THE INFLUENCE OF VIBRATORY STIMULATION ON THE NOCICEPTIVE COMPONENT OF THE LOWER LIMB FLEXION REFLEX IN MAN

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ABSTRACT

The nociceptive component of the flexion reflex (FR) has been observed by several investigators to be a reliable physiological manifestation of pain perception in man. It has thus been used as an index of spinal nociceptive activity to study analgesic mechanisms. Vibration is one analgesic procedure which has been reported to decrease subjective pain report in certain pain conditions. The objective of this study was thus to quantitatively investigate the behavior of the human FR during segmental peripheral conditioning with high-frequency vibration.

A first study was undertaken to determine the relationship between pain sensation and the FR under the paradigm developed in this laboratory. Visual analogue scale (VAS) ratings of subjective sensation were found to bear a high, linear correlation with stimulus intensity ($r=0.95$) and with FR area ($r=0.91$).

In the main study, we found segmental vibration to inhibit the FR in 5 of 9 subjects. The modulatory effects displayed a slow onset and decay, lasting beyond the 30 min vibration period. Maximum modulation occurred late into vibration, and in some cases after vibration. The direction and magnitude of modulatory effects were basically the same in both biceps femoris (BF) and tibialis anterior (TA) muscles. Our results raised the possibility that prolonged vibration could influence spinal nociceptive reflexes in ways

similar to the vibration-induced pain relief described in the clinical literature.

RESUME

La composante nociceptive du réflexe de flexion (RF) au membre inférieur s'est avérée être une manifestation physiologique fiable du percept de la douleur chez l'humain. Le RF a donc été utilisé comme indice de l'activité nociceptive spinale dans plusieurs études investigant les mécanismes de l'analgésie. La vibration est une procédure analgésique pouvant réduire le rapport subjectif de douleur dans certaines conditions douloureuses. Le but de cette étude était donc d'investiguer de façon quantitative le comportement du RF humain sous l'effet de la vibration à haute fréquence, appliquée de facon périphérique segmentaire.

Une première étude fut entreprise pour déterminer la relation entre la sensation de douleur et le RF, selon la procédure developpée dans notre laboratoire. La sensation subjective, telle que rapportée sur une échelle visuelle analogue, démontra une haute corrélation linéaire avec l'intensité du stimulus (r=.95), ainsi qu'avec l'aire du RF (r=.91).

L'étude principale révéla un effet inhibiteur de la vibration segmentaire sur le RF chez 5 sujets sur 9. Les effets modulatoires s'installaient graduellement et persistaient après la période de vibration de 30 min. La modulation maximale apparaissait vers la fin de la période vibratoire, ou même après. La direction et l'importance des effets modulatoires étaient essentiellement pareils dans le

biceps fémoral et le jambier antérieur. Nos résultats suggéraient que la vibration prolongée pourrait influencer les réflexes nociceptifs spinaux d'une façon analogue à celle décrite dans la littérature concernant le soulagement de la douleur.

DEDICATION

To my parents,

without whose unfailing confidence in me,

love and gentle guidance,

none of this would have been possible.

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PREFACE

According to section 7 of the Guidelines Concerning Thesis Preparation as specified by the Faculty of Graduate Studies and Research, McGill University (March, 1989*), the student has the option of including as part of the thesis, the text of and original paper, or papers, suitable for submission to learned journals for publication. This option has been taken for the present thesis with the authorization of the Chairman, Department of Physiology.

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The candidate has the option, subject to the approval of the Department, of including as part of the thesis the text, or duplicated published text (see below), of an original paper, or papers. In this case the thesis must still conform to all other requirements explained in Guidelines Concerning Thesis Preparation. Additional material (procedural and design data as well as descriptions of equipment) must be provided in sufficient detail (e.g. in appendices) to allow a clear and precise judgement to be made of the importance and originality of the research reported. The thesis should be more than a mere collection of manuscripts published or to be published. It must include a general abstract, a full introduction and literature review and a final overall conclusion. Connecting texts which provide logical bridges between different manuscripts are usually desirable in the interests of cohesion.

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LIST OF REFERENCES

Chapter 3: Chan, C.W.Y. and Dallaire, M., Subjective pain sensation is linearly correlated with the flexion reflex in man, Brain Res., 479 (1989) 145-150.

Chapter 4: Dallaire, M. and Chan, C.W.Y., Prolonged peripheral vibration could inhibit the nociceptive component of the lower limb flexion reflex in man. Submitted to Brain Research.

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CHAPTER ONE

GENERAL INTRODUCTION

LITERATURE REVIEW

Ever since the pioneering studies conducted by Sherrington and his colleagues in the early 1900's, the flexion reflexes have been the object of extensive studies in both animals and man. Originally investigated to acquire a basic understanding of simple neural networks, and later for their functional significance, the flexion reflexes are now being employed in the realm of human pain research. Specifically, an experimental methodology for the elicitation and recording of the flexion reflex (FR) in the lower limb has been developed, which can be applied to quantitative studies of pain (Chan and Tsang, 1985; Hugon, 1973; Willer et al., 1976).

The flexion reflex is a spinal reflex. It doesn't require the participation of suprasegmental structures (Hugon, 1973; Lloyd, 1943). Based on its latency, it has been established that the flexion reflex loop comprises at least one interneuron, which classifies it as a polysynaptic reflex (Lloyd, 1943).

Flexion reflex networks are simple, yet coordinated and purposeful (Pansky and Allen, 1980). The function of the FR is widely accepted as being protective in nature, i.e. in response to noxious stimuli, the FR results in withdrawal of the limb from the offending agent, thereby preventing further

tissue damage. The evolutionary significance and survival value of such a mechanism is obvious.

The reflex pattern typically consists in excitation of flexor muscles and inhibition of extensor muscles in the stimulated limb. The amplitude of the resulting movement depends on the intensity of the insult. A low-intensity pain will result in a small, localized movement of the limb, sufficient to remove the affected area from the stimulus. A highly painful stimulus will elicit a widespread response of all the limb flexors, causing the gross removal of the whole limb , with the limb remaining flexed for some time after removal from the stimulus ("after-discharge") (cf. Eyzaguirre and Fidone, 1975).

Pathway of the flexion reflex

As mentioned above, the FR is an injury-sensitive reflex. However, its neuronal circuits are not strictly reserved for the transmission of nociceptive impulses. In fact, its afferent limb comprises fibers of different diameters and conduction velocities mediating different somatic sensations (Hugon, 1973; Lloyd, 1943). These fibers are classified into groups (Lloyd, 1943): the large Group II fibers conveying tactile sensations from mechanoreceptors; the smaller, higher-threshold Group III fibers mediating sharp, localized pain; and the smallest, unmyelinated Group

IV fibers mediating aching, diffuse pain. The latter two fiber types arise from skin and muscle nociceptors (Kandel and Schwartz, 1985).

Upon entering the spinal cord, the FR afferents synapse with interneurons in the gray matter. These synaptic connections will shuttle the afferent information to multiple destinations involving segmental, proprio- and supra- spinal mechanisms. At the segmental level, the ipsilateral flexor motoneuron pool will be activated, resulting in limb flexion. Concurrently, the ipsilateral extensor motoneurons will be inhibited (reciprocal inhibition), while in the contralateral limb, there will be extensor excitation and flexor inhibition (crossed extensor reflex). This segmental activity is designed for smooth, coordinated withdrawal of the affected limb along with maintenance of upright posture (Kandel and Schwartz, 1985).

It has also long been observed that input from the lower limb or lower cord could lead to reflex activity respectively in the upper limb or upper cord (Kearney and Chan, 1979; Shimamura and Livingston, 1963). This implies that the FR afferent information is not confined to the affected spinal segment, but is relayed to the segments innervating the other limbs as well. In this connection, Shimamura and Livingston (1963) have investigated the pathways underlying the multi-segmental expression of reflex activity evoked by stimulating various dorsal roots and peripheral nerves in the cat.

Recording from ventral roots at different spinal levels allowed them to monitor changes in the configuration and latency of the double-burst reflex pattern observed. Their results provided evidence of two respective longitudinal sensorimotor reflex systems subtending the two bursts, and mediated by cutaneous and high-threshold muscle afferents in the cat. The first burst was found to be mediated by a propriospinal system. This system is relatively direct and is characterized by bilateral projections which frequently cross and re-cross the spinal cord. Such a system provides a neurophysiological basis for multi-segmental reflex activity upon localized segmental stimulation.

There remained, however, a major question as to the nature of the pathway mediating the late reflex component observed in the study. Shimamura and Livingston's (1963) results strongly suggested the late burst to be mediated by a long-loop pathway involving supraspinal structures. Specifically, they described a reflex system involving a relay in the bulbar reticular formation, the so-called spino-bulbo-spinal system in the cat. In man, there is no evidence suggesting that a long-loop pathway might mediate the late FR component in the lower limb. In fact, several investigators have described a lower limb FR EMG burst in spinal man similar to the long-latency burst recorded in normal subjects (Dimitrijevic and Nathan, 1968; Hugon, 1973; Roby-Brami et al., 1987).

Despite the lack of evidence for a long-loop pathway mediating the late component of the human lower limb FR, there is no doubt that propriospinal circuits are constantly modulated by descending pathways from higher brain regions. Corticospinal volleys have been shown to produce excitation of spinal interneurons (Lundberg, 1964) and primary afferent depolarization of cutaneous afferents (Willis and Coggeshall, 1978). The "dorsal reticulospinal system", arising from the medial portion of the lower brainstem and descending in the dorsolateral fasciculus of the spinal cord, is responsible for a tonic inhibition of transmission through the spinal FR pathways (Willis and Coggeshall, 1978). Moreover, the existence in man of supraspinal centers capable of modulating spinal excitability is well established (Bathien and Morin, 1972; Hugelin, 1972). In this connection, mental states such as anxiety and accrued attention have been shown by Willer and associates (1979) to be capable of exerting a modulatory influence on the flexion reflex, thus implicating limbic and reticular arousal systems - at least according to Routtenberg's (1968) hypothesis.

Electromyographic analysis of lower limb FR in man

The above information obtained from animal experiments serves as a background for the study of the FR in man. Using electromyographic (EMG) techniques, the ipsilateral lower limb

FR elicited by electrical stimulation of the sural or tibial nerve or their receptive field, has been found to typically consist of two excitatory components separated by a period of inhibition (Hagbarth, 1960; Hugon, 1973; Meinck et al., 1981). These components characteristically appear at latencies of 40-60 and 70-150 ms, respectively. The initial phase of the reflex may be excitatory or inhibitory, depending on the muscle (e.g. biceps femoris, tibialis anterior, quadriceps) investigated (Meinck et al., 1981).

The two EMG components of the FR have very distinct profiles. The short-latency component has a comparatively low threshold (Lloyd, 1943; Shahani and Young, 1971) and almost always requires a train of mild electrical pulses (i.e. temporal summation) to be elicited (Hugon, 1973; Janko and Trontelj, 1983). The late component manifests itself at higher intensities of stimulation and can be elicited by a single pulse or a train of pulses, the critical parameter here being a high stimulus intensity. Besides elicitation threshold, several arguments suggest that the early and late FR components are mediated by Lloyd's (1943) group II and group III fibers, respectively. These include compound action potential configuration in the afferent fibers, conduction velocity of afferent volleys, and latency of the respective EMG components (Hugon, 1973). The early and late EMG components of the flexion reflex have thus been dubbed RA II and RA III, respectively (Hugon, 1973). The two components

also seem to differ in their duration. The RA II response has been generally observed to be of shorter duration than the RA III response (Hugon, 1973; Shahani and Young, 1971), presumably due to less dispersion of impulses conducting along the larger fibers.

It is noteworthy that the RA II response is often absent from the biceps femoris, especially in subjects who are not relaxed (Hugon, 1973). Intense stimulation may also obscure the distinction between the two reflex bursts. Shahani and Young (1971) have found that increasing the stimulus intensity decreases the latency of both components (this effect being more pronounced in RA III) and increases their duration, often causing them to merge.

Additional supportive evidence of the identity of the flexion reflex afferents is supplied by behavioral studies. Such studies have revealed that the sensation produced by stimuli evoking the RA II reflex is of a tactile nature, akin to repetitive tapping with a reflex testing hammer (Hugon, 1973; Willer, 1983). The large, myelinated group II cutaneous fibers are known to convey such tactile, innocuous stimuli (Collins et al., 1960). Conversely, subjective reports give a strong correlation between the RA III component and sharp pricking pain (Willer, 1983; Hugon, 1973). Such pain is known to be transmitted via the fine, high-threshold myelinated group III cutaneous afferents (Collins et al., 1960). Although Kugelberg (1948) believed the RA III reflex to be due

to activation of group IV fibers, their slow conduction velocity seems to preclude their involvement in the RA III reflex (Hugon, 1973; Faganel, 1973). At the motor level, the RA II reflex is the electromyographic expression of a small amplitude muscle twitch, the functional significance of which is believed to be that of a locomotor placing reaction (Hugon, 1973); while the RA III reflex is associated with gross movement and withdrawal of the limb (Shahani and Young, 1971).

Pain research and the nociceptive flexion reflex

Various manifestations of the flexion withdrawal reflex are commonly used as behavioral tests of analgesia in animals (D'Amour and Smith, 1941; Martin et al., 1964). Of particular relevance here is the hindlimb flexion reflex, the late component of which is held to faithfully represent a nociceptive reaction. In this connection, Chung and his collaborators (1983) have reported a selective, naloxone-reversible suppression of the late component of the hindlimb FR in cat with morphine. This influence of morphine on the FR is in agreement with the effects of morphine on clinical pain, and with the morphine-induced inhibition of activity in nociceptive dorsal horn neurons described by Le Bars and his co-workers (1975, 1976). Additional evidence confirming the parallel behavior of the flexion reflex and pain comes from studies investigating the influence of peripheral afferent

conditioning on nociceptive reactions. For example, transcutaneous electrical nerve stimulation (TENS), which is used for the relief of pain in some clinical conditions, has been shown to depress the flexion reflex in cat (Chung et al., 1983; Shin et al., 1986) and in man (Chan and Tsang, 1987). These reports also concur with the inhibition of C-fiber evoked activity in primate spinothalamic tract cells observed by Chung et al. (1984) and Lee et al. (1985) upon conditioning with TENS. Conversely, Wall and Woolf (1984; Woolf and Wall, 1986) have observed a prolonged facilitation of the flexion reflex upon conditioning with selective electrical stimulation of C-fibers. They described this facilitation as strikingly reminiscent of certain types of clinical pain characterized by prolonged, poorly-defined hyperalgesia. This could explain the occasional clinical observation that TENS might enhance pain slightly.

In man, the high correlation between the RA III response and subjective pain sensation has led many investigators to believe that the flexion reflex might provide a means to gain insight into the mechanisms of pain transmission. In this regard, a methodology was developed using electrical stimulation of the sural nerve, or the distal receptive field supplied by its branches, to elicit the lower limb FR in man (Hugon, 1973; Willer et al., 1976). Willer and co-workers (1976) found that stimulation of the skin of the sole in the sural nerve's distal receptive field elicited an RA III

response without the RA II component, as well as a concomitant burning pain sensation. They also found that the threshold for eliciting the RA III response (and pain) from the distal sural receptive field is about half as much as the threshold for its elicitation from stimulating the sural nerve itself. The isolated RA III response is thought to be due to preferential activation of small-diameter nociceptive fibers which innervate pain receptors lying in the more superficial dermo-epidermal nerve plexus (Willer, 1977, et al.,1976). Indeed, the larger myelinated cutaneous afferents are located in the deep, dermal plexus, and innervate encapsulated receptors which are not very sensitive to electrical stimulation, but respond mainly to specific stimulation (Crosby et al., 1962; Winkelmann, 1960).

The possible existence of a causal relationship between the absence of RA II response and a lower RA III elicitation threshold was further pursued by Willer's group (Willer et al., 1976). They carried out a selective ischemic block of group II fibers during sural nerve stimulation and found RA III amplitude and pain sensation to increase along with the concurrent disappearance of RA II response and decrease in RA III threshold. In this connection, Hugon (1973) had already observed that both RA III response and pain sensation are facilitated by a preceding noxious stimulus (producing both RA III and pain), while precession of the test stimulus with a tactile stimulus showed an inhibitory effect. These results

agreed with Melzack and Wall's Gate Control Theory of pain (Melzack and Wall, 1965; Wall, 1978), which states that pain transmission at the spinal level is gated by the ratio of activity in large-diameter afferents versus that in small-diameter afferents.

The relevance of the flexion reflex to pain research has been further investigated by Chan and Tsang (1985). They established a methodology, using stimulation of the tibial nerve's distal receptive field, which elicits a stable, reproducible RA III reflex, allowing a quantitative analysis of this nociceptive FR component. As was the case with Willer's distal sural receptive field stimulation, they found the flexion reflex EMG under their paradigm to typically consist of an isolated RA III burst. They also observed that by requiring the subjects to maintain a constant level of tonic background contraction in their leg muscles, the FR elicited in these muscles was remarkably consistent in time as well as in magnitude. Superimposing a variable inter-stimulus interval onto this tonic background contraction also helped to circumvent the well-known habituation of the FR. Computerized averaging of the rectified and smoothed signals further allowed the collection of accurate data concerning the relationship between stimulus intensity and RA III amplitude and area values. They found both amplitude and area to increase linearly with increasing stimulus intensity. In a recent study using the aforementioned paradigm, Chan and

Tsang (1987) found the flexion reflex to be progressively inhibited by TENS in the majority of their subjects.

