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Effects of general stimulant drugs on the electrical responses of isolated slabs of cat's cerebral cortex

G. B. FRANK AND K. JHAMANDAS*

Department of Pharmacology, University of Alberta, Edmonton, Alberta, Canada

Summary

1. In the neuronally isolated cortex of the cat, local application of bemegride, picrotoxin, nikethamide, caffeine and strychnine facilitated the surface positive response of the isolated cortex and lowered the stimulus threshold for this response. Excepting nikethamide, they all produced convulsive discharge in the isolated cortex unrelated to the applied stimulus.

2. Local application of glutamate to the cortex produced spreading depression, which was sometimes preceded by spontaneous positive bursting.

3. In contrast to the " general depressants " which produce a relatively consistent pattern of effects on the electrical responses of isolated cortex, the " general stimulants ", although they all have excitatory effects on isolated cortex, each produced a greatly different type of electrical response in the isolated cortex, suggesting that several different mechanisms of action are responsible for their effects.

Introduction

In a preceding study (Frank & Jhamandas, 1970a) it was found that a number of " central depressant " drugs which possess anaesthetic-like properties produced strong central nervous system excitatory effects (excitement and convulsions) when administered alone to intact mice in high doses. On the other hand, when given after pretreating the mice with phenobarbitone, these central depressants only produced central nervous system depression. Other drugs classed as " central stimulants " also produced convulsions when given alone to intact animals, but in contrast to the " central depressants ", the stimulants either did not modify or antagonized the anaesthetic-like effect of phenobarbitone.

The " central depressants " were shown to depress the electrical responses to single stimuli of isolated slabs of cat's cerebral cortex (Frank & Jhamandas, 1970b). In the present study we have investigated the effects of some " central stimulants " on the electrical responses of isolated slabs of cat cerebral cortex.

Methods

In the present investigation the effects of central stimulants, bemegride, picrotoxin, nikethamide, caffeine and strychnine, were examined on the electrical responses of the neuronally isolated cortex in the decerebrate, unanaesthetized cat.

* Present address: Department of Pharmacology, Faculty of Medicine, University of Manitoba, Bannatyne and Emily, Winnipeg, Manitoba, Canada.

The methods and experimental procedure used were the same as those described in the preceding paper (Frank & Jhamandas, 1970b).

Results

When bemegride, picrotoxin, caffeine or strychnine was applied locally to the surface of isolated cerebral cortex in an appropriate concentration they produced spontaneous activity in the cortical slab (Figs. 1 and 2). These drug-induced electrical responses in the neuronally isolated cortex prevented satisfactory recording of the responses to brief stimuli at regular intervals. To overcome this difficulty sufficiently low drug concentrations were applied to the cortical tissues, so that either no spontaneous activity occurred or if it did occur, the interference with the evoked activity was minimal. The stimulus strength used to drive the slab was subthreshold for the full evoked response, i.e. the stimulus used was 2·0 to 2·5 V below the normal strength required to elicit a positive burst response. The effects of the locally applied stimulants on the electrical responses to single pulses were studied in this manner.

