SUMMARY

The green stems from the plant, *Ephedra Nevadensis* Wats., growing in southwestern Oklahoma, have been examined. In the preliminary analysis the percentages of moisture, total ash and acid-insoluble ash were determined. Samples of the powdered material were extracted by selective solvents. Tests for the presence of ephedrine were negative.

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Effect of Picrotoxin on the Blood Potassium of Anesthetized Animals*

By Carlos E. Cardini and Maria E. Serantes

Picrotoxin strongly antagonizes the effects of narcotics; it increases respiratory tonus, restores consciousness, etc. Advantage has been taken of this action in the treatment of barbiturate poisoning. Maloney and collaborators (1), Rosenthal and Wallach (2), and Marshall and co-workers (3), have, among others, studied this question.

During anesthesia, changes appear in the titers of the inorganic constituents of the blood, the most prominent of these being a marked decrease in plasma potassium (Marenzi and Gerschman (4), 1933). It was considered of interest to study the narcosis-induced decrease in plasma potassium as affected by the suppression of narcosis with picrotoxin. It is also to be remembered that picrotoxin, along with its antinarcotic action, possesses convulsant properties capable by themselves of inducing variations in the plasma potassium of normal dogs.

EXPERIMENTAL

The anesthetic selected, chloralose, was administered intravenously to dogs at the rate of 10 ml. of an 8% solution per Kg. After deep anesthesia had been induced, a 0.5% solution of picrotoxin (Merck) in distilled water was injected intravenously, rapidly or gradually, in a dosage varying from 0.10 mg. to 1 mg./Kg. for rapid injections, from 0.25 to 0.35 mg./Kg. in prolonged administration, distributed over 10 to 20 min. Blood was obtained from the carotid artery at intervals noted in the curves. Potassium was determined by the method of Marenzi and Gerschman (4), 1932, using blood deproteinized with trichloroacetic acid. Potassium was determined directly in plasma and whole blood. The red cell potassium content was calculated from cell volume determinations and from plasma and whole blood analyses. Red cell concentration or contents is of very little significance even when water determinations and R. B. C. counts are available.

RESULTS

Rapid Administration.—Doses of 0.10 mg. and 0.12 mg./Kg. do not induce the disappearance of narcosis. Antagonistic action begins to be evident with doses above 0.15 mg. There is a latent period which for this dosage varies between 8 and 10 min. As a rule, in 20 to 30 min., convulsions begin to appear which last to the end of the experiment. The animal is conscious throughout the entire experiment, which lasts more than an hour. With a dose of 0.25 mg./Kg., consciousness is regained within 8 to 20 min., but rather severe convulsions appear 10 min. after the injection. With a dose of 0.50

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mg., the picture is similar. With 1.0 mg., there is no latent period; immediate violent convulsions set in and the animal becomes conscious. The convulsions persist to the end.

Gradual Administration.—The results are similar to those obtained with massive dosage. In one series of experiments 0.015 mg./Kg. was injected once a minute for 20 min., a total dosage of 0.30 mg./Kg. The animal awoke in 15 to 20 min., and slight convulsions were observed. In another series, 0.025 mg./Kg. was injected every minute for 10 min.; total dosage 0.25 mg. The animal awoke at the end of the last injection and convulsions were observed that lasted until the end of the experiment.

Variations in Plasma Potassium.—The most significant experiments are represented by curves (Figs. 1, 2, 3). In Fig. 1 the descending curve resulting from the action of the anesthetic is to be

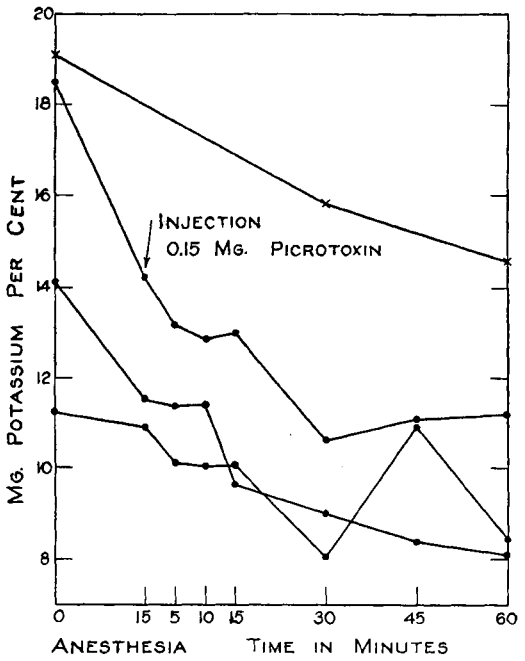

Fig. 1. (X-curve without picrotoxin).

