. The doses of cocaine studied were 3.45, 6.9, 13.8, and 27.6 mg/kg. These doses corresponded to the amount of cocaine contained in the four doses of the chloroform layer. Cocaine and the chloroform layer (via both routes) produced dose related increases in locomotor activity and dose related decreases in food consumption. The water layer (containing only trace amounts of cocaine) produced no changes in locomotor activity; however, the highest IP dose did significantly reduce food consumption. Furthermore two of the doses (one IP, one PO) of the chloroform layer produced significantly greater effects than an equivalent amount of cocaine. These data suggest that plant constitutents other than cocaine may contribute to the overall effect achieved by chewing the leaf.

Cocaine Coca Activity Food consumption Rat

THE use of *Erythroxylon coca* preparations for social, religious, therapeutic and other purposes by natives of South America has been extensively documented [4]. Many social scientists and others have visited areas to study the use of this substance, at times, using themselves as subjects. Several investigators [3, 10, 19] have reported on the relative lack of obvious toxicity and the pleasurable effects produced by these substances.

Although some clinical studies (using natives) of the pharmacological and toxicological effects of chewing coca have been reported, their conclusions were reached without the scientific rigor that should accompany contemporary clinical studies [5, 8, 15, 20]. Animal studies using the plant materials or extracts of the leaf are virtually non-existent. With the exception of a study on oral self-administration of coca leaf tea [1] no other contemporary preclinical pharmacological studies on the coca plant have been reported. In contrast to the lack of data on the plant material, there has been a large number of animal studies which delineate the pharmacologic profile of cocaine [7, 11, 12].

The present studies are designed to begin to fill this void in the knowledge of coca by exposing extracts of the plant material (some containing cocaine, others not) to the same type of rigorous scientific scrutiny that has been applied to other drug plants (i.e. marihuana, opium, belladona, etc). Specifically, the present studies are designed to assess the effects of two extracts of the coca leaf on food consumption and locomotor activity. Furthermore the extract effects were compared to cocaine dosages which were equal in cocaine content to the extract dosages.

METHOD

Subjects

The subjects were male Wistar rats (Harland Industries, Cumberland, IN) weighing between 250–300 g at the start of the experiments. Water was freely available to the subjects in the feeding experiments and freely available to the subjects in the actometric experiments except when the subjects were in the actometers. All subjects were housed individually in galvanized steel suspension cages. Ambient temperature was maintained at 21 ± 1°C and the light/dark cycle was 12 hr on, 12 hr off. Subjects in the actometric experiments had free access to food (Purina Rat Chow) except while in the actometers. The deprivation conditions of the subjects in the feeding experiments are discussed in detail below.

Apparatus

Actometric testing was carried out in circular photocell actometers described previously [14]. The actometers were

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[3]Reprint requests should be sent to Dr. John A. Bedford, Research Institute of Pharmaceutical Sciences, School of Pharmacy, University of Mississippi, University, MS 38677.

