The Figure illustrates self-stimulation during light pentobarbital anesthesia. The rat was lying on its side after having lost its righting reflex, but still withdrew the extremities to painful stimuli. 'Rewarding' brain stimulation was made contingent on producing in one group of muscles motor potentials, whose amplitude and number within a period of time exceeded a preset level, while in the contralateral group of muscles the number of discharges had to remain below a critical level. In the experiment illustrated, the rat was 'rewarded' for producing at least 96 motor potentials exceeding a certain minimal amplitude within 3 sec in its left supraspinatus muscle, while during the same interval producing not more than 2 potentials in its right set of muscles (brain stimulation is indicated by the stimulus artifacts). The rat maintained self-stimulation for several minutes at a rate of 1 stimulus per 1–3 sec. Brain stimulation was generally succeeded by a silent interval, whereupon motor activity increased progressively until reaching the criterion. Discontinuation of brain stimulation was followed by a rapid decrease and disappearance of motor activity, indicating extinction of the response. Control periods using non-contingent hypothalamic stimulation ruled out the possibility that the stimulus per se induced the motor response. The training procedure consisted of first inducing a muscle discharge by pinching of a paw, then selectively reinforcing a low rate of discharge, and gradually increasing the muscle activity criterion by shaping. Self-stimulation during anesthesia was observed in 14 experiments conducted with 6 animals, the criterion for anesthesia being the loss of the righting reflex.

We have shown in previous experiments[2,3] that operant conditioning is still possible after ablation of major forebrain structures. The present observations extend those findings in showing that operant conditioning by hypothalamic stimulation is also relatively resistant to anesthesia.

Zusammenfassung. Ratten zeigen unter leichter Pentobarbitalnarkose ein Selbstreizverhalten, wobei sie die «belohnenden» Hirnreize durch Kontraktion bestimmter Muskelgruppen auslösen. Das Lernverhalten scheint demnach zu jenen Gehirnfunktionen zu gehören, die durch Narkose relativ wenig beeinträchtigt werden.

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Ambiguous Effect of Caffeine Upon the Transmembrane Ca Current in Mammalian Ventricular Myocardium

Besides its inotropic actions, caffeine markedly prolongs the action potential duration in the mammalian myocardium (KIMOTO[1]) and induces an increase of the upstroke phase of the Ca-mediated action potentials (VERDONCK, BUSSELEN and CARMELIET[2]). This suggests that caffeine increases the Ca conductivity of the membrane, which could be one reason for the augmentation of contractile activity. In order to obtain a more precise insight into the caffeine-induced changes in Ca movements, the transmembrane Ca inward current was measured under voltage clamp conditions on isolated trabeculae and papillary muscles of the right ventricle of cats.

Methods. Voltage clamp experiments were performed using the double sucrose gap technique which has been described in detail previously[3]. To separate the slow Ca current from the fast Na current, the latter was inactivated by decreasing the resting potential by 30–50 mV. When the membrane was further depolarized from this lowered potential, the slow Ca current was elicited (REUTER and BEELER[4]).

Results and discussion. The transmembrane Ca current is markedly changed by caffeine. Figure 1 shows a typical experiment. In normal Tyrode solution with a Ca concentration of 2.2 m*M* a membrane depolarization of 70 mV triggered a Ca current of 5 μA. After 5 min of exposure to 20 m*M* caffeine, the same membrane depolarization produced a current of 11.6 μA. The current voltage relationship curve was shifted to stronger currents. In 6 other experiments, the same response was obtained and an increase in current of about 100% was observed occuring 3–8 min after the addition of the caffeine (20 m*M*) to the Tyrode solution. The augmentation in current indicates an increase in the Ca conductivity of the slow membrane channel. This caffeine effect resembles that of the catecholamines, which are knwon as very effective promotors of the Ca conductivity of the membrane, and results in a potentiation of the Ca supply of the myocardial cell. But when the time of exposure to caffeine was prolonged, the increase in Ca current did not persist. At the end of this period of persistence of the increased but constant Ca current, a gradual decrease began to occur. In the experiment, shown in Figure 1, 25 min after the addition of 20 m*M* caffeine to the Tyrode solution, this slight decrease of the Ca current was observed. Then, in the subsequent 30 min, the fall became more and more evident. Finally after 55 min the constant depolarization of 70 mV produced a Ca current of only 0.5 μA. Similarly in all the other 6 experiments, the final Ca inward current was less than the control value in normal Tyrode solution. This diminution in current was accompanied by a decrease in reversal potential. In the early phase of this second period of caffeine action, the Ca current can be temporarily restored by interruption of the continuous stimulation (Figure 2). The first Ca current elicited after a resting period of about 1 min was found to be augmented. Subsequent stimulations produced a staircase-like decrease in current so that, 4–6 pulses later, the current magnitude was similar to this existing before the interruption of stimulation.