The similar organization of flexion reflex and pain pathways is further confirmed by the observation of Willer et al. (1979) that the RA III response and pain sensation are modulated in parallel under supraspinal influences. They found stress to facilitate both pain and the nociceptive FR while mental tasks had inhibitory effects. They suggested a possible involvement of the reticular and limbic structures, which would exert global inhibitory or excitatory effects on both ascending and reflex nociceptive messages. However, in the same series of experiments, they also found evidence that under certain conditions, supraspinal influences could differentially affect pain and the flexion reflex.

Additional arguments upholding the connection between pain sensation and the FR include a shared sensitivity to opioid modulation, as evidenced by the naloxone-reversible morphine inhibition (Willer and Bussel, 1980), and increased threshold due to stress-induced analgesia (Willer and Albe-Fessard, 1980). Willer et al. (1980) also studied the effects of spatial summation on the flexion reflex and pain. They found the sural nerve elicited FR and pain to be facilitated by a preceding noxious stimulus to the peroneus superficialis nerve, while a tactile input to the same nerve depressed both.

Far from being exhaustive, the foregoing review nevertheless serves to illustrate the strong case put up by the evidence to date, concerning the potential of the human lower limb FR as a **quantitative** index of nociception. The usefulness of such an index in studies investigating analgesics and pain conditioning procedures is obvious.

A previous study (Chan and Tsang, 1987) has already investigated the influence of peripheral electrical conditioning with TENS on the flexion reflex, elicited under the paradigm developed in our lab (Chan and Tsang, 1985). The next logical step was to study the effects of a **natural** (versus electrical) conditioning procedure on the FR elicited under the same experimental paradigm. Cutaneous mechanical vibration was chosen because it is known to stimulate large diameter tactile afferents, and because it has been reported to be effective in alleviating certain types of pain. The following section reviews the literature pertinent to the pain-alleviating properties of vibration.

Vibratory stimulation as pain relief therapy

Over the past decade or so, a number of new avenues to pain control have been investigated. Among these are various modes of central and peripheral afferent stimulation, some of which have been successfully applied as pain relief therapy for certain types of clinical pain, namely dorsal column

stimulation (DCS) and more commonly, TENS. These approaches to the treatment of pain have evolved from Melzack and Wall's gate control theory, which proposed the involvement of large-diameter fibers in the modulation of pain messages transmitted by small-diameter nociceptive fibers. In this connection, mechanical vibratory stimulation offers perhaps even more promise than DCS or TENS as a pain relief therapy, since it is more selective than electrical stimulation in the activation of large-diameter afferent fibers (Eriksson et al., 1979; Talbot et al., 1968).

Despite only recent interest in the pain relief properties of vibration, it was observed as early as 1960 by Wall and Cronly-Dillon that conditioning vibratory stimulation caused an elevation in subjective threshold for pain detection in man. The same investigators found vibration to abolish the response of cat dorsal horn cells to noxious stimulation, a finding confirmed by Salter and Henry's (1986) report of a selective inhibition of nociceptive dorsal horn cells with brief (2-5 s) vibration. In the last decade, the antinociceptive properties of vibration have been further demonstrated to operate on experimental (Bini et al., 1984; Ekblom and Hansson, 1982; Pertovaara, 1979; Sherer et al., 1986, Sullivan, 1968) as well as clinical pain of oro-facial (Lundeberg et al., 1983; Ottoson et al., 1981), neurogenic, or musculoskeletal origin (Lundeberg, 1984a,b,c; Lundeberg et al., 1983, 1984). In terms of effectiveness, comparative

studies have found vibration and TENS to have similar potency for relieving pain (Duranti et al., 1988; Lundeberg et al., 1984).

The effectiveness of vibration for pain relief appears to be determined by several factors. These include type and degree of pain, site of vibration application, area of skin contact, application pressure, and frequency of vibration. Lundeberg (1984a) found vibration to be more effective in the relief of pain of neurogenic or musculoskeletal origin. Moreover, vibratory stimulation seems less effective in relieving severe, as compared to light or moderate, pain (Bini et al., 1984; Lundeberg, 1984b; Ottoson et al., 1981). The optimal site of application is generally agreed by the same investigators to be within the immediate area of pain. Occasionally, the "best site" was situated outside the painful area, on a trigger or acupuncture point (Lundeberg, 1984b; Lundeberg et al., 1984). Interestingly, Lundeberg and his associates reported an enhancement of pain in some patients when vibration was applied to areas other than the "best site". Lundeberg et al.(1984) also found larger probe areas and moderate (rather than light) pressures of application to be positive factors in determining the degree of vibration-induced pain relief. Comparing frequencies ranging from 20 to 400 Hz, the same group found high-frequency vibration between 100 and 200 Hz to yield maximal pain relief. In a study comparing the effects of 20 and 240 Hz vibration on

electrical pain threshold in the hand, Pertovaara (1979) similarly reports a significant threshold elevation only with the high-frequency stimulus. Contrasting with these reports are Ekblom and Hansson's (1982) results, showing an increase in electrical pain threshold with both 10 and 100 Hz vibration.

An interesting question at this juncture concerns the identity of the receptors mediating the pain-inhibitory effects of vibration. Several receptors, innervated by cutaneous or muscle afferents, have been shown to respond to vibratory stimulation. The elegant studies of Talbot et al. (1968) and Merzenich and Harrington (1969) have identified two subpopulations of tactile vibration-responsive afferents, on the basis of physiological and psychophysical data from primates and humans. In the frequency range below 60 Hz, Meissner corpuscles (glabrous skin) or hair follicle complexes (hairy skin) from monkeys responded maximally, and a sensation of "flutter" was reported by the human subjects. Conversely, Pacinian corpuscles were found to be exquisitely sensitive to frequencies of vibration above 60 Hz, presumably subtending the "vibration" sensation reported by the subjects. In man, microneurography has shown that both primary and secondary endings of muscle spindle, and even some Golgi tendon organs, could be activated by vibration, primary endings being more strongly driven at frequencies above 100 Hz (Burke et al., 1976). Unfortunately, many studies investigating vibration

have used frequencies between 60 and 100 Hz, which is within or near the range of overlap of frequency sensitivities for all the receptors described above.

Pertovaara's (1979) finding that 240 Hz but not 20 Hz vibration elevated pain threshold led him to postulate that Pacinian corpuscles were mediating the antinociceptive effect, based on Talbot et al. (1968) and Merzenich and Harrington's (1969) findings. The observations that greater application pressures and contact with underlying bony structures potentiate vibration analgesia (Lundeberg et al., 1984) also argue in favor of Pacinian corpuscle involvement. These factors all increase the mechanical coupling between the vibrating probe and underlying deep tissues. Yet Pacinian corpuscles are known to be widely distributed in deep structures such as subcutaneous connective tissue, ligaments, joints and periosteum. Pacinian corpuscles are also known to be sensitive to transmitted vibration from remote locations (Vallbo and Johansson, 1984), a condition more likely to occur with greater application pressure of contact with rigid structures such as bone. The likelihood of Pacinian corpuscle involvement however does not preclude recruitment of other receptors. Specifically, vibration transmitted to deep tissues could conceivably activate muscle spindle endings, the primary endings being especially responsive to high-frequency vibration, like Pacinian corpuscles. It is also likely that other cutaneous mechanoreceptors, such as Meissner corpuscles

and hair follicle complexes, would be activated as well (Merzenich and Harrington, 1969; Talbot et al., 1968).

Another interesting question regards the time course of vibratory effects, which could yield clues as to the nature of the underlying inhibitory mechanisms. Although vibration has been reported to be an effective mode of pain relief for some patients, the results are quite disparate with regard to the temporal profiles of the effects obtained. A transient inhibitory effect with brief vibration has been reported by some investigators for both experimental (Bini et al., 1984; Pertovaara, 1979) and clinical (Ottoson et al., 1981) pain. For example, Bini et al. (1984) observed immediate and complete relief of moderate pain with 20-60 s of vibration, followed by recovery of pre-vibration pain within one minute after cessation of vibration. They also reported no further increase in recovery time with vibration lasting up to 4 minutes. Pertovaara (1979) noted a transient elevation in electrical pain threshold with 15-20 min of vibration applied locally.

In contrast, Ekblom and Hansson (1982), using experimentally-induced dental pain and a longer vibration application time (30 min), observed pain relief to have a gradual onset, peaking after 20-30 min of vibration, and manifest a gradual but relatively short decay, returning to normal within 10 min post-vibration. Further studies by Lundeberg (1984a, b, c; et al., 1983, 1984), investigating the

effects of 100 Hz vibration on patients suffering from acute or chronic pain of neurogenic or musculoskeletal origin, all reported maximal pain relief after 25-45 min of vibration, and lasting up to several hours after cessation of the vibratory stimulation.

From the above results, a pattern emerges in which the duration of the pain relief seems to increase with increasing duration of the applied vibratory stimulation. This suggestion is supported by Lundeberg et al. (1984), who upon variation of vibration application time, found a clear relationship between duration of application and the degree and duration of pain relief. Such a gradual onset and slow decay of the inhibitory effect could be consistent with the release of some neuromodulator triggered by vibration.

The different results reviewed above probably imply a complex picture in which a gate controlled mechanism could be initially activated, to be supplemented subsequently by a system with a longer lasting action (e.g. a humorally-mediated system) upon persistence of the vibratory stimulation. There is also the possibility that different types of pain (e.g. acute versus chronic) are preferentially controlled by different systems (Melzack and Wall, 1982). Such hypotheses are at present mere speculation and emphasize the need for further investigation.

The existence of a "best site" of stimulation for pain relief and the short-lived effects of placebo-vibration in

some patients (Lundeberg, 1984a), suggest that the pain relief achieved using vibratory stimulation is not due to placebo effects. Lundeberg et al. (1987) recently examined this issue more systematically in a double blind study, and found the incidence of vibration-induced pain alleviation to be significantly higher than that induced by placebo among their patients. Lundeberg et al. (1984) and Hansson et al. (1986) also found vibration-induced pain relief to be unaffected by naloxone, thereby questioning the involvement of endogenous opioids as possible mediators of these effects. The results of studies using naloxone to antagonize analgesia must however be interpreted with caution. In a study comparing naloxone reversibility in low- and high- frequency acupuncture, Han and Xie (1984) have provided evidence for a dosage-dependent action of naloxone. Although we can only speculate about the mechanisms involved, it appears evident that vibratory stimulation could be an effective method of combatting certain types of acute or chronic pain. In fact, in some patients, it was shown to be equally effective as, or even superior to, other treatment modalities, presumably because of more selective activation of large-diameter fibers (Lundeberg et al., 1983; Lundeberg, 1984c).

Vibratory stimulation appears to be a prime candidate for inclusion into the existing armamentarium of pain relief techniques, an attractive factor being its non-invasive nature. Although tolerance develops with long-term use, as

is the case with TENS, decreased effectiveness of peripheral stimulation therapy may be avoided by alternating vibration with other modes of stimulation (Lundeberg, 1984a). Besides, there is also evidence that different modes of peripheral stimulation may potentiate each other's effects (Hansson and Ekblom, 1983; Duranti et al., 1988).

PROBLEM FORMULATION

It is always tempting to extrapolate the effects of conditioning procedures on "objective correlates" of pain to the subjective experience of pain. In animal studies this is a difficult gap to bridge. In man, the ideal solution entails the simultaneous measurement of subjective pain sensation with the nociceptive index under study. At the very least, the existence of a close relationship between pain sensation and other nociceptive measures investigated must be established.

An important question to adress, therefore, concerned the nature and extent of the relationship between subjective pain report and the FR elicited under our paradigm. Indeed, unless a high correlation between subjective pain sensation and the flexion reflex could be established, comparison of our results with the clinical literature would be quite tenuous. Subjective report, measured on a visual analogue scale, was thus investigated simultaneously with flexion reflex area

generated by random variations of stimulus intensity. The results of this study will be presented in Chapter 3.

In the main study, we sought to investigate the influence of natural, selective stimulation of large-diameter afferents via vibration on the human flexion reflex. That large-diameter afferents may play a role in the suppression of pain and nociceptive reactions is no longer in doubt (Besson and Chaouch, 1987). Also beyond doubt is the accumulating evidence that under certain conditions, fine-diameter nociceptive afferents also possess the capability to interfere with pain impulses, perhaps even more effectively than large afferents (Chung et al. 1983; Le Bars et al., 1979a,b; Shin et al., 1986; Woolf et al., 1980). In this regard, the relative lack of specificity of TENS with respect to the receptors/fibers it activates clouds the issue of relative contributions of different fiber types to anti-nociception.

Vibration, on the other hand, is known to selectively activate large diameter afferents, and as such, may prove helpful in determining their role in pain inhibition. One study has already investigated the influence of vibration on the flexion reflex in man (Ertekin and Akcali, 1978). The facilitatory effects they reported on the FR are at odds with those observed in clinical studies. These results either indicate a discriminative effect of vibration on the FR and pain pathways under the influence of vibration, or perhaps a masking of inhibitory effects by a predominant input to

facilitatory pathways. Indeed, vibration is known to excite muscle spindle afferents -in particular group Ia fibers, and to induce a reflex contraction in the target muscle, the so-called tonic vibration reflex. These investigators did not monitor motoneuronal excitability, and although no mention is made of visible reflex muscle activity, it is possible that a heightened excitability subliminal for the induction of muscle contraction might have been present. The flexion reflex elicitation paradigm developed in our laboratory requires a tonic background muscle contraction from the subjects (details in Chapter 2), allowing the maintenance and monitoring of the motoneuronal excitability to be attained.

The objective of the proposed study was thus to quantitatively investigate the influence of vibratory stimulation on the nociceptive component of the flexion reflex in man, under our FR-elicitation paradigm. Specifically, we wished to determine 1) whether prolonged segmental vibration produced an inhibitory or excitatory influence on the flexion reflex, and 2) the magnitude and time course of its modulatory effects. These questions were of particular interest in light of the conflicting reports about the temporal profile of its analgesic effects in the literature. Furthermore, it would be interesting to compare the results of vibration with those already obtained in our laboratory with TENS using the same FR-elicitation paradigm (Chan and Tsang, 1987).

Chapter two comprises a detailed description of the instrumentation and methodology used in the experiments. Chapters three and four constitute papers which are respectively published and submitted for publication, presented in slightly modified versions for the sake of coherence. Chapter five provides a summary of the project, followed by conclusions.

CHAPTER TWO

METHODS

This chapter comprises a detailed description of the equipment and methodology employed. It is divided in two sections: the first section describes the methods pertinent to the study investigating the relationship between flexion reflex and subjective sensation, followed by a description of the methods pertaining to the main study, investigating the influence of vibration on the flexion reflex.

I. CORRELATION BETWEEN FLEXION REFLEX AND SUBJECTIVE SENSATION

Subjects

This study was performed on 7 normal female volunteers between 23 and 30 years of age with no history of pain syndrome, neurological or neuromuscular disorders. They were given a detailed explanation of the experimental procedures both verbally and through an "Informed Consent Form" which they signed.

Elicitation and recording of the flexion reflex

The methodological details for the elicitation and recording of the flexion reflex have already been described elsewhere (Chan and Tsang, 1985, 1987) and will be detailed again here. Subjects were oriented to the laboratory and carefully briefed before the experimental session so as to dissipate unnecessary anxiety. Once informed consent was obtained from subjects, they were seated comfortably in a

semi-reclined position, with their right knee and ankle fixated in slight extension by partial casts to enhance FR excitability (Baxendale and Ferrell, 1981) (Fig. 2.1). The flexion reflex was elicited electrically with a 30 ms train of square pulses (pulse width: 1 ms, frequency: 200 Hz) delivered from a Grass 88 stimulator to the sole of the foot via constant current and stimulus isolation units (Grass CCU1 and SIU5). The cathode (Graphic Controls FC26 surface electrode) was placed on the medial plantar arch of the right foot, over the distribution of the tibial nerve's medial plantar branches (L4-L5). The anode plate was strapped just proximal to the ankle joint. Stimulus intensity was monitored with the use of a passive current probe inserted in the stimulating circuit, and displayed on a storage oscilloscope (Tektronix 5115) along with the FR EMG for visual inspection.

