The amount of radioactivity for each tissue was divided by the tissue weight (*DPM*/g tissue). The data for each tissue was pooled for the animals within the control and stress groups and subjected to statistical analysis using a two-tailed *t*-test.

The Table shows that in the control group, following injection of 1 μCi of ^{28}Mg, the uptake of ^{28}Mg by the cortex was higher than by the hippocampus, thalamus and superior colliculus. Cerebellum, medulla and pituitary gland all showed higher values than the cortex with the highest uptake occurring in the pituitary. The statistical difference in the uptake was the following: cortex-hippocampus $p < 0.01$, cortex-thalamus $p < 0.001$, cortex-superior colliculus $p < 0.02$, cortex-cerebellum $p < 0.01$, cortex-medulla $p < 0.005$ and cortex-pituitary $p < 0.01$. Stress conditions induced statistically significant increases of Mg uptake in the cortex, hippocampus and the pituitary.

We reported previously that cold stress conditions induced an increase in the uptake of ^{45}Ca in the cortex, hippocampus, cerebellum, medulla and the pituitary gland[11]. The Figure compares the uptake of magnesium and calcium under control and stress conditions in the cortex, hippocampus and pituitary following the injection of 1 μCi of either ^{28}Mg or ^{45}Ca. The relation of the levels of uptake of the two cations (magnesium and calcium) differ in the cortex and the hippocampus. Magnesium uptake levels are higher in the cortex than in the hippocampus while the reverse is true of calcium. The enhancing effect of stress on the uptake of both magnesium and calcium did not affect this relation.

The lack of change in the brain and pituitary radiolabelled inulin uptake under cold stress[11] indicated no modification of the vascular bed or extracellular space. Our results tend to confirm DOUGLAS' stimulus-secretion coupling hypothesis[12], in which the stress alters the permeability characteristics of the membrane for calcium and inulin and consequently initiates hormone release. Our current findings with ^{28}Mg suggest a change in the permeability of the blood-brain and blood-pituitary barriers for magnesium induced by cold stress. This view is further supported by reports[13] of increased uptake of rubidium-86 by the brain following i.p. injection of ^{86}RbCl in rats immersed in an ice-water bath[14].

Résumé. L'imposition de conditions hypothermiques augmente l'incorporation de ^{28}Mg dans le cerveau et la glande pituitaire du rat.

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Rhythmic Activity of the Isolated Spleen during Longterm Perfusion

During asanguinous pulsatile perfusion of isolated spleens at 36 °C up to 72 h for hematological studies, we noticed oscillations in the vascular resistance. Rhythmic changes in the splenic volume, flow and pressure are typical of the in vivo spleen circulation of cats and dogs[1–4]. We could register undulatory changes during perfusion in the flow and the pressure simultaneously in the spleens of calves, pigs or humans. In addition, we could measure the influence of phentolamine, papaverine and colchicine on this rhythm.

Materials and methods. The results of the perfusion of 21 spleens of piglets (spleen weight 20–60 g) are described. The operative procedure, perfusate composition and the perfusion system are described elsewhere[5,6]. After splenectomy and a warm ischemia time of about 5 min, the spleens were flushed with an icecold, specially balanced salt solution and cooled to 4 °C. After 90 to 120 min the spleens were connected with the perfusion system. The arterial perfusion pressure was measured with a pressure transducer and the flow with an electromagnetic flowmeter; both were continuously registered on a recorder. The arterial perfusion pressure was corrected for the pressure drop across the cannula at the given flow, and this true arterial pressure was divided by the flow per minute and 100 g splenic weight to calculate the vascular resistance. The following drugs were added to the perfusate: papaverine 0.25–5 mMol, phentolamine 2.5–5 mg/l and colchicine 0.5 mg/l (Regitin® and Colcemid® of Ciba-Geigy, Wehr, Federal Republic of Germany).

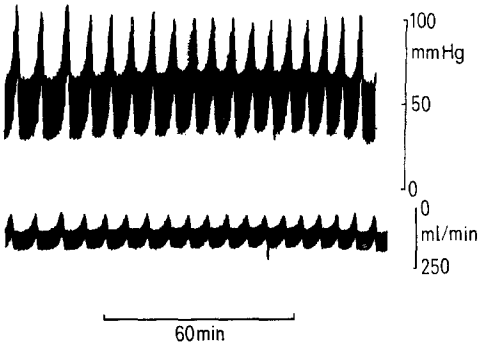

Fig. 1. Original registration of the perfusion pressure and flow of a pig spleen, 36th and 37th h of perfusion, to be read from right to left.