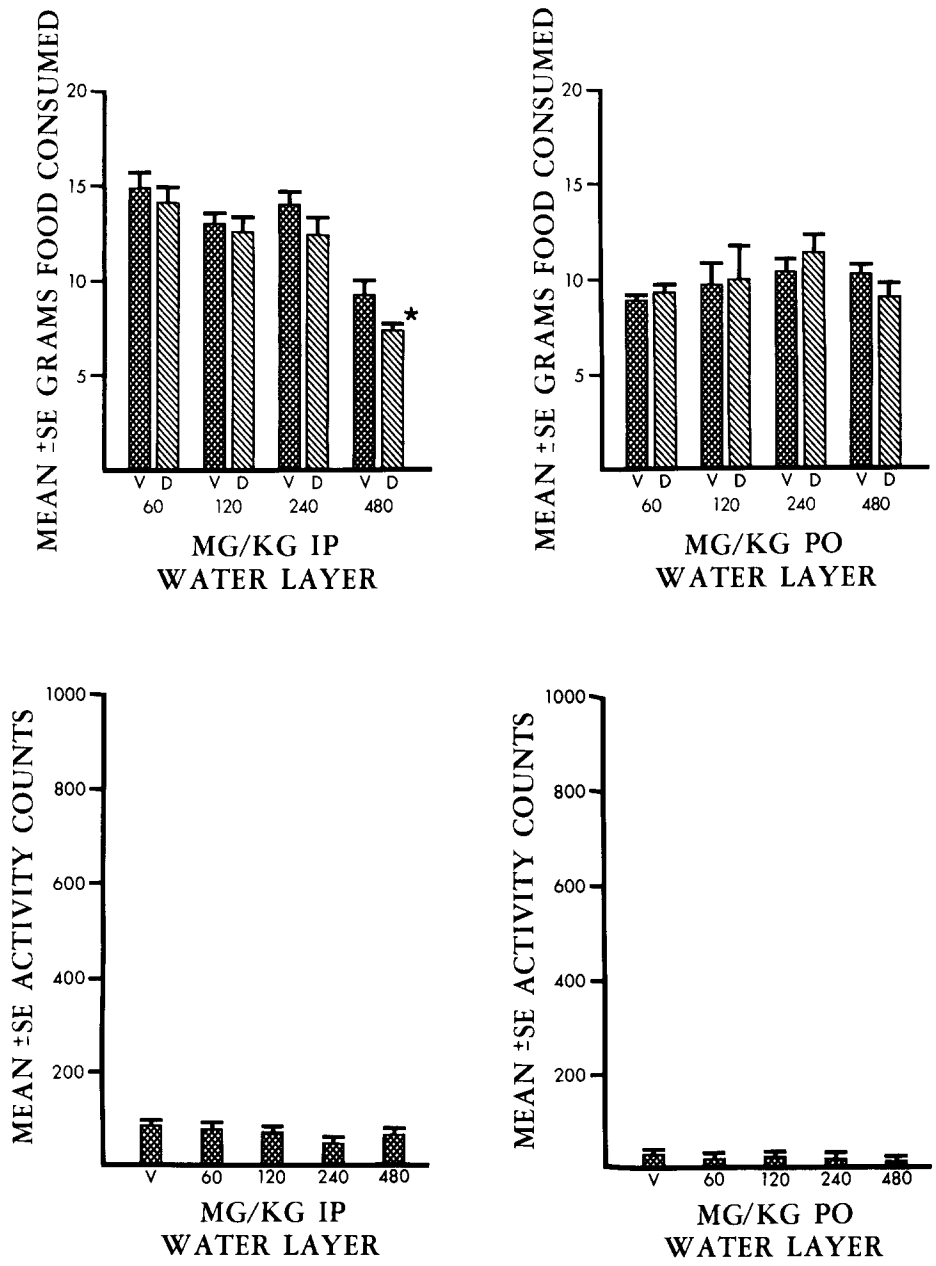

FIG. 1. The effects of the water layer pretreatments (IP and PO) on food consumption (top two graphs) and locomotor activity (lower two graphs). (D=drug treatment, V=vehicle treatment, see text for discussion). *$p \leq 0.05$.

located in a dark, temperature controlled room. A white noise generator provided a continuous auditory environment. The feeding experiments were conducted in the subjects home cage utilizing specially designed feeders and ground rat chow (Purina).

