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Acoustic Myography -The signal from contracting skeletal muscles

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Yan-Dong Yao BSc (King's College, London)

This thesis is submitted as part of requirement for a degree of Doctor of Philosophy in the University of Glasgow.

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To my family and friends who share my dreams and light the way.

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SUMMARY:

Muscular contraction is associated with low frequency transverse mechanical waves called acoustic myograph (AMG). It can be recorded by microphones and accelerometer. The origin of AMG is thought to be due to the lateral movement of the muscle as a whole. The signal is implicated in many reports as having great potential as force indicator during muscular contraction and could be use clinically in distinguishing healthy and disease, and evaluate force in muscles normally with little clinical access.

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The aim of this series of investigation was to assess the feasibility of using AMG as a force indicator, especially in situations where direct force measurement is not practical and in FES applications as fatigue indicator. Attempts were made to clarify AMG signal characteristics and assess the possible parameters which could be deployed to describe the AMG signal.

Two series of experiments were carried out: one on voluntary contractions of human quadriceps and the other on stimulated contractions of rabbit anterior tibialis. Strain gauges were used to measure force and accelerometers were used to record AMG signal from the skin surface of the thigh on human and from the muscle surface of rabbit anterior tibialis. The AMG signal was recorded between 0.5Hz and wideband frequency and sampled at 512Hz. AMG signal amplitude was calculated by both the rectify-integrated and room-mean-square methods. Frequency content of AMG signal was analyzed by Fast-Fourier-Transform method.

The studies carried out on human quadriceps were to: 1) Locate the possible optimal recording site for AMG. 2) Investigate the relationship

between AMG and force production during isometric constant force and varying force contractions. 3) Study the AMG-force relationship during sustained and intermittent contractions till fatigue. The study performed on rabbit anterior tibialis were to: 1) Investigate the AMG signal characteristic with changing muscle length both under twitch and tetanic contractions. 2) Examine the AMG signal change with stimulation frequencies both at fixed and varied muscle lengths. 3) Assess the possible association between force and AMG in stimulated contraction till fatigue by both continuous and intermittent stimulation. 4) Study the influence of fatigue on AMG-force relationship under different stimulation frequencies.

Results shown that there is no single optimal position for AMG recording on human quadriceps. But high AMG signal intensity were recorded at the mid and proximal-lateral region. The data presented in this report collaborated a linear relationship between AMG and force level in isometric contractions of human quadriceps muscle. This close link between AMG signal intensity and force production is also present in fatigue induced by sustained muscle contractions but not by intermittent contractions.

The data obtained from rabbit tibialis anterior muscle showed less systemic relationships between AMG and force production. AMG signal intensity did not display a close relationship with force under condition of changing muscle length, stimulation frequency and fatigue. There were no significant change in AMG signal median frequency during voluntary contractions and AMG signal recorded during stimulated contractions were dominated by stimulation frequency.

The good correlation between AMG and force in voluntary

contractions, shows promise as an indicator of voluntary force from isometric contractions and fatigue by sustained contractions. The possible clinical use of AMG could be in the area of assessing force output from muscles with limited access, such as paraspinal muscle and facial muscles. It could also be used in conjunction with EMG to assess the state of the muscle function in health and disease and muscle mechanic in training.

The use of AMG as force indicator in stimulated contractions, such as FES applications, requires further investigation. It did not appear to relate strongly to force output under conditions investigated. Alternative properties of AMG signal, such as power content in specific frequency range, should be investigated further for FES applications. The AMG median frequency certainly is not a good force indicator under all conditions tested in the experiment.

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CHAPTER 1

GENERAL INTRODUCTION

One of the distinctions between plants and animal kingdom, is muscular movement. Muscular movement requires precise control of muscle to generate the right amount of tension with right muscle length and/or velocity of change in muscle length, for the desired effect and the co-ordination between different muscles in the body. Movement and posture maintenance involving skeletal muscle control can effectively be carried out by the vast majority. Most of animals and human are, in fact, so good at this type of muscular control, that some of the action and strategies of movement does not enter into their conscious thinking. A START START

However, there are people, with malfunction / damage to skeletal muscle or their innervation, who cannot attempt even the simplest tasks involving skeletal muscle without some external aids. The Functional Electrical Stimulation (FES), involving the application of electrical current to elicit contraction, can bring about movements. It can restore some features of normal motor activity to paralysed persons. This method is one of the external aids currently in use to help the paraplegics, ie people with injuries to portion of the spinal cord controlling some skeletal muscles, to perform simple tasks such as feeding, standing up and sitting down and walking.

THE PROBLEM OF FATIGUE: FES has limited success in movement reconstruction in paraplegics (Pedotti & Ferrarin 1992). However, there are problems associated with this method. The bioengineering systems currently available cannot detect the onset of fatigue, when the skeletal muscle is being repeatedly stimulated by electrical current. Prolonged stimulation can cause the skeletal muscle to fatigue and thus produce less force to perform tasks. This is

potentially dangerous in case of walking and standing, when the patient could not support his/her own weight. This raised the need for a detection system for the fatigue state of skeletal muscle, to prevent over-stimulation induced muscle damage and fatigue.

THE FEED BACK SIGNAL: A detection system for fatigue should utilise a signal closely associated with the state of fatigue. There are few potential candidates, one of which is the force produced by electrical stimulation. In the fatigue state, force generation is known to fall with progressive fatigue. This decline in force is a good candidate for the detection system. Forces are often measured by means of a strain gauge, however there are cases where this type of force measurement is not appropriate, such as during walking or standing. An alternative must then be sought. Electromyography (EMG) is one possible candidate. The amplitude of the signal can be employed to determine the state of muscle activation with respect to patterns and performance. The EMG signal can be analyzed on-line by computer systems, and in the state of fatigue the rectified and integrated EMG (iEMG) signal tends to increase in relation to the force of contraction. The ratio of iEMG-Force could be used for indicating fatigue. However, EMG signal cannot be a reliable indicator due to the interference of the electrical stimulation and the possible movement of electrodes during a contraction. The frequency content of EMG signal might be an alternative. The power spectrum, the relative proportion of energy content at each frequency, shifts towards low frequencies as fatigue develops. Unfortunately, the change in frequency spectrum of EMG, thought to be capable of indicating fatigue, was observed before force

reduction and remained unchanged with further fall in force. It is possible that the frequency change is a consequence of muscular activity and not associated directly with the development of fatigue (Edwards 1981). Thus, EMG as a fatigue indicator is of limited use.

There have been some recent interest in the usage of Acoustic Myography (AMG) as an indictor of fatigue (Barry, Geiringer & Ball 1985). Using this method a mechanical signal can be recorded with no potential interference from electrical stimulation. The AMG signal is thought of as the mechanical counterpart of EMG, it can be used to reflect the muscle performance (ie. force and velocity of a contraction) more than with EMG. This is especially useful in the case of myopathies, where a large electric signal may be accompanied by only a small mechanical response. In order to assess the possibility of using AMG signal as indicator of force, questions had to be asked in regards to characteristic of AMG signal and the possible factors influencing its generation. The series of study presented in this report will further clarify AMG characteristic in normal human voluntary contractions of the lower extremities. In addition, investigation of AMG signal characteristics during electrical stimulation of skeletal muscles in rabbits, can provide a model to relate to the behaviour of muscle in both fresh and fatigued state.

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In order to improve the quality of life with FES, individuals with damaged skeletal muscle system, understanding the essential elements of normal skeletal muscle function is required. Most of muscular activities, once learned at childhood, are executed by most of us without conscious thinking. Just how does the brain function to bring about this precise control, in deciding how hard to grip a tooth brush

Figure 1. Diagrammatic representation of a motor unit, with motor neuron and the associated muscle fibres. (modified from Basmajian & De Luca 1985)



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when cleaning teeth and how fast or slow should the muscle act when combing hair?

1.1 SKELETAL MUSCLE FUNCTION

To understand the function of skeletal muscle, one must first comprehend their structures. Skeletal muscle contains many fibres, ie. hundreds of thousands in a human biceps muscle or gastrocnemius muscle. Normally muscle fibres do not act individually but in groups, or motor units. A motor unit is defined as: a motoneuron and the set of muscle fibres innervated by it. This is shown diagrammatically in figure 1. The number of fibres in a motor units may vary from muscle to muscle, ie. 5 to 6 in a human extraoccular muscle and 2000 in the medial head of gastrocnemius muscle (reviewed by Basmajian and De Luca 1985). For each motor unit to perform work, there must be the central control from the spinal cord in the form of nerve impulses, the chemical signalling across the neuromuscular junction, the propagation of electrical signal in the form of action potential across the muscle fibre membrane and the activation of the muscle myofibril to perform the contractions.

1.1.1 MOTOR UNIT TYPES:

Not all the muscle fibres have the same microscopic appearance and functional properties. They vary in diameters and oxidative capacity of the muscle fibres. The motor units can therefore be classified according to the type of muscle fibres with different enzymic / metabolic properties or the functional characters of neurons. The difference between various type of muscle fibres in a motor units can

be seen in features such as enzymic activity, glycogen content, fat content, oxidative / glycolytic capacity of fibres, fibre diameter in a motor unit and excitability at neural muscular junction. There are also differences in neural characteristics such as nerve fibre diameter, conduction velocity. These differences ultimately lead to differences in muscle mechanics, such as: peak force achievable, rate of force development and fatiguability of a motor unit. There are essentially three general groups of motor units, classified based on their mechanical response to stimulation: •type S (slow unit) has high resistance to fatigue, high excitability, small fibre diameter and generates lower peak force; etype FF (fast fatiguable unit) has little resistance to fatigue, with low excitability, large fibre diameter and can generate high peak forces. •type FR (fatigue resistant unit) is the intermediate between S and FF type motor unit. Type FR motor unit can produce high peak force and has large fibre diameter (Burke, Levin & Zajac 1971). S type motor units are best suited for posture maintenance with continuous muscle contraction at low force. FF type of motor units are best suited for quick and powerful contractions, such as jumping. There were observations of motor unit fibre type conversions with artificial stimulation, as reviewed by Salmons (1985).

The following section of the report will focus on control to motor units and the effect on the motor unit function in terms of force generation.

1.1.2 MOTOR UNIT BEHAVIOUR:

The discovery of the existence of different types of motor units

did not shed light on the activation and coordination of these motor
units in force production as a group or the control of force production.
It is logical to assume that the central nervous system is partly
responsible for the precise force generation with some feedback from
muscle being activated. There are two essential questions being asked:
1) Is there a strategy governing the activation of a motor unit? 2) Are
there rules governing activated motor unit control and regulation? The
answers are discussed in the following sections.