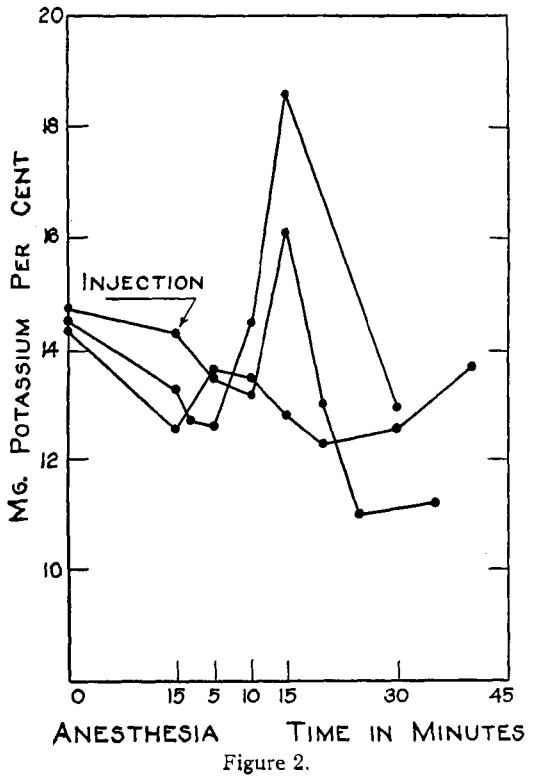

Figure 2.

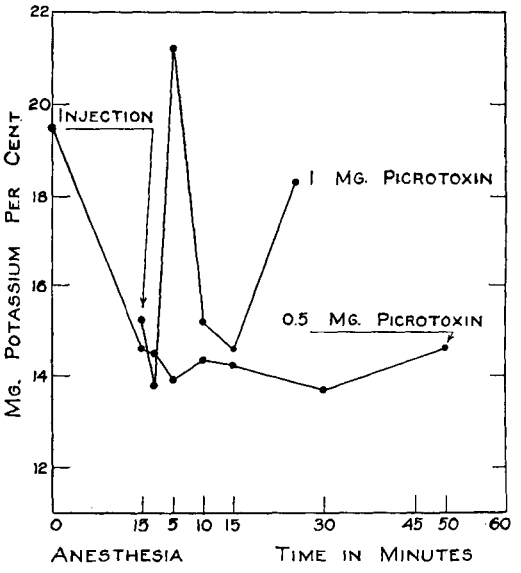

Figure 3.

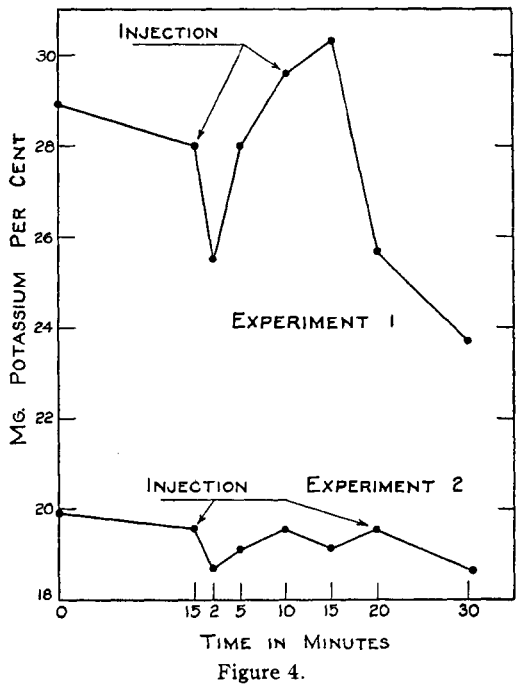

Figure 4.

TABLE I.—THE PICROTOXIN–CHLORALOSE ANTAGONISM

			Variation in Potassium in Whole Blood							Variations in Potassium Referred to Original Cell and Plasma Volume			
Exp. No.	Wt. of Dog, Kg.	Amt. of Picrotoxin Injected, Mg./Kg.	Time, Min.	Cell Volume	Plasma Volume	Blood Potassium, Mg. %	Plasma Potassium, Mg. %	Blood Cell Potassium,[a] Mg. %	Cell Volume Potassium, Mg.	Plasma Volume Potassium, Mg.	Potassium in Cells, Mg. %	Potassium in Cell Volume, Mg.	Potassium in Plasma Volume, Mg.
1	24	1.00	0	57.57	42.43	18.8	15.3	21.4	12.3	6.5	21.4	12.3	6.5
			2	51.92	49.08	26.6	13.7	40.2	20.9	5.7	36.2	20.8	5.8
			5	56.70	43.50	22.6	21.2	23.7	13.4	9.1	23.7	13.7	9.0
			10	59.00	41.00	20.2	15.2	23.7	14.0	6.2	24.0	13.8	6.5
			15	59.18	40.82	19.5	14.5	23.1	13.6	5.9	23.3	13.4	6.2
			25	58.25	41.75	21.1	18.3	23.1	13.4	7.5	23.8	13.4	7.7
2	23.5	1.00	0	39.96	60.04	20.3	18.0	23.4	9.3	10.8	23.4	9.3	10.8
			2	50.51	49.49	27.2	25.9	28.6	14.4	12.7	29.3	11.7	15.4
			5	53.57	46.43	33.0	27.8	37.4	20.1	12.8	46.1	18.4	14.7
			10	53.39	46.61	30.1	30.1	30.1	16.0	14.0	30.8	12.0	18.0
			15	54.20	45.80	28.8	26.8	30.5	16.5	12.2	32.0	12.7	16.1
			25	57.00	43.00	30.5	29.2	31.6	18.0	12.2	32.6	13.0	17.4
3	6.0	0.25	0	44.97	55.03	23.9	17.1	34.4	15.4	9.4	34.4	15.4	9.4
			7	41.49	58.51	21.2	13.9	31.6	13.0	8.1	30.2	13.6	7.6
			12	48.01	51.99	20.3	15.4	25.8	12.3	7.9	26.1	11.9	8.5
			20	48.35	51.65	21.9	15.7	28.5	13.7	8.1	29.6	13.3	8.6
			25	48.58	51.42	21.2	14.4	29.3	14.2	6.9	29.6	13.3	8.0
			30	47.97	52.03	22.4	16.0	29.4	14.1	8.3	30.3	13.6	8.8