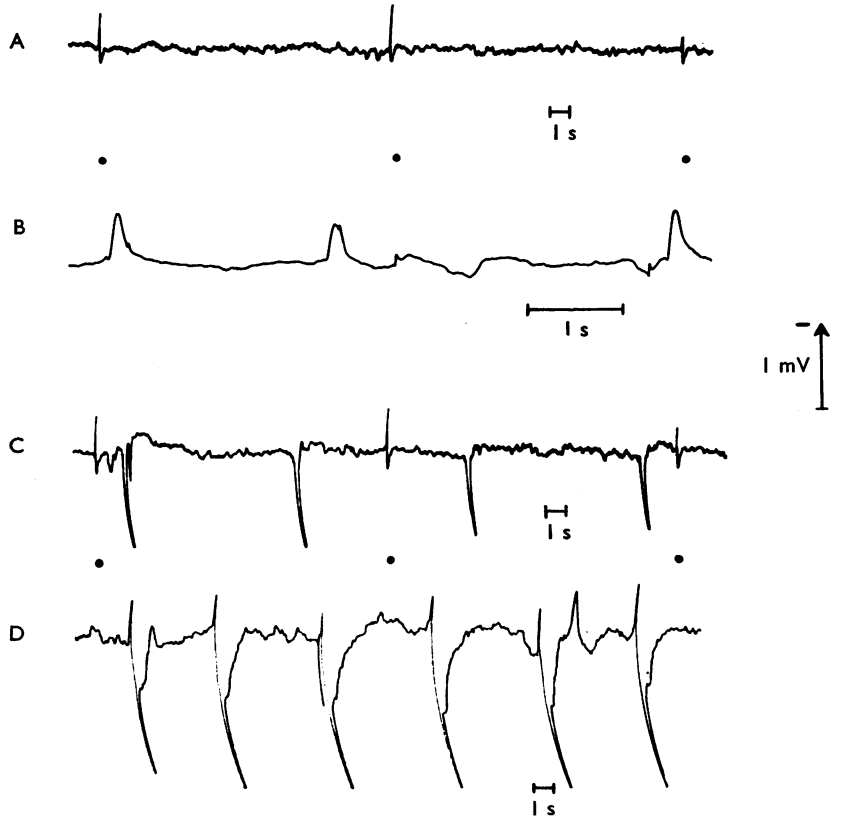

FIG. 1. Responses recorded from the surface of cat's isolated cerebral cortex. A, Control response to direct stimulation; B, spontaneous activity produced by picrotoxin (0·15% w/v); C, spontaneous activity produced by caffeine (3·0% w/v); D, spontaneous activity produced by bemegride (1·0% w/v). Dots indicate the times that stimuli were applied to the cortical slab.

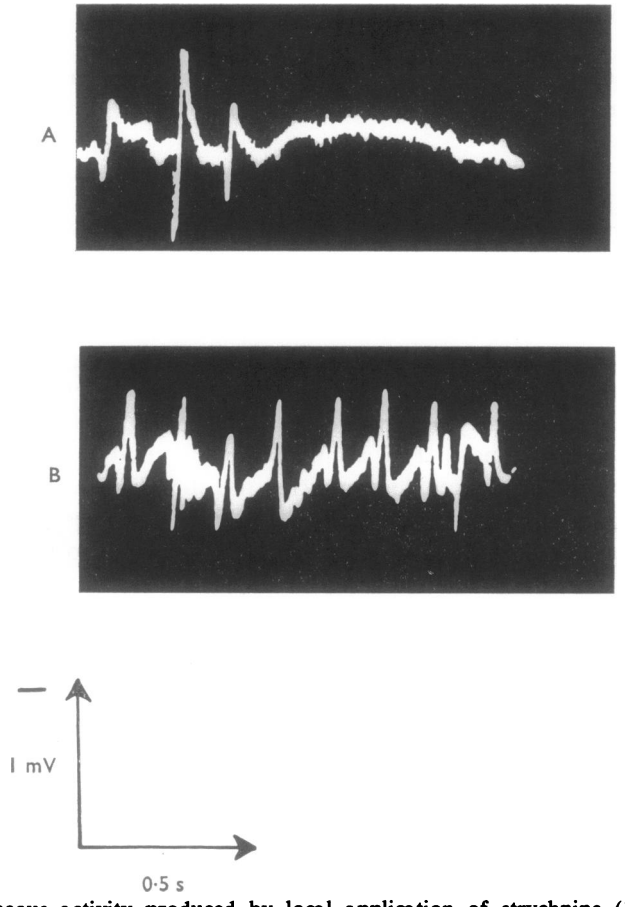

FIG. 2. Spontaneous activity produced by local application of strychnine (1% w/v) to the isolated cerebral cortex of the cat. A, after 5 min; B, after 15 min.

