Time coupling of skeletomotor discharges in response to pseudo-random transsynaptic and transmembrane stimulation

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Abstract. Firing pattern of skeletomotor neurones innervating triceps surae muscles in response to pseudo-random muscle stretching and white noise modulated transmembrane current stimulation was investigated in decerebrate cats. Pseudo-random muscle stretching (upper cut-off frequency 60 Hz, amplitude σ (standard deviation) ranging from 18.5 µm to 40 µm) was applied to triceps surae muscles. Membrane potential changes and action potentials of skeletomotor neurones were recorded intracellularly. White noise modulated current was applied through the same (recording) microelectrode. Sequences of ten identical 5 s periods of either muscle stretching or transmembrane current stimulation were applied. Skeletomotor neurones belonging to slow motor units (rheobase less than 8.5 nA) generated action potentials in response to both pseudo-random muscle stretching and transmembrane current stimulation, while firing threshold of those belonging to fast motor units could not be reached by the muscle stretches applied. Peri-spike averaging of muscle length and injected current records showed that the action potentials appeared at the peak of either depolarizing current wave or muscle stretching both preceded by a change in opposite direction (the spikes coinciding with the peak in muscle length PSA being actually elicited by muscle spindle action potentials triggered at the moment of the peak stretching velocity). Time coupling of action potentials occurred during both muscle stretching and transmembrane stimulation, being more tight in the latter case as well as when larger amplitudes of the stimuli were applied. It is supposed that discharges from muscle spindle primary endings phase-locked to small pseudo-random muscle length changes may, due to the time coupling of skeletomotor action potentials, provoke a synchronous firing of skeletomotor neurones, mostly of those belonging to slow motor units. Possible effects of such a firing pattern on the resulting muscle reflex contraction and the stretch reflex stability as well as a possibility of it being provoked by fusimotor discharges are discussed.

Introduction

Muscle spindle primary endings have been shown to exhibit phase locking: in response to cycles of periodic stimulation (sinusoidal muscle stretching) they generate action potentials occurring at a constant phase angle (Grüsser and Thiele 1968; Kröller and Grüsser 1982; Lippold et al. 1958; Matthews and Stein 1969; Poppele and Bowman 1970). At high sinusoidal stretching frequencies (muscle vibration) it results in "driving" (e.g. Brown et al. 1967) – i.e. mean discharge rate becomes equal to the sinusoidal input frequency. Recent investigations of these receptor responses to pseudo-random muscle stretches show time coupling of their action potentials to occur, depending on input signal amplitude and/or frequency (Kröller et al. 1988a; Querfurth and Grüsser 1986). The significance of this phenomenon for information transmission has been stressed (Eckhorn and Querfurth 1985; Kröller et al. 1985).

To arrive eventually to the effector – the skeletal muscle, informations encoded by muscle spindles and transmitted by their afferent fibres to the spinal cord, have to pass through skeletomotor (α) neurones. Action potentials of these nerve cells, generated in response to sinusoidal muscle stretching (Anastasijević et al. 1969; Anojić et al. 1967; Westbury 1971) as well as the resulting bursts in electrical muscle activity (Jansen and Rack 1966; Poppele and Terzuolo 1968; Tamai 1974) have been shown to appear at a fairly constant phase angle with respect to sinusoidal muscle length changes. It held true also when synaptic excitation was mimicked by sinusoidally modulated transmembrane current stimulation (Baldissera et al. 1984; Kostyukov and Kryzhanovsky 1982). At high frequency low amplitude muscle stretching (vibration) action potentials of both cat skeletomotor neurones (Homma and Kanda 1973; Homma et al. 1972) and of motor units in human jaw-closing muscles, receiving strong monosynaptic reflex excitation from muscle spindle primary endings (Desmedt and Godaux 1973), exhibited a close and
highly consistent temporal relation to the vibration cycles, while latency fluctuations of motor unit spikes in human limb muscles extended throughout the vibration cycle (Godaux et al. 1975). This absence of usually firm temporal relations was, according to the authors (Godaux et al. 1975), due to prevalence of polysynaptic excitation from muscle spindle primary endings in the tonic stretch reflex (Hagbarth and Eklund 1966).