[a] Calculated from plasma and whole blood, potassium analyses and all volume determinations.

noted; the injection of 0.15 mg. of picrotoxin does not modify this drop. This dosage is of major interest because it shows that the antagonistic action of the drug does not modify the drop in potassium; it is the dose which restores consciousness without producing convulsions.

With greater doses the restoration of consciousness is accompanied by violent convulsions; these convulsions coincide with an abrupt increase in potassium. This is not invariably so, for in some instances (Fig. 3) with a dose of 0.5 mg., which induced violent convulsions, the fall in potassium continued. The rarity of these findings leads one to attribute them to a factor other than the direct effect of picrotoxin.

In prolonged dosage (Fig. 4) there is a small drop at the beginning of the injection, then an ascending period, but in a manner much less pronounced than following the massive injection of an equal quantity of picrotoxin. The last decline is less marked than that induced by the anesthetic alone.

Variations in Distribution of Potassium between Blood Cells and Plasma.—Doses of 0.25 mg. and 1 mg. of picrotoxin per Kg. were selected for a study of the possible interchange of potassium between the blood cells and plasma. Both dosages induced definite changes in plasma potassium. The results obtained did not appear to depend on the size of the dose (Table I).

It was observed in these experiments that the blood cell volume underwent significant but variable changes. In experiments 1 and 3, there was a decrease in cell volume, followed by an increase and finally a return to its original value. In experiment 2, the cell volume increased throughout the experiment.

Significant changes in the erythrocyte count, erythrocyte water content and cell volume are known to occur in the intact animal as a result of various procedures such as exercise and epinephrine injection. The data available in these experiments are insufficient to determine these changes in the erythrocytes of the circulating blood and hence an accurate expression of erythrocyte potassium exchanges is impossible.

Large increases in plasma potassium concentration were accompanied by a significant increase in red cell potassium either on a concentration (mg./100 cc. of cells) or a cell volume content (mg./100 cc. of whole blood) basis. This increase in red cell potassium on both a concentration and a cell volume content basis apparently results from a transfer of potassium from the plasma into the cells and the addition of a large number of erythrocytes to the circulating blood due to the contraction of certain organs.

CONCLUSIONS

A clean-cut antagonism was shown between picrotoxin and the narcotic action of chloralose.

In each instance the animal awoke and remained awake throughout the course of the experiment. Along with this action picrotoxin retained its marked convulsant activity.

The smallest dose which induced suppression of narcosis without causing convulsions was 0.15 mg./Kg.

Many authors have correlated the convulsant action of picrotoxin with its antagonistic action against narcotics (Krautz and collaborators (5)); the present results show only that these actions do not necessarily occur simultaneously.

The decrease in plasma potassium, which normally occurs during anesthesia, is modified only when convulsions are induced, which would indicate that the suppression of narcosis does not modify this decrease but renders it less intense.

At the beginning of convulsions, marked changes in the blood appear.

In most experiments there is an abrupt increase in potassium, coinciding with the beginning of muscular contractions; the potassium values then become lower as this behavior continues.

This abrupt increase is very similar to that produced by the intravenous administration of epinephrine. It may in part be attributed to the liberation of muscle potassium during the spasms and to the indirect effect of the discharge of epinephrine by bulbar action, which also produces a hyperglycemia, already emphasized by various workers studying the action of picrotoxin. This would be in agreement with the mechanism of the regulation of potassium as demonstrated by Houssay, Marenzi and Gerschman (6), 1936. It is interesting to note that picrotoxin displays this antagonism in a latent period of 5 or more min., except in very high doses where the effect is almost immediate.

The changes in the whole blood potassium are principally referable to changes in the potassium concentration in the cells and cell volume, because the change in plasma potassium content in terms of whole blood volume is very small. Possibly during the muscle and organ contractions, there is an abrupt entry of red cells into the blood; in addition, a transfer of potassium to the cells very probably occurs, because a constant increase greater than the experimental error inherent in this indirect determination was observed in the red cell potassium concentration.

Results obtained with other inorganic constituents prove that the action of picrotoxin does not greatly modify the variations in them induced by anesthesia, as studied by Marenzi and Gerschman (7) (1933), except that magnesium here tends to increase, whereas ordinarily it decreases during anesthesia.

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