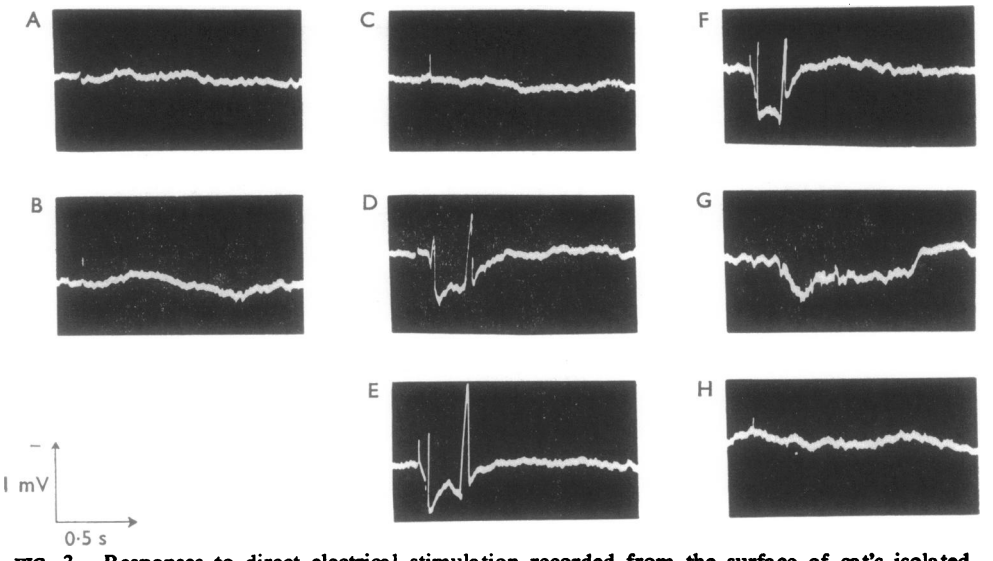

FIG. 3. Responses to direct electrical stimulation recorded from the surface of cat's isolated cerebral cortex before and after the local application of bemegride (0·5% w/v). Control responses, A and B. Responses after drug application, C, 30 s; D, 60 s; E, 3 min. The filter paper strip containing the drug was removed after E. Responses after removal of the drug, F, 10 min; G, 20 min; H, 35 min.

Bemegride. Bemegride (Fig. 3) was applied in concentrations of 0·025% or 0·05% in six separate experiments. In these doses, bemegride did not affect the surface negative response but it greatly increased the size of the positive burst response. The latter potentiated response was characterized by the presence of large negative spikes along its course. Responses to electrical stimuli recovered to their original level in 45 to 60 min after removal of the drug.

Picrotoxin. Picrotoxin (Fig. 4) in concentrations of 0·05 or 0·1% (three experiments) produced a marked increase in the excitability of the cortical slab. These changes consisted initially of large negative spikes. Several min later positive bursting occurred and epileptiform discharges, lasting 1–3 s, were superimposed on the positive bursts ; these occurred even during the resting interval between the two