Drugs and Solutions

Drug solutions were prepared on the morning of use. Cocaine dosages were calculated on the basis of the hydrchloride salt and were prepared using sterile normal saline as the diluent. Cocaine hydrochloride flakes U.S.P. were obtained from Mallinkrodt Chemical Corp. (St. Louis, MO). 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. All water soluble compounds (e.g., water soluble alkaloids, quaternary ammonium compounds, sugars, cyclitols, glycosides, etc.) were contained in the water layer while the chloroform layer contained the water insoluble constituents (e.g., alkaloids, terpenes, sterols, fatty acids, etc.). Injections of dosages of the

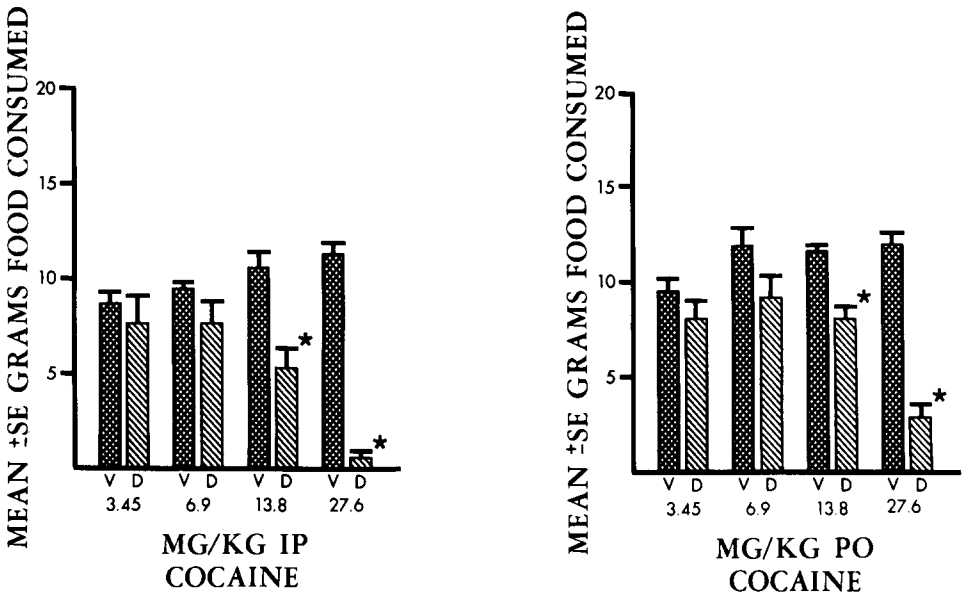

FIG. 2. The effects of the chloroform layer pretreatments on food consumption. The left graph presents the results of the IP pretreatments while the right hand graph presents the results of the PO pretreatments. (D=drug treatment, V=vehicle treatment, see text for discussion). *$p \leq 0.05$.

water layer were prepared using sterile water while the chloroform layer was suspended in sterile water using two drops each of Tween 60 and Arlacel per ml of water. Four dosages (60, 120, 240, 480 mg/kg) of the residue of each layer were tested using both IP and PO routes. The PO (oral) dosing was carried out using a rodent gavage needle. The dosages of cocaine HCl tested were 3.46, 6.90, 13.8, and 27.6 mg/kg IP and PO. These dosages corresponded to cocaine content of the four dosages of the chloroform layer that were tested. Cocaine content of the two extracts was determined by gas chromatography using a method developed in our laboratory [18].

Actometric Testing Procedure

Following a four day laboratory acclimation period, the subjects were randomly assigned to individual groups (n=10). On the afternoon prior to testing the subjects food was removed (18 hr prior to test time) in order to control for deprivation state since this may influence locomotor activity. A session consisted of a 30 min determination of baseline activity (recorded at 15 min intervals) followed by a 60 min test period during which activity was recorded at 15 min intervals. Between the baseline and test segments of a session the subjects were appropriately dosed. Injection volumes for both IP and PO treatments were based on 2 ml/kg of body weight. Each dose of each of the three treatments and the vehicle were administered to 10 subjects by each route. The vehicle control for cocaine treatments was sterile normal saline. The vehicle controls for the water and chloroform layers were distilled water and the Tween 60, Arlacel, water mixture, respectively. Statistical comparisons were accomplished by a two-way analysis of variance and by a Duncans New Multiple Range Test [6].

