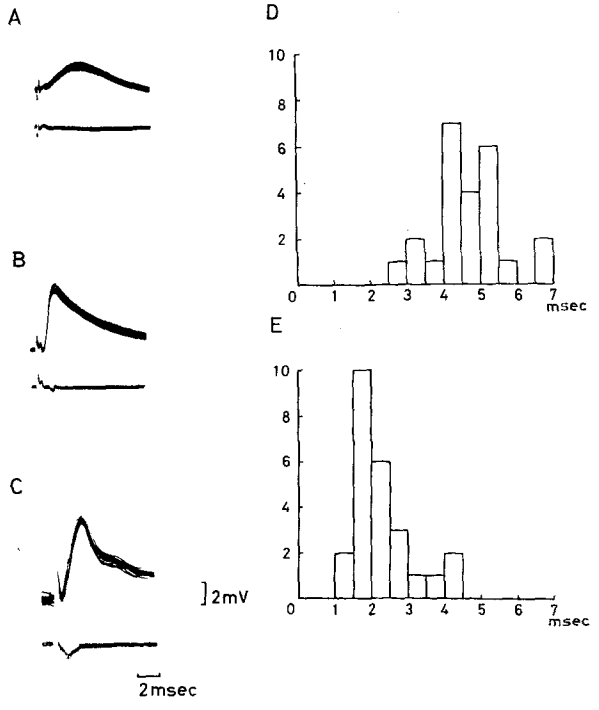
Sprouting of Cortico-Rubral Synapses in Red Nucleus Neurones After Destruction of the Nucleus Interpositus of the Cerebellum

In view of the very poor possibilities of regeneration of nerve cells in the central nervous system (CNS) in higher animals[1], the capacity for compensation by the remaining CNS following lesions must be regarded as surprisingly good. Little, however, is known about functional changes and their morphological correlates serving this compensation. As far as lesions in the peripheral nervous system are concerned, it is well established that sprouting from remaining intact motor axons is able to re-innervate denervated endplates[2]. Recent detailed morphological evidence indicates that a similar mechanism may operate also in the CNS[3]. This communication reports physiological evidence of sprouting in CNS (cf. also[4]). Histological and physiological investigations have revealed that giant neurones in the cat's red nucleus (RN) receive projections mainly from 2 sources, i.e. the nucleus interpositus (IP) of the cerebellum and the sensorimotor cortex (SM) of cerebrum. Axons from the former impinge on the somatic membrane, while those from the latter innervate the distal dendritic membranes of the cells[5-7]. Electrophysiologically, distal dendritic synaptic input is characterized by the dendritic cable properties, which cause a much slower time course of these dendritic excitatory postsynaptic potentials (EPSPs) than of somatic EPSPs[5,8]. Furthermore, hyperpolarizing currents injected from the recording microelectrode can increase the amplitude of EPSPs by increasing the membrane potential relative to the equilibrium potential of EPSP. Because of the dendritic cable properties, EPSPs caused by synaptic contacts on the dendrites are much less sensitive to membrane potential displacement (at the soma) than somatic ones[5]. By those methods we will test electrophysiologically the possibility of sprouting of intact cortico-rubral fibres on the somatic membrane after interruption of the IP input. 2 groups of nembutalized adult cats were used; one control group of normal animals (n = 13,24 RN cells) and 1 test group with IP lesions (n = 5,26 RN cells). In the latter group the left IP was destroyed electrolytically (subsequently histologically verified) 2 weeks before the acute experiment. The general procedures of intracellular recording from RN cells were the same as those employed previously[5]. During the acute experiments the sensorimotor cortex, the cerebral peduncle (at the stereotaxic coordinates of A 8.5, L 6 and H 3.7) and the ventrolateral nucleus of thalamus (VL) were stimulated (the two latter by pairs of acupuncture needles, which were insulated except at the tips). In normal cats stimulation of the VL nucleus cause a monosynaptic fast rising EPSP in RN neurones due to collateral activation of the interposito-rubral projection[9]. By stimulation of the VL nucleus the degree of destruction of IP could thus be physiologically determined during the experiments. RN neurones were identified antidromically by stimulation of the contralateral C_1 spinal segment.

The typical slow CP-EPSP and fast IP-EPSP in a normal cat are illustrated in Figure A–B. In contrast, the CP-EPSP shown in Figure C, from a cat with chronic IP destruction, has a much faster rise time and also larger amplitude than that seen in normal cats. There is no simple decay following the rapid rise and early summit; instead there is rather a later top as if the slow EPSP was superimposed on the fast one. Correspondingly, spike potentials following CP stimulation were usually initiated with shorter latencies than in normal cats. Moreover, CP stimulation caused prominent field potentials after IP destruction, while such fields were quite small in normal cats. Figure D–E illustrates the frequency distribution of the rise time of the EPSPs (time to peak) as measured from the onset of the EPSP to the top after correcting for the extracellular field potentials. It is clearly seen that the time to peak of the CP-EPSPs in chronic cats (Figure E) is much faster than in normal cats (Figure D). Similar fast-rising EPSPs could be induced by stimulating the SM. In addition the CP (and SM) EPSPs became more sensitive to membrane potential displacement as would be expected if the fast-rising part indeed represented newly formed terminals on the soma. The most straight forward interpretation of these results apparently is to assume that dendritic corticorubral synapses sprout to form synaptic contacts with the denervated somatic membrane.



