be concluded that a distal site continues to secrete acid during metabolic induction of urinary alkalinization. This observation is consistent with the hypothesis that at least a fraction of distal sodium reabsorbtion takes place by way of an exchange for hydrogen ions and therefore with the hypothesis advanced by PITTS and LOTSPEICH[1].

Although, no clear cut site of alkalinization could be detected in these studies, it is suggested that these data are consistent with proximal alkalinization. Such a process could be obliterated by the distal acidifying action as the proximal urine passed this site after release of the ureteral clamp[4]. Since the tubule is relatively permeable to CO_2, it is recognized that alkalinization could occur by concentrating the urine and increasing the ratio of HCO_3^- to H_2CO_3. Examination of the stop-flow curves show that the process of acidification takes place rather far distally, and it is considered unlikely that alkalinization secondary to concentration more distal than the locus of acidification is the main mechanism involved in producing high urinary pH. Such a process could occur, however, at the proximal level. A process of proximal alkalinization would be a corollary to that of proximal acidification demonstrated by GOTTSCHALK et al. under certain conditions[5].

The fact that it was not found possible to maintain a urinary pH above 7 when mannitol containing NaCl was infused is in agreement with the results reported by VAN SLYKE and EVANS[6]. They found that loading of normal dogs with NaCl and glucose induced a fall in pH of the urine. The acidification was attributed to be a consequence of reduction of plasma HCO_3^- concentration secondary to dilution. Examination of their data, however, shows that a considerable drop in pH was found before the drop in plasma HCO_3^- became significant. Furthermore, it would be difficult to explain why mannitol containing NaCl should cause more dilution than mannitol alone in these experiments. It may be that increasing the sodium load is responsible for increasing tubular exchange of H^+ for Na^+[7].

Zusammenfassung. Bei Hunden, die wegen Infusion von $NaHCO_3$, Na-Laktat oder NaOH alkalischen Harn produzierten, wurden «stop-flow»-Versuche durchgeführt. Es konnte gezeigt werden, dass unter diesen Umständen distal eine Ansäuerung des Tubulusharns stattfindet, so dass die Alkalisierung in den proximalen Abschnitten stattfinden muss.

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Department of Physiology, Medical College of Virginia, Richmond (Virginia U.S.A.), February 9, 1962.

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Mechanism of Action of Cocaine and Amphetamine in the Brain

The central stimulant action of cocaine is to a great extent similar to that of the amphetamines. Cocaine and amphetamines cause similar changes in the EEG pattern, while evoking alertness and suppressing appetite[1] to a similar degree.

The amphetamines are structural analogues of catecholamines (arterenol and dopamine); there exists a rigid structure-activity relationship[2]. This supports the suggestion that amphetamines may mimic the action of catecholamines on specific receptors in the brain[3] and that they may therefore have a direct arterenergic mechanism of action.

The central action of cocaine seems to be related to its sympathetic action since its stereo-isomer pseudococaine, although having identical local anaesthetic properties with cocaine, is devoid of peripheral sympathetic activity and of a central stimulant action[4]. Furthermore, the central stimulant action of cocaine and that of amphetamine is selectively antagonized by sympatholytic drugs such as dibenzyline and piperoxane. There is no structural relationship between cocaine and catecholamines and amphetamines so that a direct catechol-like action seems improbable. Alternatively an indirect arterenergic action might underlie the action of cocaine in the brain, that is, an action by release of catecholamines. Experiments will be conducted to test this hypothesis.

Reserpine causes a release of catecholamines (CA) and finally a depletion of long duration[5]. This would imply that indirect arterenergic drugs that act by releasing CA, should no longer be active after depletion of CA by reserpine, whereas those directly occupying the specific receptors ought still to exert their normal effect.

Spontaneous motor activity of groups of two mice was measured with the cumulative recording procedure[6]. About 1 or 2 h after the mice had been placed in the cage and their exploratory behaviour had subsided, they were injected either with *d*-amphetamine (5.62 and 10 μMol/kg in succession) or *l*-cocaine (31.6 and 56.2 μMol/kg) (see Figure 1a and 2a). Cocaine induces a similar increase in motor activity as amphetamine, which is, however, of short duration. Amphetamine in an equally active dose has an action 2–4 times longer. The animals used in this type of experiments were injected with 0.5 mg/kg reserpine twice a day over 3 consecutive days starting 5 days after the previous test doses of amphetamine or cocaine. On the fourth day the same doses of cocaine and amphetamine were administered as given previously and again motor activity was recorded (see Figure 1b and 2b). Similar results were obtained when both amphetamine and cocaine were injected in the same group of two mice. For average results see Table.