Anorexic Testing Procedures

Following a 4 day acclimation period during which the subjects had 24 hr access to chow (biscuit form) the subjects were randomly assigned to experimental groups (n=10). The subjects were then given 1 hr/day access to ground chow for 7 consecutive days. The amount of food consumed was measured after each days access period. On the next day all subjects within a test group were weighed and then dosed with the vehicle appropriate to their test condition and immediately given 1 hr access to the ground chow. On the following day the same procedure was followed except that the appropriate test preparation, dose and route were administered prior to access to chow. Statistical comparisons between control data and drug data were accomplished via either Mann-Whitney U or the Wilcoxon Matched-Pairs Signed-Ranks nonparametric test [17].

RESULTS

All three test compounds produced significant effects on activity or food consumption or both at one or more of the doses tested. The least active of the three treatments was the water layer. Figure 1 presents the effect of all water layer treatments on both activity and food consumption. The water layer produced virtually no change in activity scores following any of the dose/route treatments. This was generally true for the feeding data also; however, a significant reduction in consumption was observed with the 480 mg/kg IP pretreatment.

The remaining two treatments produced pronounced effects on both behaviors studied following both routes of administration. Figure 2 presents the effects of cocaine (IP and PO) on food consumption. Significant reductions in food consumption compared to vehicle controls occurred with doses of 13.8 and 27.6 mg/kg following both the IP and PO routes. Figure 3 presents the effects of the chloroform layer (IP and PO) on food consumption. Significant reductions in consumption were observed following IP and PO pretreatments with 240 and 480 mg/kg of the chloroform layer. Finally, a comparison between treatments revealed that the 240 mg/kg IP dose and the 480 mg/kg PO dose of the

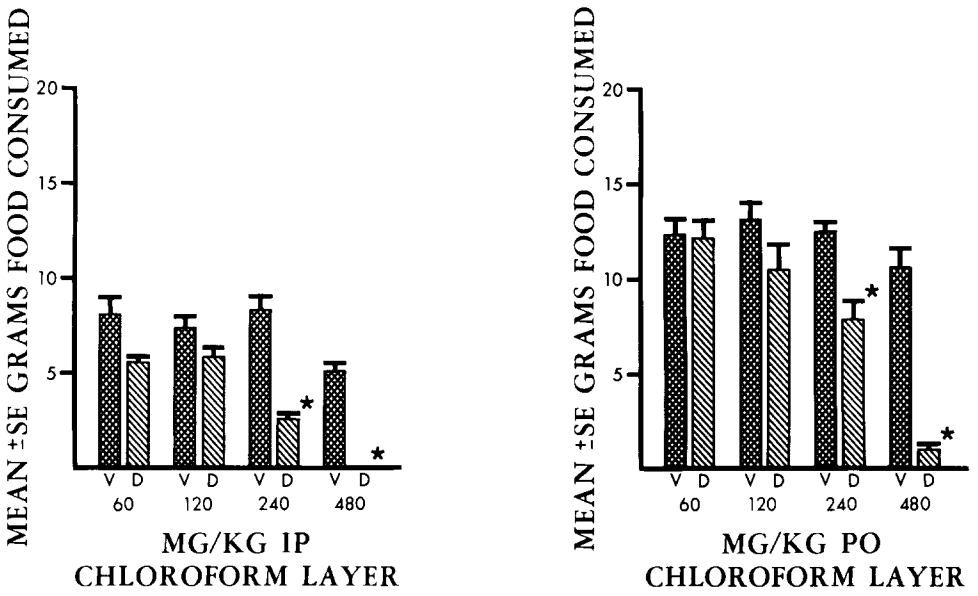

FIG. 3. The effects of cocaine HCl pretreatments on food consumption. The left hand graph presents the results of the IP pretreatments while the right hand graph presents the results of the PO pretreatments. (D=drug treatment, V=vehicle treatment, see text for discussion). *$p \leq 0.05$.