To avoid anticipatory reactions (Gaebelin et al., 1974) and habituation (Dimitrijevic et al., 1972), stimuli were delivered at intervals varying randomly between 10 and 20 s. In order to enhance FR excitability (Jenner and Stephens, 1982) and again to prevent its habituation (Kearney and Chan, 1979), subjects were required to maintain a tonic background contraction (10% of maximum voluntary contraction) in the biceps femoris. This was achieved by first asking the subject to perform a maximal contraction of that muscle. The EMG signal from the biceps femoris (BF) was filtered and displayed on an oscilloscope (Tektronix 2213), for visual feedback to

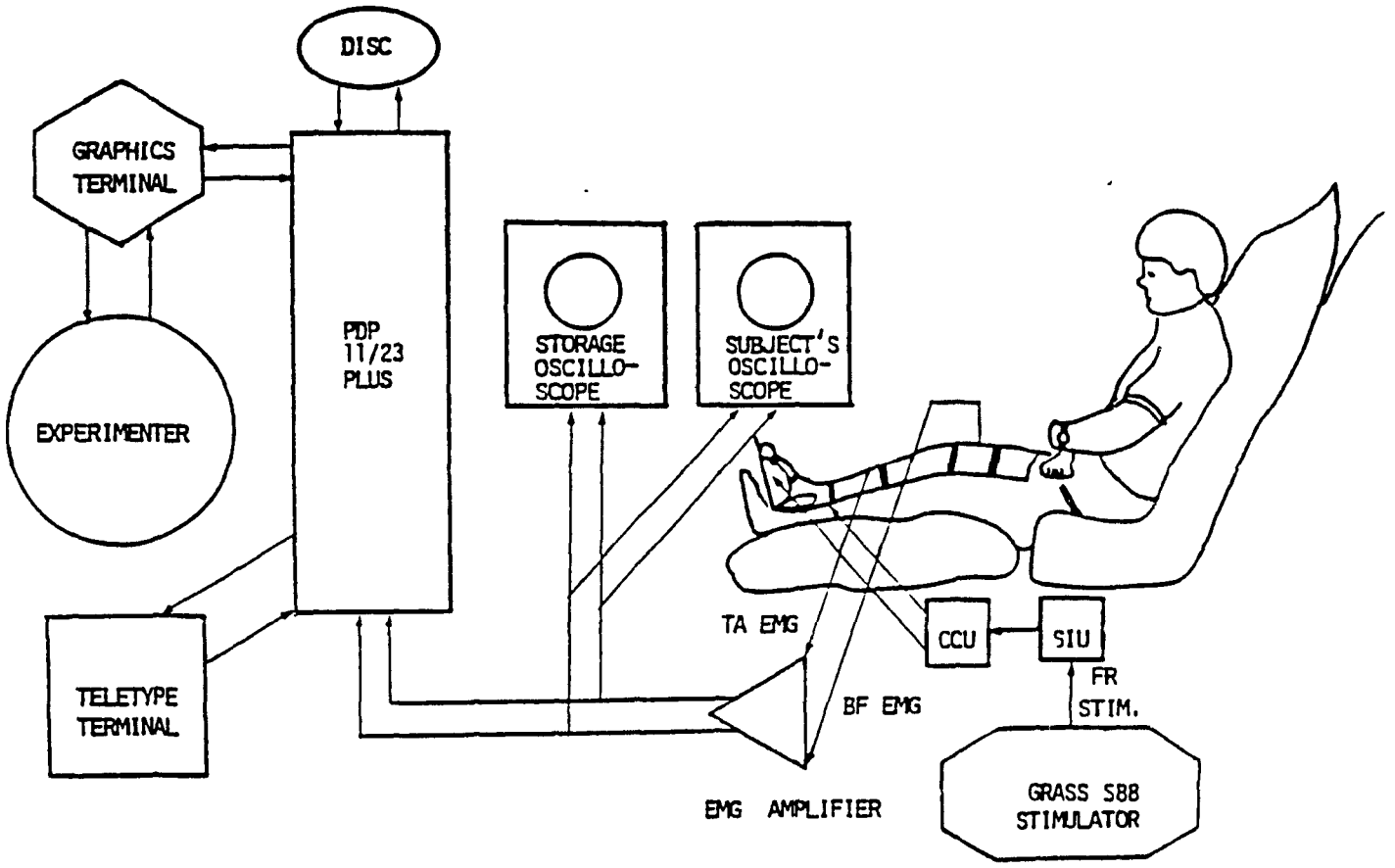


Fig. 2.1 EXPERIMENTAL SET-UP

the subject. Three such maximal efforts were required of the subject, with a brief rest period between each exertion. The mean of the three voltage readings was taken as 100% maximal voluntary contraction (MVC), and the desired background contraction level (i.e. 10%) was calculated in volts. With the muscle in the relaxed state, the oscilloscope trace was then repositioned by the experimenter in such a way that an appropriate contraction by the subject would bring the trace to superimpose on a fixed "target" line on the graticule representing the required level of contraction. In order to maintain a stable level of motoneuronal excitability, on-line computer monitoring via a PDP 11/23 plus microprocessor ensured that the stimulus was delivered only when the subject kept the contraction within the specified limits (for 2 consecutive seconds) via oscilloscope feedback.

Raw EMG recordings were obtained from bipolar skin electrodes (Graphic Controls 1801) placed on the carefully prepared skin overlying the motor points of the right BF (Basmajian and Blumenstein, 1980). These EMG signals were then amplified and bandpass filtered (10-500 Hz) before being fed into a PDP 11/23 plus microprocessor. The amplified and filtered EMG's were sampled at 1.5 kHz for 100 ms prior to and 300 ms after the FR stimulus, and displayed on the storage oscilloscope for visual monitoring.

Measurement of subjective sensation

Subjects were asked to estimate the intensity of each stimulus by moving a cursor along a VAS connected to a linear potentiometer (Ottoson et al., 1981). The scale consisted of a 20 cm horizontal line drawn on a white sheet of paper (Fig. 2.2). The left extreme of the scale was labelled "threshold intensity" and the right extreme, "maximally tolerable intensity". Each subject's perception threshold was determined using the limits method, and tolerance level was obtained by gradually increasing stimulus intensity to a maximally bearable limit. Stimuli of varying intensities were then delivered to the subject for a period of 5 to 10 min before re-testing threshold and tolerance intensities. Ten stimuli each at the previously determined perception threshold and tolerance level intensities were delivered to the subject, who was instructed to rate these stimuli on the VAS. If the ratings still corresponded to threshold and tolerance intensities, the experiment proceeded. If not, threshold and tolerance determination procedures were begun anew.

Once the subjective sensations corresponding to the two extremes of the VAS were well anchored in the subject's mind, threshold and tolerance stimulus intensities were normalized as 0% and 100% respectively. The range between these two intensities was then divided into 10% increments, yielding a total of 11 stimulus intensities. Each stimulus intensity was presented 10 times for suitable averaging, in a totally

Fig. 2.2 VISUAL ANALOGUE SCALE

randomized order. The VAS ratings were recorded by the experimenter from a digital-display voltmeter measuring the output from the VAS potentiometer and placed out of the subject's field of vision. Subjects were instructed to return the VAS cursor to the left extreme of the scale after each estimate, to prevent biases due to comparison of sequential stimuli.

Data acquisition and analysis

Off-line, the EMG traces were rectified and smoothed using a 75 Hz low-pass filter. The 10-response ensembles collected at each stimulus intensity were averaged, and the (FR) area above mean baseline level was extracted for each average and expressed as a percentage of the maximum area value obtained in a given subject. FR areas were taken over time windows ranging from 60 to 200 msec, depending on each subject's EMG activity pattern. Arithmetic means for the VAS estimates at each stimulus intensity were also expressed as percentages of tolerance level values. The data for all subjects were then pooled. Finally, correlation coefficients were computed and linear regression lines drawn for the relations between (a) FR EMG area and stimulus intensity, (b) subjective sensation and stimulus intensity, and (c) FR EMG area and subjective sensation.

II. INFLUENCE OF VIBRATION ON FLEXION REFLEX

Subjects

In total, 16 normal, healthy subjects with no history of pain or neuromuscular disorders were screened for flexion reflex stability and willingness to tolerate a high stimulus intensity repetitively. Nine subjects (all female, aged 16-28) participated in the full investigation. The seven remaining subjects were excluded from the main study either because their control FR values were unstable (n=5), the experimental session was prematurely terminated (n=1), or the subject's pain tolerance level could not be achieved (n=1).

Elicitation and recording of the flexion reflex

Basically, the FR elicitation/recording procedure used in this study is the same as the one described above for the first study. The only differences are:

a) In this study, the intensity of the stimulus was set to the maximum that the subject could tolerate. Also, once set, it remained constant throughout the experiment.

b) Recordings were obtained from both the biceps femoris and tibialis anterior (TA). As in the previous study, subjects were required to maintain a tonic background contraction of 10% maximum voluntary contraction (MVC) in the biceps femoris. The level of contraction to be maintained in the tibialis anterior was 20% MVC.

Conditioning vibratory stimulation

The conditioning stimulus consisted of a 240 Hz sinusoidal signal (Wavetek function generator 182A), amplified (Ling Dynamic Systems TPO 25) and delivered through an electromagnetic vibrator (Ling Dynamic Systems 101) to the skin area corresponding to the L5 dermatome on the lateral aspect of the right lower leg. In order to avoid the transmission of extraneous vibration to the subject, the vibrator was mounted on a stand and not in contact with the table. The probe contacting the skin was made of plastic and had a round, flat surface area of 3.5 cm^2. It was applied perpendicularly to skin surface with light to moderate pressure. The intensity of the vibratory stimulus was determined by gradually increasing the probe excursion until the subject would perceive a distinct but gentle "buzzing, vibrating" sensation in the leg. The amplitude and shape of the probe motion were recorded by means of a piezoelectric accelerometer (PCB 303A; PCB 480B power unit) mounted to the probe and displayed on the storage oscilloscope.

Data acquisition and analysis

The magnitude of the flexion reflex was measured throughout the experimental session by collecting 15- or 20-response ensembles at some 6-8 min intervals. Each subject received 2-3 control ensembles prior to, 3-4 ensembles during, and 1-3 ensembles after the 30-min vibration period. The

number of ensembles delivered varied according to the subject's adeptness at keeping the background muscle contraction within the specified limits and ability to tolerate the stimulus. Off-line, the EMG traces were rectified and smoothed using a 75 Hz low-pass filter, and an average signal was obtained from each ensemble. The mean RA III area and S.D. were then computed for each ensemble, and expressed as percentage of the mean of all control responses in a given subject to allow intersubject comparisons. The mean of the pooled control values in each subject was then statistically tested against the ensemble mean showing maximum modulation, according to Dunnett's procedure for multiple comparisons (Dunnett, 1964). Only ensemble averages recorded during the vibration period were considered for statistical analysis, unless they were found to be non-significant. In such cases, the post-vibration averages were also tested to uncover possible slow-course changes peaking after cessation of vibration. A p-level of less than .05 was considered significant. The Pearson product-moment correlation coefficient (r) was then computed to establish the relationship between BF and TA with regard to FR EMG area.

CHAPTER THREE*

SUBJECTIVE PAIN SENSATION IS LINEARLY CORRELATED WITH THE FLEXION REFLEX IN MAN

* Note that this chapter is a slightly modified version of an already published short communication with the same title by Chan, C.W.Y. and Dallaire, M., Brain Research, 479 (1989) 145-150. In addition, an abstract has been published: Dallaire, M. and Chan, C.W.Y., Is subjective pain sensation correlated with the flexion reflex in man?, Neurosci. Abs. 13 (1987) 109.

SUMMARY

In an attempt to define the relationship between physiological and psychological correlates of pain during low-level voluntary contraction, electrical stimuli between threshold and maximally tolerable intensities were delivered to the sole of the foot in 7 normal subjects. The two measures used to assess the response magnitude are: (1) the flexion reflex in the ipsilateral biceps femoris (BF), and (2) the estimate of perceived intensity reported on a visual analog scale (VAS). Our results showed that both BF FR area and VAS ratings bore a direct linear relationship with stimulus intensity and with each other, suggesting that at least under our paradigm, the sensory component of a nociceptive stimulus may already be largely set at the spinal interneuronal level.

INTRODUCTION

Pain is often measured using psychophysical scaling techniques. The main weakness of subjective measures, however, resides in their potential susceptibility to contamination by a multitude of factors external to the immediate pain sensation. These factors include anxiety, expectations, past experiences, ethnic background, situational

context - all of which may contribute, to varying degrees, to confound subjective pain ratings.

It is, therefore, desirable to objectively assess psychophysical pain measures by simultaneously measuring some physiological correlate of nociception. The lower limb flexion reflex (FR), elicited through electrical stimulation, has in recent years been proposed as such an index (Bromm and Treede, 1980; Chan and Tsang, 1987; Hugon, 1973; Willer, 1977). Specifically, the nociceptive (RA III) component of the FR has been found to have the same threshold as that of pain sensation (Hugon, 1973, Willer, 1977), and to be modulated in parallel upon conditioning (Bromm and Seide, 1982).

In this context, Bromm and Treede (1980) have investigated the relationship existing between pain sensation, assessed by a visual analog scale, and the electrically elicited withdrawal reflex in an upper limb muscle, the extensor digitorum. They reported a *power* function to be the best descriptor between the two indices. Using a similar electrical stimulus, Willer et al. (1984) examined the subjective pain measured on a 10-point numerical scale, and the FR recorded in a lower limb muscle, the biceps femoris, as a function of stimulus intensity. They found both the reflex activity and subjective pain rating to increase *linearly* with increasing stimulus intensity, the threshold and maximum values of both measures occurring at nearly identical

stimulus intensities (measured in mA). The question of "power" versus "linear" fit between psychological and physiological correlation of pain therefore remains to be resolved.

Furthermore, no study has so far considered the effect of voluntary contractions on the relationship between subjective pain sensation and the FR, when the latter is measured during a constant level of tonic contraction. This question is intriguing from the standpoint that descending systems involved in voluntary motor control have been found to modulate sensory inputs. For example, the pyramidal tract fibers are known to exert presynaptic inhibition of first order sensory neurons (Lundberg, 1964).

In order to address the above issues, the objective of this study is to define the relationship that exists among stimulus intensity, subjective perception, and the lower limb flexion reflex recorded during a constant level of low tonic background contraction. We therefore simultaneously measured subjective sensations obtained from a VAS, and the electromyographic (EMG) activity of the flexion reflex evoked in the BF, as a result of electrical stimulation applied to the sole of the foot. As was done in previous studies (Bromm and Treede, 1980; Willer et al., 1984), the electrical stimulus presented to the subjects ranged from "perception threshold" to "pain tolerance" level. However, because no change in relationship was reported when subjective estimate

switched from pre-pain to pain (Bromm and Treede, 1980, Willer et al., 1984), we made no attempt to define individual "pain threshold".

A preliminary report of the study has been briefly communicated in an abstract form (Dallaire and Chan, 1987).

METHODS

The study was performed on 7 normal female volunteers between 23 and 30 years of age with no history of pain syndrome, neurological or neuromuscular disorders. They were given a detailed explanation of the experimental procedures both verbally and through an "Informed Consent Form" which they signed.

The methodological details for the elicitation and recording of the flexion reflex have already been fully described in a previous paper (Chan and Tsang, 1985). Subjects lay comfortably in a semi-reclined position. Their right knee and ankle were fixated in partial casts to enhance FR excitability (Baxendale and Ferrell, 1981). The stimulus used to elicit the FR consisted of a 30 msec train of square pulses (1 msec duration, 200 Hz internal frequency). The cathode (a Graphic Controls FC26 surface electrode) was placed on the medial plantar arch of the right foot, and the anode was strapped just proximal to the ankle joint.

To avoid anticipatory reactions (Gaebelin et al., 1974) and FR habituation (Dimitrijevic et al., 1972), the interstimulus interval was varied randomly between 10 and 20 sec. Subjects were also required to maintain a tonic background contraction (10% of maximal voluntary contraction) in the right BF muscle. Such a background contraction has been shown to enhance FR excitability (Jenner and Stephens, 1982) and prevent FR habituation (Kearney and Chan, 1979). In an attempt to ensure a stable level of motoneuronal excitability, on-line computer monitoring via a PDP 11/23 plus microprocessor ensured that the stimulus was delivered only when the subject kept the contraction within the specified limits via oscilloscope feedback.

Raw EMG recordings were obtained from bipolar skin electrodes (Graphic Controls FC26) placed on carefully prepared skin overlying the motor points of BF (Basmajian and Blumenstein, 1980). These EMG signals were then amplified with a gain of 10,000 and bandpass filtered (10-500 Hz) before being fed into a PDP 11/23 plus microprocessor. The amplified and filtered EMGs were sampled at 1.5 KHz for 100 ms prior to and 200 ms after the FR stimulus, and simultaneously displayed on a Tektronix 5115 oscilloscope for visual inspection.

Subjects were asked to estimate the intensity of each stimulus by moving a cursor along a VAS connected to a linear potentiometer (Ottoson et al., 1981). The scale consisted of a 20 cm horizontal line drawn on a white sheet of paper. The

left extreme of the scale was labelled "threshold intensity" and the right extreme, "maximally tolerable intensity". Each subject's perception threshold was determined using the limits method, and tolerance level was obtained by gradually increasing stimulus intensity to a maximally bearable limit. Stimuli of varying intensities were then delivered to the subject for a period of 5 to 10 min before re-testing threshold and tolerance intensities. Ten stimuli each at the previously determined perception threshold and tolerance level intensities were delivered to the subject, who was instructed to rate these stimuli on the VAS. If the ratings still corresponded to threshold and tolerance intensities, the experiment proceeded. If not, threshold and tolerance determination procedures were begun anew.

Once the subjective sensations corresponding to the two extremes of the VAS were well anchored in the subject's mind, threshold and tolerance stimulus intensities were normalized as 0% and 100% respectively. The range between these two intensities was then divided into 10% increments, yielding a total of 11 stimulus intensities. Each stimulus intensity was presented 10 times for suitable averaging, in a *randomized* order. The VAS ratings were recorded by the experimenter from a digital-display voltmeter measuring the output from the VAS potentiometer. Subjects were instructed to return the VAS cursor to the left extreme of the scale after each estimate,

to prevent biases due to comparison of sequential stimuli.

