microelectrode, including those which are closest to its tip, discharge in 2–3 successive bursts, within 60–70 msec after the stimulus. The probability of occurrence of neuronal discharge is maximum at the times which correspond to the highest negative values of the first derivative (the steepest negative slopes) of the gross response. This is illustrated in Figure 1. Tracing D shows a single gross evoked response, obtained from an unanesthetized, unrestrained, animal, and tracing E shows its first derivative. The tracing marked '10 up' shows all the spikes in the corresponding neuronal response, whose amplitude was above 10 μV. At the points marked 'X', it can be seen that the highest concentration of spikes corresponds to the negative peaks of the first derivative of the gross response. In the later part of the response, from 70 to 125 msec after the stimulus, much of the neuronal discharge occurs further away from the tip of the microelectrode (tracings 10, 20, 30), and the correspondence between the highest probability of discharge and the negative slopes of the gross response is not as consistent as it is in the early part of the response.

Discussion. A question arises as to whether the concentration of neuronal spikes, at the times which correspond to the steepest negative slopes of the gross response, is due to an increase in the number of neurons discharging at those times or to an increase in the frequency of discharge of the neurons involved, or to both. By separating the spikes according to amplitude it can be seen (Figure 1) that, during the times at which the negative slope in the gross response is steepest, a higher concentration of spikes occurs in all amplitude ranges, indicating that more neurons are active at those times. When the activity of a single neuron is studied it is found that both the probability of discharge as well as the frequency of discharge of that particular neuron, are greatest at the times which correspond to the steepest negative slope of the gross response. This is shown in Figure 2, in which A is the average gross evoked response, B is its first derivative and C and D are the frequency of discharge and the time histogram corresponding to the activity of a single neuron in the neuronal response. For this single neuron, the greatest probability of discharge as well as the highest frequency of discharge occur at the times at which the negative peaks of the first derivative of the gross response are highest.

It can therefore be concluded that, in the early part of the response, both the number of active neurons and their frequency of discharge are highest when the negative slope of the gross response is steepest[10].

Résumé. Il a paru intéressant de préciser les relations de temps et de phase entre la réponse évoquée massive et la réponse des neurones du corps genouillé latéral. Des microéléctrodes ont été implantées dans le corps genouillé latéral du chat et des réponses ont été évoquées par stimulation visuelle. On a pu démontrer que la probabilité aussi bien que la fréquence de la décharge neuronique varient en raison de la première dérivée de la réponse massive à ondes lentes.

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Selective Blockade by Mephentermine of Reserpine-Induced Serotonin Depletion

Previous reports have shown that the concurrent administration of reserpine (1 mg/kg) and mephentermine (1 mg/kg) produces behavioral excitation in rabbits and dogs[1,2]. The excitation produced in rabbits lasts about 30 min[1]. No stimulation occurs when either drug is given separately. The pharmacological data indicate the stimulation induced by the drug combination is central in origin and depends on the presence of brain biogenic amines[1].

The present biochemical study was undertaken to determine whether mephentermine might hasten the release by reserpine of brain norepinephrine and serotonin, thereby, increasing the concentration of 'free' neurohumor that may excite neurons. The results were unexpected and suggest that the excitation caused by the combined administration of reserpine and mephentermine is produced by the selective release of brain norepinephrine. To our knowledge, this is the first demonstration that a sympathomimetic amine will inhibit the release of brain serotonin caused by reserpine and leave unaffected the release of brain norepinephrine.

In this study behavioral excitation was produced in rabbits by giving i.v. 1 mg/kg of reserpine phosphate followed 7 min later by 1 mg/kg of mephentermine (Wyamine). One group of rabbits was sacrificed 20 min after the drug combination. These animals were hyperactive. A second group was killed 40 min after the injections; these rabbits were previously hyperactive, but were calm at the time of sacrifice. A third group of rabbits was pretreated with 100 mg/kg of iproniazid 24 and 2 h before the drug combination (reserpine and mephentermine) was given and sacrificed 1 h after the drug combination; these animals were hyperactive throughout the period of observation. Iproniazid alone does not cause hyperactivity, but it is known to prolong the stimulation studied herein[1].

Control experiments consisted of the following: one group of rabbits was given 2 injections of saline, a second group was given reserpine and saline, and the third group was administered saline and mephentermine. The injections were given 7 min apart and the animals killed 20 and 40 min after the 2 injections (Figures 1 and 2) by a

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rapid i.v. injection of pentobarbital (30 mg/kg). The brainstem was isolated (caudate, putamen and pineal were excluded) and rapidly frozen with dry ice for later analysis. The methods for determining norepinephrine (NE) and serotonin (5-HT) are given in detail elsewhere[3,4] and proved to be highly specific for these compounds.

