over the last 3 days (Table II). An analysis of variance using square root transformations was performed. No significant differences were found between sex or diet groups at the 0.05 level. The data suggest, however, that diet may have a differential effect on the activity level of male and female rats ($F = 2.14$, $df = {}^{5}/_{60}$, $p < 0.10$). When male and female activity scores were combined, differences between diet groups were not apparent[8].

Table II. Mean activity scores over the last 3 days for rats on varied diets

	Diet					
	Protein			Fat		Purina
	10%	25%	60%	29% unsaturated	31% saturated	
Male	411	604	497	600	329	579
Female	535	635	728	435	770	501
Total	946	1239	1325	1035	1099	1080

Résumé. Nous avons mesuré l'activité spontanée de rats mâles et femelles recevant des rations contenant 10%, 25% et 60% de protéine et 29% de graisses non-saturées ou 31% de graisses saturées. Les rats ont été maintenus isolés dans des cages à stabilimètre 6 jours de suite. On a comparé les données obtenues pendant les 3 derniers jours. On n'a pas trouvé de différences significatives dans l'effet des différents régimes, ni pour les mâles ni pour les femelles.

A. M. SEIFERT and J. MAYER

Department of Nutrition, Harvard School of Public Health, Boston (Massachusetts, USA), 5th December 1966.

[8] This work was supported in part by a grant from the National Institute of Neurological Diseases and Blindness, No. NB 01941, of the National Institutes of Health, U.S. Public Health Service, Bethesda, Maryland, and the Fund for Research and Teaching of the Department of Nutrition, Harvard School of Public Health, Boston, Massachusetts.

Responses of Primary Muscle Spindle Endings at Constant or Changing Muscle Length to Variations in Fusimotor Activation

A systematic analysis has been undertaken of the responses of muscle spindle endings to defined changes in fusimotor activation. This study was prompted by the fact that the fusimotor and the skeletomotor systems are often found to be activated in parallel, as is the case in various reflex contractions[1-3] (for further references see e.g. MATTHEWS[4]) and in the physiologically-induced movements in respiration[5-8]. Recent work suggests that coactivation of fusimotoneurones may also occur in voluntary contractions in man[9]. In the case of strong fusimotor activation in parallel with the contraction of the main muscle, the discharge rate of both primary and secondary endings may increase during the phase of shortening in contraction[5,8].

The responses were studied of primary and secondary spindle endings to systematic variations of static and dynamic fusimotor activation, both with constant muscle length and during length change in pace with the variations in fusimotor stimulation. This preliminary note concerns results obtained with primary endings. A full report will be published elsewhere.

Methods. In 12 cats, stimulations were performed, in ventral root filaments, of single fusimotor fibres to muscle spindles in the soleus and lateral gastrocnemius muscles, the discharge of single muscle spindle endings being recorded in dorsal root filaments. Impulse frequency was measured by an 'instantaneous frequency meter'[10]. Periodic changes of muscle length with triangular waveform (see trace c in Figures 1 and 2) were generated and monitored as previously described[11]. The designation of a fusimotor fibre as 'dynamic' or 'static' followed the same criteria as in a previous paper[10]. In order to be able to subject single fusimotor fibres to increasing and decreasing stimulus frequencies, the stimulator was triggered from a voltage-to-frequency converter with a linear input-output relation. This device, in turn, was fed by a low-frequency generator (Hewlitt Packard Model 202 A) delivering triangular (see trace d in Figures 1 and 2) or sinusoidal voltage waves. The range of stimulus frequencies of the fusimotor fibres was chosen to lie either between 20–400 shocks/sec or between 60–180 shocks/sec.

Results. Primary endings at constant muscle length. Within the range of 60–180 shocks/sec, there was a rather linear relation between the frequency of stimulation of static and dynamic fusimotor fibres and the discharge rate of the primary endings. Stimulus frequencies below 30–40 shocks/sec did not produce any significant increase in discharge rate. When the rates of stimulation exceeded 180 shocks/sec, the corresponding increments in afferent discharge rate became successively smaller and approached its maximal level. Both static and dynamic fibres yielded their maximal responses at stimulus frequencies of 250–300 shocks/sec. However, considerably