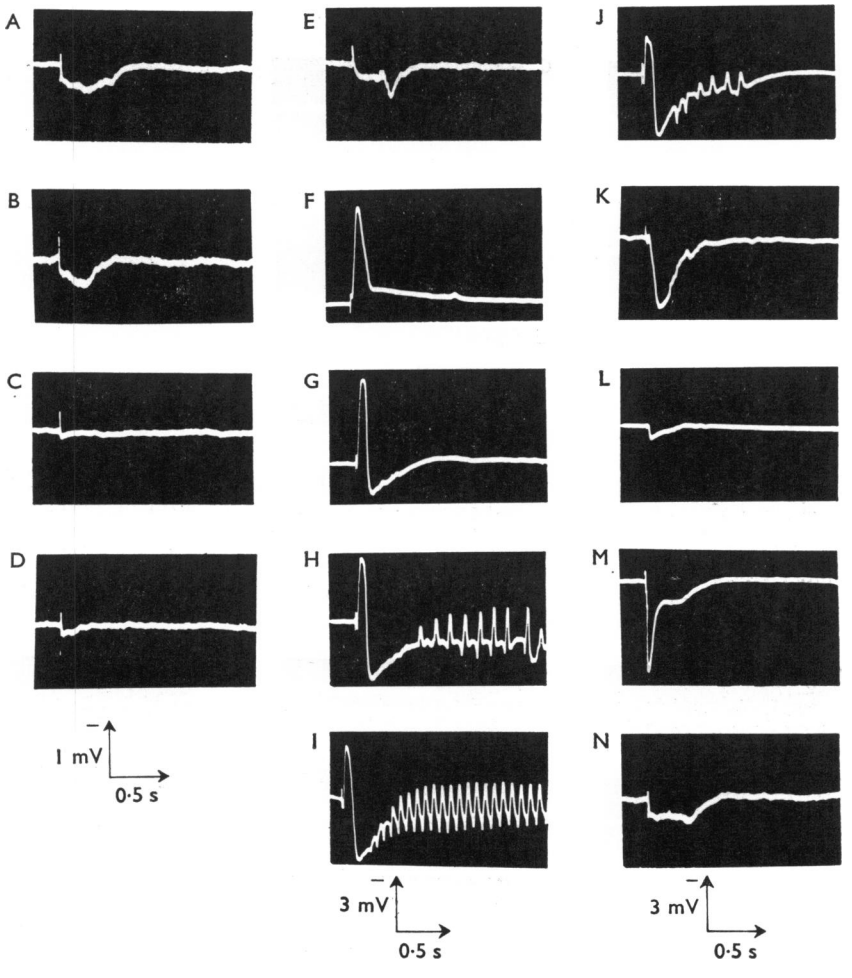

FIG. 4. Responses to direct stimulation recorded from the surface of cat's isolated cerebral cortex before and after local application of picrotoxin (0·1% w/v). Control responses ; A and B to suprathreshold stimuli ; C and D to threshold stimuli. Responses to subthreshold stimulus after drug application, E, 5 min ; F, 8 min ; G, 11 min ; H, 15 min ; I, 20 min. The filter paper strip containing the drug was removed from cortex after I. Responses after removal of the drug, J, 60 min ; K, 120 min ; L, 130 min ; M, 150 min ; N, 5 h.

stimuli. The response of the slab to electrical stimulation was reduced after this period of intense excitation and it took 4–5 h after drug removal before the control responses could be reproduced.

Caffeine and nikethamide. These drugs (Figs. 5 and 6) produced very similar changes in the excitability and electrical responses of the cortical slabs. They had little effect on the surface negative response in the slabs undergoing stimulation with subthreshold stimuli, but they both increased the amplitude of surface positive responses. Nikethamide was applied to the slab in concentrations of 5% or 10% (six experiments) and caffeine in concentrations of 2% or 3% (six experiments). The recovery from the effects of nikethamide took place in 30–60 min and from caffeine in 60–90 min. Nikethamide did not produce spontaneous discharges when applied in concentrations up to 10%, but caffeine did produce such activity when applied in a concentration of 3%.