A–C. Upper traces are intracellular responses in RN neurones, while the lower traces show the corresponding field potentials recorded at a just extracellular position. A and B illustrate a CP-EPSP and an IP-EPSP respectively (same cell) from a normal cat. C shows a CP-EPSP after IP destruction. Time and voltage calibrations for all intra- and extra-cellular responses are shown at C. The histograms in D (normal cats) and E (after IP lesion) illustrate the frequency distribution (number of cells on the ordinate) of the 'time-to-peak' of CP-EPSPs (in msec, abscissa).

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It may be argued, however, that a change of the cable properties could give similar results to those described above. In order to test this possibility the electrical properties of RN cell membranes were assessed by analysing the membrane transients induced by current pulses through the microelectrode. No significant changes were found between normal cats and those with chronic IP lesion. The possibility of attributing the change of the time course of CP-EPSPs following IP destruction to any change of the electric membrane properties thus seems refuted. A denervation supersensitivity could possibly be responsible for an increase of amplitude of the CP-EPSPs, but fail to explain the change in their time course.

Further experiments, including a study of the time course of sprouting, must however be performed before it is possible to relate the present findings with the well-known compensation after cerebellar lesions.

Résumé. Après destruction chronique de la projection rubrale (somatique) interpositionnelle, on constate par examen électrophysiologique que le système cortico-rubral dendritique est de toute évidence capable, par bourgeonnement, de rétablir des contacts synaptiques avec la région «dénervée» des cellules RN.

N. TSUKAHARA, H. HULTBORN[10] and F. MURAKAMI

Department of Biophysical Engineering, Faculty of Engineering Science, Osaka University, 1-1 Machikaneyama, Toyonaka, Osaka (Japan), 30 July 1973.

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Inactivation of Prostaglandin E_1 by Lungs of the Foetal Lamb

Prostaglandins of E and F types undergo substantial inactivation during passage through the pulmonary vascular bed of the adult animal[1-7]. Most probably, the loss of activity results from degradation of the compounds by specific enzymes endogenous to lung tissue[6]. We report here that lungs of foetal lambs possess similar inactivating properties.

Material and methods. Suffolk foetal lambs of known gestational age (111 to 142 days; term is 147 days) were exteriorized with the mother under methoxyflurane-nitrous oxide anaesthesia. Placental circulation was preserved while spontaneous ventilation was prevented by covering the head with a saline-filled rubber glove. Newborn (11–12 days from birth) and 6-month-old lambs were anaesthetized with sodium pentobarbital (30 mg/kg given i.v.), and anaesthesia was maintained with a mixture of methoxyflurane-nitrous oxide-air under positive pressure ventilation.

The great vessels were exposed through a left thoracotomy and the ductus arteriosus of foetal and newborn lambs was ligated. A polyethylene tube was inserted into the main pulmonary artery (foetal and newborn lambs) or in the right atrium of older animals (*pre-pulmonary catheter*). A second catheter was introduced retrogradely into the left common carotid artery and positioned in the ascending aorta just above the aortic valve (*post-pulmonary catheter*). Systemic blood pressure was recorded from the thoracic aorta using a catheter advanced from a femoral artery. A Statham strain gauge (P 23 Db) coupled to a Brush chart recorder model 200 served for this purpose. Blood gases and pH were measured with an Instrumentation Laboratory analyzer model 113 in samples from the thoracic aorta.

Prostaglandin E_1 (PGE_1) solutions were made up in saline by dilution of a 10 mg/ml ethanol-water (95:5 by vol.) stock solution. Concentrations varied between 4 and 64 µg/ml depending on the weight of the animal. PGE_1 was infused alternately into the pre- and post-pulmonary catheters at a variable rate (0.02–3.88 ml/min) with a Harward pump model 940. Each infusion lasted 2 min and 15 to 30 min intervals were allowed between administrations. In each experiment, the response to the first infusion was excluded from the final results to minimize possible errors due to tachyphylaxis.

Results and conclusion. Occlusion of the ductus arteriosus in foetal lambs produced a partially reversible fall in systemic blood pressure. After stabilization, the systolic blood pressure ranged from about 50 mm Hg at 111 gestation days to 70–100 mm Hg near term. The blood pressure was between 102 and 125 mm Hg in the newborn and 6-month-old lamb. In all animals, PGE_1 had hypo-

Pulmonary inactivation of prostaglandin E_1

Animal	Weight (kg)	Loss of activity (%)	Mean loss (± S.E.)
Foetal lamb	2.2	62	—
111, 113 gestation days	2.5	52	
Foetal lamb	3.1	76	
126–142 gestation days	3.9	56	
	3.3	64	72 ± 5[a]
	3.4	62	
	4.2	86	
	4.9	89	
Newborn lamb	4.6	87	
11, 12 days	5.1	97	84 ± 6[a]
Six-month-old	17	69	
lamb	33	85	

In each experiment, sequential dose-response curves were obtained with PGE_1 infused into the aorta and pulmonary artery (or right atrium). The percentage inactivation is given by [100-($D_a/D_{pa} \times 100$)] where D_{pa} and D_a are the doses of PGE_1 required to produce an equal fall in systemic blood pressure by the pre- and post-pulmonary routes, respectively. Rates of infusion into the pre- pulmonary line were 0.38–9.50 µg/kg/min (foetal lambs) and 0.37–7.60 µg/kg/min (newborn and 6-month-old lambs). PO_2 of thoracic aorta blood was 15–26 mm Hg (mean 21) in foetuses and 92–160 mm Hg in animals after birth. Values of pH below 7.25 were corrected with a slow i.v. infusion of a 7.5% sodium bicarbonate solution. [a] Difference between means is not significant using Student's *t*-test.

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