[1] R. GRANIT and B. R. KAADA, Acta physiol. scand. *27*, 130 (1953).
[2] E. ELDRED, R. GRANIT and P. A. MERTON, J. Physiol. *122*, 498 (1953).
[3] C. C. HUNT and S. W. KUFFLER, J. Physiol. *113*, 283 (1951).
[4] P. B. C. MATTHEWS, Physiol. Rev. *44*, 219 (1964).
[5] V. CRITCHLOW and C. v. EULER, J. Physiol. *168*, 820 (1963).
[6] T. A. SEARS, J. Physiol. *174*, 295 (1964).
[7] G. EKLUND, C. v. EULER and S. RUTKOWSKI, J. Physiol. *171*, 139 (1964).
[8] C. v. EULER and G. PERETTI, J. Physiol. *187*, 59 (1966).
[9] K.-E. HAGBARTH and Å. B. VALLBO, Acta Soc. Med. upsal. In press (1967).
[10] P. B. C. MATTHEWS, J. Physiol. *169*, 58P (1963).
[11] G. LENNERSTRAND and U. THODEN, Experientia (1967).

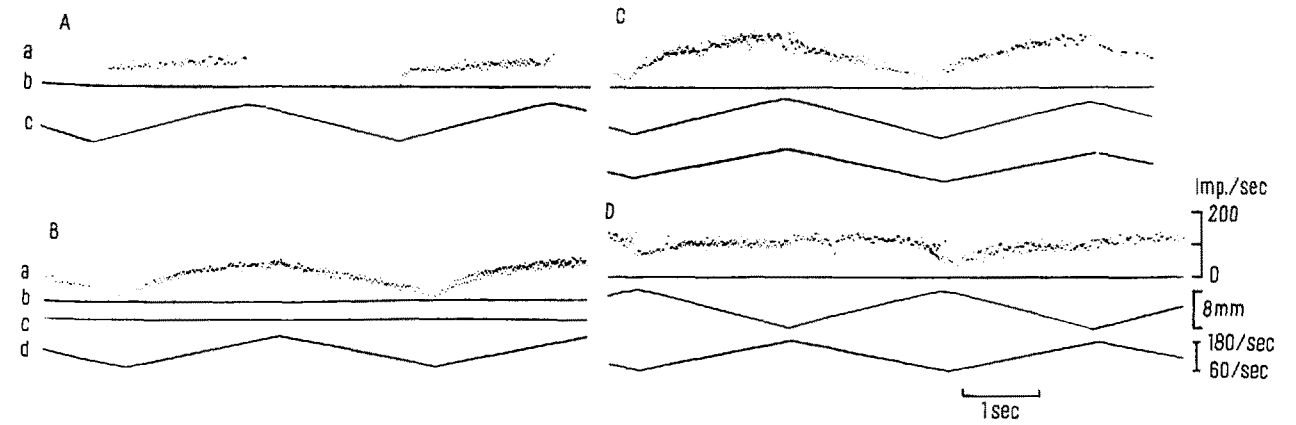

Fig. 1. Static fusimotor effects on a primary ending. Trace (a) is the impulse frequency record, (b) the zero line for the impulse frequency registration, (c) the muscle length registration with length increasing upward, and (d) the registration of stimulus frequency. The velocity of length change is 4 mm/sec and the stimulation frequency range 60–180 shocks/sec. The calibrations are the same in (A), (B), (C) and (D). (A) shows the response of the ending to muscle length changes alone, (B) varied fusimotor stimulation alone, (C) muscle length increment in phase with increasing fusimotor stimulation and (D) muscle length decrement in phase with increasing fusimotor stimulation.