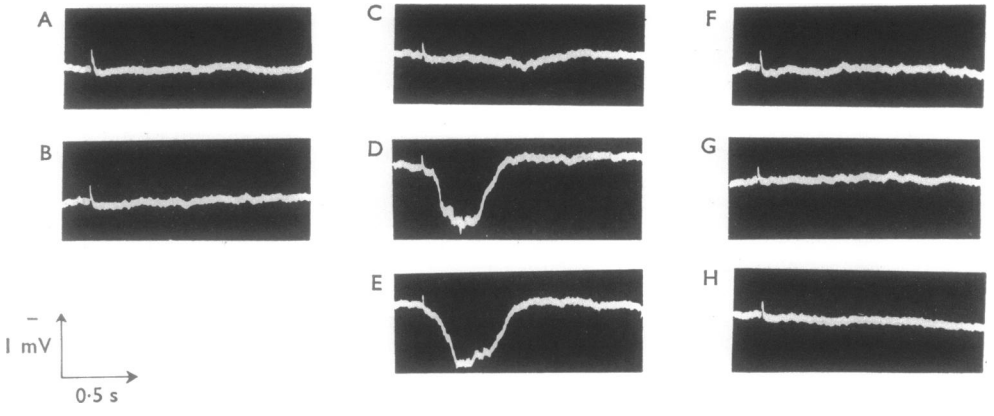

FIG. 5. Responses to direct electrical stimulation recorded from the surface of cat's isolated cerebral cortex before and after the local application of caffeine (2% w/v). Control responses, A and B. Responses after drug application, C, 3 min ; D, 4 min ; E, 6 min. The filter paper strip containing the drug was removed after E. Responses after removal of the drug, F, 20 min ; G, 40 min ; H, 50 min.

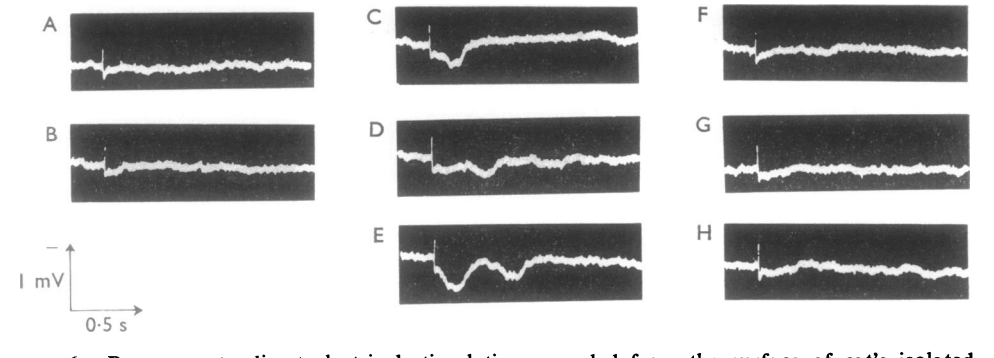

FIG. 6. Responses to direct electrical stimulation recorded from the surface of cat's isolated cerebral cortex before and after the local application of nikethamide (10% w/v). Control responses, A and B. Responses after drug application, C, 1 min ; D, 2 min ; E, 3 min. The filter paper strip containing the drug was removed after E. Responses after removal of the drug, F, 15 min ; G, 20 min ; H, 22 min.

Strychnine. Strychnine (Fig. 7), when applied to the cortical surface in concentrations of 0·1 or 0·2% (six experiments), produced effects which were similar to those produced by bemegride (Fig. 3). The positive burst response produced after drug application had a prolonged duration and was accompanied by large negative spikes. Recovery of cortical responses to control levels took place in 30–60 min after drug removal.

Glutamic acid (sodium salt). When applied in concentrations from 1 to 4%, glutamic acid (sodium salt) (eight experiments) induced spreading depression in the cortical slab (Fig. 8). All excitability was lost during the spreading depression. Normal activity returned in 30–50 min after removal of the drug. In two out of the eight experiments, the application of glutamate (2% w/v) produced a transient stimulation consisting of positive bursts before the start of the spreading depression. When glutamate was applied to the cortical slabs in lower concentrations which did not produce the spreading depression, it did not have any effect on the evoked responses.