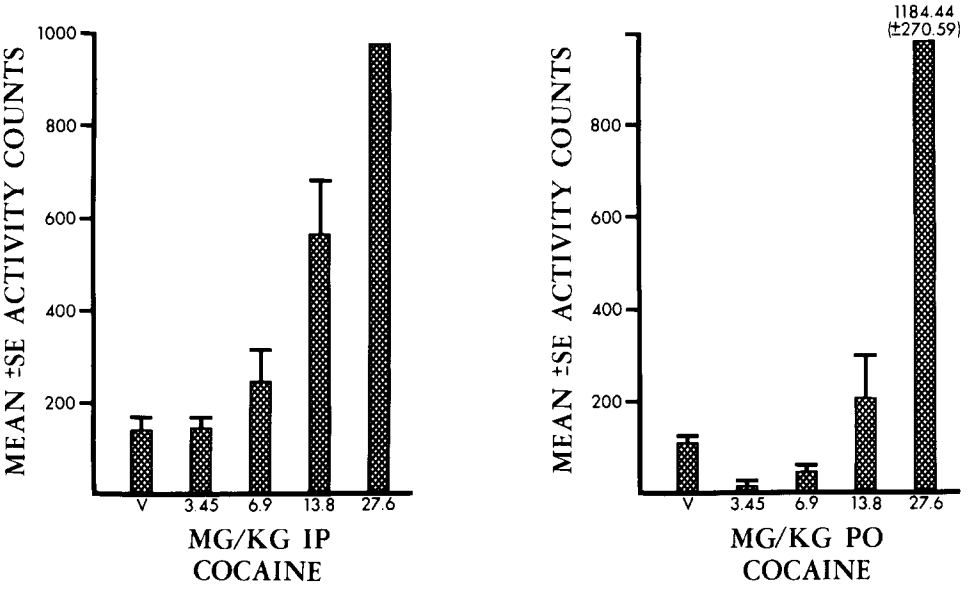

FIG. 4. The effects of cocaine HCl pretreatments on locomotor activity. The left hand graph presents the results of the IP pretreatments while the right hand graph presents the results of the PO pretreatments. The reader is referred to the text for a discussion of the statistical treatment of these data. (V=saline).

chloroform layer produced a greater reduction in consumption than did doses of cocaine (13.8 and 27.6 mg/kg) equivalent to the amount of cocaine contained in these two treatments.

Locomotor activity scores were observed to increase systematically with the dose of cocaine and of the chloroform layer. Figures 4 and 5 present the effects of cocaine and the chloroform layer on locomotor activity scores. Significant increases were obtained with each treatment-route combination. An overall significant treatment main effect was observed with cocaine and the chloroform layer via both routes. In addition, when subjected to a Duncan's Multiple Range Test [6] several treatment means were found to differ from one another. In the case of cocaine, 27.6 mg/kg IP was found to differ from 13.8 mg/kg IP which in turn was found to differ from all other PO treatment means. With the chloroform layer (Fig. 5) 240 and 480 mg/kg IP were found to