Figure 1 clearly shows that mephentermine alone failed to reduce the concentration of brain NE. Mephentermine also failed to excite these animals. In contrast, reserpine alone, or in combination with mephentermine, lowered the concentration of NE. Also the magnitude of this reduction was similar in these 2 groups of animals at 20 and 40 min. Nevertheless, the animals given the drug combination were hyperactive at the 20 min period, whereas those given reserpine alone were not.

Pretreatment with the monamine oxidase inhibitor iproniazid prevented the rapid drop in NE obtained with reserpine and mephentermine given in combination. These animals were hyperactive 1 h after the drug combination and the mean concentration of norepinephrine in these rabbits was 314 $\pm$ S.E. 40 ng/g fresh brain weight. This value is only slightly less than those obtained for control animals that had not been given drugs.

Figure 2 shows that reserpine alone caused a loss of brain 5-HT, but that mephentermine alone had no such effect. However, mephentermine clearly prevented the loss of 5-HT that is normally caused by reserpine.

The fact that mephentermine blocked the loss of 5-HT but not the release of NE caused by reserpine, suggests that the excitation evoked by these 2 drugs in combination is related to the selective release of NE. The finding that 5-HT levels remained within normal limits following reserpine and mephentermine, whether the animal was excited (at 20 min) or not (40 min) supports this hypothesis. It is also apparent that when 5-HT and NE was released simultaneously by reserpine alone no excitation occurred. Presumably in this case 5-HT is a physiological antagonist to the actions of free NE. Since mephentermine blocks the release of 5-HT by reserpine, the stimulation seen at 20 min would be due to the selective release of NE which disappears at 40 min because the NE levels are below a critical amount for excitation.

This hypothesis is supported by the findings of others. Rats stressed by electrostimulation show a significant reduction of brain NE, but not of acetylcholine or 5-HT[5]. Morphine produces excitement in cats, reduces brain levels of NE, but does not change 5-HT concentrations[6]. Cold stress in rats and rabbits greatly reduces the ability of reserpine to lower brain 5-HT, abolishes the sedative effect of reserpine, but does not affect the rapid release of NE[7]. Also, the precursor of NE, but not of 5-HT, have analeptic effects in reserpinized mice[8], cats[9] and rabbits[1]. These results are consonant with the hypothesis[6] that NE is normally a neurohumoral mediator for ergotropic and 5-HT for trophotropic activity. Iproniazid, according to this concept, prolongs the excitation caused by the drug combination by preventing the metabolism, but not the release of NE.

Regardless of the mechanism responsible for the excitation studied, the data clearly indicate that the sympathomimetic agent, mephentermine, negates the ability of reserpine to lower brain 5-HT without affecting its ability to release NE.

Zusammenfassung. Die kombinierte i.v. Verabreichung von Reserpin (1 mg/kg) und Mephentermin (1 mg/kg) bewirkte Erregung in Kaninchen. Diese Erregung ist verbunden mit einem Verlust an Noradrenalin (NE) im Gehirn, nicht jedoch mit einem Verlust an Serotonin (5-HT). Aus den Ergebnissen lässt sich schliessen, dass die beobachtete Erregung von einem momentanen Ungleichgewicht zwischen NE und 5-HT im Gehirn herrührt.

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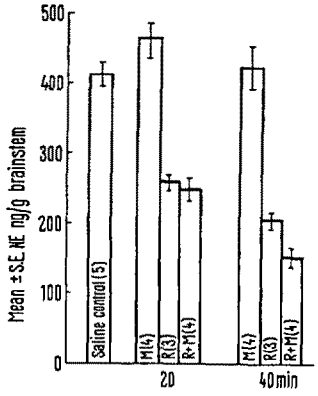

Fig. 1. Influence of mephentermine (M), reserpine (R) and the combined effect of reserpine and mephentermine (R + M) on the concentration (ng/g wet weight) of norepinephrine in the brainstem of rabbits at 20 and 40 min intervals. Numbers in parentheses represent the No. of animals in each group.

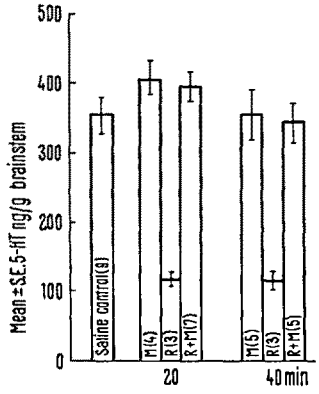

Fig. 2. Effects of mephentermine, reserpine and the combination of reserpine and mephentermine on brainstem 5-HT levels. Abbreviations as in Figure 1. Note the failure of reserpine to reduce 5-HT in the presence of mephentermine.

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