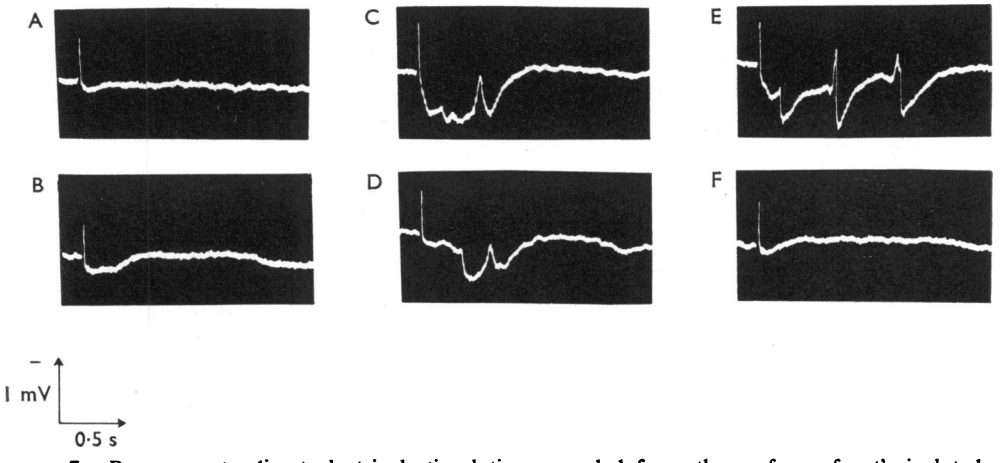

FIG. 7. Responses to direct electrical stimulation recorded from the surface of cat's isolated cerebral cortex before and after local application of strychnine (0·1% w/v). Control responses, A and B. Responses after drug application, C, 15 s ; D, 30 s. The filter paper strip containing the drug was removed after D. Responses after removal of the drug, E, 15 min ; F, 20 min.

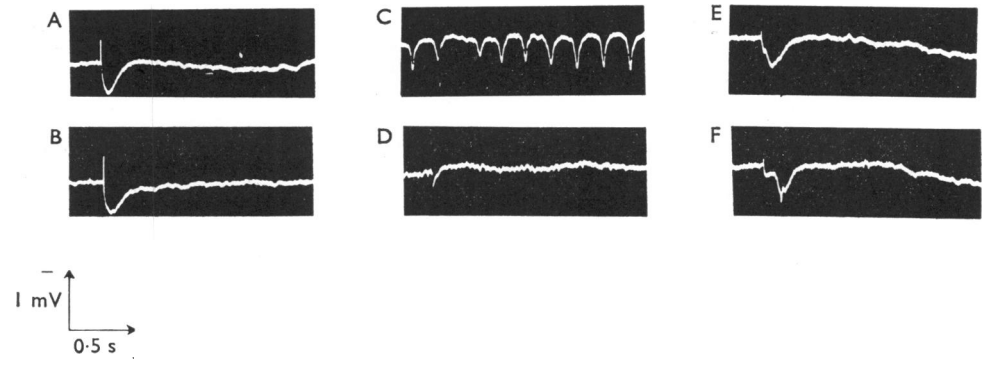

FIG. 8. Responses to direct electrical stimulation recorded from the surface of an isolated slab of cat's cerebral cortex before and after local application of L-glutamic acid (2% w/v). Control responses, A and B. Responses after drug application, C, 15 s ; D, 45 s. The filter paper strip containing the drug was removed from the cortex after D. Responses after removal of the drug, F, 25 min ; G, 65 min.