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RECRUITMENT AND DECRUITMENT:

For a motor unit to function, there must be control of activation and de-activation. The activation of a motor unit at a certain force level is termed recruitment and the subsequent deactivation is decruitment. There has been substantial investigation of individual motor unit properties and the coordinated characteristics of motor neurone pool. There are a few general characteristics:

A) Order of recruitment: Motor units with small motor neurons were observed to be recruited before those with large motor neurons and the lower threshold motor units has slower conduction velocity. This phenomena is called the "size principle". (Henneman, Somjen & Carpenter 1965, Milner-Brown, Stein & Yemm 1973).

B) Contraction type dependence: During a fast force increasing contraction in ballistic movement, recruitment is dependent on rate and strength of force (De Luca, Lefever, McCue & Xenakis 1982). The force threshold of a motor unit shifts down with increasing rate of force output and the peak of twitch force occurs at approximately the same muscle tension irrespective of the force rate (Budingen and Freund 1976). In a non-ballistic contraction, recruitment is force dependent,

but independent of muscle and the rate of force change. In a constant force isometric contraction, the recruitment can change with time during one contraction, implying the recruitment of new motor units throughout a contraction (reviewed by Basmajian and De Luca 1985). 1. N. 1.

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C) Muscle dependence: In some muscles all motor units are recruited at relatively low force levels, while other continues to recruit motor units up to maximal force: In first dorsal interosseous, recruitment of all motor units was shown to be completed at force level of 50%MVC; while the deltoid muscle continues to recruit motor units up to 80%MVC, as shown by De Luca et al (1982) with data represented in figure 2. There were reports of recruitment of motor units up to 80%MVC in biceps brachii, 70%MVC in brachialis and 50%MVC in adductor pollicis. It is apparent form these reports that recruitment of new motor units is only one strategy in muscle force generation.

D) Phase dependency: In a decreasing force contraction, the motor units are de-activated or decruited in the opposite order in which they were recruited. The recruitment and decruitment is well demonstrate in figure 2. The recruitment and decruitment sequences can change in the same muscle when performing tasks of different orientation (Person 1974). This is probably due to changing mechanical factors and the recruitment sequence is constant in any one plane of movement.

FIRING RATES:

After recruitment of a motor unit, the mechanical output of that motor unit is controlled by the firing rate of the neuron innervating it. Firing rate is the reciprocal of the time between two adjacent

Figure 2. Recuitment and decruitment of 8 motor units from the human deltoid and first dorsal interosseous (FDI) muscles during force-varying contractions of 40 and 80%MVC. Subjects were divided into four groups: normal, long-distance swimmers, powerlifters and pianists. Recruitment was only observed up to 52%MVC in FDI, but 80%MVC in the deltoid. (modified from De Luca et al 1982a)



discharges of motor unit. The firing rate of motor units can be affected by the following factors:

A) Muscle type: In smaller muscles, such as those in adductor pollicis, the firing rates of motor units reach relatively higher values than motor unit in biceps brachii muscle (Kukulka and Clamann 1981).

B) Force level of a contraction: Low threshold motor units began firing earlier with low firing rate, then increase firing rate slowly to reach a maximum at relatively low force level. The higher the recruitment threshold of the motor unit, the less the motor unit increased its firing rate with increasing force (Person and Kuddina 1972). The minimal firing rate of a motor unit increased linearly with the threshold of recruitment and the motor unit near the muscle surface had higher thresholds of recruitment than those deep in the muscle (Clamann 1970). It is also known that an active motor unit could increase firing rate slightly with increasing force. A high correlation between force and firing rate of a motor unit was demonstrated. As the force level decreased, the firing rate decreased to 30-40% of the preferred rate before becoming inactive (De Luca et al 1982). の一般になって

Motor units can contract with isolated twitches and tetanus. Motor units with higher recruitment threshold and short contraction time can achieve full tetanus at a relative high firing rate, and the opposite is true of motor units with lower recruitment threshold (De Luca 1982).

C) Contraction type: In a strenuous isometric contraction, the firing rate of relatively high threshold motor unit abruptly doubled in first dorsal interosseous muscle (De Luca et al 1982), as illustrated in figure 3. This represents an extra mean of tapping into the resources for



Figure 3. The firing activity of two high force-threshold motor units. The dash line is the motor unit behaviour and the solid line is the force output, during a triangular force-varying contraction of the first dorsal interosseous (FDI). (modified from De Luca et al 1982a)

force generation. In a sustained contraction of healthy muscles, the firing rate of motor units have tendency to decrease independently of force output of the muscle (Person and Kudina 1972, De Luca et al 1982). This is shown in figure 4. Neuronal adaptation processes and or/ decrease in the excitation to the muscle were implicated. The same phenomenon was observed during force-varying isometric contractions and the firing rate at decruitment is less than that at recruitment during a force-varying contraction. Furthermore, the firing rates of the fast motor unit motoneuron decreased, and the slow-twitch motoneuron did not (Kernell and Monster 1982). Thus, firing rate is at least in part a property of the motoneuron. It is also observed that a decrease in fast motor unit firing rates but not slow motor units were observed in force varying contractions (Bigland-Ritchie, Furbush & Woods 1986) A CALL AND A

1.1.3 FATIGUE

The term "muscle fatigue" was generally used to describe many conditions, such as metabolic failure as result of marathon running or signal transmission failure resulting from strong contraction that occluded the muscle. The most appropriate definition comes from Edwards (1981), which states "muscular fatigue is a failure to maintain the required or expected force". Fatigue sometimes even begin before decrease in force is visible. Fatigue can be a failure of any, or the combinations, of links in the command chain. The links can be grouped: 1) The upper motor neurone pool in motor cortex, which is in the highest position of command chain for muscle contraction, possibly being influenced by feedback from muscle, tendon and joint (Gandevia, Allan, Butler & Taylor 1996). 2) the action of the lower motor neuron



Figure 4. The firing records of four concurrently active motor units (dash line) are shown superimposed on the force output (continuous line), during a constant-force isometric abduction of the deltoid. (modified from De Luca et al 1982b)

pool in spinal cord, which directly governs motor units activity; 3) the excitation-contraction coupling processes that transmitting the signal to muscles and the metabolic and enzymic process providing energy to the contractile mechanism. Two types of fatigue are generally studied: the central fatigue, which is the failure at the sites of upper and lower motor neurons; while the peripheral fatigue occurs after the impulses being transmitted to the pre-synaptic area of neuromuscular junctions.

CENTRAL FATIGUE:

This type of fatigue is the failure of neural drive, resulting in either a reduction in the number of functioning motor units, or the reduction in motor unit firing frequency.

It is commonly believed that central fatigue does not occur in prolonged intermittent submaximal contractions. This can be tested by comparing the force of a maximal voluntary contraction with that obtained by supermaximal tetanic stimulation of the motor nerve, which is rather painful for the subjects and could result in muscle tendon damage. A safer, more elegant and commonly used method is that of 'twitch interpolation', which involves the application of single maximal stimuli to the appropriate motor nerve during a voluntary sub-maximal contraction, as shown in figure 5 (Merton 1954)

Evidence of central fatigue was demonstrated during sustained MVC contractions of quadriceps muscles. It was observed that there is a greater tendency for central fatigue to develop as time passes in a well motivated subject (Bigland-Ritchie, Jones, Hosking & Edwards 1978). Grimby, Hannerz & Hedman (1981) provided evidence to support the existence of central fatigue, which affects motor unit with



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short contraction time and high axonal conduction velocity more significantly than those with long contraction time and low axonal conduction velocity. Furthermore, there seemed to be elevated recruitment thresholds for motor units with short contraction time and high axonal conduction velocity with fatigue. A Contraction of the second second

PERIPHERAL FATIGUE:

This type of fatigue is the failure of force generation of the whole muscle, as a consequence of impaired neuromuscular transmission, or failure of muscle action potentials; or the impaired excitationcontraction coupling. Force generation was impaired in adductor pollicis muscle during sustained MVC till fatigue and blood flow occlusion advances this (Merton 1954). The action potential recorded from the surface of the muscle appeared to be unchanged (Merton 1954, Bigland-Ritchie 1981). Therefore, fatigue in this study was due to impaired transmission beyond the spinal cord and not at the neuromuscular junction.

Fatigue can also be a consequence of failure of action potential propagation from the post-synaptic endplate. This may stem from the accumulation of potassium ions near the muscle fibre, leading to a loss of membrane potential and a change in waveform of action potential (Sojaard, Adam & Saltin 1985). This change in action potential due to high extracellular potassium concentration is similar to the change in human adductor pollicis muscle action potential after fatigue, shown in figure 6 (Bigland-Ritchie, Jones & Woods 1979), where the action potential is slowed and the amplitude decreased and ultimately failed.

Other metabolic changes within the muscle fibre can contribute



Figure 6. Changes in action potential waveform in human adductor pollicis. The diagram shown action potential after 1 and 20 seconds stimulation at 50 Hz. (modified from Bigland-Ritchie et al 1979)
to development of fatigue. The accumulation of hydrogen ion and lactate is known to inhibit the action of enzymes and limit rate of energy supply in fatigue. Hydrogen ion accumulation is thought to inhibit calcium activation of actomyosin ATPase (Fitts 1994). Furthermore, induced alkalemia can prolong and acidemiamay reduce endurance in dynamic exercise.

In terms of force generation, the depletion of metabolic substrate and the accumulation of its end products, together with the increase in temperature and the limited enzymic activity, twitch tension is therefore reduced in individual fatigued motor units and, rise and relaxation of twitch takes progressively longer (Edwards 1981).

FATIGUE DUE TO ELECTRICAL STIMULATION:

It was shown that failure of propagation of electrical activity at and beyond the neuromuscular junction (ie. peripheral fatigue) develops at a rate dependent on the frequency of stimulation in human muscles. There are two categories of peripheral fatigue: the high frequency fatigue with stimulation frequency being about 80Hz and the low frequency fatigue of about 20Hz (Edwards 1977). Low frequency fatigue is the selective loss of forces at low frequency of stimulation and is thought to be the result of impaired excitation-contraction coupling (Grimby & Hannerz 1977). A good example is the fatigue induced by submaximal contractions ie. 19 to 30 Hz stimulation. The effect of this low frequency fatigue is more noticeable in a stretched muscle ie. during eccentric contractions (Newham, Mills, Quigley & Edwards 1983)

On the contrary, high frequency fatigue is the selective loss of

force at high stimulation frequencies (Bootterman, Graf & Tansey 1992). This type of fatigue is believed to be a result of failure of neuromuscular junction transmission and / or membrane excitation and propagation of electrical signals (Edwards 1977). This type of fatigue can also be obtained by cooling muscle and inducing ischaemia. This results in a reduction of maximal force output of a muscle be it voluntary or electrically elicited.

