

Skeletomotor and Fusimotor Control of Gastrocnemius Muscle from Deiters' Nucleus¹

A fast pathway extends from Deiters' nucleus to extensor muscle spindles², and a monosynaptic connection exists between Deiters' neurones and α -extensor motoneurons³. The presence of this projection in the cat might lead one to expect that the direct control of Deiters' nucleus on α -motoneurons prevails over the indirect control through the γ -loop. Experiments were therefore performed to discover the extent of the vestibulospinal influences from Deiters' nucleus on skeletomotor and fusimotor neurones.

Methods. Muscle tension and impulse frequency in single spindle afferents from the gastrocnemius muscle were recorded in 17 cats anaesthetized with pentobarbitone sodium (Nembutal) (35 mg/kg i.v.) or decerebrated at precollicular level. Deiters' nucleus was stimulated stereotaxically with rectangular pulses, 0.5 msec in duration and at frequency above and below the threshold for the motor response. The voltage used (1.3 V) was just suprathreshold for producing the typical enhancement in the extensor tonus of the ipsilateral forelimb⁴. Both limbs were completely denervated except the left gastrocnemius muscle.

Results. 38 spindle receptors, whose afferent fibres conducted impulses at 22–101 m/sec, were excited by repetitive stimulation of Deiters' nucleus. Threshold activation was always obtained at a frequency of stimulation (80–100/sec) which caused no tension change in the muscle fixed in isometric condition. Extrafusal contraction occurred at higher frequency of stimulation (100–120/sec). However, both the extrafusal and intrafusal responses increased with rising frequency of stimulation from 100 to 500/sec (Figure 1). The relative changes of the spindle discharge produced by Deiters' stimulation with respect to the prestimulation values did not increase, however, but actually decreased with increase in muscle extension from 0 to 10 mm. Section of the ipsilateral dorsal roots

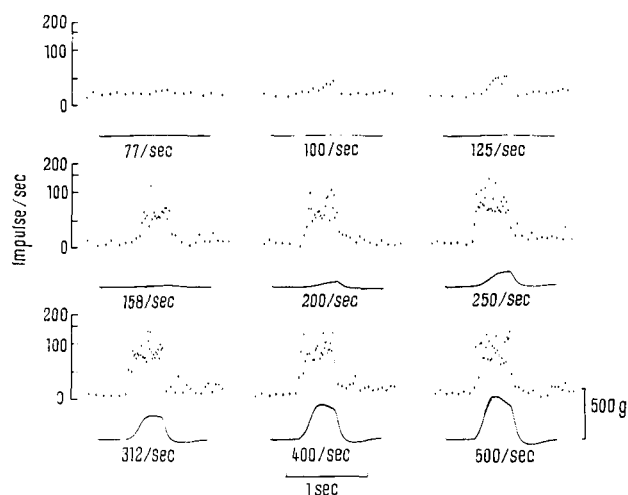


Fig. 1. Effects of repetitive stimulation of the lateral vestibular (Deiters') nucleus on a gastrocnemius muscle spindle receptor (conduction velocity of afferent fibre: 101 m/sec) and on contraction of the gastrocnemius muscle (initial muscle extension: 4 mm). Decerebrate cat. Upper traces — spindle discharge. Each dot represents an impulse. The value of its ordinate gives the instantaneous frequency of this impulse. Lower traces — muscle tension. Deiters' nucleus was stimulated with 0.5 msec rectangular pulses of 2 V at the frequencies indicated at the bottom of each record.

L6–S2 reduced the slope of the tension curve, without greatly affecting the peak of the contraction, thus indicating that the extrafusal muscle contraction persisted even after interruption of the γ -loop. The spindle receptor discharge, on the contrary, was slightly enhanced. The responses of muscle spindle receptors to stimulation of Deiters' nucleus at different frequencies persisted also in slack muscle, in spite of any possible unloading effect that might have been due to shortening of the extrafusal muscle fibres.