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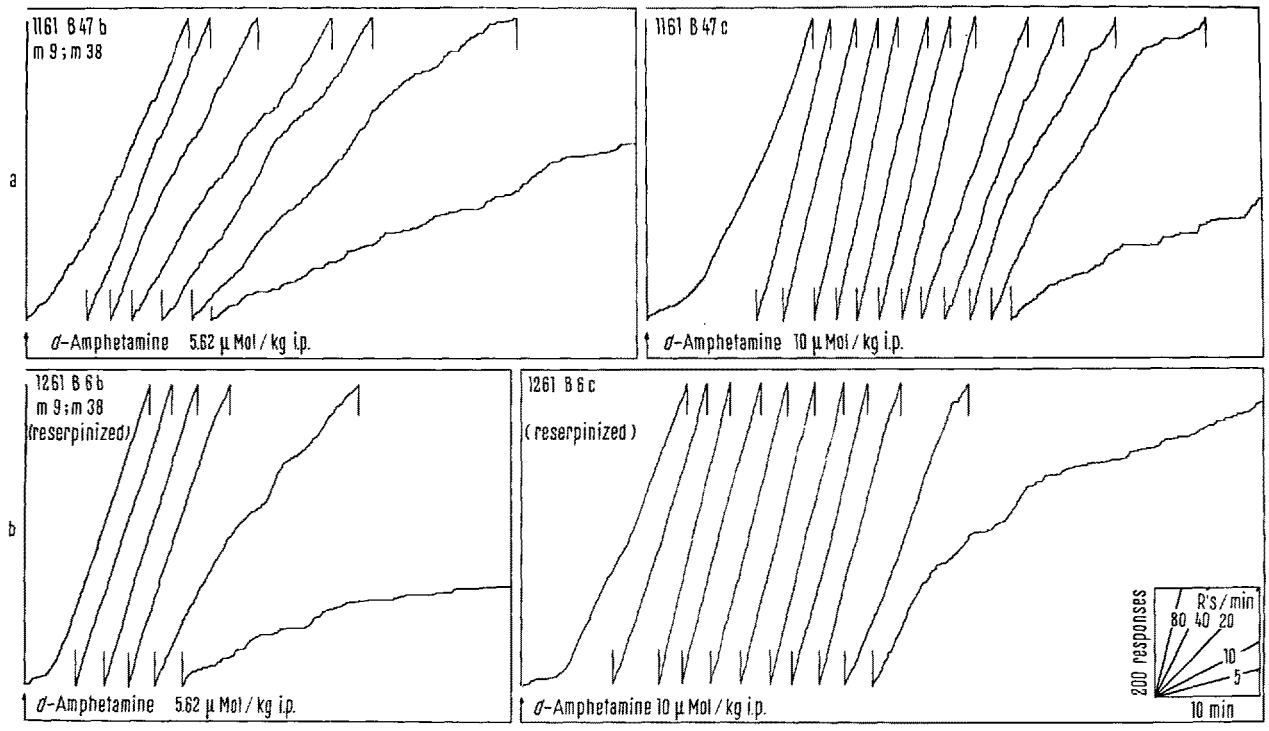

Fig. 1. (a) Cumulative record of the action of *d*-amphetamine (5.62 and 10 µMol/kg) on the motor-activity in a group of two mice. The animals were injected after they had been accustomed to their new environment. (b) Cumulative record of the action of *d*-amphetamine (5.62 and 10 µMol/kg) in the same group of mice 8 days later after they had been reserpinized with 0.5 mg/kg reserpine twice a day over 3 days prior to this experiment. Note that there is practically no difference in activity in experiment a and b. This implies that depleting CA-stores by reserpine does not affect the action of amphetamines. It is therefore concluded that amphetamine mimics CA by directly occupying CA receptors in the brain

The average effect of psychomotor stimulants on locomotor activity in untreated and reserpinized mice

Drug	Normal mice ED_{50} Mol/kg	Reserpinized mice Ratio $\frac{\text{effect after}}{\text{effect before}}$ (%)
d-amphetamine	5.0	80–140[a]
dl-methyl-amphetamine	3.6	40–120[a]
l-cocaine	25	0
dl-benzyl-amphetamine	55	0

[a] When the effect after reserpine was less than before, maximum effect could always be obtained by increasing the dose of the stimulant.