EMG AND FATIGUE:

As muscle fatigue develops, the attempt to maintain force output is achieved by increasing rate of activation of the active motor units (Kernell, Ducati & Sjoholm 1975). This is reflected in the increase EMG signals. A useful indicator of fatigue was that of the ratio between EMG and force. Signals can be examined for their energy content in different frequency range, which is called power frequency spectrum. The power spectrum of EMG are typically with multiple peaks, and spread over frequency range between 2 to 250Hz. Mean and median frequency, which is the theoretical point equally dividing the power content, can be calculated. This mean frequency could be taken as mean firing rate of motor units in muscle with homogeneous fibre type.

In fatigue states, a shift towards low frequency spectrum can be seen in the surface EMG (Bigland-Ritchie, Johnson, Lippold & Woods 1982). EMG is thought to have two components, the motor unit spike train and the motor unit action potential. The observed shift in the power spectrum could be attributed to both. The decrease in motor unit firing rate in sustained contractions was demonstrated by Bigland-

Ritchie, Dawson, Johansson & Lippold (1986). Bigland-Ritchie et al (1979) contributed evidence of failure of muscle membrane and thus slowing and decrease in peak amplitude of motor unit action potential, as shown in figure 6. The slowing of motor unit activation potential rise time and the decrease in amplitude may contribute to the shift towards the lower frequency range of EMG spectrum.

A recent signal thought to be related to force generation in muscle had arouse interest. It is the subject of investigation here reported: acoustic myography (AMG).

1.2 AMG HISTORY

Acoustic myography (AMG) is defined as the study and recording of waveforms from contracting skeletal muscles, which sometimes is referred to as muscle sound or sound myogram. This sound or wave emitted during a contraction was first documented and demonstrated by Francesco Maria Grimaldi (1665) in his book Physicomatheis de lumine. He described in detail the sounds heard by pressing a thumb in his ears, as that of a low rumble character. He attributed this to "the hurrying motion of animal spirits". The subject of AMG stayed as an mere observation until more than 100 years passed, the investigation was taken one step further by Wollaston (1810), who had examined the sound characteristics in a more scientific manner. He experimented by placing his thumbs in his ears and clenched his fists. The sound quality, as he noted is one of low rumble and the loudness increases with increase exertion. These characteristic of acoustic myogram led him to suggest the possible use

of muscle sound as force indicator. To classify the sound he heard scientifically, he artificially simulated the sound by rubbing a pencil across a board with evenly distributed notches; and had a carriage driven across the uniform cobblestones on a London street. He then calculated the frequency of the sound to be about 23Hz.

In 1885, Herroun and Yeo (1885) examined the sound signal recorded during stimulated contractions and compared it with that of voluntary contractions. They demonstrated the characteristic of the sound are the same in voluntary and stimulated contractions, and the muscle sound detectable on elbow flexors was very similar in character with the first heart sound.

After those studies, there were not many interest in investigation of AMG partly due to the lack of adequate experimental tools and contamination from ambient vibrations. It was in 1948, Gordon and Holbourn (1948) studying muscles controlling eyelid and jaw movements, found an increase in AMG frequency with force level. They were the first to suggest AMG as a mechanical counter part of EMG. In addition, they examined the frequency content of AMG signal, which led to the conclusion implying sound due to the asynchrony of motor units and the position of the recording is not important. In the previous sections on EMG and fatigue, it is evident that EMG is a recordable electrical signal, which could indicate electrical activity in the muscle. However, this electrical nature of EMG is susceptible to interference from electrical stimulation pulses, which make EMG a less favoured candidate as force indicator in an FES application. AMG signals, on the other hand, is a mechanical signal

less likely to be contaminated by stimulus artifacts, and had a linear relationship with force during voluntary isometric contractions (Stokes & Cooper 1992).

Further interest in the subject of AMG can be divided into two broad categories: those concern with voluntary and stimulated contractions in human and work to elucidate the nature of AMG production and the construction of working animal models. The literature concerning both will be reviewed in Chapter 2 and 3 separately.

1.3 AIM OF THIS INVESTIGATION

The aim of this series of investigation was to assess the feasibility of using AMG as a force indicator, especially in situations where direct force measurement is not practical and in FES applications as fatigue indicator. Attempt were made to clarify AMG signal characteristics and examine the possible parameters could be deployed to describe the AMG signal.

CHAPTER 2

EXPERIMENT ON HUMAN QUADRICEPS

2.1 INTRODUCTION

Investigations of AMG signals accompanying human muscle contractions have been reported in the literature and can be divided into three main areas: a) experiments with sustained isometric contraction at constant force b) experiments which allow muscle length change c) experiments with contractions producing fatigue.

2.1.1 ISOMETRIC CONTRACTIONS:

THE INTENSITY OF AMG:

Many authors have reported AMG increase with intensity of isometric contraction in many muscles. The relationship between AMG and force is described differently by different authors (table 1). Barry et al (1985) reported of a linear AMG-force relationship in mid range of force level investigated, but a non-linear relationship does exist at lower and higher forces range. However, this report did not indicate the range of percentage maximal voluntary force (MVC) tested. They may well have examined a small force range.

The force range was very well classified by Orizio and co-workers (1989b). They were working on the human biceps brachii muscle with isometric contraction in range of 10% to 100%MVC in steps of 10%. The result shown in figure 7, displays a parabolic relationship between intensity of AMG and force level up to 80%MVC and then a sharp decrease in AMG resulted as force level increases further in short isometric contractions.



Figure 7. Mean integrated AMG signal recorded by a contact sensor during isometric contractions from human biceps brachii. The force is increased in steps of 10% up to maximal voluntary force. (modified from Orizio et al 1989a)

Author	Muscle	Device	%MVC	AMG-F	Yr.
Lammert	BB	Accelerometer	20-100	S-shape	1976
Oster	BB	Microphone	0-50	Linear	1980
Barry	BB	Microphone	0-50	Linear	1985
Stokes	ES	Microphone	10-100	Quadratic	1988
Orizio	BB	Microphone	10-100	Parabolic	1989
Maton	BB	Microphone	10-100	Quadratic	1990
Zwarts	BB	Microphone	20-100	Linear	1991
Rouse	ТВ	Microphone	20-100	Linear	1991
Zhang	QF	Accelerometer	20-100	Linear	1991
Stile	Jaw	Microphone	0-30	nonlinear	1991
Stoke	QF	Microphone	20-100	Liner	1991
Zhang	QF	Accelerometer	20-80	Linear	1992
Stoke	АР	Microphone	0-100	Curve	1992
Smith	QF	Microphone	20-100	Nonlinear	1993
Smith	QF	Microphone	20-100	Linear	1993

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TABLE 1. Literature reports on AMG and force relationship during isometric contractions of human muscles. The abbreviation used were : biceps brachii (BB), Erector spinae (ES), triceps brachii (TB), Adductor pollicis (AP) and quadriceps femoris (QF)

Other non-linear relationship between AMG and force is reported in biceps brachii, erector spinae, adductor pollicis and quadriceps muscles. (table 1).

The most widely reported AMG-force relationship is that of linear nature throughout the entire force range up to MVC. A number of studies

were carried out on the human quadriceps which confirm this relationship (Stokes & Dalton 1991a, Zwarts & Keidel 1991, Zhang, Frank, Rangayyan & Bell 1991 & 1992). It was also observed that this same linear relationship between AMG and force persisted even after fatiguing activity (Stokes & Dalton 1991b; Zwarts & Keidel 1991), as illustrated in figure 8. This same linear relationship between force and AMG was also shown in a single unfused and fused stimulated contractions, as shown by Barry (1990).

The relationship between the AMG and force level is less predictable in a smaller muscle. In the jaw elevator muscles (Stile & Pham 1991), AMG amplitude tended either to increase monotonically with bite force, or to increase to a maximum value and remain nearly constant or decrease with additional increase in force in the range of 0-30%MVC.

THE FREQUENCY CONTENT OF AMG:

The analysis of AMG frequency started very early, with Wollaston (1810) being the first one to estimate AMG frequency to be about 23Hz. A range of peak frequency, mean and median frequencies are reported. The literature is summarised in table 2. The muscle examined were diverse and the force range and the device used in the reports were very different. But mostly, the AMG signal were concentrated in below 40Hz.

Working on the small abductor pollicis brevis and abductor digiti minimi muscle of the hand with electrical stimulation on the median or ulnar nerve, AMG was recorded from evoked twitches (Barry 1991); the sound



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Figure 8. Relationships between force and integrated electromyogram (iEMG), force and acoustic myogram (iAMG) for human quadriceps before (open circle) and after fatigue (closed circles). (modified from Stokes & Dalton 1991)

1st	Muscle	Device	%MVC	Freq.	Yr.
Author			i	(Hz)	
Oster	BB	Microphone	10-50	25±2.5	1980
Rhatigan	BB	Angiograph	0-100	15±4.2	1986
Maton	BB	Microphone	0-100	15,5-22,2	1989
Wee	BB	Microphone	0-20	11,3	1989
Orizio	BB	Microphone	10-100	10-22	1990
Mealing	RF	Microphone	80	7.5-10	1990
Rouse	TB	Microphone	20-100	12-15	1990
Mealing	00	Microphone	MVC	22±5	1991
Mealing	RF	Microphone	80%	10.8±3	1990
Zhang	RF	Accelerometer	20-80	11-19	1992
Dalton	BB	Microphone	0-50	6-14.1*	1993
Dalton	BB	Microphone	0-50	6.9-10**	1993
Dalton	QF	Microphone	10-100	7.1-16.9	1993
Herzog	VL	Accelerometer	70	40±7	1994
Herzog	RF	Accelerometer	70	25±9	1994

TABLE 2. Summary of literature reports on the AMG frequency during contractions of human muscles. The abbreviation used were: biceps brachii (BB), rectus femoris (RF), triceps brachii (TB), orbicularis oris (OO), quadriceps femoris(QF) and vastus lateralis (VL).

was found to be of biphasic nature with two frequency components: a low frequency (<10Hz) and a high frequency (>25Hz) component, as illustrated in figure 9. The two different components of sound are thought to be due to two types of movement of the muscle (See Chapter 3 introduction section on work performed on animals): The large, low



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Figure 9. The evoked acoustic signal from the abductor pollicis brevis muscle after supramaximal stimulation of the median nerve at the wrist (top trace) and the same signal filtered with a 30Hz high-pass filter to eliminate the large, biphasic component of the signal (bottom trace). Arrow indicate the points taken from the latency and rise-time measurements; the bar indicates the amplitude measurement. (modified from Barry 1991)

frequency component due to bulk movement of the muscle and the high frequency component due to the resonant vibration of the muscle (Barry and Cole 1988b). The analogy of the sound produced by ringing a bell was used to explain AMG frequency. The amplitude of low frequency sound is thought to be determined by the contraction force (eg, the bell swings farther when hit harder), and the high frequency of the sound is related to muscle geometry, stiffness and contractile properties, which determine the resonant frequency of the muscle.