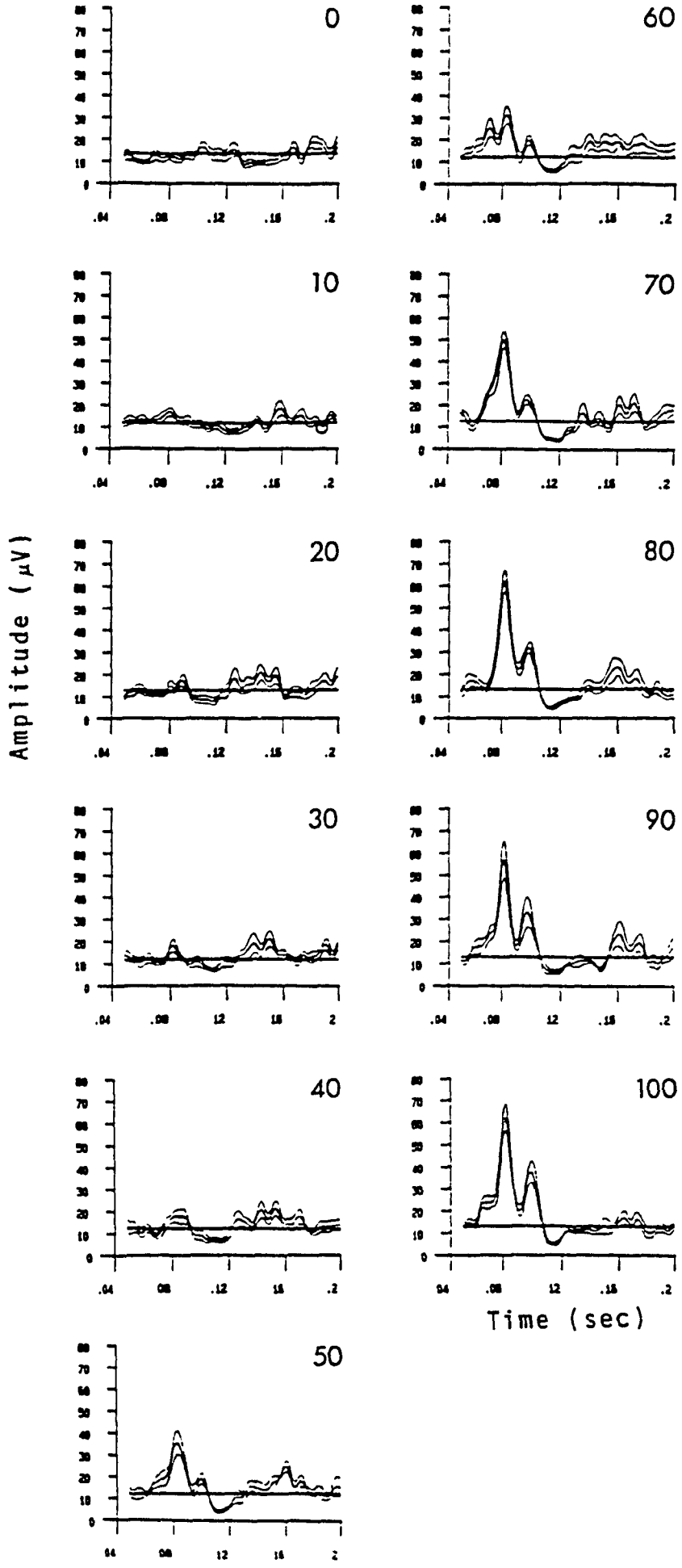
Off-line, the EMG traces were rectified and smoothed using a 75 Hz low-pass filter. The 10-response ensembles collected at each stimulus intensity were averaged, and the (FR) area above mean baseline level was extracted for each average and expressed as a percentage of the maximum area value obtained in a given subject. FR areas were taken over time windows ranging from 60 to 200 msec, depending on each subject's EMG activity pattern. Arithmetic means for the VAS estimates at each stimulus intensity were also expressed as percentages of tolerance level values. The data for all subjects were then pooled. Finally, correlation coefficients were computed and linear regression lines drawn for the relations between (a) FR EMG area and stimulus intensity), (b) subjective sensation and stimulus intensity, and (c) FR EMG area and subjective sensation. A more detailed description of the procedures has been given in Chapter 2.

RESULTS

Fig. 3.1 illustrates the gradual increase in the FR EMG area in the BF as a function of stimulus intensity in subject ML. Each plot shows the mean $\pm$ 1 S.E.M., and the mean baseline level for 10 responses of the rectified and smoothed EMG signal, at 10% increments of the stimulus intensity relative to the maximum tolerable intensity (=100% tolerance;

Fig. 3.1. EMG response elicited in the biceps femoris (BF) of subject ML as stimulus intensity increased from perception threshold (=0% tolerance; top plot on left) to maximum tolerance (=100% tolerance; bottom plot on right). y-axis, EMG amplitude in μV; x-axis, time in seconds.

Each plot represents the mean (thick line) $\pm$ 1 S.E.M. (thin line), and the mean baseline level for 10 responses of the rectified and smoothed EMG signal. Note the increase in the RA III component of the flexion reflex (at a latency of some 70 ms) as stimulus intensity reached tolerance level.


0
60
10
70
20
80
30
90
40
100
50
Amplitude (μV)
Time (sec)

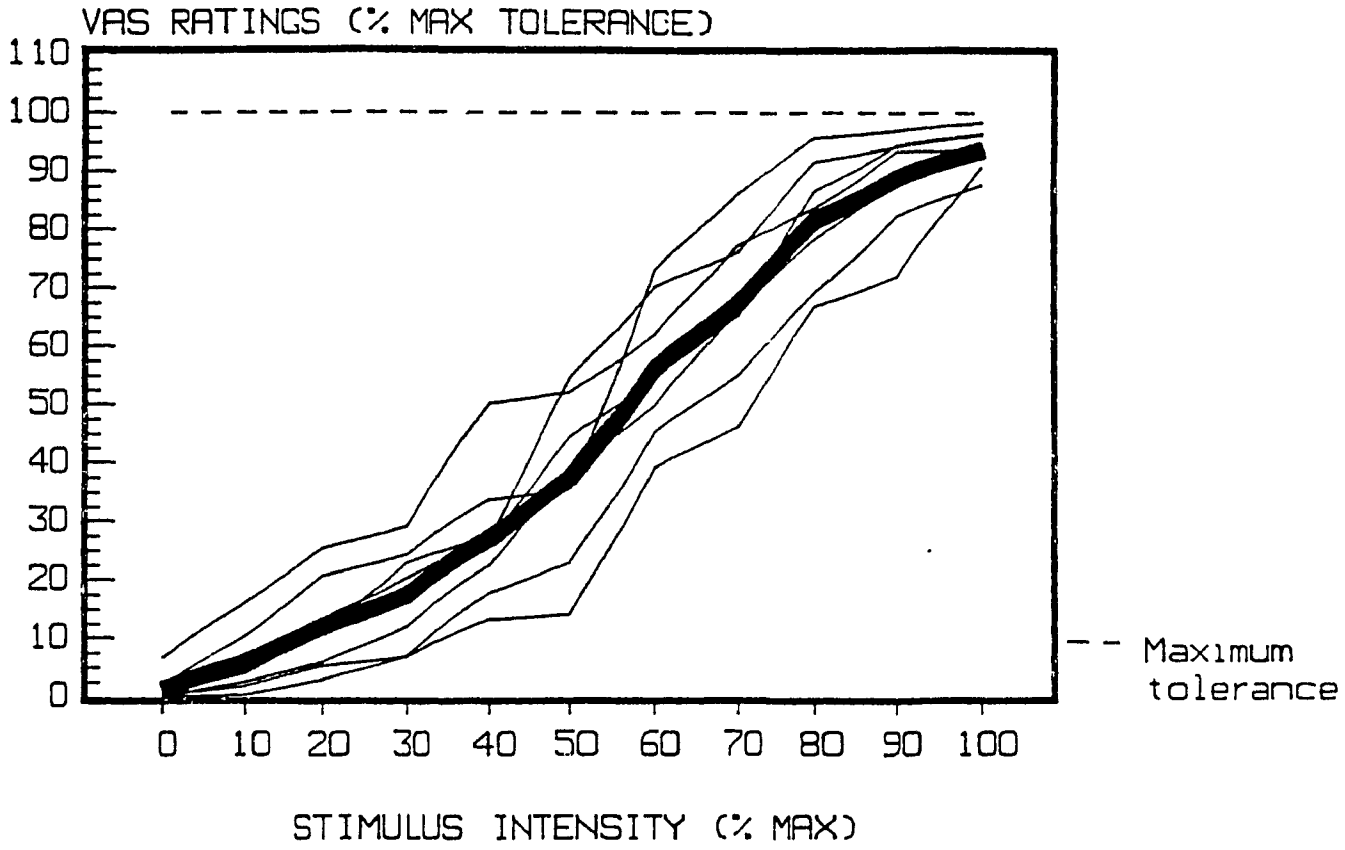
bottom plot on right). Note that the RA III component of the FR was barely visible at about 70 msec when the stimulus intensity was at 10% of maximum tolerance intensity (second plot on left). When the stimulus intensity was increased from 10% to 100% tolerance level, the peak amplitude of the RA III response could also be seen to increase from 17 μV (second plot on left) to 62 μV (bottom plot on right).

From the standpoint of the EMG response pattern: subject ML exhibited EMG activity with temporal parameters in strong agreement with those of subjects participating in our previous studies (Chan and Tsang, 1985, 1987). In sum, six of the seven subjects exhibited a distinct RA III burst with onset latencies between 60 and 70 msec, and a return to baseline between 100 and 120 msec. However, no preceding RA II component was observed in any of the subjects studied. In one subject, the RA III burst had an onset latency of 110 msec and lasted until 160 msec. Longer latency bursts (beyond 120 msec) were also found in some subjects, especially at higher stimulus intensities, and occasionally fused with the RA III response.

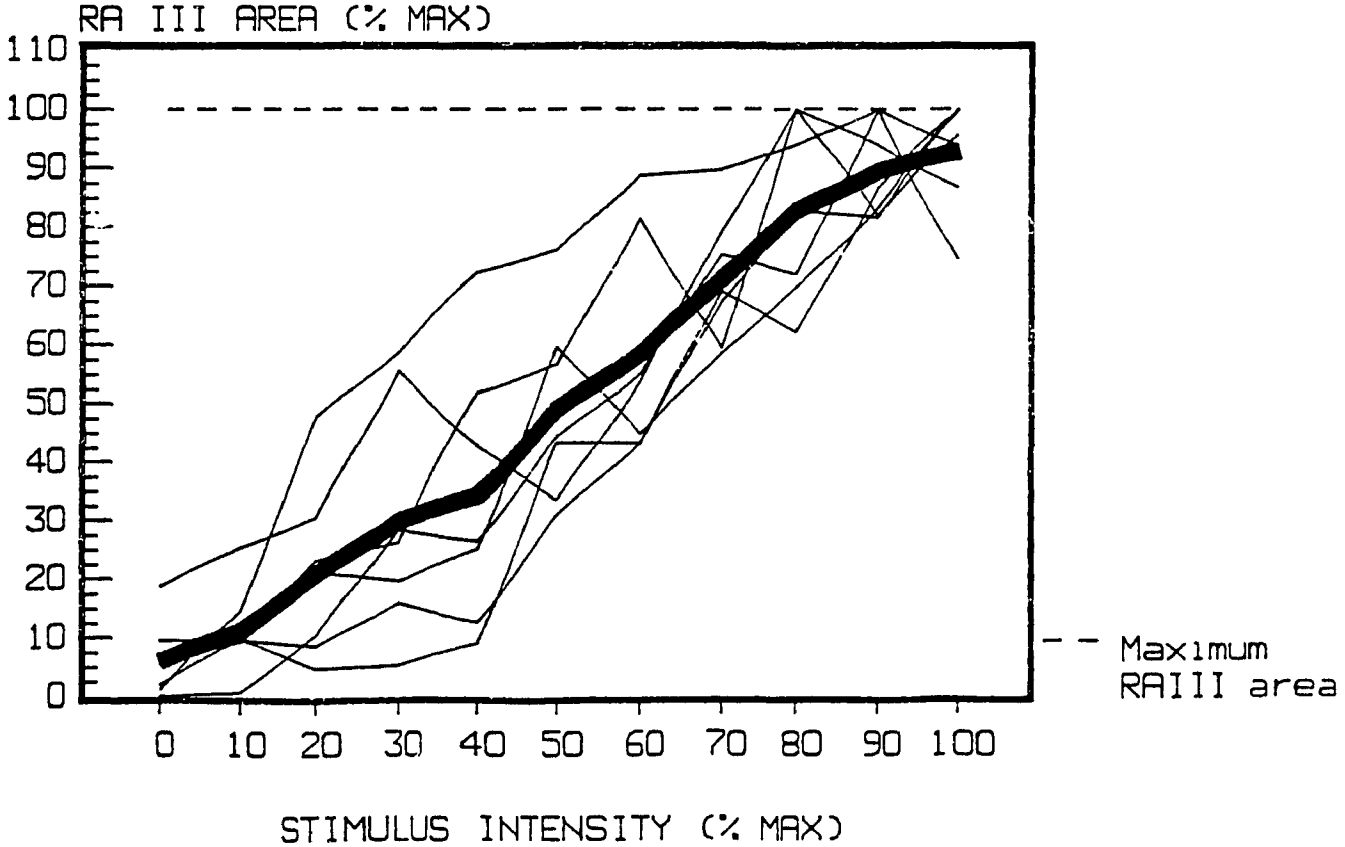
Fig. 3.2 depicts the recruitment curves for subjective sensation (*top*) and flexion reflex EMG area (*bottom*) as a function of stimulus intensity. The thin lines represent the data from individual subjects, superimposed with the average curves from the pooled data (heavy lines). All values are

Fig. 3.2. Parallel increase in subjective sensation (A) and RA III area (B) with increasing stimulus intensity. All data points normalized with respect to maxima (dotted lines). Thin lines represent individual subjects, thick lines are averaged data (n = 7).

A. SUBJECTIVE SENSATION vs STIMULUS INTENSITY



B. RA III AREA vs STIMULUS INTENSITY



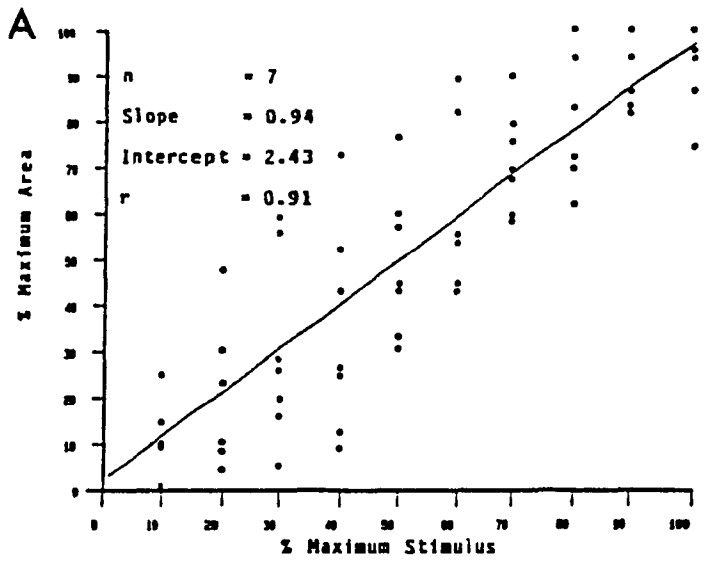
shown normalized with respect to maxima which are set at 100%. Interestingly, the subjective sensation increased in magnitude with increasing stimulus intensity in a similar, but smoother, way in comparison to FR area.

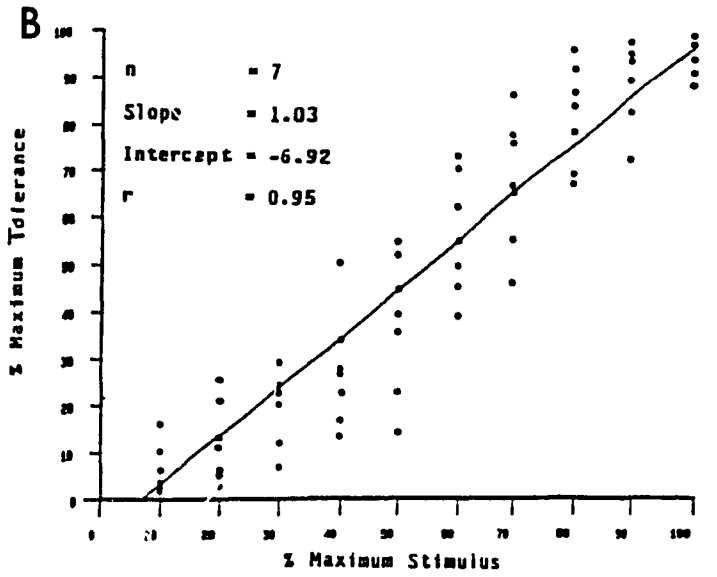
To determine whether a systematic relationship existed between FR area and subjective estimates on the one hand, and stimulus intensity on the other, the data for all 7 subjects were pooled, and linear regression lines drawn through the points. Fig. 3.3 A and B show the regression lines drawn respectively for FR area and subjective estimates as a function of stimulus intensity. Note that each point on the graphs represents 10 data points. Correlation coefficients were high for both measures, with r values of 0.91 for FR area and 0.95 for subjective estimates. An addtional finding is that the slopes of the regression lines were close to unity, being 0.94 and 1.03 for FR area and subjective estimates, respectively. Of special interest is Fig. 3.3C, illustrating the fact that subjective estimates were also correlated with FR area. The correlation coefficient of this linear regression line has a high value of r=0.84 and a slope of 0.87.