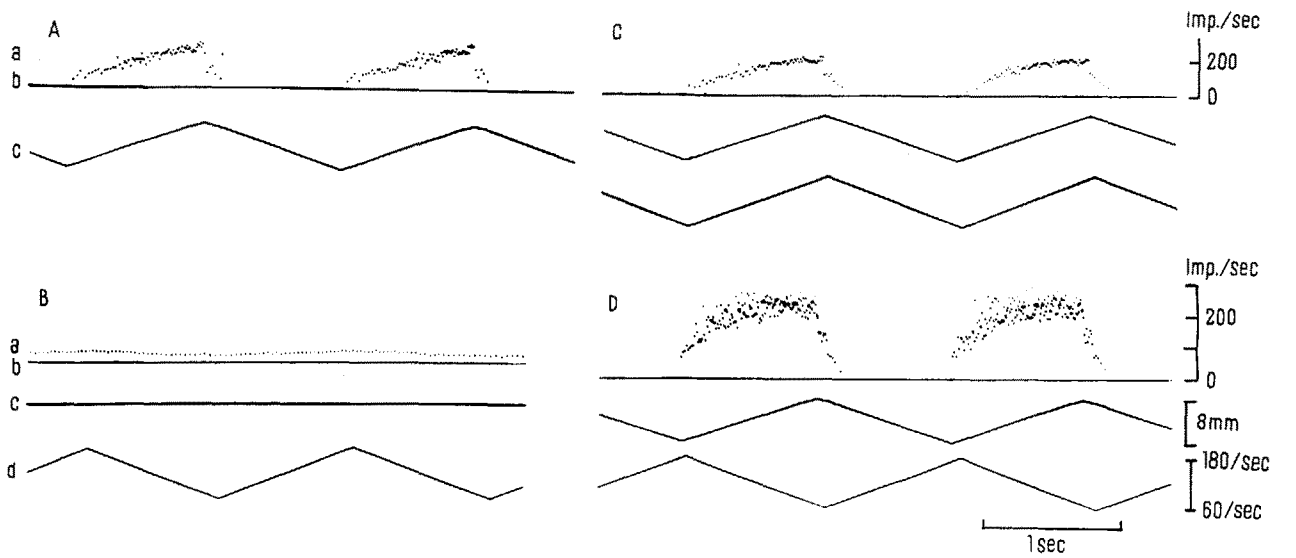

Fig. 2. Dynamic fusimotor effects on a primary ending. For explanation of traces a, b, c and d see Figure 1. The impulse frequency calibration in (C) is different from that in (A), (B) and (D). Other calibrations are the same in (A), (B), (C) and (D). The velocity of length change is 8 mm/sec and the range of fusimotor stimulation frequency is 60–180 shocks/sec. (A) shows the response of the primary ending to muscle length changes alone, (B) to varied fusimotor stimulation alone, (C) length increment in phase with increasing stimulation frequency and (D) length decrement in phase with increasing stimulation frequency.

higher discharge rates were reached in response to static fusimotor stimulation of both types described[9] than was obtained in response to dynamic fusimotor stimulation at the same stimulus frequency (Figure 1 B to be compared with Figure 2 B).

In order to determine phase shifts in the stimulus-response relations the endings were subjected to sinusoidally-modulated fusimotor stimulation. At sinus frequencies above 0.5 c/sec the response of the ending lagged behind the fusimotor stimulation. The phase lag increased with increasing frequency of sine wave stimulation. Primary endings showed roughly the same phase-lag irrespective of whether a 'dynamic' or 'static' fibre was stimulated.

When static fusimotor stimulation was modulated sinusoidally at increasing sinus frequencies, the corresponding variations in discharge rate of the primary ending could be recognized even at 16 c/sec, the highest modulation frequency applied. With dynamic fusimotor stimulation, on the other hand, the ending could seldom follow modulation frequencies above 4 c/sec.

Primary endings during changes of muscle length. In view of the above results, it was of interest to find out whether the 2 fusimotor systems differed in their ability to produce an increase in the discharge rate of the primary ending during muscle shortening, of the kind seen in respiratory movements, for example. Experiments were therefore performed in which were combined muscle length changes and varying fusimotor stimulation of the primary endings. The muscle length changes imposed on the spindles were periodic and of triangular waveform. The changes of stimulus frequency (between 60 and 180 shocks/sec) followed suit either in phase (Figures 1 C and 2 C) or out of phase (Figures 1 D and 2 D).

Typical results are given in Figures 1 and 2. Provided single fusimotor fibres were used, an increase in spindle discharge frequency during muscle shortening could only

be obtained if the fibre stimulated belonged to the static type (Figure 1 D). Simultaneous activation of 2 static fusimotor fibres or of 1 dynamic and 1 static fibre converging onto the same spindle ending, gave similar results. On the other hand, at the rates of change of muscle length and fusimotor stimulation frequency used, stimulation of a single dynamic fusimotor fibre, or of 2 converging fibres for that matter, could never increase or even maintain the impulse frequency during muscle shortening (Figure 2 D).

The findings thus suggest that the increase in impulse frequency of the primary ending seen during contraction in natural movements requires static fusimotor activation[12].

Zusammenfassung. Statische oder dynamische γ-Fasern primärer Muskelspindelendigungen in den Fussextensoren der Katze wurden mit sich ändernden Frequenzen gereizt: Nur Stimulation statischer γ-Fasern mit steigender Frequenz bei gleichzeitiger Muskelverkürzung erhöht die Entladungsgeschwindigkeit primärer Endigungen, wie sie in physiologisch induzierten Bewegungen gesehen wird.

G. LENNERSTRAND and U. THODEN

Nobel Institute for Neurophysiology, Karolinska Institutet, Stockholm 60 (Sweden), 27th December 1966.