Change in AMG signal frequency with force was reported. Orizio, Perini & Veicsteinas (1989c) investigated AMG signal during isometric contractions in biceps brachii muscle. They reported that the peak frequency of AMG increased with increasing force level and the frequency spectrum became bimodal above 30%MVC, furthermore the mean frequency and relative power content increased with increasing force. When compared with known motor unit behaviour in this muscle, they concluded that the AMG frequency can be a non-invasive tool for reflecting the activation patterns of motor units.

2.1.2. DYNAMIC CONTRACTIONS:

The above studies investigated isometric contractions, but many daily activities involve muscle performing dynamic contractions in which muscle change length. This more complex mechanical situation may affect the accompanying AMG signal.

One systematic study on AMG during dynamic contractions was carried out by Dalton and Stokes (1991). They worked with human biceps brachii muscles during submaximal dynamic contractions. It was observed that both the concentric and eccentric contractions produced linear relationship between AMG and force levels. But the slope of the AMGforce regression line is different, with AMG showing greater activity during concentric contractions. The data are displayed in figure 10. Based on result from simultaneous recording of EMG signal from the biceps brachii muscle, and the subsequent analysis on AMG frequency spectrum (Dalton & Stoke 1993), they concluded that AMG could be used as force monitoring signal during dynamic contractions and the signal content is linked to motor unit behaviour.

2.1.3. FATIGUING CONTRACTIONS:

INTENSITY OF AMG: Fatigue had been found to associate with changes in AMG signal amplitude. Among the few systematic experiments Orizio, Perini & Veicsteinas (1989b) recorded AMG from biceps brachii muscle during sustained isometric contractions till exhaustion at four specific force levels: 20%, 40%, 60% and 80%MVC. Results showed a 5 fold increase in AMG for force level of 20%MVC; 4.5 fold decrease at 60 and 80%MVC, while the 40%MVC contractions produced no significant change in AMG intensity, as shown in figure 11. Dalton, Comerford & Stokes (1992) studied the rectus femoris during intermittent fatiguing contractions. The AMG decreases linearly between 75% to 60%MVC, then remains relatively unchanged. If muscle contraction continued, when force falls to 52%MVC the AMG begins to increase once more. This is illustrated in figure 12. The different behaviour of AMG signal were attributed to the different muscle activation strategy.

THE FREQUENCY CONTENT OF AMG: Very few studies have



Figure 10. Integrated acoustic myographic activity (iAMG) at different force levles during concentric (closed circles) and eccentric (open circles) contractions of the biceps brachii muscle in eight normal subjects (mean and standard deviation). (modified from Dalton and Stokes 1991)

Figure 11. Acoustic myogram (iAMG) signal during sustained isometric contractions of human biceps muscles at force levels of 20%, 40%, 60% and 80%MVC. (modified from Orizio et al 1989)



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Figure 12. Acoustic myographic signal (iAMG) changes with progressive force decline during intermittent isometric contractions sustained till fatigue. The initial force was 75%MVC. (modified from Dalton et al 1992)

been carried out on the AMG frequency spectrum during fatiguing contractions. Orizio, Perini & Veicsteinas (1989a,b) reported that the power spectrum density range first enlarges and then reduces during sustained isometric contractions of 20% and 80%MVC in biceps brachii. The reduction was associated with the approaching fatigue. In addition, at lower force levels (20% and 40%MVC) there was no change in mean frequency and relative power content during contraction till exhaustion. In the higher force ranging between 60% and 80%MVC, both mean frequency and relative power content increase at first and then decrease with time, peaking at the first half of the whole contraction time. Mealing et al (1990), also performed frequency analysis on AMG during fatigue of human quadriceps isometric contraction at 80%MVC. They found a wide frequency range with 7.5 and 10Hz peaks. In contrast, Zwarts & Keidel (1991) found that the spectrum of AMG did not change with fatiguing test, but the peak frequency of the spectrum was less pronounced. Again, the authors attributed the AMG frequency change to motor unit activation patterns.

2.1.4. AIM

The aim of this study is to investigate the suitability of AMG as a force monitoring signal in FES applications. This allows 3 immediate aim to be identified:

a) The identification of the optimal position for AMG signal recording from a muscle.

b) The characterisation of the AMG signal and its relationship with force generation.

c) The investigation of possible changes in AMG signal

characteristics as fatigue develops.

2.2 METHODS

2.2.1 SUBJECTS AND COMMON PROCEDURES

Subjects: The human subjects were healthy male and female subjects recruited within the Department of Physiology. They were age between 24 and 39 years. They had no known history of neurological disease or skeletal muscular abnormalities. The nature of the experiments was explained to them and they understood that they could withdraw from the experiment at any stage. Experiments were not performed within three days of the subjects' performing heavy exercise to avoid interference with muscle fatigue. The number of participants in each subset of experiments will be detailed in the following sections on experimental protocols. *Common procedures:* The subjects sat on a stool with the upper part of the body leaning slightly forward and arms folded, while performing isometric contractions of the quadriceps. The hip knee and ankle joints were at about 90 degree. Force generated by quadriceps contractions was recorded with a strain gauge (detail see section 2.2.2 on force recording) mounted on the stool . The muscle sound was recorded with an accelerometer (for detail see section 2.2.2 on AMG recording) held on the surface of the skin on the thigh. This is shown in figure 13. Three maximum voluntary contractions were performed at the beginning and the end of each set of experiments. AMG generated

Figure 13. Photograph of the human experimental setup: The subject sits upright on a stool, with a strain gauge R250 (A) mounted to record force production by knee extension. The AMG signal was recorded by an Entran Accelerometer (B) fixed on skin of subject by double-sided adhesive tape.

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were displayed on an oscilloscope. A line of target force was marked on the oscilloscope, and actual force produced was displayed simultaneously, so the subjects could have some visual feedback on their performance. The level of the target forces were set in accordance with the maximum voluntary contraction on the day of the experiment. The subjects were repeatedly invited to verbalise their perception of the effort of muscle contraction and any sensations as to avoid the onset of muscle fatigue.

2.2.2 SIGNAL RECORDING

Two signals were recorded during each experiment, they are level of force produced in contractions and the accompanying AMG signal. *AMG recording:* AMG signal was measured by means of accelerometer (Entran EGA-F-100 with flange). This accelerometer operates in temperature range of -40 to 120°C, with 2.02mv/g sensitivity. It operates unidirectionally, by means of the strain gauge method. It had a linear response between 10 to 600 Hz as shown in figure 14 top graph. Above 600Hz, the response is non-linear but this lies well beyond the range of AMG frequency. The small size (3mm x 5mm), as shown in figure 14 bottom photo, together with its high sensitivity and good frequency response range, made it more superior than the commercially available Tandy and Hewlett-Packard (HP) piczoelectric microphones (Baxendale & Yao 1992). It's compactness enables secure attachment to be achieved with relative ease and ensure signal was free from mechanical artefact. For example: changes in contact pressure as the tension of cuff holding microphones in place

Figure 14. The frequency response of Entran accelerometer in a range of 0-2750Hz (top graph). The insert is an enlarged section of frequency response between 0-300Hz. The photograph shown an Entran accelerometer fixed on the skin of a subject by double-sided adhesive tape.





(Smith & Stokes 1993) and sounds generated by movement between skin and sensor.

Force recording: Forces produced by isometric contractions were measured by strain gauge (supplied by R.S.Components, UK): RS 250Kg load cell was used in the human experiments. This strain gauge operates within temperature range of -30 to 70° C, when calibrated in compression mode. It has capacity up to 250 Kilogram, with accuracy being 0.05%, as demonstrated in figure 15.

2.2.3 DATA RECORDING AND STORAGE:

The AMG and force signals coming from the transducers was amplified, with AMG signal being filtered between 0.5Hz to >50,000Hz. These analogue signals were converted to digital signals via an intelligent processor (PCM-8) and than recorded on the video track on a videotape for storage, as demonstrated photographically in figure 16 and schematically in figure 17. The equipment specifications are as followed:

a) AC-DC amplifier and Filters;

The amplifiers (Neurolog NL 106 AC-DC amplifier) were manufactured by Digitimer Ltd. These amplifiers allow operation in both AC and DC with DC offset adjustment and gains of up to 100 times. They operate with a low frequency cut-off in AC mode of 2Hz. The high frequency response is beyond 30kHz

The filter employed in the experiments was NL 125-126 filter (Neurolog, Digitimer Ltd.) It has two active filter sections to control the high and low pass characteristics. The low frequency cut-off ranges are:



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Figure 15. Calibration curve of strain gauge (R250) used for force recording during all experiments on human subjects.

Figure 16. Photograph of the equipment used in this series of experiment. The signal from accelerometer and the strain guage could be visualised on an oscilloscope (E), concurrent filtered by Neurolog systems (D) and passed on to the PCM-8 (C) for data channelling and eventual storage. The data could digitised by the 1401 system (B) and analysed by an PC computer (A).





Figure 17. Schematic representation of method used in data acquisition and storage.

DC, 0.5Hz - 5kHz continuously and the high frequency cut-off ranges are wide band (>50kHz) and 5Hz - 50Hz, they are continuously adjustable. The gain within the pass band is 1.

b) PCM-8 A/D VCR recorder adapter

The PCM-8 is an instrument which permits the use of widely available and inexpensive video cassette recorders (VCR) to record up to eight channels of analogue laboratory data. In the experiments described in this thesis only 4 channels recording mode was used. In the four channel mode , the sampling rate was 22 KHz. The converted digital data modulates a video carrier in a format compatible to NTSC video. The video encoded data is then recorded by any VCR through the video input. Data recorded on video tapes in such a way can be played back on a VCR connected to the PCM-8, by decoding the video signal. The original analogue signals are then available on the analogue outputs of the PCM-8 at the same amplitude as recorded, for display or further processing. The PCM-8 records data on the video track of a video tape, there are still two low grade channels available for recording audio signal during experiment, this made the logging of experiment conditions more secure.