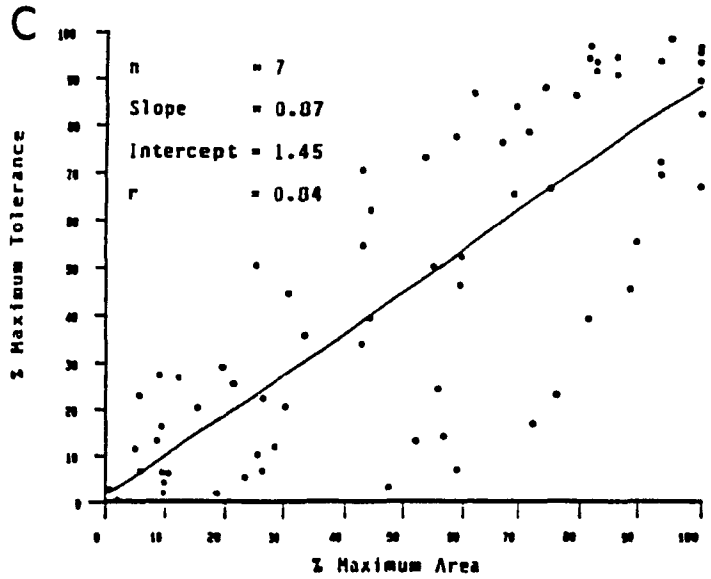
DISCUSSION

There are previous reports showing the amplitude of the FR to be linearly related to electrical stimulus intensity in

Fig. 3.3. Linear regression lines drawn through the pooled data from 7 subjects. All data were expressed as percentages of their maximum value for each subject, and each point plotted represents the mean of 10 data points. High correlations were found between: (A) FR area and stimulus intensity (r=0.91); (B) subjective estimate and stimulus intensity (r=0.95); as well as (C) subjective estimate and FR area (r=0.84).


A
n = 7
Slope = 0.94
Intercept = 2.43
r = 0.91
% Maximum Area
% Maximum Stimulus


B
n = 7
Slope = 1.03
Intercept = -6.92
r = 0.95
% Maximum Tolerance
% Maximum Stimulus


C
n = 7
Slope = 0.87
Intercept = 1.45
r = 0.84
% Maximum Tolerance
% Maximum Area

the extensor digitorum (Bromm and Treede, 1980) and the BF muscles (Willer et al., 1984) in the relaxed state. The present investigation of the behaviour of the FR in the BF further demonstrates the existence of such a linear relationship between EMG area and the electrical stimulus intensity, with a high correlation coefficient of r=0.91, during a low level of tonic background contractions. These findings are also in agreement with our preliminary report of a strong correlation between both the area and peak-peak amplitude of the RA III component of the FR in lower limb muscles and stimulus intensity, under the same experimental paradigm (Chan and Tsang, 1985).

What about the relationship between subjective sensation and the intensity of electrical stimulus? Stevens (1957) investigated several perceptual continua and found them to obey a power function of the form:

$$re=a.S^{n},$$

where re is the measured reaction and S represents stimulus intensity. The technique he used was one of "magnitude estimation", where subjects were presented with a standard stimulus, to be used as a reference against which subsequent stimuli were to be compared and rated numerically. Using the same technique, several investigators (Ekman et al., 1964; Goldberger and Tursky, 1976; Sternback and Tursky, 1964, 1965, reported the relationship between subjective sensation and electrical stimuli to obey Stevens' power law with exponents

greater than 1. Using a VAS, we found the psychophysical function for electrical stimuli to be adequately described by a straight line, with a correlation coefficient of r=0.95 for the pooled data from 7 subjects. Our results with electrical stimulation of the median arch, which is innervated by the distal branches of the tibial nerve, are in agreement with the findings of Willer et al. (1984) with surface stimulation of the sural nerve behind the lateral malleolus. Bromm and Treede (1980), using a magnitude estimation procedure similar to that used by Stevens, also reported subjective sensation to be linearly related to the intensity of electrical stimulus applied to the finger, with a mean correlation coefficient of 0.95 across 15 subjects. They further showed that the same data could be fitted by a power function having an exponent of 1.44. It thus appears that using double logarithmic scales in the linear approximation may introduce a bias towards a higher exponent. In addition, Cross et al. (1975) found the magnitude of the exponent to vary according to the technique used to estimate it.

Our last finding was the existence of a high correlation (r=0.84) between subjective pain ratings and FR EMG area in the BF during a constant level of tonic contraction. Again, this result is in agreement with that of Bromm and Treede (1980) who also observed a high correlation between these two measures in the *relaxed* extensor digitorum. In other words, the same linear relationship between subjective estimate and a

physiological correlate (FR EMG area) appears to hold true, regardless of whether a lower limb knee flexor (BF) or an upper limb extensor (extensor digitorum) is involved. These observations point to a similar central processing mechanism regardless of the site of origin of both spinal afferent and efferent pathways (cervical versus lumbo-sacral innervation of upper and lower limb muscles respectively). Furthermore, a low level of tonic background contraction (10% of maximum voluntary contraction) does not affect the linear relationship found between subjective rating and FR area.

The parallel behaviour of the perceptual and spinal reflexive limbs of the CNS nociceptive pathways implies that, under our experimental paradigm, the sensory component of pain may already be "set" at the spinal interneuronal level. It would therefore be interesting to determine if, and to what extent, these two measures of pain will remain parallel under various conditioning procedures. In this context, Willer et al. (1979) found pain sensation and the BF FR to co-vary under conditions of stress and accrued attention related to a mental task, while noxious homotopic and heterotopic electrical stimuli caused the two responses to dissociate. Bromm and Seide (1982), investigating the influence of tilidine (morphine agonist) and prazepam (benzodiazepine) on the simultaneously measured withdrawal reflex and pain sensation, observed the two measures to be modulated in parallel by the drugs. Future experiments will investigate the influence of

peripheral somatosensory stimulation on both subjective pain sensation and FR under our paradigm.

CHAPTER FOUR*

PROLONGED PERIPHERAL VIBRATION COULD INHIBIT THE NOCICEPTIVE COMPONENT OF THE LOWER LIMB FLEXION REFLEX IN MAN

* This chapter is a slightly modified version of a short communication with the same title by Dallaire, M. and Chan, C.W.Y. submitted to Brain Research.

SUMMARY

The aim of this investigation was to determine the influence of prolonged, segmental peripheral vibration on the nociceptive component (RA III) of the lower limb flexion reflex (FR) in man. The FR was electrically elicited from the sole of the foot in 9 normal subjects and recorded from the biceps femoris (BF) and tibialis anterior (TA) muscles. Ensemble averages of 15 or 20 responses were collected at intervals prior to, during, and after a 30-min period of high frequency (240 Hz) vibration applied to the ipsilateral lower leg (L5). Vibration induced a gradual inhibition of the FR in 5 of 9 subjects, which lasted beyond the conditioning period in both BF and TA. Our results raise the possibility that prolonged vibration could influence spinal nociceptive reflexes in ways similar to the vibration-induced pain relief described in the clinical literature.

INTRODUCTION

In recent years, the lower limb flexion reflex (FR) has received considerable attention as an index of nociception in both animal and human models. In man, methodologies for electrically eliciting the FR have evolved, focusing specifically on the RA III component (Hugon, 1973) of its electromyographic (EMG) response, and studied mainly in the biceps femoris (BF) and tibialis anterior (TA) muscles (Chan

and Tsang, 1985; Willer, 1983). The RA III burst has since been demonstrated to have the same threshold as pain (Willer, 1977); to covary with pain sensation as a function of stimulus intensity (Chan and Dallaire, 1989; Willer et al., 1984); and to be modulated in parallel with pain sensation under the influence of certain peripheral conditioning procedures (Willer et al., 1980, 1984) and supraspinal influences (Willer et al., 1979). In addition, clinical pain control procedures such as morphine administration (Willer and Bussel, 1980) and transcutaneous electrical nerve stimulation (TENS) have been found to depress the FR (Chan and Tsang, 1987).

Peripheral conditioning therapies such as TENS have been reported to relieve certain types of clinical pain. These modalities evolved from Melzack and Wall's gate control theory, which proposed that the transmission of impulses travelling in fine-diameter nociceptive afferents might be gated at the spinal dorsal horn by activity in large-diameter afferents (Melzack and Wall, 1965). The exact mechanisms underlying TENS analgesia have yet to be elucidated. Evidence to date points to at least partial involvement of large-diameter non-nociceptive afferents in the pain-relieving effects of TENS, but there is still some debate as to the type(s) of afferents stimulated (Eriksson et al., 1979; Janko and Trontelj, 1987; Pomeranz and Paley, 1979), and the relative contribution of each fiber type to pain relief (Shin et al.,1986; Sjolund, 1985; Woolf et al., 1980). Vibration, however, is known to selectively activate large diameter,

rapidly-conducting, non-nociceptive primary afferents (Hunt, 1961; Hunt and McIntyre, 1960; Johansson et al., 1982). Moreover, brief vibration has been shown to inhibit experimentally-induced nociceptive reactions in animals (Salter and Henry, 1986; Wall and Cronly-Dillon, 1960). In man, vibration could cause a decrease in the subjective report of experimental (Ekblom and Hansson, 1982; Pertovaara, 1979) and clinical pain (Hansson and Ekblom, 1983; Lundeberg, 1984; Lundeberg et al., 1984; Ottoson et al., 1981). One previous study (Ertekin and Akcali, 1978) reported a facilitation of the human flexion reflex upon conditioning with vibration. However, the authors made no mention of the duration of application of the vibratory stimulus. This investigation was thus undertaken to study the effects of prolonged, segmental vibration on the lower limb flexion reflex in man, in an attempt to determine whether these effects parallel those reported in previous studies investigating the effects of vibration on clinical and experimentally-induced pain.

METHODS

Sixteen (16) healthy subjects with no history of pain or neuromuscular disorders were screened for flexion reflex stability and willingness to tolerate a high stimulus intensity repetitively. Of these 16 subjects, nine (all female, aged 16-28) participated in the full study.

The methodological details for the elicitation and recording of the flexion reflex have already been described elsewhere (Chan and Dallaire, 1989; Chan and Tsang, 1985, 1987). Subjects were carefully briefed before the experimental session to obtain informed consent and to dissipate unnecessary anxiety. During the experiment, they were seated comfortably in a semi-reclined position, with their right knee and ankle fixated in slight extension by partial casts to enhance FR excitability (Baxendale and Ferrell, 1981). The flexion reflex was elicited electrically with a 30 ms train of square pulses (pulse width: 1 ms, pulse frequency: 200 Hz) delivered from a Grass 88 stimulator to the sole of the foot. The cathode was placed on the medial plantar arch of the right foot, over the distribution of the tibial nerve's medial plantar branches (L4-L5). The anode was strapped just proximal to the ankle joint. The intensity of the stimulus was set to the maximum that the subject could tolerate, which was in the 50-60 mA range in the subjects investigated. Stimulus intensity was monitored with the use of a passive current probe inserted in the stimulating circuit, and displayed on a storage oscilloscope (Tektronix 5115) along with the FR EMG for visual inspection. The sensation evoked by the electrical stimulus was described as a sharp, pricking electrical pain, localized to the skin area under the stimulating cathode.

To avoid anticipatory reactions (Gaebelin et al., 1974) and FR habituation (Dimitrijevic et al., 1972), stimuli were

delivered at intervals varying randomly between 10 and 20 s. In order to enhance FR excitability (Jenner and Stephens, 1982) and again to prevent its habituation (Kearney and Chan, 1979), subjects were required to maintain a tonic background contraction in the muscles investigated (10% and 20% of maximum voluntary contraction in BF and TA, respectively). To maintain a stable level of motoneuronal excitability, on-line computer monitoring via a PDP 11/23 plus microprocessor ensured that the stimulus was delivered only when the subject kept the contraction within the specified limits via oscilloscope feedback.

The conditioning stimulus consisted of 240 Hz vibration delivered through an electromagnetic vibrator (Ling Dynamic Systems 101) to the skin area corresponding to the L5 dermatome on the lateral aspect of the right lower leg. The probe contacting the skin was made of plastic and had a round, flat surface area of 3.5 cm^2. It was applied with light to moderate pressure and with a peak-peak excursion such that the subject experienced a soft "buzzing, vibrating" sensation. In the subjects investigated, such a sensation corresponded to a probe displacement ranging between 120 and 200 μm (peak-peak). The amplitude and shape of the probe motion were transduced from a piezoelectric accelerometer (PCB 303A) mounted to the probe and displayed on the storage oscilloscope.

Raw EMG recordings were obtained from bipolar skin electrodes placed on the carefully prepared skin overlying the motor points of the right BF and TA (Basmajian and Blumenstein, 1980). These EMG signals were then amplified and bandpass filtered (10-500 Hz) before being fed into a PDP 11/23 plus microprocessor. The amplified and filtered EMG's were sampled at 1.5 kHz for 100 ms prior to and 300 ms after the FR stimulus. The magnitude of the flexion reflex was measured throughout the experimental session by collecting 15- or 20- response ensembles at some 6-8 min intervals. Sets of stimuli were delivered prior to, during, and up to 30 min after a 30-min vibration period.

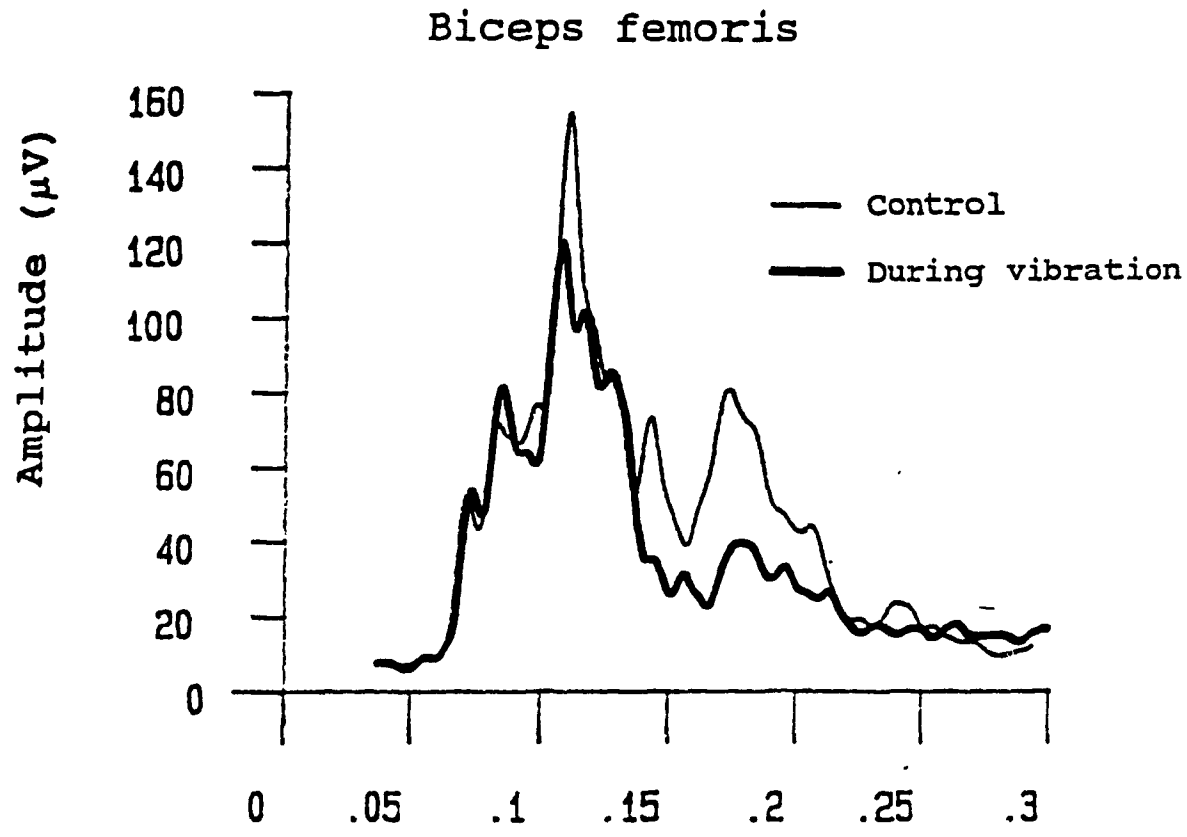
Off-line, the EMG traces were rectified and smoothed using a 75 Hz low-pass filter, and an average signal was obtained from each ensemble. Area computations were performed over time windows ranging anywhere from 40 to 290 ms, according to individual EMG patterns, and for EMG response at 1 S.D. above the baseline level resulting from the tonic background contraction. The mean RA III area and S.D. were then expressed as percentage of the mean of all control responses in a given subject to allow intersubject comparisons. For each subject, the mean of the pooled control values was statistically tested against the ensemble mean value showing maximum modulation, according to Dunnett's (1964) procedure for multiple comparisons. Only ensemble averages recorded during the vibration period were considered for statistical analysis, unless they were found to be non-

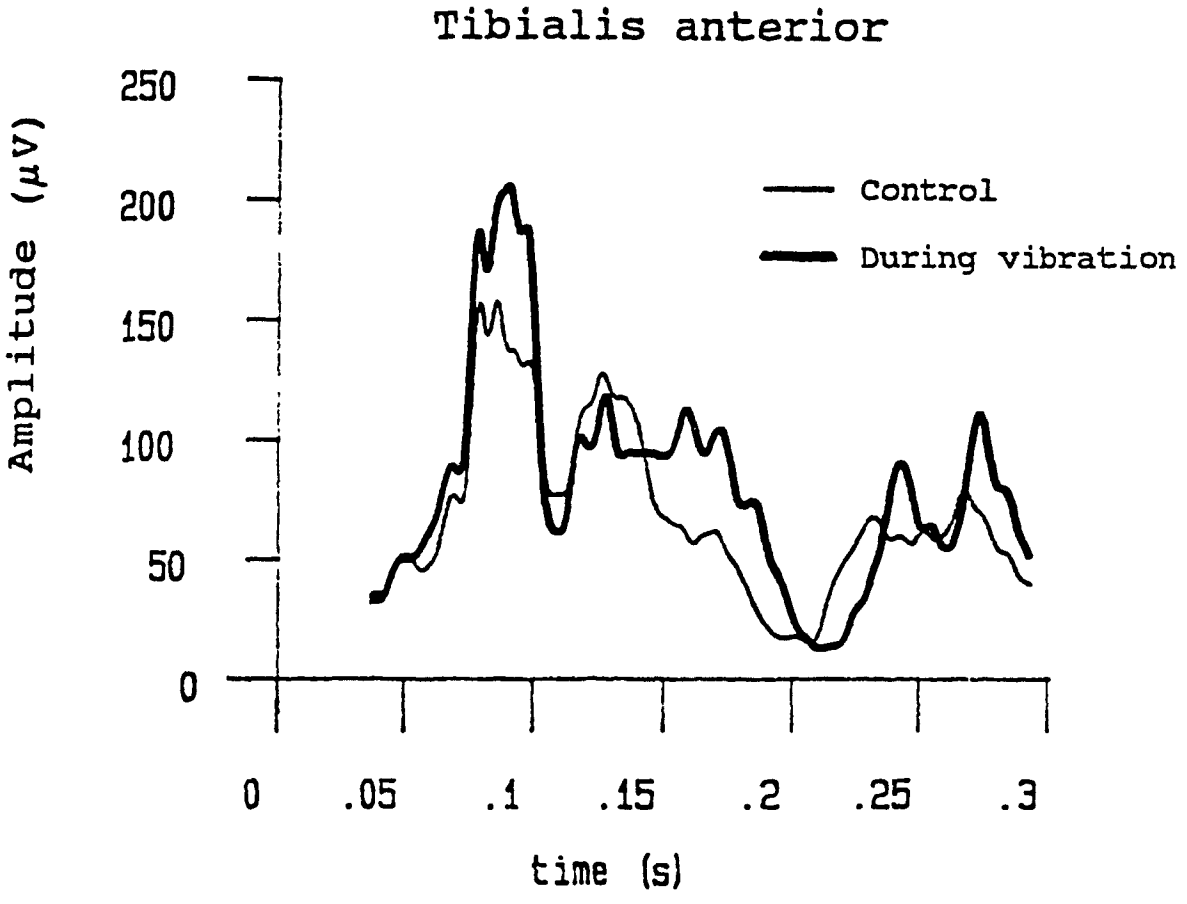
significant. In such cases, the post-vibration averages were also tested to uncover possible slow-course changes peaking after cessation of vibration. A p-level of less than .05 was considered significant. The Pearson product-moment correlation coefficient (r) was then computed to verify the parallel modulation FR EMG area in BF and TA. A more detailed description of the procedures has been given in Chapter 2.