It may be seen from these Figures that pre-treatment with reserpine does not appreciatively affect the response to amphetamine but that the action of cocaine is completely annulled. These results thus strongly indicate that cocaine has an *indirect* arterenergic mechanism of action, presumably by releasing catecholamines from their stores. On the other hand, it seems to be proved that amphetamine has a *direct* arterenergic mechanism of action.

A further proof of the indirect arterenergic action of cocaine was provided by the following experiment. Groups of 2 mice were pre-treated with reserpine over a period of 4 days (total amount 4 mg/kg reserpine) after which a test dose of *l*-cocaine (56.2 µMol/kg) was given, which, according to the previous experiments, appeared to be ineffective. 3 h later, the animals were loaded with L-DOPA (200 µMol/kg), a compound that can pass the blood brain barrier and that can be converted into catecholamines. 1 h after the L-DOPA administration cocaine was again injected which now appeared to exert a practically normal action (see Figure 2c). The effectivity of cocaine in reserpinized mice could, however, not be regained by administration of 5-HTP (a precursor of 5-HT) instead of L-DOPA. These experiments thus prove the indirect arterenergic action of cocaine, that is acting by releasing catecholamines from stores in the brain.

Amphetamine-like compounds such as methamphetamine appear to have a direct mechanism of action (see Table). Derivatives of amphetamines which have large substituents on the nitrogen atom, as for instance benzylamphetamine, are antagonized by pre-treatment with reserpine and therefore have an indirect arterenergic mechanism of action, in the same way as cocaine (see Table).

On the basis of their mechanism of action, amphetamine-like drugs or psychomotor stimulants may be divided into a group with a *direct* arterenergic mode of action, and a group with an *indirect* arterenergic mode of action.

Zusammenfassung. Die Ähnlichkeit der zentral stimulierenden Wirkung des Kokains und des Benzedrins wurde besprochen und weiter untersucht. An mit Reserpin vorbehandelten Mäusen wurde die Wirkung des Benzedrins nicht geändert; die Wirkung des Kokains aber wurde völlig blockiert.

Aus diesen Befunden wurde geschlossen, dass die psychomotorischen Stimulantien oder Weckamine in zwei Klassen eingeteilt werden können. Klasse I: Substanzen

