Cold Stress Induced Changes in the Uptake and Distribution of Radiolabelled Magnesium in the Brain and Pituitary of the Rat

Even though magnesium content of the nervous system was studied in various mammalian species[1–9], there still is relatively little information about the transport of this cation into brain tissue[10]. In the present note we report the effects of cold stress conditions on the uptake of radiolabelled magnesium by certain brain areas and by the pituitary gland.

Magnesium-28 is an isotope emitting both γ- and β-radiation with a half-life of 21.3 h. The specific activity of ^{28}Mg as $^{28}MgCl_2$ was 350 μCi of $Mg^{28}Al^{28}$ per mg of stable magnesium at the time of experiment.

Adult white male rats (250–300 g) were studied under normal (control) conditions (32 animals) and under conditions of cold stress (32 animals). The stress consisted of drenching the animals in cold water at 4 °C for 30 min.

Under pentobarbital anaesthesia each animal received into the carotid artery a dose of 1.0 μCi of ^{28}Mg (in 0.2 ml of Ringer's solution buffered with 4 mM HEPES buffer to a pH of 7.56). Due to the short half-life of ^{28}Mg the injection solution was calibrated to be 1 μCi/0.2 ml (SA = 1 μCi/3.49 μg Mg) at the beginning of the experiment. The injection which was given 5 min after cold stress was followed by decapitation in 15 sec.

Following each decapitation, the brain was quickly dissected free and the following tissues placed into tared scintillation vials and weighed: cortex, hippocampus, thalamus, superior colliculus, cerebellum, medulla and the pituitary. 1 ml aliquots of tissue solubilizer (Soluene – 350, Packard) were added to each tissue vial and the tissues digested within 2 h in a water bath at 55 °C. 10 ml aliquots of a scintillation mixture (Dimilume, Packard) were then added to each vial and the vials subjected to scintillation counting. The β-radiation of ^{28}Mg was measured with the Beckman LS-200 scintillation counter within 10 h following the start of the experiment.

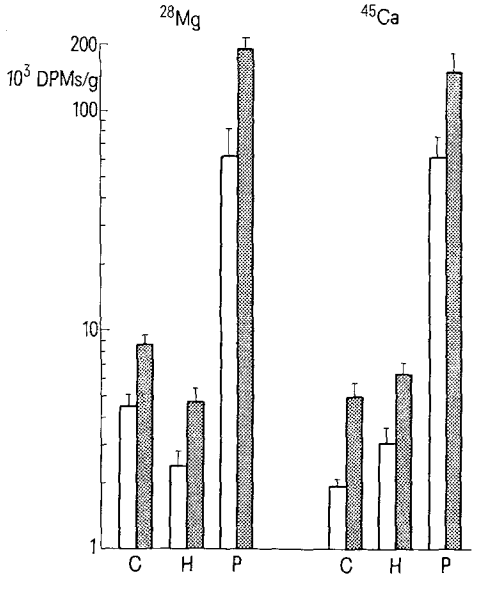
The following formulae were used:

$$(1)\quad N_t = N_o e^{(-0.6935/T)}$$

where N_t = radioactivity remaining after time t; N_o = initial radioactivity; T = half-life ($T_1/_2$) expressed in min; t = elapsed time in min; and e = base of natural logarithm.

$$(2)\quad \frac{B\text{-}CPM - B\text{-}BKG}{BQF} \times \frac{1}{N} = DPM$$

where, $B\text{-}CPM$ = raw counts for each tissue vial in channel B; $B\text{-}BKG$ = background counts in channel B; BQF = efficiency of ^{28}Mg in the B-channel; and N_t = same as above – (i.e., activity left after time t).



Uptake of ^{28}Mg (left side) and ^{45}Ca (right side) by cortex (C), hippocampus (H) and pituitary gland (P) following intra carotid injection of 1 μCi of ^{45}Ca or ^{28}Mg. Values of radioactivity are expressed in DPMs/g of wet tissue as means ± S.E.M. of 32 samples. □, control; ▦, cold stress.

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Uptake of ^{28}Mg by different brain areas and pituitary gland in rats following intra carotid injection of 1 μCi of ^{28}Mg (3.490 μg Mg in 0.2 ml Ringer's solution)

Tissue	Control	Stress	Statistical significance
Cortex	4,500 ± 450	8,500 ± 1,000	$p < 0.005$
Hippocampus	2,400 ± 470	4,800 ± 800	$p < 0.015$
Thalamus	800 ± 270	4,000 ± 1,800	NS
Superior colliculus[a]	2,300 ± 800	7,800 ± 2,700	NS
Cerebellum	8,600 ± 1,200	11,300 ± 1,400	NS
Medulla[a]	16,200 ± 2,700	17,800 ± 2,900	NS
Pituitary gland[a]	62,200 ± 20,700	192,600 ± 37,000	$3 < 0.01$

The radioactivity is expressed as DPMs/g of wet tissue. Values are expressed as means ± S.E.M. of 32 samples or [a]16 samples.

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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5 February 1974.

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[14] Research for this paper was supported by NSF No. GB 27740 X. The authors are grateful for the technical assistance of Mrs. M. TURNER, Mrs. E. ROVNER and Mrs. L. Macko.

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).

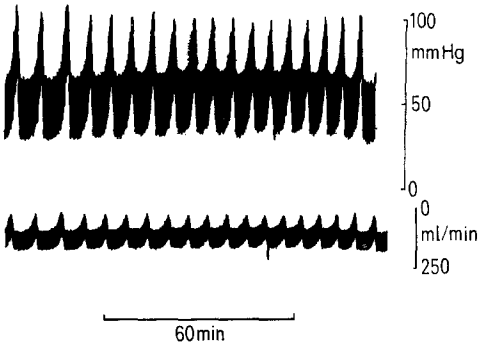

Fig. 1. Original registration of the perfusion pressure and flow of a pig spleen, 36th and 37th h of perfusion, to be read from right to left.

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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