Discussion

Although many drugs which have effects on the central nervous system cause convulsions when injected by themselves into intact white mice, it has been possible to separate these drugs into two classes based on their ability (" general depressants ") or lack of ability (" general stimulants ") to potentiate the anaesthetic-like effect of concurrently administered phenobarbitone (Frank & Jhamandas, 1970a). When their effects on isolated slabs of cat's cerebral cortex were investigated (Frank & Jhamandas, 1970b ; and the present study), it was found that the " general depressants " depressed the responses to electrical stimulation and that the " general stimulants " facilitated or potentiated the responses to electrical stimulation (Table 1). Thus when studied in two different species on two different types of responses, this separation into two distinct classes was maintained ; none of the drugs placed in one of the classes when studied in intact white mice had to be placed in the other class when its effects on isolated cortex was investigated. This finding is not at all surprising provided one assumes that the classification is based on a fundamental distinction in the mechanisms of action of these two groups of drugs. That this finding was merely a fortuitous accident seems to be a far less probable explanation of the results. The mechanisms which underlie the stimulant action of the analeptics and the stimulant action of the depressants apparently differ considerably from each other. The depressants which have been shown to produce only an inhibition of

TABLE 1. *Classification of drugs acting on the central nervous system based on their effect on cortical neurones*

I General Depressants	II General Stimulants
Depress the responses of isolated cortical slabs to electrical stimuli and increase the stimulus threshold.	Facilitate the responses of isolated cortical slabs to electrical stimuli. Reduce the stimulus threshold.
Group A	Group A
Class 1. Reduce surface negative and surface positive responses. No stimulation or facilitation of these responses. e.g. Diphenhydramine Promethazine Gammahydroxybutyrate Gammabutyrolactone Gamma aminobutyric acid Hyoscine Diazepam Meprobamate Local anaesthetics* Tetrodotoxin*	Class 1. Stimulate cortex directly. Increase amplitude of surface negative and surface positive responses. Produce spiking during burst activity. e.g. Bemegride Strychnine
Class 2. Reduces surface negative and surface positive response in a low concentration. Produces spontaneous activity in a high concentration. e.g. Chlorpromazine	Class 2. Stimulates cortex directly. Increases amplitude of the surface negative and the surface positive responses. No spiking during burst activity. e.g. Caffeine
Class 3. Depresses cortical neurones upon topical application but stimulates cortical neurones upon intravenous injection. e.g. Pethidine	Class 3. No direct stimulation of isolated cortex. Facilitates only surface positive response. No spiking during burst activity. e.g. Nikethamide
	Class 4. Long duration of stimulant activity. Produces large negative spikes followed by positive bursts and strong epileptiform after discharges. e.g. Picrotoxin.
	Group B
	Initial stimulation followed by spreading depression. e.g. Glutamate

* Results from previous work (see **Discussion**).

activity at the neuronal level, may produce stimulation in the intact animals indirectly by inhibiting inhibitory neurones in the central nervous system. The stimulants on the other hand may cause excitation by directly activating excitatory neurones in the central nervous system or by blocking synaptic activity in specific inhibitory pathways.

It was suggested some time ago that both general and local anaesthetics acted by a single fundamental mechanism of action at the cellular level both in the central nervous system and on peripheral excitable tissues (Inoue & Frank, 1962). This mechanism of action consisted of a depression of excitability by the suppression of the increase in sodium conductivity which normally follows an adequate stimulus. In subsequent studies it was shown that both local and general anaesthetics had similar effects in intact white mice and on isolated slabs of cerebral cortex (Frank & Sanders, 1963). The present series of investigations represents an extension of the original suggestion of Inoue & Frank (1962) referred to above. The results obtained would suggest that all the drugs here classified as " general depressants " have the potential ability to produce an anaesthetic-like effect by an action on the central nervous system. Among the drugs classed as general depressants, all those tested so far (see Frank, 1968 ; Frank & Jhamandas, 1970a) have been shown to depress excitability in skeletal muscle or nerve fibres by suppression of sodium conductivity at concentrations comparable to therapeutic dose levels. It remains to be seen if the remainder of the " general depressants " also have this specific effect on sodium conductivity. Among the " general stimulants " it has been shown that leptazol lacks this specific effect on sodium conductivity (Inoue, Pinsky & Sanders, 1967) ; it remains to be seen if the other " general stimulants " also lack this effect.

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