RESULTS

Fig. 4.1 illustrates changes in the rectified, smoothed EMG signals under the modulatory influence of vibration. The average FR EMG at the time of maximum modulation (thick trace) is superimposed upon the average EMG from the pooled control trials (thin trace) for comparison. An example of flexion reflex depression observed in the BF of subject SC is shown in Fig. 4.1A. The maximum inhibition occurred around 25 minutes into vibration, with an RA III area of 6.47 $\pm$ 0.91 μV*s (mean $\pm$ S.D.), down from the control value of 8.83 $\pm$ 1.35 μV*s. The change represented a 27% decrease in FR, which was found to be statistically significant ($p<.01$). Fig. 4.1B shows an example of flexion reflex facilitation with vibration. The EMG traces depicted here were collected from the TA of subject SL, which displayed maximum RA III potentiation 20 minutes into the vibration period. The area value in this case went up by 37%, from 6.99 $\pm$ 1.81 (control)

Fig. 4.1. Comparison of control (pre-vibration) FR EMG response with that recorded at time of maximum modulation by vibration (25 and 20 minutes into vibration respectively in Fig. A and Fig. B). Thin traces (control) represent the average of 45 rectified, smoothed EMG signals, and thick traces (vibration) represent the average of 15 such signals. The RA III from BF in subject SC (top) displayed the more common inhibitory influence of vibration. An example of FR potentiation during vibration was observed in TA of subject SL (bottom)


Biceps femoris
Amplitude (μV)
160
140
120
100
80
60
40
20
0
Control
During vibration
0
.05
.1
.15
.2
.25
.3


Tibialis anterior
Amplitude (μV)
250
200
150
100
50
0
Control
During vibration
0
.05
.1
.15
.2
.25
.3
time (s)

to 9.55 ± 2.47 *u*V*s (vibration), proving significant at the p<.01 level.

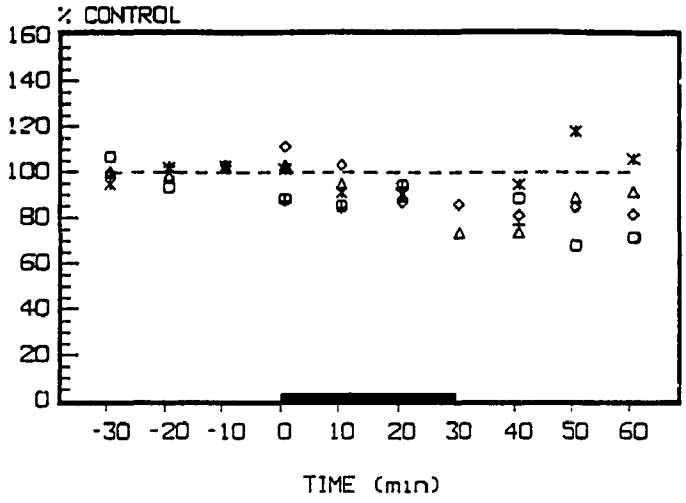
The normalized data from all subjects were grouped according to direction of flexion reflex modulation and plotted against time. Figures 4.2 and 4.3 illustrate the inhibitory and facilitatory effects, respectively. Note the stability of the control averages and the generally progressive onset of modulatory changes peaking late during the vibration period or shortly thereafter. Also noteworthy is the persistence of modulatory effects post-vibration, with a slight tendency towards returning to control level. Considering the limited data and shortness of the post-vibration collection period, we feel it premature at this point to discuss the meaning and extent of the tendencies observed during that time period.

Vibration had an inhibitory effect on the FR in five subjects and potentiated it in two others. Two subjects showed no response to vibration. Although not all changes were significant, the BF and TA responses were modulated in parallel in all of these subjects (Table 4.1). In three subjects (TR, LD, GL), the level of FR inhibition failed to reach significance during the 30 minutes of vibration, but peaked in the post-vibration period, whereby significance was demonstrated. The group of subjects displaying inhibition had a mean RA III area of 77.7 ± 8.4% in BF and 81.1 ± 10.5% in TA during and following vibration. The two subjects in whom

Fig. 4.2. Inhibitory influence of vibration on the EMG area of RA III response in 5 of 9 subjects. Top plots are data from individual subjects, each point representing 15 or 20 FR recordings. Bottom plots are group data (mean $\pm$ 1 S.D.). Left- and right-hand columns refer to biceps femoris (BF) and tibialis anterior (TA), respectively. All data are normalized with respect to the mean of the pooled control trials (dotted lines). Note the stability of control averages and the gradual onset of inhibition.

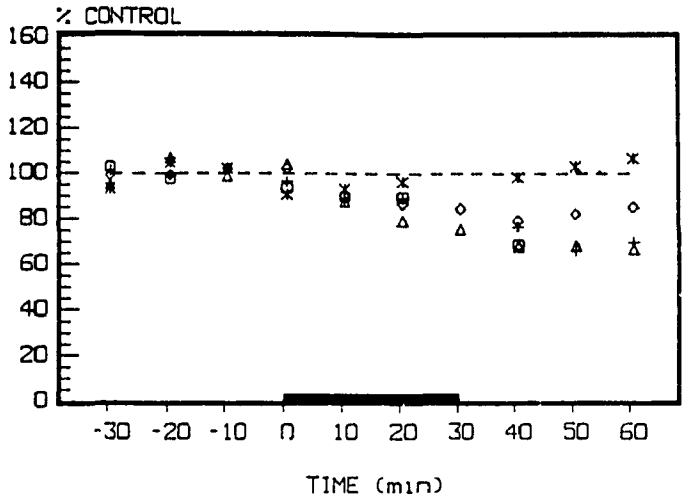
INHIBITION OF BF FLEXION REFLEX

A. INDIVIDUAL DATA

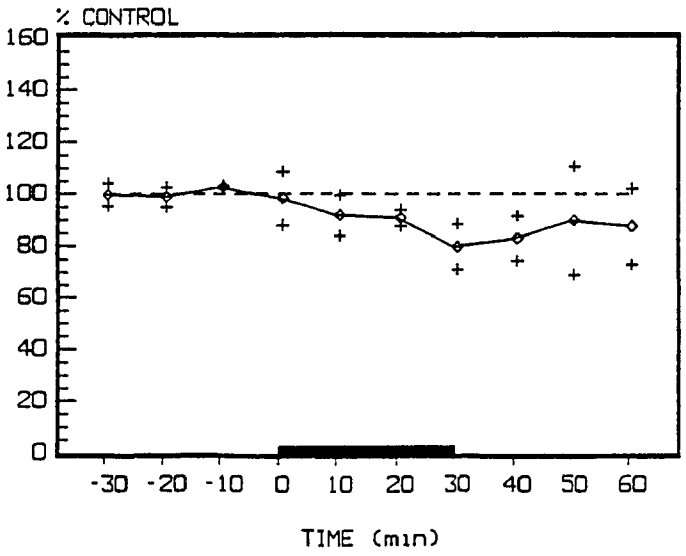


INHIBITION OF TA FLEXION REFLEX

A. INDIVIDUAL DATA



B. GROUP DATA



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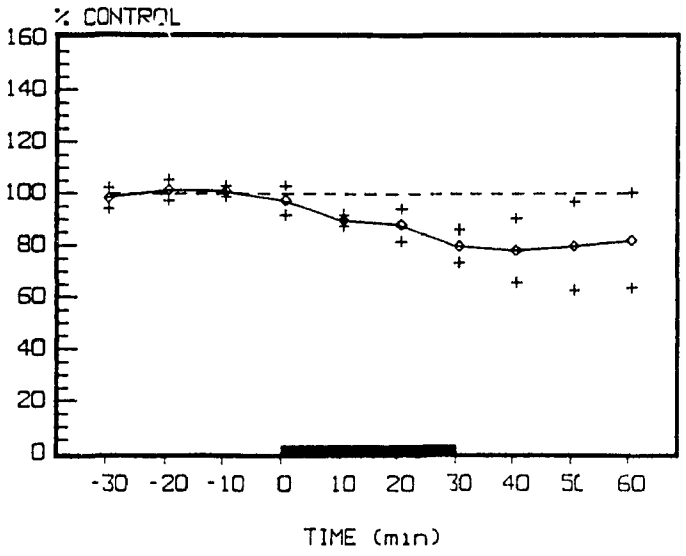


n=5

+ S.D.

-- Control value

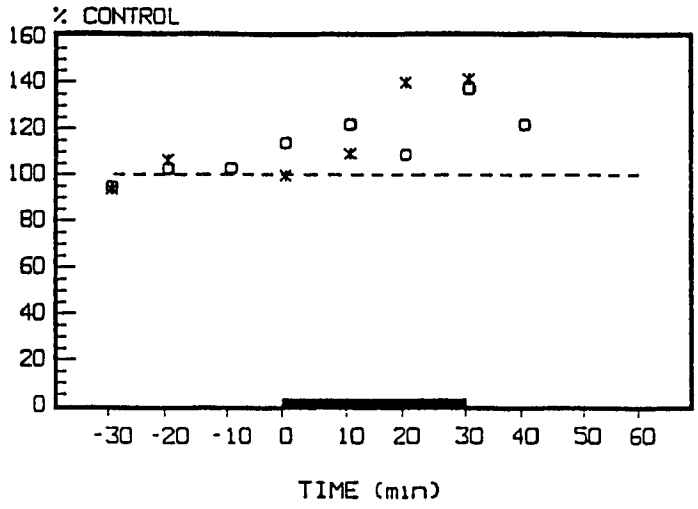
—— Group mean

▬ 30 min of vibration

Fig. 4.3. Facilitatory influence of vibration on RA III in 2 of 9 subjects. Explanations are the same as in Fig. 4.2.

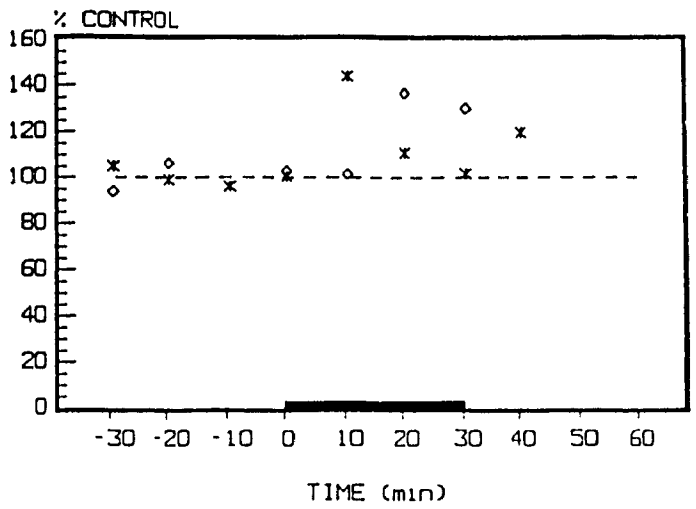
FACILITATION OF BF FLEXION REFLEX

A. INDIVIDUAL DATA

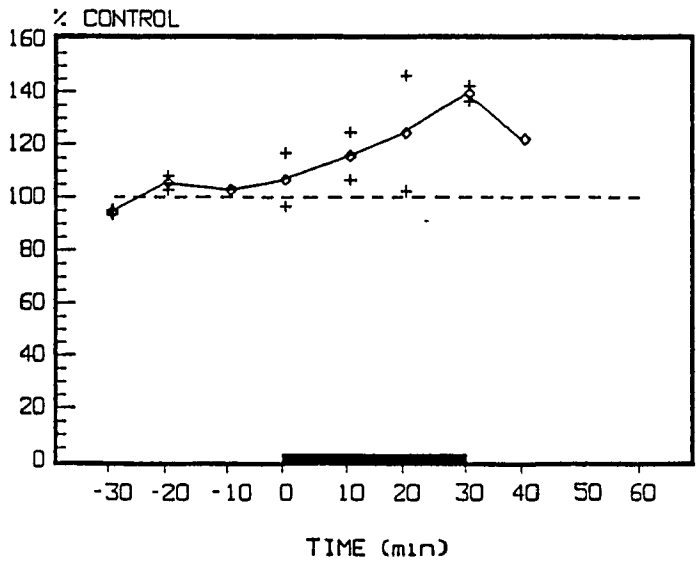


FACILITATION OF TA FLEXION REFLEX

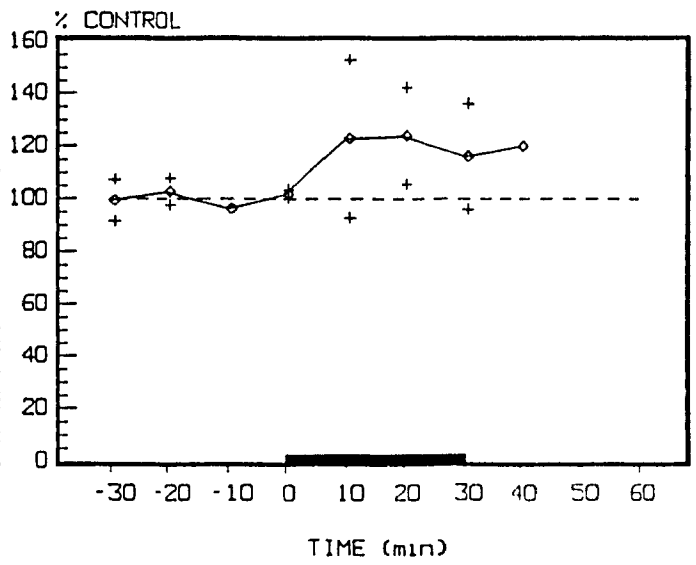
A. INDIVIDUAL DATA



B. GROUP DATA



B. GROUP DATA



n=2

\+ S.D.

-- Control value

—— Group mean

▬ 30 min of vibration

TABLE 4.1

INTRA-SUBJECT COMPARISON OF MAXIMUM MODULATION OF FR RA III AREA WITH CONTROL VALUES IN BF AND TA DURING AND AFTER SEGMENTAL VIBRATION TO THE LOWER LEG

A. BICEPS FEMORIS

Subject	Effect	Control ($uV*s$)	Maximum modulation ($uV*s$)	Maximum modulation Time of occurrence (min)	p-value
TR	Inhibition	5.8 ± 2.4	4.9 ± 1.5	30[a]	.05
LD	Inhibition	4.7 ± 1.4	4.0 ± 1.7	40[a]	.01
JF	Inhibition	1.5 ± 0.3	1.4 ± 0.2	25	.05
SL	Facilitation	3.4 ± 0.8	4.9 ± 1.5	25	.01
AS	No effect	9.5 ± 2.8	10.6 ± 3.3	15	ns
SC	Inhibition	8.8 ± 1.4	6.5 ± 0.9	25	.01
SCA	Facilitation	4.9 ± 0.7	6.8 ± 0.8	25	.01
MC	No effect	7.6 ± 1.4	8.4 ± 1.1	25	ns
GL	Inhibition	7.3 ± 1.9	6.2 ± 1.4	35[a]	.05

B. TIBIALIS ANTERIOR

Subject	Effect	Control ($uV*s$)	Maximum modulation ($uV*s$)	Maximum modulation Time of occurrence (min)	p-value
TR	Inhibition	5.9 ± 1.4	5.3 ± 1.7	25	ns
LD	Inhibition	8.6 ± 1.9	7.6 ± 1.6	40[a]	.01
JF	Inhibition	3.3 ± 0.7	3.0 ± 0.4	5	ns
SL	Facilitation	7.0 ± 1.8	9.6 ± 2.5	20	.01
AS	No effect	7.3 ± 1.5	7.9 ± 1.3	5	ns
SC	Inhibition	11.2 ± 2.0	8.4 ± 1.2	25	.01
SCA	Facilitation	9.5 ± 1.2	13.7 ± 3.2	10	.01
MC	No effect	10.2 ± 1.4	11.2 ± 1.6	5	ns
GL	Inhibition	6.2 ± 1.0	5.2 ± 1.2	25	.05

Values are mean ± 1 S.D.
ns = non-significant at p=.05
a = maximum modulation occurring post-vibration

vibration caused a potentiation of the FR, displayed slightly more marked changes in RA III than those observed in the "inhibition group", increasing to 139.2 ± 2.9% and 140.4 ± 5.1% for BF and TA respectively (Table 4.2).