Factors contributing to the activation of spindles on stimulating Deiters' nucleus were investigated in 8 cats. Activation of spindles persisted after extrafusal muscle paralysis induced by intravenous injection of 0.7–1.2 mg/kg of gallamine triethiodide (Flaxedil, May & Baker Ltd.). Paralysis of γ -fibres with 0.25–0.50% solution of

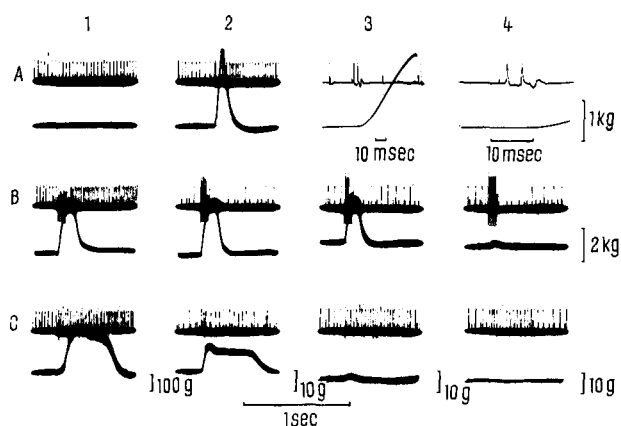


Fig. 2. Persistence of spindle receptor discharge from the gastrocnemius to repetitive stimulation of Deiters' nucleus, following procaine block of γ -efferents and extrafusal muscle paralysis with Flaxedil. Decerebrate cat. Upper traces: spindle discharge (conduction velocity of afferent fibre: 72 m/sec). Lower traces — muscle tension (initial muscle length: 4 mm).

A₁ — spontaneous discharge; A₂ — single shock stimulation of the gastrocnemius nerves at 1.56 times α -threshold (0.05 msec pulse duration). A₃, A₄ — same as A₂, but at higher sweep speeds, to show the direct spike and the early discharge. B — effects of repetitive stimulation of the gastrocnemius nerve at 300/sec, 0.05 msec, 8.3 times α -threshold. C — effects of repetitive stimulation of Deiters' nucleus at 500/sec, 0.5 msec, 2.2 V. B₁, C₁ — controls showing the spindle acceleration following stimulation of γ -efferents (B₁) and the spindle afferent receptor discharge to Deiters' stimulation (C₁). B₂, C₂ — after local procaine block of the γ -efferents distally to the stimulating electrode, note selective abolition of the γ -response to peripheral nerve stimulation in the absence of any depression of the tension curve (B₂). Both the skeletomotor and the fusimotor responses to Deiters' stimulation are depressed but not abolished (C₂). B₃, C₃ and B₄, C₄ — records taken from a series of consecutive responses following i.v. injection of Flaxedil. The fusimotor response to Deiters' stimulation persists in spite of the extrafusal paralysis. Note the high sensitivity of the myograph in C₂, C₃, C₄. Calibration of 1 sec applies to all records except A₃, A₄.

¹ This investigation was supported by PHS research grant No. NB-02990-05 from the National Institute of Neurological Diseases and Blindness, N.I.H., Public Health Service (USA).

² R. GRANIT, O. POMPEIANO, and R. WALTMAN, *J. Physiol.* 147, 385 (1959).

³ S. LUND and O. POMPEIANO, *Experientia* 21, 602 (1965).

⁴ A. BRODAL, O. POMPEIANO, and F. WALBERG, *The Vestibular Nuclei and their Connections, Anatomy and Functional Correlations* (Oliver and Boyd, Edinburgh, VIII, 1962).

procaine hydrochloride⁵ which prevented the spindle acceleration to repetitive stimulation of the gastrocnemius nerves (at 200-300/sec, 0.05 msec, 6-10 times α -threshold), abolished in most units the afferent discharge to stimulation of Deiters' nucleus. The spindles tested, therefore, were activated through γ -fibres. In some receptors, however, excitation from Deiters' nucleus persisted in spite of procaine blockage and gallamine administered at the height of γ -paralysis (Figure 2). The persistence of spindle activation in the presence of γ -nerve blockage and extra-fusal muscle paralysis shows that in some gastrocnemius spindles the effect from Deiters' nucleus is also mediated by fusimotor fibres larger than γ -efferents.