In the studies of skeletomotor responses, however, particular attention has not been paid to the phenomenon of close time coupling of skeletomotor action potentials to the exciting stimulus. Its importance might be less obvious than that of the time coupling of afferent discharges carrying information to the central nervous system. However, if present, it might affect the resulting muscular contraction due to the known properties of skeletal muscles (dependence of muscle contraction on the succession of stimulating impulses (Gurfinkel and Levik 1973; Parmiggiani et al. 1982), "catch-property" (Burke et al. 1970)), as well as the stability of the stretch reflex considered as a parallel multi-loop feedback (Koehler and Windhorst 1981; Ögützöreli and Stein 1976; Stein and Ögützöreli 1978).

During the last ten years white noise analysis (Marmarelis and Marmarelis 1978) has been widely used in studies of the responses of non-linear physiological systems. Random stimulation is more like natural, shortens the time required for recording (which is particularly important for intracellular recording but also to avoid effects introduced by non-stationarity of spike trains) and, being provided by mathematical background for analysis of input-output relations in non-linear systems, does not impose constraints in amplitude and/or frequency range of the input signal applied. Reflex responses of cat skeletomotor neurones to pseudo-random muscle stretching and white noise modulated transmembrane current stimulation have been investigated recently in our laboratory. Findings concerning the generation and time coupling of skeletomotor action potentials will be described in this paper.

**Material and methods**

Experiments were performed on nine adult decerebrated cats. Operative procedure till decerebration was carried out under halothane anaesthesia. The right hind limb was completely denervated except for the triceps surae muscles. L4-S2 laminectomy was performed to uncover the spinal cord surface for microelectrode insertion. Ventral roots were left intact and no curarizing agent was administered.

**Recording of skeletomotor neurone action potentials**

Membrane potential changes and action potentials of skeletomotor neurones innervating triceps surae muscles were recorded intracellularly. Type of the motor unit to which the impaled motoneurone belonged was estimated on the basis of the rheobase current (Zengel et al. 1985): boundaries between the three motor unit types (slow, fast resistant and fast fatiguing) were placed at 8.5 and 15 nA respectively.

**Pseudo-random muscle stretching**

The distal tendon of the triceps surae muscles was attached to the movable arm of an electromagnetic puller (Pye Ling 406). Output signal from the white noise generator (with a shift register) was fed to the power amplifier of the puller by a length servosystem. In order to attain a steep attenuation of gain beyond the upper cut-off frequency (Stein et al. 1986) low pass filters were incorporated. A force transducer was attached between the puller and the muscle tendon for muscle tension recording (data not used in this work). Muscle length changes were monitored by Tektronix 015-0168-00 displacement transducer which also provided the negative feedback signal for muscle stretching servocontrol. The magnitude of the random stretch is given by the standard deviation (σ) of the normally distributed displacement. It was about 1/6 of the peak-to-peak displacement. Up to three different amplitudes, ranging from 110 μm to 240 μm peak-to-peak, were applied. The muscles were prestretched to the length corresponding to that attained at 90 deg ankle joint angle. The upper cut-off frequency of the noise, being limited by the stretch apparatus, was set at about 60 Hz while the lower cut-off frequency (0.25 Hz) was defined by the duration of recording (5 s, the first one discarded, see below).

Main characteristics of the stimuli (amplitude probability density, power spectra) were checked regularly with a Hewlett Packard computer (sampling interval 1 ms, sampling period 4096 ms). Schemes of the stimulus apparatus and recording, as well as the main properties of the stimuli are shown in Fig. 1.

**White noise-modulated transmembrane current stimulation**

The output signal of the white noise generator was injected intracellularly through the recording micropipette. Two different amplitudes, in the range from 4 to 64 nA peak-to-peak dependent on the rheobase of each particular cell, were applied. The lower cut-off frequency (0.25 Hz) was defined by the duration of recording, while the upper cut-off frequency of the noise was about 1280 Hz. It was not cut down to the values comparable to the upper cut-off frequency of pseudo-random muscle stretching. This made the comparison of skeletomotor responses more difficult but offered the possibility of observing the effects of higher input signal frequencies than those obtainable with the stretching apparatus. Current records were sampled at a rate of 4 kHz.

**Experimental protocol and analysis of records**

Sequences of ten identical 5-s periods of either pseudo-random muscle stretching or intracellular stimulation were applied. Records of skeletomotor action potentials and the corresponding input signals were