2.2.4 SIGNAL PROCESSING AND ANALYSIS:

Data stored on a videotape can be retrieved by play back on a normal VCR via the PCM-8. Signals were digitised via a processor (CED 1401) and then analysed by Spike2 software on a PC computer, see details in figure 18.

a) CED 1401: An intelligent processor (CED 1401, by Cambridge Electronic Design, UK) was used to capture data played



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Figure 18. Schematic representation of method used in data retrieval and processing.

back from the FM tapes via PCM-8 (described above) onto a computer for analysis.

b) Computer and its software: The computer used for data capturing was a Viglen 386PC with software (Spike2 data capturing and processing program) supplied by Cambridge Electronic Design. Spike2 is a software package designed to capture and process both event and wave form data. The data capturing section of the program permits sampling up to 18 data channels, but for the purpose of these experiments only two wave form channels were used. The wave form channels can be sampled at different rates: ie. 512Hz for AMG and 170Hz for force level. The data processing part of the program can analyse Spike2 data files interactively or using user-written scripts. It permits the display of any section of the recorded data in different formats: raw data, frequency contents etc.. In some of the analysis performed on the data a signal average program (Sigavg by CED, Cambridge UK) was used, which can perform on line capturing of data and average the data with user controlled sampling rate, data point number of data repetition for averaging.

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2.2.5 ETHICAL COMMITTEE APPROVAL:

Experiments performed on human subjects involved in this study, had approval from the Ethical Committee of the Western Infirmary, Glasgow.

2.2.6 EXPERIMENTAL PROCEDURES:

a) AMG map of isometric contractions:

Following the aim of this investigation, the first priority was to identify the optimal recording site for AMG signal. Five healthy subjects participated in this



experiment and all recording was collected for analysis. A grid (28 to 34 points depending on the circumference and length of thigh of the subjects) from which AMG recording was going to be made had to be constructed. A line was drawn on each subject's thigh between the top of the patella and the tip of the false pelvis near the hip joint. This line was then divided into four equal portions with perpendicular lines, on which points were marked with 3 cm intervals according to the circumference of the thigh. This grid is represented in figure 19, where the position denoted by negative numbers are on the lateral site of the thigh and the positive number denote the medial sties. Three recordings of AMG signal accompanying isometric contractions at the same given force level were made at these mapping points by moving the accelerometer to each position. AMG mapping were performed as five subsets at force level of 10, 20, 40, 60 and 80% MVC respectively, with each subset of experiment performed as a whole on the same day and at least 5 day interval between subsets. Three contractions of maximal voluntary contraction were performed at the beginning and the end of the experiment from an identical position on the map and fatigue could be detected as a drop in maximal force production. Fatigue was

avoided by 1 minute interval rest between changing positions.

b) Force AMG relationship in isometric contractions: In this experiment, the subjects were requested to perform two sets of contractions of 0 to 100% MVC in steps of 10%MVC: 1) isometric force holding contractions lasting 5 to 10 seconds; 2) isometric triangular force varying contractions with which there were 10 seconds increase in force generation to match the target force level and 10 seconds gradual relaxation back to resting level. At each force level, three contractions were made with at least 10 seconds interval between them. Any contraction with force level sustained less than 5 seconds was rejected and repeated. The AMG signal was recorded at the mid point of the central line between the top of the patella and the hip bone, as there seemed to be no clearly defined optimal position for AMG recording in the AMG mapping test. and the source of the

c) Fatiguing tests with isometric contractions: In this series of experiment, two set of tests were performed. The first set of test was designed to investigate the AMG signal during a continuous maximal voluntary isometric contractions in the quadriceps muscle. The subjects performed three maximal voluntary contractions to familiarise with the experiment. Then, they were instructed to sustain a maximal voluntary contraction till exhaustion or till they were asked to relax (the cut off point was 50% MVC force output for the analysis).

The second set of the test were performed a week after the first test, to avoid interference fatigue effecting the outcome the second test. This test involved subjects performing bursts of MVC till exhaustion or
on reaching the same cut off point as in test one. Each burst of MVC was sustained for 15 seconds, with 15 seconds of relaxation time in between. Care was taken to ensure that during these fatiguing protocols, unlike in brief isometric contractions, the subjects had no visual feed back to their performance. This was introduced to eliminate subconscious recruitment of other muscle groups during a contraction.

2.2.7 ANALYSIS OF SIGNALS

AMG signals: The AMG signal was sampled at rate of 512Hz. This is performed based on the fact that minimum of two points per sinusoid is required by the Fast Fourier Transform (FFT) calculation method, to analyse data. Sampling at rate of 512Hz, will ensure adequate analysis of AMG signal up to 256Hz. AMG has diminishing signal content beyond 60Hz (Goldenberg, Yack, Cerny & Burton 1991, Dalton and Stokes 1993). By using 512 Hz as sampling rate, over 95% of signal content could be obtained and analysed.

The signal was band pass filtered between 0.5Hz and 120Hz and amplified when necessary with Neurolog NL106 and NL125 modules. The signal is subsequently full wave rectified and integrated or subjected to root-mean-square calculation by a computer program. The time constant used was 0.5 seconds. The amplitude of AMG signal was measured during a period of steady state of isometric force, and the values presented was of AMG peak to peak amplitude and that of true AMG signal without the background noise at resting state.

Frequency content of AMG signal was analysed with Spike2 computer software. This program incorporated Fast Fourier Transform (FFT) method of analysis to establish the frequency content. A section of AMG lasting about 0.5 seconds was selected during the steady state of muscle force for analysis. The FFT calculation was performed on 256 points. The median frequency was measured by specifying a frequency range between two frequencies and the program shown the median frequency value to be the frequency beyond and below which the power content is equal in value.

For analysis of sustained isometric contractions, the rectified and integrated, and the root-mean-squared analysis of AMG amplitude were performed by continuous running method and the program selects slices of 0.5 second AMG signal for analysis at defined time intervals, of which median frequency analysis was not permitted by the program.

Force measurements:

The force signal was digitised by CED 1401 and stored in a PC. The minimum resolution of time and force was 8 micro-seconds and 0.0005%MVC, respectively. Forces were measured by cursor and pointers.

2.2.8 STATISTICAL METHODS:

Force, AMG(RI) and AMG (RMS) were normalised to control values for individual subject and subsequently expressed in percentages of maximum. The group means were calculated and standard deviation of data from mean was evaluated. The students t-test was performed in all experimental data. Where lines were fitted to the data, a least square fit method was used within SigmaPlot for Windows. This determined the relation between AMG and force. In addition, rank and sign tests were deployed where appropriate.

2.3 RESULTS

2.3.1 AMG MAP:

Following the aim of this investigation, the first priority was to identify the optimal recording site for AMG signal. A map was constructed, using 28 or 34 points, depending on the circumference and length of thigh of subject. The recording points are presented in figure 19. AMG recordings were made sequentially from these points.

Five male subjects made repeated contractions at 10, 20, 40, 60 and 80%MVC, with accelerometer attached to each position on the map. The results are presented in the following sections:

A>. AMG INTENSITY MAP:

The AMG recorded during a single contraction is shown in figure 20. In this figure, the contraction force was that of 80% MVC, and the contraction lasted for about 10s. There was strong AMG signal associated with movement of the muscle at the beginning and the end of a contraction. AMG signal was only analysed during steady state force, which is normally the middle 5s of data during a contraction. The signals were rectified and integrated to give an "easy" measure of AMG intensity. This method was deployed for subsequent analysis of contractions recorded from each point of the map. The AMG intensity are presented with the same format as in figure 19. A map of one subject at contraction force level of 40%MVC is shown in figure 21, where colour codes for the AMG signal intensities. There were 16 AMG intensity levels presented in figure 21, and each level was multiples of 6.25% of maximal AMG signal intensity recorded at that particular force level. The data was normalised with maximal AMG intensity as 100% and minimal AMG intensity as 0% before contraction period. With this mean of presentation,

Figure 20. Graphical representation of force and AMG data collected during one isometric contraction at 80% maximal voluntary forces.

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Figure 21. A graphic display of data from one subject. The colour represents the corresponding AMG signal intensity on that position, as exhibited in figure 19. These data are taken from a series of contractions at 40%MVC. The position of maximal AMG intensity occurs at p4(-1), indicated by *.

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Figure 22. 3 dimensional representation of AMG signal intensity recorded from different position on left thigh. The data were mean values of AMG in 80%MVC isometric contractions from 5 subjects.



the difference can be maximised. In figure 21, the position with highest AMG intensity recorded coded by the lightest colour. It is at position p4(-1) on the grid. A 3 dimensional graph is presented in figure 22, showing mean data of all subjects performing 80%MVC. There is a general high AMG intensity region at the mid and distal end towards the hip. There is very little AMG at both medial and lateral boundary of the leg.

When data from all six subjects and of all contraction levels were collected, analysed and presented with the same method as before in figure 21, a few characteristics emerges:

a) Concerning individual subjects:

1) Data from individual recording points were processed by calculating root-mean-

square (RMS) and rectify-integrating (RI) values of AMG. There significant were no differences in values obtained by the two calculating methods at the same force. 2) The sites of the maximal recorded AMG signal varied with different degree of exertion up to 80%MVC, as shown in figure 23. ln this



Figure 23. The positions from which the highest AMG intensity were recorded at 10, 20,40, 60 & 80%MVC. The value in the graph represent the force level at time of recording.

particular subject, the highest AMG signal intensity was recorded at

position p2(+2) at 10%MVC; p4(-1) at 20%MVC; p4(-2) at 40%MVC; p2(+2) at 60%MVC and p3(-3) at force level of 80%MVC. The movement of the highest AMG intensity point from one position to another with changing force, does not seemed to follow a fixed pattern in individual subjects tested. Neither does the movement of the site of second highest AMG intensity position follow any deducible pattern in the subject. There were no fixed relationships between the position of the highest and second highest AMG intensity sites within individuals tested.

3) In general, the distinction between sites is less at a lower percentage of the maximal voluntary contraction, and more prominent with higher exertion. This is illustrated in figure 24, where the mapping colours tends to be darker at 10%, 20%MVC, in comparison to those at 40%, 60% and 80%MVC.