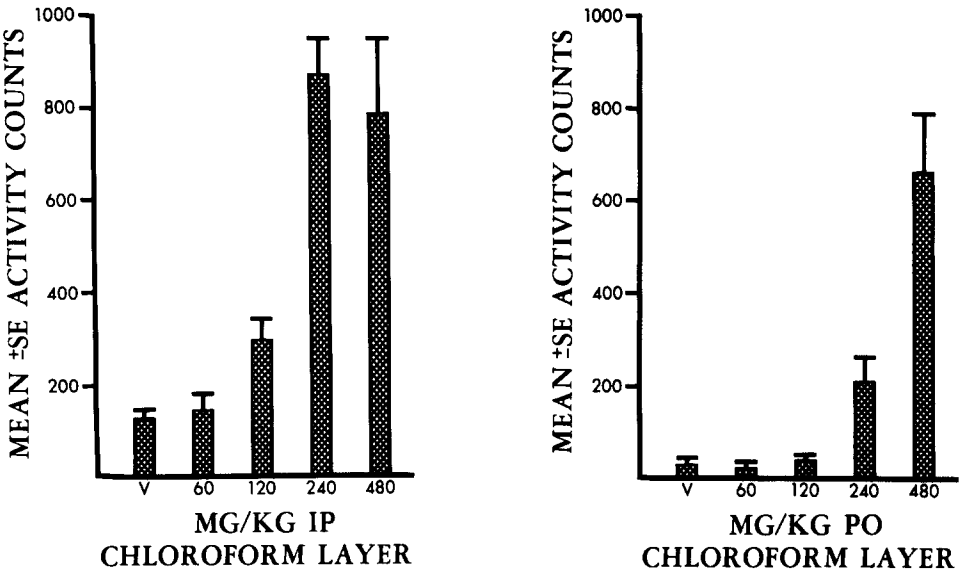

FIG. 5. The effects of the chloroform layer pretreatments on locomotor activity. The left graph presents the results of the IP pretreatmnets while the right hand graph presents the results of the PO pretreatments. The reader is referred to the text for a discussion of the statistical treatment of these data. (V=vehicle treatment, see text for discussion).

differ significantly from the remaining two IP treatment means. Finally, the 480 mg/kg PO dose of this extract was observed to produce significantly greater activity scores than all other PO treatments with this extract.

DISCUSSION

Although the water layer was found to be inactive when tested in the locomotor activity paradigm, a significant reduction in food consumption was observed following IP pretreatment with 480 mg/kg dose. The dose effect curve for this compound when administered IP indicates that it may contain an active constituent of either low potency or of small quantity. This substance may possess some anorexic activity at dosages which do not alter locomotor activity. Although this compound was not completely free of cocaine, the trace amounts of cocaine contained in the doses tested was considerably below that amount of cocaine shown to be ineffective in reducing food consumption. Further testing of this compound is underway in order to determine whether larger doses will produce greater reductions in food consumption.

The remaining two treatments (cocaine, chloroform layer) were found to be active in both paradigms, via both routes at one or more of the doses tested. Cocaine was shown to produce a significant reduction in consumption following both IP and PO treatments with the 13.8 and 27.6 mg/kg doses. The effects observed with the IP route are consistent with other published reports [2]. The quantitatively similar effects observed between the IP and PO treatments are quite striking, since until recently it was felt that cocaine was hydrolyzed in the liver or gastrointestinal tract rendering it ineffective [16]. These data clearly demonstrate that cocaine is active orally in suppressing food intake, but more importantly, its potency is similar when given either IP or PO. A recent paper comparing the effects of cocaine in humans in which the same dose was administered orally and intranasally reported similar effects [19]. These authors reported that cocaine given by the oral route was "at least as effective as the same dose given intranasally". The similarity in the magnitude of the effects of IP and PO treatments on food consumption was also compared in the locomotor activity paradigm. The highest dose of each compound produced similar increases in activity. With the lower doses the IP treatment tended to produce greater increases in activity than equal PO doses but these differences were not statistically significant.

The chloroform layer produced somewhat similar effects to that of cocaine on food consumption. Significant reductions in consumption were observed following the 240 and 480 mg/kg IP and PO treatments. Again, as with the cocaine treatments the IP and PO routes produced qualitatively and quantitatively similar effects. These data strongly suggest, that although the natives of South America typically chew the coca leaves, perhaps some or much of the effect they achieve is due to swallowing the juices. Furthermore, the local vasoconstrictor effects of cocaine in the oral cavity may reduce the regional blood flow and in fact, decrease its systemic absorption in the oral cavity. Finally, the finding that two of the chloroform layer treatments (240 mg/kg IP and 480 mg/kg PO) produced a significantly greater reduction in food consumption than did that amount of cocaine contained in these treatments alone, suggests that this extract may contain some plant constituents other than cocaine that may contribute to the behavioral pharmacology of coca. A recent report [4] emphasizes that, "of the 14 alkaloids known to be present in the coca leaf, almost all research to date has concentrated only on the cocaine alkaloid." In addition, others have speculated that these alkaloids may contribute the pharmacological actions of the leaf [9].