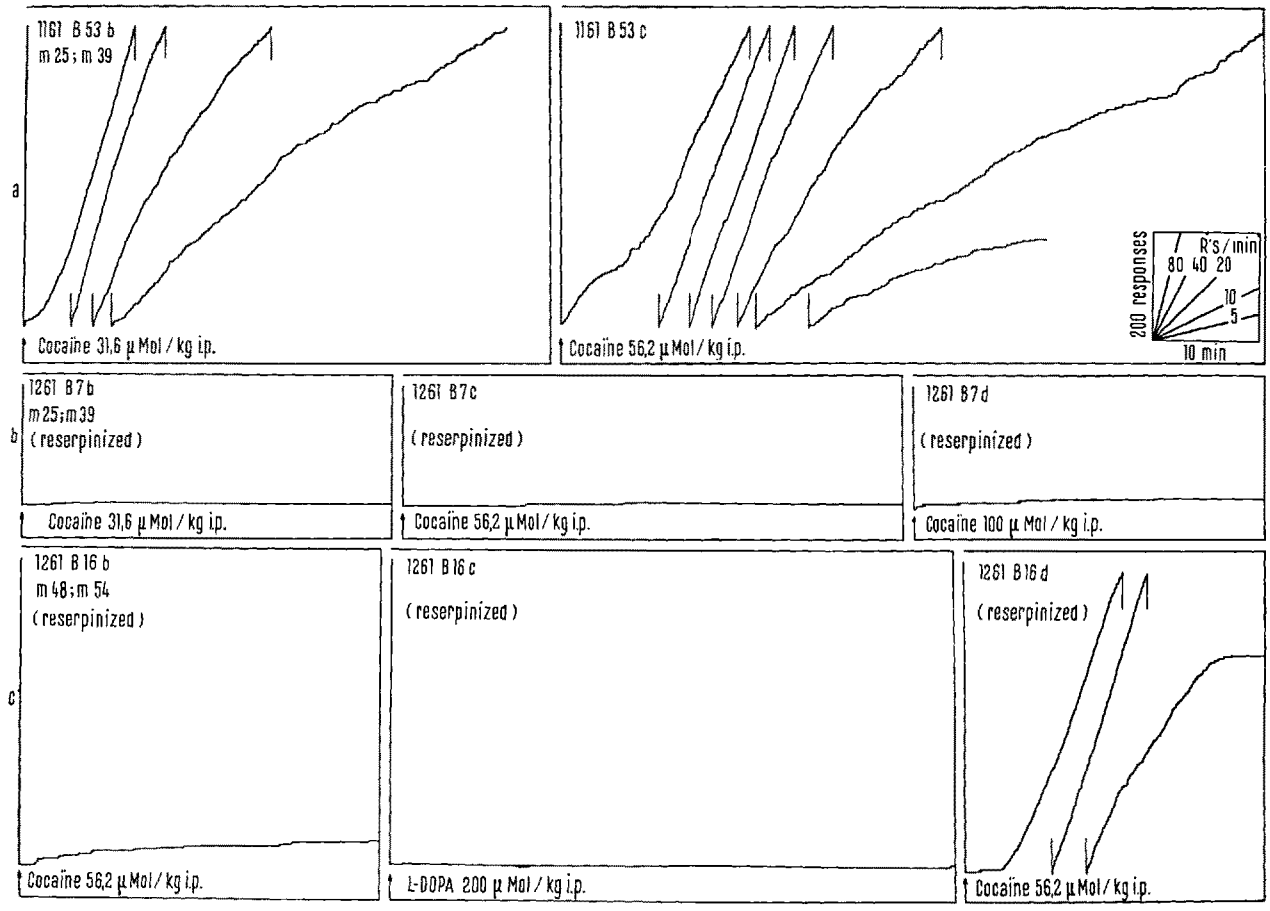

Fig. 2. (a) Cumulative record of the action of *l*-cocaine (31.6 and 56.2 µMol/kg) on the motor-activity in a group of two mice in a similar experiment as in Figure 1a. Note that *l*-cocaine causes a similar increase in locomotor activity as amphetamine though of less duration. (b) Cumulative record of the action of *l*-cocaine in the same group of two mice after they had been reserpinized in a similar way as those of Figure 1b. Note that the action of cocaine is completely abolished by pre-treatment with reserpine and that even higher doses remain ineffective. It must be concluded that cocaine can only act when CA-stores are sufficiently filled and that therefore cocaine acts by releasing CA from the stores and so causes the free CA concentration to rise in the brain. (c) Cumulative record of the action of *l*-cocaine on the motor-activity in a group of two reserpinized mice (0.5 mg/kg twice a day over 4 days). Cocaine is ineffective. After 3 h rest the mice are loaded with 200 µMol/kg L-DOPA. DOPA as such does not increase motor-activity, however L-DOPA may be converted into catecholamines in the brain so that stores now become refilled. A test dose of 56.2 µMol/kg *l*-cocaine 1 h after the application of L-DOPA does exert a practically normal increase in motor-activity. It must be concluded that cocaine has an indirect arterenergic action by releasing CA from stores.

mit einem direkten arterenergischen Wirkungsmechanismus, das heisst Mimetika der Katecholamine (zum Beispiel Benzedrin und Pervitin); Klasse II: Substanzen mit einem indirekten arterenergischen Wirkungsmechanismus, das heisst Wirkung durch Freisetzung der Katecholamine (zum Beispiel Kokain und N-Benzyl-Benzedrin).

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The Effect of Radioprotective Chemicals on Phosphorescent Emission of Riboflavine in Oxygenated and Deoxygenated Solutions

Recent studies, reviewed by GRAY[1] suggest that the role of oxygen in radiation damage may be connected with metabolic events in the cell, which are oxygen dependent. Furthermore it appears that at the site of radiation damage consumption and depletion of oxygen occurs *in vivo*, as demonstrated by BOAG and DEWEY[2] with *Serratia marcescens*; using a single 2 µ sec pulse of irradiation in the presence of oxygen, the bacteria were almost as insensitive as if irradiated continuously with the same dose under anaerobic conditions. Also, the studies of HUTCHINSON[3,4] have shown that for monotopic action, energy transfers

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