4) The intensity of AMG recorded, at each position in the map in each individual tested, displays a general trend of increase in value with increasing force output. Figure 25 demonstrates this point with data from one individual subject, who performed three isometric contractions at 5 force levels up to 80%MVC. The data were collected at positions p1(0), p2(0), p3(0), p4(0) and p5(0) on the constructed map. There are trends of increase in AMG signal intensity with increasing exertion at each position, which is highly representative of all subjects tested. When a linear regression line is fitted to the data, the formula are given as followed: at position p1(0) Y=0.508X+15.026 (r²=0.470); at p2(0) Y=0.378X+20.646 (r²=0.5556); at p3(0) Y=0.536X+19.485 (r²=0.6094); at p4(0) Y=0.568X+23.768 (r²=0.3137); at p5(0) Y=0.501X+26.371 (r²=0.3749). The gradients of the regression lines are all statistically differ significantly from each

Figure 24. Graphical representation of iAMG signal intensity from various sites (up to 35 points) on surface of left thigh during isometric contraction. The left hand side of the data representing the direction of the hip, while the right hand side to the top of patella. Data are of contractions at force levels of 10, 20, 40, 60 and 80% maximal voluntary contractions in ascending order. The intensity of the AMG signal were presented by 16 colour codes, and light colours denote higher intensity.



Figure. 25. AMG signal recorded over five positions along the long axis of quadriceps in one subject. The subject performed isometric contractions at force level of 10%, 20%, 40%. 60% and 80%MVC. The formulae for regression lines are given as followed: at position p1(0) Y=0.508X+15.026 (r²=0.470); at p2(0) Y=0.378X+20.646 (r²=0.5556); at p3(0) Y=0.536X+19.485 (r²=0.6094); at p4(0) Y=0.568X+23.768 (r²=0.3137); at p5(0) Y=0.501X+26.371 (r²=0.3749). The gradients were statistically tested and all are found to be significantly different from zero (p<0.05) and significantly different from each other (p<0.01).

other. The gradient of the regression line is slightly higher at position p4(0) than the rest for this subject.

b) Concerning collective data;

1) Whilst differences can be clearly seen between adjacent recording positions in figure 24. When the data are pooled these differences could be cancelled out. There was no significant difference between the means of AMG signal intensities at neighbouring positions on the map, for a given force level. The data were analysed by both the ranking test and student t-test, as in the case of the individual data.

2) The positions from which the highest intensity were recorded, are shown in figure 26. These positions were not the same in all subjects at any given force level: for example, contractions of 10%MVC yield highest AMG intensity at position p2(2), p3(0), p4(-1), p4(1) and p1(0) in different subjects. This is also true for the second highest AMG recording position. It becomes difficult to assess the general trend in movement of the highest AMG intensity points with changing force: ie. subject 1 in figure 26 shown positions of highest AMG intensity recorded changed from p2(2) at 10%MVC, to p4(-1) at 20%MVC, p4(-2) at 40%MVC, p2(2) at 60%MVC and finally back to p3(-3) at 80%MVC.

3) When all data were pooled together with mean and standard deviation calculated as well as ranks from ranking test, there is no predominant optimal AMG recording site across all subjects. In fact, the inter-subject variations were very large at each recording point for all level of force.

4) Similar to the individual data shown in figure 25, figure 27 displayed the collective data from the longitudinal positions p1(0), p2(0), p3(0), p4(0) and p5(0) of all 5 subjects examined. The first order linear

Figure 26. Diagrammatic representations of positions from which the highest AMG intensities were recorded. Each diagram represents data from one individual subject. The position of the value, eg 10% indicates the highest AMG intensity during contractions at 10%MVC. Other maxima are plotted using the same convention.

Figure 27. AMG signal recorded during isometric contractions, at p1(0), p2(0), p3(0), p4(0) and p5(0) of the map. The number of subjects involved is 5. They all performed isometric contractions at force level of 10%, 20%, 40%, 60% and 80%MVC. The data was that of rectified and integrated AMG signal. The first order linear regression fit to the data are as follows: Y=0.499X+11.584 (r²=0.5575) at p1(0); Y=0.392X+17.025 (r²=0.3373) at p2(0); Y=0.676X+15.471 (r²=0.5475) at p3(0); Y=0.810x+17.377 (r²=0.6155) at p4(0) and Y=0.763X+15.32 (r²=0.6358) at p5(0). The gradients of these regression lines are all statistically significantly different from zero (p<0.05). The gradients are all significantly different (p<0.01).

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regression fit to the data are as follows: Y=0.499X+11.584 (r^2 =0.5575) at p1(0); Y=0.392X+17.025 (r^2 =0.3373) at p2(0); Y=0.676X+15.471 (r^2 =0.5475) at p3(0); Y=0.810X+17.377 (r^2 =0.6155) at p4(0) and Y=0.763X+15.32 (r^2 =0.6358) at p5(0). The gradients of these regression lines are all statistically significantly different from zero (p<0.05). They differ significantly between each other. As in the figure 25, the gradient of regression line at position p4(0) is higher than at p1(0), p2(0), p3(0) and p5(0).

5) The same increase of AMG with increasing exertion are observed at all positions on the map. Examples of data from positions at the lateral and medial sites are displayed in figure 28. Similar to data obtained from positions on the central longitudinal line, AMG intensity recorded from all lateral and medial sites showed increases with increasing force at all positions. Statistical analysis yield regression lines for force and AMG intensity relationships in figure 28: Y=0.568X+12.754 at p4(-4); Y=0.529X+19.182 (r²=0.389) at p4(-3); $(r^2=0.523)$ Y=0.683X+15.674 (r²=0.59) at p4(-2); Y=0.775X+17.666 (r²=0.646) at p4(-1); Y=0.789X+16.304 (r²=0.657) at p4(0); Y=0.644X+17.296 Y=0.704X+16.1 (r²=0.702) $(r^2=0.635)$ at p4(1); at p4(2); Y=0.607X+12.187 (r²=0.658) at p4(3); Y=0.439X+12.651 (r²=0.436) at p4(4). All gradient of lines are significantly different form zero (p < 0.05) and are significantly different from each other. The steepest gradient remained at p4(0).

Formula for the regression lines of force-AMG relationship at all positions were tabulated in Table 3. All gradients were found to be significantly different from zero (p<0.05) and between each other. The gradient of AMG-force relationship is steepest at position p4(0) and

Figure 28. AMG signal recorded, over seven positions on the transverse direction of position 4(0), on human quadriceps muscle from five subjects (n=5). The subjects were performing isometric contractions at 10%, 20%, 40%, 60% and 80% maximal voluntary contractions. The AMG signal were rectified and integrated. The regression lines have formulae: Y=0.568X+12.754 (r²=0.523) at p4(-4); Y=0.529X+19.182 (r²=0.389) at p4(-3); Y=0.683X+15.674 (r²=0.59) at p4(-2); Y=0.775X+17.666 (r²=0.646) at p4(-1); Y=0.789X+16.304 (r²=0.657) at p4(0); Y=0.644X+17.296 (r²=0.635) at p4(1); Y=0.704X+16.1 (r²=0.702) at p4(2); Y=0.607X+12.187 (r²=0.658) at p4(3); Y=0.439X+12.651 (r²=0.436) at p4(4). All gradient of lines are significantly different form zero (p<0.05) and are significantly different form zero (p<0.05).

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Table 3. The formula of first order regression lines fitted to iAMG signal intensity and force data of all subjects (n=5), while performing 10%, 20%, 40%, 60% and 80% maximal voluntary contractions. They were statistically tested and no significant difference existed between gradients of regression lines of adjacent positions.

	u			y = 0.499x + 11.58				
	y = 0.587x + 22.07	y = 0.548x + 28.93	y = 0.569x + 23.67	y = 0.572x + 21.26	y = 0.594x + 23.46	y = 0.744x + 21.69	y = 0.704x + 20.54	
y = 0.52x + 16.766	y = 0.674x + 15.77	y = 0.567x + 19.75	y = 0.69x + 17.86	y = 0.725x + 15.32	y = 0.707x + 16.39	y = 0.633x + 17.63	y = 0,434x + 14.71	y = 0.274x + 15.96
y = 0.568x + 12.75	y = 0.529x + 19.18	y = 0.683x + 15.67	y = 0.775x + 17.67	y = 0.789x + 16.3	y = 0.644x + 17.30	y = 0.704x + 16.1	y = 0.607x + 12.19	y = 0.439x + 12.65
				y = 0.763x + 15.32				
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seemed to adhere to a trend of increase along the longitudinal direction, with the lowest value recorded from top of the patella, where the intercept of line on axis was minimum. At the transverse line from position 2(0), both gradient of regression lines and the intercept on axis increases laterally and medially away from the central longitudinal line. Unlike recordings made from 2(0) transverse line on the map, signals recorded from transverse line 3(0) displays a different trend: while the intercept of the regression lines increases at positions laterally and medially away from position 3(0), the gradients of the lines decreases. The link between gradient, intercept of formula and that of position in the map changes again at transverse line from 4(0) on the map. At positions laterally and medially away from position 4(0), both gradient and intercept of the formula decreased.

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B>. AMG FREQUENCY MAP:

Frequency content of AMG signal accompanying isometric contractions is analysed by Fast Fourier Transform (FFT) method. Figure 29 illustrates the frequency content of AMG signal from one contraction of 20%MVC at p3(0). The AMG has been bandpass filtered between 0.5 and 120Hz, to minimise artefact. The 50Hz peak could be an artifact from the power source. The greater part of the signal lies between 5 to 35 Hz, with very little content beyond 60Hz. This particular spectrum contains two peaks at about 8 and 28 Hz.

The frequency content of AMG signal recorded during repeated contractions at 80%MVC from each position on the map, are illustrated in figure 30. Each spectrum shows power frequency contents in range of 2 to 60Hz. The power content in those frequency spectra are in agreement with the AMG intensity map from the same subject at similar force levels, a state of the second second

Figure 29. Power spectrum of a AMG signal accompanying one contraction. The power content of signal was concentrated in two ranges: between 5-15Hz and from 20-35Hz. There are very little power beyond 40Hz. The big peak at 50Hz is believed to be interference from power supply.

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Figure 30. Frequency spectrum of AMG signal recorded during mapping test of an individual subject (DM), while performing isometric contraction at level of 80% maximal voluntary contraction. The analysis was performed with FFT size being 256 and the maximum of the Y-axis is 0.00041.

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as shown in figure 24. As illustrated in figure 29, no AMG signals contained significant power above 60Hz for any given position and at any force. AMG power frequency content, as seen in figure 30, is greater at the mid region of the thigh than other positions. In addition, the signal recorded from these regions showed multiple peaks in the spectrum more than any other position and the peaks seemed to be in the range of 5-45Hz. These characteristics persist at all force level tested. There were dominant low frequency signals at the patella and near patella sites, ranging from 2 to 15Hz, peaking at about 8 to 12Hz. Again these characteristics did not vary much with changing force. The power spectra of AMG signals recorded from same positions were similar with repeated trials.