Maximal modulatory effects of vibration occurred between 15 and 30 minutes into vibration, or within 10 minutes post-vibration in most cases, with the effect setting in gradually. It must be kept in mind that the "time of maximum modulation" is an approximation representing a time bin of 6-8 min, during which the response ensembles were collected, as previously mentioned in Chapter 2.

The group data in Figs. 4.2 and 4.3 clearly show the parallel time course of flexion reflex modulation in BF and TA. Table 4.2 compares the changes in FR among all the subjects who responded to vibration. The values shown represent FR area in BF and TA at time of maximum modulation, normalized with respect to each subject's control value. Interestingly, the RA III was modulated to the same extent in both muscles. In order to quantify the relationship between these two muscles, the Pearson correlation coefficient (r) was computed. As expected, the result confirmed a high correlation, with an r value of 0.81.

DISCUSSION

Natural activation of large-diameter afferents through vibratory stimulation has been shown to induce a reduction in

TABLE 4.2

INTER-SUBJECT COMPARISON OF FR MODULATION BETWEEN BF AND TA FOLLOWING SEGMENTAL VIBRATION TO THE LOWER LEG

SUBJECT	BF (% CONTROL)	TA (% CONTROL)
TR	76.8*	89.4
LD	67.6**	65.8**
JF	90.1*	90.7
SL	141.3**	136.8**
SC	73.3**	75.2**
SCA	137.2**	144.0**
GL	80.7*	84.2*
FR INHIBITION:	n = 5 mean = 77.7 S.D. = 8.4	n = 5 mean = 81.1 S.D. = 10.5
FR FACILITATION:	n = 2 mean = 139.2 S.D. = 2.9	n = 2 mean = 140.4 S.D. = 5.1

* $p < .05$
** $p < .01$

clinical pain (Hansson and Ekblom, 1983; Lundeberg, 1984; Lundeberg et al., 1984) and an elevation in experimental pain threshold (Ekblom and Hansson, 1982; Pertovaara, 1979). Under our experimental paradigm, the main influence of vibration on the human lower limb FR was an inhibition of the nociceptive RA III component, a finding consistent with the results of previous studies. Several neurophysiological investigations (Gregor and Zimmermann, 1972; Handwerker et al., 1975; Salter and Henry, 1986; Wagman and Price, 1969), using transient, non-noxious conditioning stimuli, have also described a characteristically inhibitory effect of large-diameter afferents on the response of spinal neurons to nociceptive stimulation in animals.

An observation worthy of notice is the fact that none of the subjects reported feeling any appreciable change in the intensity of the test stimulus during or after the vibration period. We found this surprising in light of the growing body of evidence pointing to common pathways subtending pain perception and the nociceptive flexion reflex. However, Willer et al. (1979) have shown that the behavior of the human flexion reflex can be dissociated from that of pain perception under certain circumstances. Furthermore, it is possible that the mild influence of vibration on the flexion reflex found in this study would also require systematic measurements of subjective perception to uncover changes in this variable.

Ideally, placebo trials should have been conducted. Lundeberg et al. (1987) have investigated the placebo effect

in a double-blind study comparing actual vibration with placebo vibration in alleviation of chronic pain. Although significantly more patients reported pain relief with actual vibration (i.e. 48%), placebo vibration alleviated pain in 34% of their patients. This result is consistent with previous double-blind studies (Evans, 1974; Thorsteinson, 1978). Lundeberg et al. (1987), however, caution that the achievement of true placebo stimulation is doubtful when studying peripheral stimulation techniques.

It is commonly reported that peripheral conditioning procedures activating large-diameter afferents, such as vibration and low-intensity TENS, could potentiate pain and/or nociceptive reactions in a minority of patients/subjects (Chan and Tsang, 1987; Ekblom and Hansson, 1985; Lundeberg, 1984a,b). We observed such a potentiation on the FR RA III in two of our subjects, a proportion consistent with the findings of previous studies. In an investigation similar to ours, Ertekin and Akcali (1978) reported a marked facilitation of the human BF flexion reflex with vibration applied locally in the immediate vicinity of the test stimulus, and lasting for as long as 20-25 min beyond the vibration. Unfortunately, the duration of application of vibration was not specified. They were not able to explain this facilitation in terms of sensitization/dishabituation phenomena. However, the fact that their subjects reported no concomitant increase in pain sensation raises the likelihood that the efferent motor

pathways of the FR might have been facilitated independently from the afferent pathways.

The question remains as to the identity of the receptors mediating vibration-induced inhibition of pain and nociceptive reactions. Several studies have documented the responsiveness of both superficial and deep receptors to mechanical vibrations. Among the cutaneous and subcutaneous receptors, Pacinian and Meissner corpuscles, as well as hair follicle complexes, have been found to be particularly responsive to vibration (Merzenich and Harrington, 1969; Talbot et al., 1968). In the frequency range below 60 Hz, Meissner corpuscles (glabrous skin) or hair follicle complexes (hairy skin) from monkeys responded maximally, and a sensation of "flutter" was reported by the human subjects. Conversely, Pacinian corpuscles were found to be exquisitely sensitive to frequencies of vibration above 60 Hz, presumably mediating the "vibration" sensation reported by the subjects. In muscle, muscle spindles and even Golgi tendon organs are known to be sensitive to vibration (Eklund and Hagbarth, 1965; Hagbarth, 1973). In this connection, Brown et al. (1967) have found vibration to be a more selective stimulus for muscle spindles than Golgi tendon organs. In man, microneurography has shown that both primary and secondary endings of muscle spindle could be activated by vibration, primary endings being more strongly driven at frequencies above 100 Hz (Burke et al., 1976).

Among the receptor types responsive to high frequency vibration, the Pacinian corpuscle has been proposed as the most likely candidate to mediate vibration-induced analgesia (Lundeberg et al., 1984; Pertovaara, 1979). This hypothesis is consistent with the observations by Lundeberg's group (Lundeberg, 1984a,b,c; et al., 1984) and Ottoson et al. (1981) that pain relief was enhanced when the vibratory stimulus was applied so that contact was made with underlying bony structures. As is well known, Pacinian corpuscles are widely distributed in subcutaneous connective tissue, periosteum, joints and tendons, all of which are more likely to receive transmitted vibration if it is applied to rigid structures such as bone. However, despite using a vibration frequency thought to preferentially activate Pacinian corpuscles, it is impossible to rule out involvement of other cutaneous and muscle mechanoreceptors.

The relatively small probe area and the site and pressure of vibration application used in this study may have recruited only a restricted population of mechanoreceptors. A sub-optimal input to the hypothesized anti-nociceptive mechanisms might explain the relatively weak, albeit significant, inhibition of RA III in our subjects. Indeed, parameters of the vibratory stimulus have been shown to be important determinants of the effectiveness of vibration in pain alleviation (Lundeberg et al., 1984). Hence, the findings reported here may not apply under different sets of parameters.

Other factors which may account for differences in extent and time course of vibratory effects reported in the literature could relate to the intensity and type of pain. It has been reported that vibration is more effective at relieving light to moderate pain (Bini et al., 1984; Lundeberg, 1984b). In our concern to reproduce a noxious sensation and to ensure a stable flexion reflex, subjects were encouraged to reveal their true maximal tolerance level with regard to the electrical stimulus delivered to them. It is possible that such a strong stimulus may have rendered the flexion reflex less amenable to afferent modulation by vibration. Furthermore, the electrical stimulus used in the present study evoked a sharp, transient pain sensation described in the literature as characteristic of Aδ fiber activation, which is believed to mediate acute pain. Our results may therefore not extend to chronic pain conditions.

Our last finding is the gradual onset of the FR modulation by vibration, typically peaking 20-30 min into vibration, and in some cases after its cessation. Although the period of post-vibration FR-testing was too short to observe a return to control values, it allowed us to observe the persistence of the FR modulation. These results concurred with those of clinical pain studies reporting a slower time course of vibration effects with increasing duration of application (Lundeberg et al., 1984; Ottoson et al., 1981). Such a slow time course has also been observed in our

laboratory in a previous study investigating the influence of TENS on the nociceptive flexion reflex (Chan and Tsang, 1987). Bearing in mind the limited number of subjects fully investigated (n=9), the present results should be considered as preliminary. As such, further studies are needed to address the exact nature and extent of the mechanisms involved here. The slow time course of effects observed in this study would, however, be consistent with the gradual build-up of some neuromodulatory substance.

CHAPTER FIVE

SUMMARY AND CONCLUSIONS

This research project was undertaken to investigate the influence of cutaneously-applied vibration on the human lower limb flexion reflex. The rationale was that testing the output of a known nociceptive reflex with a natural tactile input such as vibration, might yield valuable clues as to the neurophysiological mechanisms underlying pain alleviation through peripheral conditioning.

The first study was conducted to establish the nature of the relationship between subjective pain sensation and the flexion reflex. One previous study had already reported a power function to best describe the relationship between pain sensation and the flexion reflex in the upper limb extensor digitorum muscle (Bromm and Treede, 1980). Another study found pain sensation and the lower limb flexion reflex to be linearly related (Willer et al., 1984). These investigations, however, had been performed on muscles in the relaxed state. The possibility that the low background muscle contraction required of the subjects under the FR elicitation paradigm developed in this laboratory might modify the relationship between the two nociceptive measures therefore made it necessary to describe this relationship under our paradigm.

An electrical stimulus was delivered to the sole of the subject's foot to elicit the flexion reflex, which was recorded electromyographically from the biceps femoris muscle. The subject rated the sensation evoked by each stimulus by moving a cursor along a VAS consisting of a 20 cm line drawn on a white sheet of paper. At the beginning of the

experiment, the subject's perception threshold and maximal tolerance were determined. This perceptual range was then divided in 10% increments, and 10 stimuli were delivered at each stimulus intensity, in randomized order. The 10-response ensembles of VAS ratings and RA III area values were averaged and normalized with respect to their respective maxima. The data for all subjects (n=7) were pooled and linear regression lines drawn through the scatter plots. Our results showed that both BF FR area and VAS ratings bore a direct linear relationship with stimulus intensity and with each other, with high correlation coefficients, r=0.91, 0.95 and 0.84 respectively for each function.

In the main experiments, the flexion reflex was elicited with a maximally tolerable electrical stimulus to the subject's foot sole, and recorded from BF and TA with surface EMG electrodes. Fifteen- or twenty- response ensembles were collected at some 6-8 min intervals throughout the experiment. Ensembles were collected prior to, during, and for up to 30 minutes after a 30-min vibration period. The frequency chosen for the vibratory stimulus was 240 Hz, in an attempt to preferentially activate Pacinian corpuscles. Vibration was applied segmentally, to the ipsilateral L5 dermatome.

Area values for the RA III were computed off-line from the averaged ensembles and normalized with respect to the control mean. The area value at time of maximal FR modulation was tested against the control value for significance. Out of nine subjects, the FR was inhibited in five, facilitated

in two, and unchanged in two. The direction and time course of modulation was parallel in BF and TA. The effects of vibration had a slow onset and persisted beyond the vibration period.

The gradual onset and offset of FR inhibition induced by vibration in the present study are in agreement with what has generally been reported in the clinical literature (Ekblom and Hansson, 1982; Lundeberg 1984a,b,c; Lundeberg et al., 1983, 1984). Some researchers have also reported transient effects (Bini et al., 1984; Ottoson et al., 1981; Pertovaara, 1979). However, these studies have used short vibration application periods (20 s to 5 min). In this regard, Lundeberg (1984b) has demonstrated that the duration of pain relief by vibration is related to the duration of application. The time course of vibration effects in this study is also consistent with that observed in a previous study in this laboratory investigating the influence of TENS on the FR (Chan and Tsang, 1987). Whether or not vibration and TENS share the same antinociceptive mechanisms is still a matter of debate. Nonetheless, the slow time course of effects observed in this laboratory for both conditioning procedures would be consistent with the slow release and build-up of some neuromodulatory substance. Despite some reports implicating opioids in TENS analgesia (Chapman and Benedetti, 1977; Salar et al., 1981; Sjolund and Eriksson, 1979; Sjolund et al., 1977), no such claims have been made concerning vibration-induced analgesia. In fact, naloxone has so far been shown

to be unable to reverse the antinociceptive effects of vibration (Hansson et al., 1986; Lundeberg, 1984b; Salter and Henry, 1987). Caution must be exercised, however, when interpreting the results of studies using naloxone to antagonize analgesia. Naloxone has been shown to preferentially bind with μ-receptors (Iwamoto and Martin, 1981; Lord et al., 1977). Also, dynorphins, enkephalins and β-endorphins display distinct affinity patterns for the various known opioid receptors (Hollt, 1983). The reversibility of a given analgesic procedure could therefore be a function of the opioid system(s) implicated and of naloxone doses administered. Evidence for such dosage-dependent effects of naloxone has already been reported by Han and Xie (1984), in an investigation comparing naloxone reversibility in low- and high- frequency acupuncture analgesia. They found that the analgesic effects of these two modalities of acupuncture could be blocked by low (0.5 mg/Kg) and high (24 mg/Kg) doses of naloxone, respectively.

SIGNIFICANCE OF THE STUDY

Nociceptive reflexes in animals and humans have provided researchers with a universal tool to study and compare neurophysiological mechanisms of nociception across different species. In addition, it is generally believed that nociceptive reflexes can faithfully reflect the human

experience of pain. The above arguments, however, are only valid if a high correlation between a given nociceptive reflex and pain sensation can be demonstrated.

Our systematic investigation of the lower limb flexion reflex (FR) and of subjective pain report as a function of varying stimulus intensities established a high correlation between these two nociceptive indices. Although previous studies have reported a parallel modulation of flexion reflexes and pain sensation (Bromm and Treede, 1980; Willer et al., 1984), the susceptibility of the FR to different elicitation protocols made it essential for us to investigate its relationship with subjective sensation under our experimental paradigm. urthermore, the methodology for recording subjective pain report developed in this study can be applied to future studies, so that the modulatory effects of pain conditioning procedures on the flexion reflex and pain sensation can be assessed simultaneously.

In the second study, we proceeded to investigate the influence of vibratory stimulation on the flexion reflex. A previous study in our laboratory had already demonstrated the inhibitory influence of TENS on the FR (Chan and Tsang, 1987). Although preliminary, the results of the present study indicate that vibration could also induce an inhibition of the FR. One particularly important question concerned the temporal profile of vibration-induced FR modulation. The inhibitory effects triggered by vibration displayed a slow onset and decay, not unlike that observed in the above-

mentioned TENS study. This finding is interesting when we consider that the exact mechanisms mediating the inhibitory effects of TENS and vibration are as yet unknown. Could these different peripheral inputs tap into the same antinociceptive systems? Or do they act through distinct, slow-course pain control mechanisms? A better understanding of the endogenous pain control mechanisms will allow optimal exploitation of existing peripheral pain conditioning procedures, and perhaps spur the development of new approaches to pain alleviation.

REFERENCES

Basmajian, J.V. and Blumenstein, R. (1980). Electrode Placement in EMG Biofeedback. Williams and Wilkins, Baltimore.

Bathien, N. and Morin, C. (1972). Variations comparees des reflexes spinaux au cours de l'attention intensive et selective. Physiol. Behav. 9: 533-538.

Baxendale, R.H. and Ferrell, W.R. (1981). The effect of knee joint afferent discharge on transmission in flexion reflex pathways in decerebrate cats. J. Physiol. (Lond.) 315: 231-242.

Besson, J.M. and Chaouch, A. (1987). Peripheral and spinal mechanisms of nociception. Physiol. Rev. 67: 67-186.

Bianconi, R. and Van Der Meulen, J.P. (1963). The response to vibration of the end organs of mammalian muscle spindles. J. Neurophysiol. 26: 177-190.

Bini, G., Cruccu, G., Hagbarth, K.E., Shady, W. and Torebjork, E. (1984). Analgesic effect of vibration and cooling on pain induced by intraneural electrical stimulation. Pain 18: 239-248.

Bromm, B. and Seide, K. (1982). The influence of tilidine and prazepam on withdrawal reflex, skin reaction and pain rating in man. Pain 12: 247-258.

Bromm, B. and Treede, R.D. (1980). Withdrawal reflex, skin resistance and pain rating due to electrical stimuli in man. Pain 9: 399-454.

Brown, M.C., Engberg, I. and Matthews, P.B.C. (1967). The relative sensitivity to vibration of muscle receptors of the cat. J. Physiol. 19: 773-800.

Burke, D., Hagbarth, K.E., Lofstedt, L. and Wallin, B.G. (1976). The responses of human muscle spindle endings to vibration of non-contracting muscles. J. Physiol. (Lond.) 261: 673-693.

Chan, C.W.Y. and Dallaire, M. (1989). Subjective sensation is linearly correlated with the flexion reflex in man. Brain Res. 479: 145-150.