In summary, the present paper clearly demonstrates that extracts of the coca leaf have significant biological activity. The activity of the chloroform layer may be solely attributable to the cocaine content of this extract; however, there was some indication that this extract was more potent than a similar amount of cocaine. The ability of the water layer

to reduce food consumption suggests that the plant may contain water soluble constituents which exhibit anorexigenic activity.

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REFERENCES

1. Altshuler, H., A. Law and D. Najar. Studies of Erythroxylon Coca: Self-Administration of Aqueous Extracts. *Proc. west. Pharmac. Soc.* **20:** 351, 1977.
2. Bedford, J. A., R. F. Borne and M. C. Wilson. A Comparative Behavioral Profile of Cocaine and Norcocaine in Rats and Monkeys. *Pharmac. Biochem. Behav.*, in press.
3. Cardenas, M. Psychological Aspects of Coca Addiction. *Bull. Narcot.* **42:** 6, 1952.
4. Carroll, E. The Plant and Its Use in Cocaine: 1977, edited by R. Peterson and R. Stillman. NIDA Res. Monogr. **13:** 35–45, 1977.
5. Chamochumbi, N. Coca Effects in the Metabolism of the Addicted. *Revta Farmac. Med. exp.* **2:** 9, 1949.
6. Edwards, A. L. *Experimental Design in Psychological Research.* Holt, Rinehart and Winston, Inc., New York, 1968.
7. Ellingwood, E. and M. Kilbey. *Cocaine and Other Stimulants.* Plenum Press, New York, 1977.
8. Gutierrez-Noriega, C. and V. Zapata-Ortiz. Physiological and Pathological Observations in Coca Addicts. *Revta Farmac. Med. exp.* **1:** 1, 1948.
9. Gutierrez-Noriega, C. and V. Zapta-Ortiz. The Intelligence and Personality of the Coca Addicted. *Rev. Neuropsiquiat.* **13:** 22, 1950.
10. Martin, R. T. The Role of Coca in History, Religion and Medicine. In: *The Coca Leaf and Cocaine Papers,* edited by G. Andrews and D. Solomen. Harcort, Brace, Joyanovich, New York, 1975.
11. Mortimer, W. C. *History of Coca, the Devine Plant of the Incas.* 1901. Reprinted by AND/OR Press, San Francisco, 1974.
12. Mule, S. *Cocaine: Chemical, Biological, Clinical, Social and Treatment Aspects,* SRC Press, Cleveland, 1976.
13. Peterson, R. and R. Stillman. Cocaine, 1977: NIDA Research Monograph No. 13, U.S. Government Printing Office, Washington, 1977.
14. Pickens, R. and W. Crowder. A recorder of Locomotor Activity. *Am. J. Psychol.* **80:** 442–445, 1967.
15. Risemberg, M. F. Accion de la Coca y la Cocaine en Sepetos Habituados. *Rev. Med. exp.* **3:** 132, 1944.
16. Ritchey, J. M. Central nervous System Stimulants. In: *The Pharmacological Basis of Therapeutics,* edited by L. A. Goodman and A. Gilman. New York: The Macmillan Company, 1965.
17. Siegal, S. *Non-Parametric Statistic for the Behavioral Sciences,* McGraw-Hill, New York, 1956.
18. Turner, C. E., C. Y. Ma and M. A. Elsohly. Constituents in Erythroxylon coca I: gas chromatographic analysis of cocaine from three locations in Peru. *Bull. Narcot.* **31:** 171–76, 1979.
19. VanDyke, C., P. Jatlow, J. Ungerer, P. G. Barash and R. Byck. Oral cocaine; Plasma concentrations and Central Effects. *Science* **200:** 211–213, 1978.
20. Weil, A. Letters From Andrew Weil. *J. Psychedelic Drugs* **7:** 215, 1976.