Some authors calculated a single median frequency rather than examine the whole frequency spectrum (Herzog, Zhang, Vaz, Guimarares & Janssen 1994, Mealing & McCarthy 1991). The median frequencies of the signal power spectrum were analysed and calculations made with data from all subjects at all positions. An example of results is shown in figure 31. The median frequencies of AMG have less variances at position p4(0)at five level of forces tested. The relationships between AMG median frequency and force could be fitted well with first order regression lines at all recording positions. However, statistical analysis yields insignificant difference between regression line gradients and zero (p>0.1). Thus, the AMG median frequency did not change significantly with varying force at all force levels tested.

2.3.2 FORCE-AMG RELATIONSHIP:

The results from section 2.3.1 showed that there is no single

Figure 31. The median frequency of AMG signal recorded, over five positions on the longitudinal direction at 1(0), 2(0), 3(0), 4(0) and 5(0) of map of the left thighs (n=5). The subjects performed isometric contractions at force level of 10%, 20%, 40%, 60% and 80%MVC. The intensity was that of rectified and integrated AMG signal. The change in AMG median frequencies with force are insignificant (p>0.1) at all five positions.

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optimum recording site of the quadriceps AMG, but the signal intensity recorded from the middle of the thigh were generally more intense. Consequently, position 3(0) of the map was chosen for all subsequent experiments.

The relationship between AMG and force production during isometric contractions was investigated during both steady isometric contraction and during which the force was ramped up and down at a constant rate. The results are presented as follows:

A>. ISOMETRIC CONSTANT FORCE CONTRACTIONS:

Figure 32 illustrates AMG data recorded during isometric constant force contractions at different force levels taken from one subject. Figure 32a was recorded over a relaxed muscle. It shows some low intensity vibration, which may represent some basic muscle tone or sounds from other sources. Figure 32b to 32h shows signals recorded from active muscle with increase force output. There were characteristic high amplitude bursts associated with knee extension movements at the beginning and the end of the contractions. The signals were more consistent after the first second of the contraction and the analysis were only carried out on signals during mid-contraction. The intensities of the AMG signal accompanying contractions observably increase in amplitude as progressively higher forces are reached.

The relationship between RI and RMS AMG intensity and force is displayed in figure 33, which shows AMG collected from one subject during a series of isometric contractions held at progressively greater forces up to MVC. The AMG were rectified and integrated with a time constant of one seconds or the root mean square AMG calculated. RI AMG (as shown in Figure 33A) and RMS AMG (as shown in figure 33B) Figure 32. AMG recorded from quadriceps of a normal subject during brief isometric force holding contractions with the shows the signal recorded from a resting muscle; a) resting b) 10%, c) 40%, d) 60%, e) 70%, f) 80%, g) 90% and h) 100% knee joint at 90°. The subject used visual feedback to regulate the force achieved at the requested target force. Trace a) of maximal voluntary contraction. Subjects were allowed 10-15 sec. rest in between contractions.

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Figure 33. The relationship between AMG recorded from mid point of quadriceps and force. The AMG was recorded from one subject (BM), who made a series of contractions each lasting 5-10 seconds at different force levels between 10 to 100%MVC. The data points plotted show full wave rectified AMG integrated with a time constant of 1 second (graph A) and root mean square AMG (graph B). Data could be fitted by straight lines with similar coefficients: Y = 0.7186X + 27.0758 (r²= 0.859) for graph A; Y = 0.7328X + 29.7253 (r²=0.815) for graph B. Both gradients are significantly different from zero (p<0.01) and different from each other (p<0.01).



are plotted against force and it clearly demonstrates a linear positive relationship (p<0.001) exists between AMG intensity and force. The data could easily be fitted with the formulae

Y=0.7186X+27.0758 (for RI data)

and Y=0.7328X+29.7253 (for RMS data)

for a single straight line which had similar coefficients (r^2 = 0.859 for RI data and r^2 =0.815 for RMS data respectively). There was significant difference between the result obtained by RI method and those by RMS methods (p<0.01).

When the data collected from six subjects were pooled and plotted against force in figure 34, the data could be fitted by lines with formula of

Y=0.6684X+14.8648, r²=0.680 for RI data;

and Y=0.7223X+14.1024, r²=0.716 for RMS data

and the slope and the coefficients of the two line fit formula were very similar, but the gradients of the lines are significantly different (p<0.01).

B>. ISOMETRIC TRIANGULAR FORCE VARYING CONTRACTIONS:

The subjects were also instructed to perform isometric triangular force varying contractions up to a peak target force. They reached peak force about 10 seconds after the beginning of the contraction and then reduced the force to zero in another 10 seconds. Samples of AMG signals, taken from one subject, accompanying the force varying contractions to different peak force levels are displayed in figure 35. As in figure 32a, there was some low intensity vibration existed when muscle was at rest. There were smaller and less prominent initial bursts of AMG activity associated with posture adjustment before and after contractions than in isometric hold contractions. The "envelope" of the raw AMG does not

Figure 34. The relationship between AMG recorded and force, in recordings made from six normal subjects, each of who made repeated isometric contractions at up to 10 force levels. A. shows rectified integrated AMG and B. shows root-mean-square AMG. Data could be fitted by straight lines with formulae: Y = 0.6684X + 14.8648 (r²=0.680) for graph A and Y = 0.7223X + 14.1024 (r²=0.716) for graph B. Both gradients are significantly different from zero (p<0.01).



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generated a linear force ramp up to a specified force and then down to zero. The upward arrows indicate the beginning of peak force achieved in the other ramps are: a) resting b) 10%, c) 40%, d) 60%, e) 70%, f) 80%, g) 90% and h) 100% of the contraction. The downward arrows indicate the end. Trace A shows the record obtained from a resting muscle. The Figure 35. AMG signal recorded from quadriceps of a normal subject during isometric contractions, in which the subject maximal voluntary contraction. Subjects were instructed to have 10-15 seconds rest between contractions.

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seem closely related to the force profile at the lower force range (figure 35b-e) up to 70%MVC. However, at forces above 70%MVC there is a marked increase in AMG amplitude as the force reaches its peak. The intensity of the signal increases with increase force up to the target level and then gradually declined with relaxation.

The force level measured was that of the peak force developed and AMG signals during isometric triangular force varying contractions were analysed both by RI method and RMS method. The result is graphically displayed in figure 36, which is a plot of RI (graph A) and RMS AMG (graph B) against the peak force achieved for each force varying contraction in one subject. As shown in this figure, the AMG signal increase with peak force output as in isometric hold contractions, there were no significant difference between results obtained from both of the analysis methods. The data could be fitted with straight lines of formulae:

Y=0.90263X+10.9722, r²=0.927 for RI data

and Y=0.8535X+12.8297, r²=0.947 for RMS data

with similar coefficients. In this case the analysis has revealed changes in the AMG in the lower force range, which was not obvious in the signal envelope, as in figure 35. Statistical analysis showed that the gradients are significantly different from zero (p<0.01)

AMG data for all six subjects were collected and analysed by both RI and RMS methods. The results were then pooled and RMS and RI AMG were plotted against peak force in the triangular force varying contractions in figure 37. As in figure 34, the AMG increase in amplitude with increasing force output. The data were fitted with single straight line of the formula:

Y=0.62376X+15.2768, r²=0.682 for RI data;

Figure 36. The relationship between AMG and peak quadriceps force. The AMG was recorded from one subject (BM), who made a series of isometric triangular force varying contractions, each lasting 5-10 seconds, to different peak force levels. The data points plotted show full wave rectified AMG integrated with a time constant of 1 second (Graph A) and root mean square AMG (graph B). The regression lines fitted to the data have formulae: Y=0.90263X+10.9722, r²=0.927 for graph A and Y=0.8535X+12.8297, r²=0.947 for graph B. The gradients of the lines are significantly different from zero (p<0.01). The data were analysed from traces presented in figure 35.



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Figure 37. The relationship between AMG and peak force. The AMG signal was recorded from six subjects during isometric triangular force contractions, each lasting 5-10 seconds, to different peak force levels. The data points plotted show full wave rectified AMG integrated with a time constant of 1 second (graph A) and root mean square AMG (graph B). The regression lines to the data have formulae: Y=0.62376X+15.2768, $r^2=0.682$ for graph A and Y=0.6285X+13.4385, $r^2=0.683$ for graph B. The gradients of the lines are significantly different from zero (p<0.001).



FORCE (%MVC)

and Y=0.6285X+13.4385, r^2 =0.683 for the RMS data with similar coefficients. Again, there is not much difference in the results obtained by the two different method of analysis, though the pooled data fitted less well than that from single subjects.

The triangular-force-varying contractions were of two phases: the contracting phase and the relaxing phase. Data from a single triangular-force-varying contractions were analysed and the relationship between RI AMG and force production calculated. AMG signal during one contraction up to 100%MVC peak force is graphically displayed in figure 38. The figure showed data from both the contracting (closed circles) and relaxing (open circles) phases. There are trends of proportional change in AMG intensity with changing force output. The fitted regression lines to the data have formulae: Y=0.57578X+ 29.2888 (r²=0.6915) for the force rising phase; and Y=0.45629X+28.1202 (r²=0.63954). The gradients of the regression lines are significantly different from zero (p<0.01). And the one from contracting phase is steeper than that of relaxing phase (0.57578 vs. 0.45629) with statistical test value p<0.01.

The data from all subjects and all contraction levels tested, were analysed, collected and displayed together in figure 39 (force range between 10%MVC to 50%MVC) and figure 40 (force range between 60%MVC and 100%MVC). When linear regression lines were fitted to the data, they demonstrated consistently higher gradients of AMG-force relationship existed during contracting phase than that of the force relaxing phase (p<0.01). The difference persists in contractions of all peak-force level tested, as shown in figure 41. The rate of AMG change with force during contracting phase did not change with peak force in a way that is significantly different from that during the relaxing phase

Y=0.57578X+29.2888 (r²=0.6915) for the force rising phase; and Y=0.45629X+28.1202 (r²=0.63954). The gradients of Figure 38. The relationship between AMG and quadriceps force during one isometric triangular force contractions up to the regression lines are significantly different from zero (p<0.01) and the gradients difference between contracting phase 100%MVC. Filled symbols show integrated AMG changes with force during contracting phase and open symbols show AMG intensity changes with force level during force relaxing phase. The fitted regression lines to the data have formulae: and relaxing phase is significantly different from each other (p<0.01).



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Figure 39. Graphic display of data from all four subjects collected during isometric triangular force contractions, as shown in figure 36, up to target peak force levels of 10, 20, 30, 40 and 50%MVC. The fitted lines were of first order regression.



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Force (%MVC)

Figure 40. Graphic display of data from all four subjects collected during isometric triangular force contractions of quadriceps up to specific target peak forces, as shown in figure 36, of 60, 70, 80, 90 and 100%MVC. The fitted lines were of first order regression.