Riassunto. La stimolazione ripetitiva del nucleo di Deiters produce la contrazione del muscolo gastrocnemio,

che si accompagna ad una scarica afferente fusale. La risposta dei recettori fusali dipende non soltanto dall'attività delle fibre γ , ma anche da quella di fibre fusimotorie di più grande diametro, probabilmente di natura α .

G. CARLI, K. DIETE-SPIFF,
and O. POMPEIANO

Istituto di Fisiologia dell'Università di Pisa, Centro di Neurofisiologia e Gruppo d'Elettrofisiologia del C.N.R., Sezione di Pisa (Italy), April 26, 1966.

⁵ P. B. C. MATTHEWS and G. RUSHWORTH, *J. Physiol.* 135, 263 (1957).

Two Forms of the Effect of α, α' -Dipyridyl and 1,10-Phenanthroline on Synthesized Hydroxyproline-Deficient Collagen

We have shown in our previous work^{1,2} that some chelating agents inhibit the hydroxylation of proline-C¹⁴ and thus modify the biosynthesis of collagenous proteins in skin slices of chick embryos. By application of α, α' -dipyridyl (1 mM) collagen was formed which did not contain hydroxyproline, but its proline-C¹⁴ activity was doubled when compared with normal collagen of control samples. By the effect of 1,10-phenanthroline of the same concentration, however, a complete inhibition of proline hydroxylation as well as a partial decrease of proline-C¹⁴ incorporation into collagen occurred. Due to a certain structural similarity of both chelating agents, this contradictory effect was surprising because it pointed to a different mechanism of interference into the sequence of processes involved in the synthesis of the collagen molecule. Therefore, we were interested to find out how the above-mentioned different effect on the hydroxylation of proline and on its incorporation into collagen depends on the concentration of the chelating agent used.

Chick embryo skin slices were incubated for 2 h at 37°C in a Krebs-Ringer bicarbonate buffer with 5 μ C proline-C¹⁴ and with appropriate concentration of chelating agents. After isolation, purification and acid hydrolysis of collagen and non-collagenous proteins (purity checked by proline/hydroxyproline ratio -- see Table), the specific activity of proline-C¹⁴ and hydroxyproline-C¹⁴ was determined according to PETERKOVSKY and PROCKOP³. As non-collagenous proteins are considered all proteins which precipitate in hot TCA. The collagenous proteins remain in the supernatant. For further methodical details see ¹. The results (Table) proved unequivocally that α, α' -dipyridyl within the concentration range of 0.1-1 mM inhibits completely the hydroxylation of proline, whose activity in the collagen molecule was, however, the double

¹ J. HURÝCH and M. CHVAPIL, *Biochim. biophys. Acta* 97, 361 (1965).

² M. CHVAPIL, J. HURÝCH, B. ČMUCHALOVÁ, and E. FHRlichOVÁ, Paper presented at the International Symposium on the Biochemistry and Physiology of Connective Tissue, Lyon 1965.

³ B. PETERKOVSKY and D. J. PROCKOP, *Anal. Biochem.* 4, 400 (1962).

Effect of different concentrations of α, α' -dipyridyl and 1,10-phenanthroline on hydroxylation and incorporation of proline-C¹⁴ into collagenous and non-collagenous proteins

Substance	Collagenous proteins				Non-collagenous proteins		Proline/hydroxyproline (amount)
	Hydroxyproline-C ¹⁴		Proline-C ¹⁴		Proline-C ¹⁴		
	cpm/ μ M	Residual activity	cpm/ μ M	Residual activity	cpm/ μ M	Residual activity	
Control	14,000	100%	16,400	100%	60,000	100%	1.18
α, α' -Dipyridyl							
1 mM	143	1	31,800	194	61,800	101	1.17
0.5 mM	630	4.5	27,900	170	69,800	117	1.15
0.1 mM	1,980	14	29,500	180	71,100	118	1.15
1,10-Phenanthroline							
1 mM	35	0.25	5,900	37	15,900	26	1.28
0.5 mM	360	3	11,000	70	35,700	59	1.10
0.1 mM	734	5	15,000	94	60,300	100	1.08
0.05 mM	494	4	19,400	118	45,500	76	1.20