Chan, C.W.Y. and Tsang, H. (1985). A quantitative study of flexion reflex in man: Relevance to pain research. In: Advances in Pain Research and Therapy. Eds. Fields, H.C., Dubner, R. and Cervero, F. Raven Press, New York. Vol. 9, pp. 361-370.

Chan, C.W.Y. and Tsang, H. (1987). Inhibition of human flexion reflex by low intensity, high frequency electrical nerve stimulation (TENS) has a gradual onset and offset. Pain 28: 239-253.

Chapman, C.R. and Benedetti, C. (1977). Analgesia following transcutaneous electrical stimulation and its partial reversal by a narcotic antagonist. Life Sci. 21: 1645-1648.

Chung, J.M., Fang, Z.R., Cargill, C.L. and Willis, W.D. (1983). Prolonged, naloxone-reversible inhibition of the flexion reflex in the cat. Pain 15: 35-53.

Chung, J.M., Fang, Z.R., Cargill, C.L. and Willis, W.D. (1984). Prolonged inhibition of primate spinothalamic tract cells by peripheral nerve stimulation. Pain 19: 259-275.

Crosby, E.C., Humphrey, T. and Laner, E.W. (1962). Correlative Anatomy of the Nervous System. Macmillan, New York.

Cross, D.V., Tursky, B. and Lodge, M. (1975). The role of regression and range effects in determination of the power function for electric shock. Percept. Psychophys. 18: 9-14.

Dallaire, M. and Chan, C.W.Y. (1987) Is subjective pain sensation correlated with the flexion reflex in man? Neurosci. Abs. 13: 109.

D'Amour, F.E. and Smith, D.L. (1941). A method for determining loss of pain sensation. J. Pharmacol. Exp. Ther. 72: 74-79.

Dimitrijevic, M.R., Faganel, J., Gregoric, M., Nathan, P.W. and Trontelj, J.K. (1972). Habituation: Effects of regular and stochastic stimulation. J. Neurol. Neurosurg. Psychiat. 35: 234-242.

Dimitrijevic, M.R. and Nathan, P.W. (1968). Studies of spasticity in man. 3. Analysis of reflex activity evoked by noxious cutaneous stimulation. Brain 91: 349-368.

Dunnett, C.W. (1964). New tables for multiple comparisons with a control. Biometrics 20: 482-491.

Duranti, R., Pantaleo, T. and Bellini, F. (1988). Increase in muscular pain threshold following low frequency-high intensity peripheral conditioning stimulation in humans. Brain Res. 452: 66-72.

Ekblom, A. and Hansson, P. (1982). Effects of conditioning vibratory stimulation on pain threshold of the human tooth. Acta Physiol. Scand. 114: 601-604.

Eklund, G. and Hagbarth, K.-E. (1965). Motor effects of vibratory muscle stimuli in man. Electroenceph. Clin. Neurophysiol. 19: 619.

Ekman, G., Frankenhauser, M., Levander, S., and Mallis, I. (1964). Scaling of unpleasantness of electrical stimulation.Scand. J. Psychol. 5: 257-261.

Eriksson, M.B.E., Sjolund, B.H. and Nielzer, S. (1979). Long term results of peripheral conditioning stimulation as an analgesic measure in chronic pain. Pain 6: 335-347.

Ertekin, C. and Akcali, D. (1978). Effect of continuous vibration on nociceptive flexor reflexes. J. Neurol. Neurosurg. Psychiat. 41: 532-537.

Evans, F.J. (1974). The placebo response in pain reduction. Adv. Neurol. 4: 289-296.

Eyzaguirre, E.R. and Fidone, S. (1975). Physiology of the Nervous System. Year Book, Chicago.

Gaebelin, J., Tayor, S.P. and Borden, R. (1974). Effects of an external cue on psychophysiological reaction to a noxious agent. Psychophysiol. 11: 315-320.

Goldberger, S.M. and Tursky, B. (1976). Modulation of shock-elicited pain by acupuncture and suggestion. Pain 2: 417-429.

Gregor, M. and Zimmermann, M. (1972). Characteristics of spinal neurones responding to cutaneous myelinated and unmyelinated fibres. J. Physiol. 221: 555-576.

Hagbarth, K.-E. (1960). Spinal withdrawal reflexes in the human lower limbs. J. Neurol. Neurosurg. Psychiat. 23: 222-227.

Hagbarth, K.-E. (1973). The effect of muscle vibration in normal man and in patients with motor disorders. In: New Developments in Electromyography and Clinical Neurophysiology. Ed. Desmedt, J.C. Karger, Basel. Vol. 3, pp. 444-462.

Han, J.-S. and Xie, G.X. (1984). Dynorphin: important mediator for electroacupuncture analgesia in the spinal cord of the rabbit. Pain 18: 367-376.

Handwerker, H.O., Iggo, A. and Zimmermann, M. (1975). Segmental and supraspinal actions on dorsal horn neurons responding to noxious and non-noxious skin stimuli. Pain 1: 147-165.

Hansson, P. and Ekblom, A. (1983). Transcutaneous electrical nerve stimulation (TENS) as compared to placebo TENS for the relief of acute oro-facial pain. Pain 15: 157-165.

Hansson, P., Ekblom, A., Thomsson, M. and Fjellner, B. (1986). Influence of naloxone on relief of acute oro-facial pain by transcutaneous electrical nerve stimulation (TENS) or vibration. Pain 24: 323-329.

Hollt, V. (1983). Multiple endogenous opioid peptides. Trends Neurosci. 6: 24-26.

Hugelin, A. (1972). Bodily changes during arousal, attention and emotion. In: Limbic System, Mechanisms and Autonomic Function. Ed. Hackman, C.H. C.C. Thomas, Springfield, pp.202-218.

Hugon, M. (1973). Exteroceptive reflexes to stimulation of the sural nerve in normal man. In: New Developments in Electromyography and Clinical Neurophysiology. Ed. Desmedt J.C. Karger, Basel. Vol. 3, pp. 713-729.

Hunt, C.C. (1961). On the nature of vibration receptors in the hind limb of the cat. J. Physiol. 155: 175-186.

Hunt, C.C. and McIntyre, A.K. (1960). An analysis of fibre diameter and receptor characteristics of myelinated cutaneous afferent fibres in cat. J. Physiol. 153: 99-112.

Iwamoto, E.T. and Martin, W.R. (1981). Multiple opioid receptors. Med. Res. Rev. 1: 411-440.

Janko, M. and Trontelj, J.V. (1983). Flexion withdrawal reflex as recorded from single human biceps femoris motor neurones. Pain 15: 167-176.

Janko, M. and Trontelj, J.V. (1987). Transcutaneous electrical nerve stimulation: A microneurographic and perceptual study. Pain 9: 219-230.

Jenner, J.R. and Stephens, J.A. (1982). Cutaneous reflex response and their central nervous pathways studied in man. J. Physiol. (Lond.) 333: 405-419.

Johansson, R.S., Landstrom, U. and Lundstrom, R. (1982). Responses of mechanoreceptive afferent units in the glabrous skin of the human hand to sinusoidal skin displacements. Brain Res. 244: 17-25.

Kandel, E.R. and Schwartz, J.H. (1985). Principles of Neuroscience. 2nd ed. Elsevier North-Holland, New York.

Kearney, R.E. and Chan, C.W.Y. (1979). Reflex response of human arm muscles to cutaneous stimulation of the foot. Brain Res. 170: 214-217.

Le Bars, D., Dickenson, A.H. and Besson, J.M. (1979a). Diffuse noxious inhibitory controls (DNIC) - I. Effects on dorsal horn convergent neurones in the rat. Pain 6: 283-304.

Le Bars, D., Dickenson, A.H. and Besson, J.M. (1979b). Diffuse noxious inhibitory controls (DNIC) - II. Lack of effect on non-convergent neurones, supraspinal involvement and theoretical implications. Pain 6: 305-327.

Le Bars, D., Guilbaud, G., Jurna, I. and Besson, J.M. (1976). Differential effects of morphine on responses of dorsal horn lamina V type cells elicited by A and C fibre stimulation in the spinal cord. Brain Res. 115: 518-524.

Le Bars, D., Menetrey, D., Conseiller, C. and Besson, J.M. (1975). Depressive effects of morphine upon lamina V cell activities in the dorsal horn of the spinal cat. Brain Res. 98: 261-277.

Lee, K.H., Chung, J.M. and Willis, W.D. (1985). Inhibition of primate spinothalamic tract cells by TENS. J. Neurosurg. 62: 276-287.

Lloyd, D.P.C. (1943). Neuron patterns controlling transmission of ipsilateral hind limb reflexes in cat. J. Neurophysiol. 6: 293-315.

Lord, J.A.H., Waterfield, A.A., Hughes, J. and Kosterlitz, H.W. (1977). Endogenous opioid peptides; multiple agonists and receptors. Nature (Lond.) 267: 495-499.

Lundberg, A. (1964). Supraspinal control of transmission in reflex paths to motoneurones and primary afferents. Prog. Brain Res. 12: 197-221.

Lundeberg, T. (1984a). Long-term results of vibratory stimulation as a pain relieving measure for chronic pain. Pain 20: 13-23.

Lundeberg, T. (1984b). Vibratory stimulation for the alleviation of pain. Amer. J. Chin. Med. 12: 60-70.

Lundeberg, T. (1984c). A comparative study of the pain alleviating effect of vibratory stimulation, transcutaneous electrical nerve stimulation, electroacupuncture and placebo. Amer. J. Chin. Med 12: 72-79.

Lundeberg, T., Nordemar, R. and Ottoson, D. (1984). Pain alleviation by vibratory stimulation. Pain 20: 25-44.

Lundeberg, T., Ottoson, D., Hakansson, S. and Meyerson, B.A. (1983). Vibratory stimulation for the control of intractable chronic orofacial pain. In: Advances in Pain Research and Therapy. Ed. Bonica, J.J. Raven Press, New York. Vol. 5, pp. 555-561.

Lundeberg, T., Abrahamsson, P., Bondesson, L. and Haker, E. (1987). Vibratory stimulation compared to placebo in alleviation of pain. Scan. J. Rehab. Med. 19: 153-158.

Martin, W.R., Eades, C.G., Fraser, H.F. and Wikler, A. (1964). Use of hindlimb reflexes of the chronic spinal dog for comparing analgesics, J. Pharmacol. Exp. Ther. 144: 8-11.

Meinck, H.M., Piesur-Strehlow, B. and Koehler, W. (1981). Some principles of flexor reflex generation in human leg muscles. Electroenceph. Clin. Neurophysiol. 27: 650-654.

Melzack, R. and Wall, P.D. (1965). Pain mechanisms: a new theory. Science 150: 171-179.

Melzack, R. and Wall, P.D. (1982). The Challenge of Pain. Penguin Books, Markham.

Merzenich, M.M. and Harrington, T. (1969). The sense of flutter-vibration evoked by stimulation of the hairy skin of primates: comparison of human sensory capacity with the responses of mechanoreceptive afferents innervating the hairy skin of monkeys. Exp. Brain Res. 9: 236-260.

Ottoson, D., Ekblom, A., and Hansson, P. (1981). Vibratory stimulation for the relief of pain of dental origin. Pain 10: 37-45.

Pansky, B. and Allen, D.J. (1980). Review of Neurosciences. MacMillan, New York.

Pertovaara, A. (1979). Modification of human pain threshold by specific tactile receptors. Acta Physiol. Scand. 107: 339-341.

Pomeranz, B. and Paley, D. (1979). Electroacupuncture hypalgesia is mediated by afferent nerve impulses: An electrophysiological study in mice. Exp. Neurol. 66: 398-402.

Roby-Brami, A. and Bussel, B. (1987). Long-lasting spinal reflex in man after flexor reflex afferent stimulation. Brain 110: 707-725.

Routtenberg, A. (1968). The two-arousal hypothesis: reticular formation and limbic system. Psychol. Rev. 75: 51-79.

Salar, G., Job, I., Mingrino, S., Bosio, A. and Trabucchi, M. (1981). Effect of transcutaneous electrotherapy on CSF β-endorphin content in patients without pain problems. Pain 10: 169-172.

Salter, M.W. and Henry, J.L. (1986). Differential effects of peripheral vibration on nociceptive versus non-nociceptive neurones in the lumbar dorsal horn of the cat. Soc. Neurosci. Abs. 12: 376.

Salter, M.W. and Henry, J.L. (1987). Evidence that adenosine mediates the depression of spinal dorsal horn neurones induced by peripheral vibration in the cat. Neuroscience 22: 631-650.

Shahani, B.T. and Young, R.R. (1971). Human flexor reflexes. J. Neurol. Neurosurg. Psychiat. 34: 616-627.

Sherer, C.L., Clelland, J.A., O'Sullivan, P., Doleys, D.M. and Canan, B. (1986). The effect of two sites of high frequency vibration on cutaneous pain threshold. Pain 25: 139-140.

Shimamura, M. and Livingston, R.B. (1963). Longitudinal conduction systems serving spinal and brain-stem coordination. J. Neurophysiol. 26: 258-272.

Shin, H.K., Kim, J. and Chung, J.M. (1986). Inhibition and excitation of the nociceptive flexion reflex by conditioning stimulation of a peripheral nerve in the cat. Exp. Neurol. 92:335-348.

Sjolund, B.H. (1985). Peripheral nerve stimulation suppression of C-fiber-evoked flexion reflex in rats. J. Neurosurg. 63: 612-616.

Sjolund, B.H. and Eriksson, M.B.E. (1979). The influence of naloxone on analgesia produced by peripheral conditioning stimulation. Brain Res. 173: 295-301.

Sjolund, B., Terenius, L. and Eriksson, M. (1977). Increased cerebrospinal fluid levels of endorphins after electro-acupuncture. Acta Physiol. Scand. 100: 382-384.

Sternback, R.A., and Tursky, B. (1964). On the psychophysical power function in electric shock. Psychonomic Sci. 1: 217-218.

Sternback, R.A., and Tursky, B. (1965). Ethnic differences among housewives in psychophysical and skin potential responses to electric shock. Psychophysiol. 1: 241-246.

Stevens, S.S. (1957). On the psychophysical law. Psychol. Res. 64: 153-181.

Sullivan, R. (1968). Effect of different frequencies of vibration on pain-threshold detection. Exp. Neurol. 20: 135-142.

Talbot, W.H., Darian-Smith, I., Kornhuber, H.H. and Mountcastle, V.B. (1968). The sense of flutter-vibration: comparison of the human capacity with response patterns of mechanoreceptive afferents from the monkey hand. J. Neurophysiol. 31: 301-334.

Thorsteinson, G., Stonnington, H.H., Stillwell, G.K. and Elveback, L.R. (1978). The placebo effect of transcutaneous electrical stimulation. Pain 5: 31-41.

Vallbo, A.B. and Johansson, R.S. (1984). Properties of cutaneous mechanoreceptors in the human hand related to touch sensation. Human Neurobiol. 3: 3-14.

Wagman, I.H. and Price, D.D. (1960). Responses of dorsal horn cells of M. mulatta to cutaneous and sural nerve A and C fiber stimuli. J. Neurophysiol. 32: 803-817.

Wall, P.D. (1978). The gate control theory of pain mechanisms. A re-examination and re-statement. Brain 101: 1-18.

Wall, P.D. and Cronly-Dillon, J.R. (1960). Pain, itch and vibration. Arch. Neurol. 2: 365-375.

Wall, P.D. and Woolf, C.J. (1984). Muscle but not cutaneous C-afferent input produces prolonged increases in the excitability of the flexion reflex in the rat. J. Physiol. (Lond.) 356: 443-458.

Willer, J.C. (1977). Comparative study of perceived pain and nociceptive flexion reflex in man. Pain 3: 69-80.

Willer, J.C. (1983). Nociceptive flexion reflexes as a tool for pain research in man. In: Motor Control Mechanisms in Health and Disease. Ed. Desmedt, J.E. Raven Press, New York. pp. 809-827.

Willer, J.C., Bathien, N. and Hugelin, A. (1976). Evaluation of central and peripheral pain mechanisms by nociceptive flexion reflex in man. In: Advances in Pain Research and Therapy. Eds. Bonica, J.J. and Albe-Fessard, D. Raven Press, New York. Vol. 5, pp. 131-135.

Willer, J.C., Boureau, F., and Albe-Fessard, D. (1979). Supraspinal influences on nociceptive flexion reflex and pain sensation in man. Brain Res. 179: 61-68.

Willer, J.C., Boureau, F. and Albe-Fessard, D. (1980). Human nociceptive reactions: Effects of spatial summation of afferent input from relatively large diameter fibers. Brain Res. 201: 465-470.

Willer, J.C. and Bussel, B. (1980). Evidence for a direct spinal mechanism in morphine-induced inhibition of nociceptive reflexes in humans. Brain Res. 187: 212-215.

Willer, J.C. Roby, A. and Le Bars, D. (1984). Psychophysical and electrophysiological approaches to the pain-relieving effects of heterotopic nociceptive stimuli. Brain 107: 1095-1112.

Willis, W.D. and Coggeshall, R.E. (1978). Sensory Mechasisms of the Spinal Cord. Plenum Press, New York.

Winkelmann, R.K. (1960). Nerve Endings in Normal and Pathologic Skin. Thomas, Springfield, Ill.

Woolf, C.J., Mitchell, D. and Barrett, G.D. (1980). Antinociceptive effect of peripheral segmental electrical stimulation in the rat. Pain 8: 237-252.

Woolf, C.J. and Wall, P.D. (1986). Relative effectiveness of C primary afferent fibers of different origins in evoking a prolonged facilitation of the flexor reflex in the rat. J. Neurosci. 6: 1433-1442.