[12] This work was supported by the Swedish Medical Research Council (Project No. 14X-544-02), and by Karolinska Institutet (Reservations anslaget för främjande av ograduerade forskares vetenskapliga verksamhet).

Primary Afferent Depolarization of Trigeminal Fibres Induced by Stimulation of Brain Stem and Peripheral Nerves

It has recently been shown that in chronic cats the excitability of trigeminal afferent fibres increases synchronously with the rapid eye movements (REM) of desynchronized sleep and at the moment of arousal[1]. Since synchronous potentials have been recorded during REM from brain stem regions[2], the hypothesis might be advanced that the latter structures are actively responsible for other phenomena temporally related to REM, as, for example, for primary afferent depolarization (PAD) of trigeminal afferents. Evidence for PAD of trigeminal fibres have already been reported in acute experiments following stimulation of other trigeminal afferents[3] and cortical areas[4].

The aim of this investigation has been to test the excitability of trigeminal afferents when conditioned by electrical stimulation of brain stem structures at bulbar, pontine and mesencephalic levels. In addition the conditioning effect of peripheral nerve stimulation was tested.

Method. The experiments were carried out in curarized acute cats under nembutal anaesthesia (25–30 mg/kg). The excitability of central terminals of trigeminal fibres was tested using the WALL technique[5] with the aid of a stainless steel microelectrode (about 100,000 Ω) stereotaxically introduced into the nucleus trigemini sensibilis principalis (NV snpr)[6] at 6–8 mm rostrally to the obex. The antidromic test response evoked by monopolar microelectrode stimulation was monophasically recorded from the killed end of the ipsilateral supraorbital nerve after the eye was enucleated (Figure A). Mineral oil was used to cover the nerve into the orbital cavity. Conditioning stimuli were applied, through stainless steel microelectrodes (about 100,000 Ω) stereotaxically introduced into the brain stem at various planes (from A6 to P11 and from midline to lateral 6, contralaterally to the trigeminal electrode) after the tentorium was removed. Bipolar silver electrodes were used for peripheral nerve – infraorbital (IO), superficial radial (SR), dorsal interosseous (DI), superficial peroneal (SP), deep peroneal (DP) – stimulation. The position of both test and conditioning microelectrodes in the brain stem was carefully controlled at the end of each experiment with Nissl stain technique and the map constructed on this histological evidence.

Results. (A) Single shock stimulation (1/sec, 0.01–0.5 msec, 30–40 V) of the main sensory trigeminal nucleus evoked in the ipsilateral supraorbital nerve an antidromic response with 0.6–0.8 msec latency (Figure A test). Conditioning electrical stimuli (4 impulses at 300/sec, 50–100 μsec, 3–12 V) applied to the controlateral half of the brain stem (medulla, pons and mesencephalon) increased the amplitude of the antidromic test response recorded from the supraorbital nerve (Figure A conditioning). Figure B shows the time course of the curve obtained by conditioning stimulation of pontine reticular formation (n. reticularis pontis caudalis) as compared with stimulation of the infraorbital nerve. It can be seen that PAD evoked by pontine stimulation starts with a longer latency and reaches a maximum (at 50 msec interval) later than that induced by ipsilateral infraorbital nerve stimulation. Figure C shows the conditioning effect induced by stimulation at 3 intensities (12, 6 and 3 V) of a pontine region (P4) at various depths and at different medio-lateral levels. It can be seen that a PAD was present when both medial and lateral regions were stimulated. However, a higher effect was obtained by stimulation of the contralateral trigeminal tract (as shown in c, c1, d, d1) as compared with stimulation of more medially situated regions. Usually 1–2 impulses were enough to give maximal PAD from the trigeminal tract, while at least 4 impulses were required to induce maximal reticular conditioning. A PAD was always present when the central core of the brain stem reticular system was stimulated, a maximal

[1] F. BALDISSERA, G. BROGGI and M. MANCIA, Experientia *22*, 754 (1966).
[2] D. C. BROOKS and E. BIZZI, Archs ital. Biol. *101*, 648 (1963).
[3] I. DARIAN-SMITH, J. Neurophysiol. *28*, 695 (1965).
[4] I. DARIAN-SMITH and T. YOKOTA, J. Neurophysiol. *29*, 185 (1966).
[5] P. D. WALL, J. Physiol., Lond. *142*, 1 (1958).
[6] J. EISENMANN, S. LANDGREN and D. NOVIN, Acta physiol. scand. *59*, suppl. 214 (1963).