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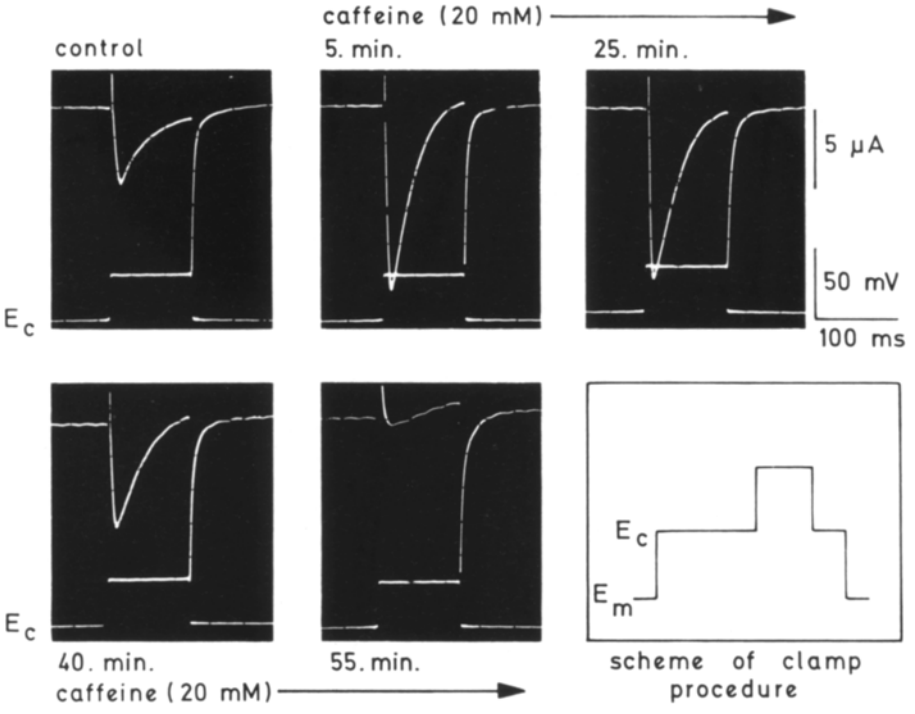

Fig. 1. Measurement of the Ca inward current in a trabecula from the right ventricle of a cat before and after addition of caffeine (20 mM) to the Tyrode solution. The upper beam of each pannel is the current registration (downward deflections indicate inward current) and the lower beam shows the membrane potential. The scheme demonstrates the clamp procedure. By a first depolarizing clamp step starting from the resting potential (E_m), the membrane potential is decreased by 40 mV in order to inactivate the fast Na system. In the original registration, this conditioning clamp step is out of screen. A further depolarization (second clamp step in the scheme) starts from the conditioning potential (E_c) and triggers the Ca inward current. Extracellular Ca concentration 2.2 mM, temperature 32 °C, pH, 7.35.

It is not considered that the gradual decrease in Ca current following a longer time of exposure to 20 mM caffeine is caused by a diminution in the Ca conductivity of the slow membrane channel. This decrease, as well as the 'staircase' phenomenon, is more likely to result from a lessening of the transmembrane Ca concentration gradient due to an intracellular accumulation of free Ca. This suggestion is based upon the fact that caffeine impairs Ca sequestration by an action at the intracellular Ca stores (Weber and Hertz[5], Nayler and Hasker[6] Blinks et al.[7], Homburger and Antoni[8]) and, in addition, blocks the Ca efflux (Shine and Langer[9]).

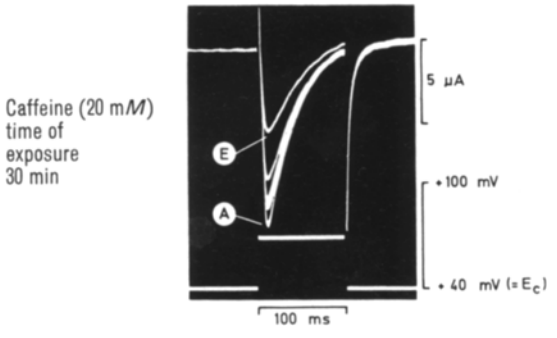

Fig. 2. Measurement of the Ca current in the presence of 20 mM caffeine (time of exposure 30 min). Staircase-like phenomenon when the membrane is continuously stimulated (frequency 20/min) after a period of rest. The first stimulation triggers the Ca current indicated by A. After 5 stimulations a steady state is reached and Ca current indicated by E is elicited. E_c means conditioning potential (40 mV less negative than the resting potential). Extracellular Ca concentration 2.2 mM, temperature 32 °C, pH 7.35.

Zusammenfassung. Coffein (20 mM) wirkt auf den transmembranären Ca^{++}-Strom der Warmblütermyokardfaser biphasisch. Zunächst nimmt der Ca^{++}-Strom zu, was auf einer Verbesserung der Ca^{++}-Leitfähigkeit des langsamen Membrankanals beruht. Nach längerer Einwirkungsdauer von Coffein kommt es dagegen zu einer Abnahme des Ca^{++}-Stroms. Als Ursache hierfür wird vor allem eine Verminderung des transmembranären Konzentrationsgradienten für Ca^{++} – infolge eines Anstiegs der freien Ca^{++}-Konzentration im Zellinnern – postuliert.

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