Results. After the first few hours of perfusion, a rather constant vascular resistance was attained and maintained for several hours; but then, sudden increases in the perfusion pressure and decreases in the flow occurred. After 1 to 2 min, the previous constant values were reached again. During the following hours of perfusion, these

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changes became more frequent and resulted in a constant rhythm (Figure 1). These oscillations were constant and were maintained in some spleens until the 72th h of perfusion, as long as there were no exchanges of the perfusate or drugs added. There were differences in the frequency of oscillation in different spleens; an individual spleen, however, had a rather constant rhythm. The mean frequency of these oscillations was 11.5 per h; that means a period of 5 min and 13 sec per wave. The average duration of perfusion until a constant rhythm was attained was 19 h. The mean peak in the vascular resistance was 145% of the previous constant value. In 15 of 21 pig spleens, such oscillations were recorded. Two slightly enlarged human spleens from patients with Hodgkin's disease had a similar rhythm. Two greatly enlarged human spleens from patients with malignant lymphoma (splenic weight 3200 g and 1850 g) had a constant vascular resistance throughout perfusion. Some spleens of calves perfused under slightly different conditions showed a similar rhythm but were not fully evaluated. There seemed to be no difference in frequency or amplitude whether the perfusate consisted of 10% or 80% pooled pig serum, or whether dextran was added to it. Even in spleens with badly-preserved lymphatic cells there was a constant rhythm. By adding papaverine (0.2–0.4 mMol) the vascular resistance decreased but the amplitude and the frequency showed no alteration (Figure 2). By increasing the dose to 5 mMol, however, the oscillations completely disappeared. After adding the α-receptor blocking agent phentolamine (2.5–5 mg/l perfusate), the vascular tone and the amplitude were diminished (Figure 3). A dose of 7.5 mg/l suppressed the oscillations completely. 2 to 3 h later, however, the rhythm began again. In 13 spleen perfusions 0.5 mg/l colchicine was added to the perfusate to block mitoses. In 12 of these cases the perfusion pressure increased an average of 25 mm Hg. In spleens which had oscillations when colchicine was added, these oscillations stopped.

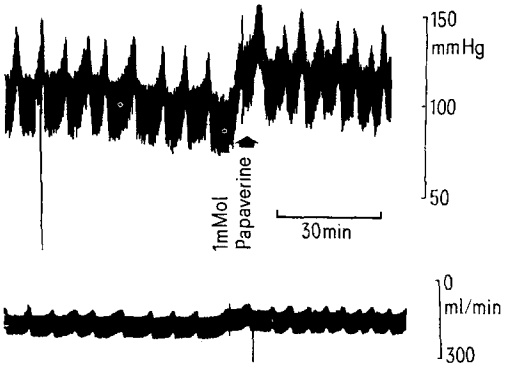

Fig. 2. Changes of the perfusion pressure and flow of a pig spleen caused by 1 mMol papaverine, 48th of perfusion, to be read from right to left.

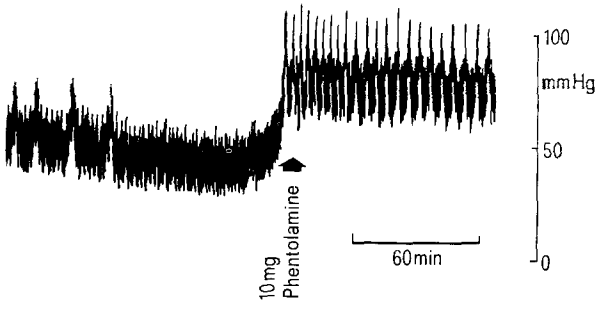

Fig. 3. Influence of 10 mg phentolamine on the perfusion pressure and flow of a pig spleen, 36th to 38th h of perfusion, to be read from right to left.

Discussion. Rhythmic activity of the spleen has, to date, only been described for the cat and dog spleen[1–4]. Those spleens have a predominant reservoir function. The spleens of pigs and calves have less muscular trabecula and the human spleen seems to have practically no reservoir function[7]. The functional significance of these oscillations remains undetermined. The relatively late appearance of the oscillation during normothermic perfusion could depend on the perfusion procedure. In cat and dog spleens, the frequency of these oscillations was 1 to 2 per min in vivo[1–4]. The 5–10 times less frequent oscillations in our system may be due to species differences, or may be only the result of the in vitro conditions. There are no data in the literature on the vascular resistance during these oscillations; only data for the flow or for the pressure. Much is known about the effect of drugs on the spleen vasculature[8], but very little about the effect on these oscillations. The effect of decreasing only the vascular tone by small doses of papaverine, and of the suppression of the oscillations by the α-blocking agent phentolamine, seems to indicate that the rhythmic activity is produced by a rhythmic release of adrenergic substances. There are adrenergic nerve terminals, as shown by fluorescence microscopy[9], distributed among the small arteries in the spleen; however, it is difficult to explain how all these terminals can be coordinated in an isolated spleen to release adrenergic substances simultaneously. There is no explanation for the increase in vascular tone with colchicine. The purpose of normothermic spleen perfusion was to establish a model to study the lymphatic system. The rhythmic activity observed in these perfused spleens was a side effect, but it demonstrates that, with normothermic organ perfusion, one can study physiological phenomena such as this rhythm under controlled and almost natural conditions.

Zusammenfassung. Bei der normothermen Perfusion von isolierten Schweine-, Kälber- und Menschenmilzen für maximal 72 Stunden wurden rhythmische Änderungen des Gefässwiderstands registriert. Der Einfluss von Papaverin, Phentolamin und Colchizin auf diese Rhythmik wird beschrieben.

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