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(p>0.05).

C>. FREQUENCY

The frequency spectra of AMG signal in isometric holding contractions were compared with that of isometric force varying contractions performed by the same subject. In both conditions, the power spectrum of AMG signals shown multiple peak frequencies at all force level tested. Data from maximal contractions are shown in figure 42. The AMG spectrum is simpler during steady force contractions (figure 42A) than during triangular contractions (figure 42B) where multiple peaks appears. There was no significant difference between the rate of AMG median frequencies change with force during constant force and force varying contractions.

2.3.3 FATIGUING CONTRACTIONS:

Fatigue is defined as the inability to maintain forces. This is evident in figure 43 and 44 for sustained isometric contraction near maximal voluntary contraction, and both figure 45 and 46 for intermittent contractions till fatigue. In figure 43, the force was well maintained in the initial 35 seconds (125-160 in recording time) and there was steady AMG signal associated. After this initial phase, the force progressively declines, starting at about 175s. However, the AMG signal shown an earlier decline than force at about 160s. During the last phase of contraction between 190 to 220 seconds, there are additional efforts in the force record, which is clearly shown in figure 44c. With each additional effort, there is a burst of AMG activity. There is an overshoot of force over the recording range in figure 44a.

The muscles tested were able to continue force production for a

Figure 41. Gradients of regression lines fitted AMG-force relationship in contracting (graph A) and relaxing phases (graph B), during isometric force varying contractions. The contractions reaches peak force between 10% to 100%MVC. Regression lines of the data have formulae: $Y=1.2*10^{-5}X+2.22*10^{-3}$ (r²=0.0134)) for the force rising phase and $Y=9.6*10^{-6}X+1.22*10^{-3}$ (r²=0.005). There is no significant change in the slopes of both phases with increasing peak force level (p>0.1)



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Figure 42. Power spectrum of AMG signal recorded, from same subject, during isometric force holding (graph A) and isometric force varying contraction (graph B). The contraction level was 100%MVC.


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maximal effort. The end point was reached when the subject could no longer sustain force production above 50% of Figure 43. Force and AMG data of an individual subject, during a sustained isometric contractions till fatigue with the original maximal voluntary contraction or was not able to continue the test (see text for detail) i

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Figure 44. Sections of 10s data taken from traces presented in figure 43 at about time 0s (trace a), 178s (trace b), 209s (trace c) during one sustained contraction till fatigue with maximal effort. The end point was reached when the subject could no longer sustain force production above 50%MVC.



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Figure 45. Force and AMG signals recorded during intermittent isometric contractions till fatigue. The contractions were of 15 seconds duration and there were 15 seconds of rest in between contractions. The test was terminated when subjects could no longer sustain force production above 50% of the initial force or was not able to continue the test.



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Figure 46. Single contractions extracted from traces presented in figure 45 at about time 0s (trace a), 490s (trace b) and 910s (trace c). The top traces are force records. These declines progressively with successive contractions. The bottom traces show associated changes in AMG signal evelopes.



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longer time by intermittent isometric contractions than sustained isometric contraction. This is shown in figure 45, where force production was continued up to 900 seconds. The decline in force was almost immediate in figure 45 and there is visible decrease in AMG amplitude in the middle of contraction from this subject. Similar to the sustained contraction till fatigue, there are additional efforts, which reduces with progressive fatigue. There are also bursts of AMG activity with these additional efforts. Figure 46 displays traces of force and AMG records form contractions at the beginning, middle and end of the test period. This figure show greater declined in force and the accompanying decline in AMG during the first half of the test and slower rate of decline in the second half. The force recorded during each contraction declines with time. In addition, there are ripples on the force record. These ripples have an initial high frequency and amplitude and then slow and diminishes with each successive contraction.

ANALYSED DATA OF FATIGUING TEST BY SUSTAINED CONTRACTIONS:

Collective data of five subjects from sustained isometric MVC contractions till fatigue is displayed in figure 47. All force level were normalised with the initial force as 100%MVC. The force shown different degree of potentiation in different subject at the initial phase. The duration of the potentiation period is different between subjects. The force then declines with time, with different rates in individual subject. The time course of fatigue were also different in different subject: the shortest time to reach fatigue stage was about 80 seconds, while the longest time was about 200 seconds. It was noted that the rate of force decline was greater

Figure 47. Graphical representation of changes in force (circles), AMG intensity (squares) with time, during sustained isometric contraction of maximal effort till fatigue. These are data from 5 individual subjects. Data were normalised to the initial values of AMG intensity (rectified and integrated) and force levels and were analysed by continuous running average with 1 second time constant.



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in subjects with the shortest fatiguing time course with an higher initial absolute force level (AMH); and the reverse is true for subject with the lowest initial force output (PR).

The AMG signal intensity recorded are normalised to the initial value of AMG as 100%. The AMG change with time follows different trend in different subject: some shown a clear trend of decline (graph AMH, DM and PR); while others had data with huge scatter.

Data were then analyzed, shown as ratio of AMG-force in 48. A single regression line could be fitted to the data. The data could be fitted by a line with formula Y= $8.9*10^{-4}X+0.904$ (r²= $3.7*10^{-3}$). The slope of this line is not significantly different from zero. Figure 48 displays AMG-force ratio increasing at a fast rate after 170s. These data comes from only one individual with very scattered data points.

ANALYSED DATA OF INTERMITTENT FATIGUING TEST:

The data of five subjects, were analysed for force level and AMG intensity during intermittent form of fatiguing test. The initial force was the maximal effort. Data from individual are displayed in figure 49. The data were normalised with the initial value as 100% and fitted with third order of regression lines.

During intermittent fatiguing test, force decreased with progressive fatigue in each subjects under investigation, even though the time taken to fatigue an subject is not the same: some reached the end points at about 800 seconds in the protocol with fast rate of force decline, while other could carried on performing till 2500 seconds into the test and maintain relatively higher force level at the end point. In all cases, there were no force potentiation during intermittent isometric contractions, unlike during
Figure 48. Summary of AMG-force ratio during sustained contraction till fatigue in 5 subjects. The initial force was the maximal effort from the subjects. The data could be fitted by a line of formula Y=8.9*10⁻⁴X+0.904 (r²=3.7*10⁻³). The slope of this line is not significantly different from zero (p>0.05).





Figure 49. Five sets of individual data showing changes in force (circle) and AMG intensity (square) with time, during intermittent isometric contraction till fatigue. The initial force was the maximal effort each subject could produce. Data were normalised to the initial values of iAMG intensity and force levels.

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sustained isometric contractions. The forces recorded from all subjects declined progressively with time. However, the rate of force declines were varied, with subjects having the highest absolute force had fastest rate of force decline.

The AMG intensity recorded shown inter-subjects variation: some shown clear trend of steady increase with time, some shown a decreasing trend and others had relatively little change with time.

The degree of AMG association with force is shown in figure 50, as the ratio of AMG to force. There was less scatter in the data at the initial stage of test, but the degree of data scatter increases with each successive contraction. This graph demonstrated tendency of increasing AMG-force ratio with progressive contractions. The data could be fitted by a line of formula $Y=6.56*10^{-4}X+0.9665$ (r²=0.4179). The slope of this line is significantly different from zero.

FREQUENCY CONTENT OF AMG SIGNAL:

The 128 points Fast Fourier Transform analysis of 5 seconds data, yield predominant peak frequencies of AMG signal between 6 to 12Hz, with most peaks landing on 10Hz in sustained 100%MVC contraction till fatigue, as shown in figure 51. There was very little power in frequency spectrum beyond 25Hz. In this figure there is little evidence of change in the peak frequency of AMG spectra with progressive fatigue. However, there were changes in the total AMG signal power content with time. The total power diminishes in AMG recorded from this individual subject. Concurrent observation shown that there were ripples on force records at about 10Hz frequency and this phenomena persisted throughout the whole test.

Figure 50. Summary AMG-force ratio change with time in five subjects. The subjects performed intermittent isometric contractions till fatigue. The initial force was a maximal effort. The data could be fitted by a line of formula Y=6.56*10⁻ $^{4}X+0.9665$ (r²=0.4179). The slope of this line is significantly different from zero (p<0.001).



AMG - Force ratio

Figure 51. Frequency spectra of AMG signal intensity during 5 second episodes at different stages of the fatiguing test from one subject. The subject was fatigued by sustaining maximal effort. Data were taken at 2, 22, 42, 62 and 82 seconds into the test. It ended at time 85 seconds.



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The frequency spectra of AMG signal produced with intermittent form of fatiguing test, example from one subjects is displayed in figure 52. In this figure there were very distinct narrow peaks between 8 and 12 Hz, which changed in amplitude with progressive fatigue. There were other less well defined and broad band frequency peaks between 18-35Hz, with less power content per bin, which were not so apparent in data from sustained contractions.

The median frequencies of AMG signals were analysed for both form of fatiguing test, and the results displayed in figure 53. During the sustained contraction till fatigue, there is no common trend in AMG median frequency change with time: some increased and others decreased. This is equally true for intermittent contractions till fatigue. うちの、町をかかいからないたいを見るという

Figure 52. Frequency spectra of AMG signals recorded from one subject. The subject was exerting intermittent maximal effort till fatigue. The analyzed bursts of AMG were taken from first, 10th, 20th, 30th and 40th contractions. The end point of fatigue was reached when force declined to 50%MVC or when subjects no longer able to produce contractions.



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Figure 53. Graphical display of median frequencies of AMG signal changes with time during sustained contraction and intermittent contraction till fatigue. The initial force was from an maximal effort. Five subjects took part in both of tests.



2.4 DISCUSSION

It was the original aim of this study to identify the optimal positions or conditions for recording AMG; investigate AMG characteristics and its relation to force of contraction; assess the possibility of AMG as an indicator of muscle fatigue. The results obtained, from this series of human study, shown that there is a good correlation between AMG signal intensity and force generation in isometric force holding and force varying contractions. AMG signal intensity was found to be proportional to force within any force varying isometric contraction. AMG was found to be tightly associated with force production during sustained isometric contractions till fatigue of near maximal voluntary contraction force. However, the close association between AMG and force level was not observed in the intermittent isometric contractions till fatigue. The AMG signal frequency was complicated and the median frequency of AMG was not associated with force production.

2.4.1 MATERIAL AND METHODOLOGY:

The Entran accelerometer was chosen due to the relatively small size and weight of the device and its good frequency response range. In addition its sensitivity is 400 times greater in one direction than others. Thus, it is not affected by the stretch on the skin at the recording site. This recording device can be easily attached on to the skin surface with double sided adhesive tape and is not sensitive to the method of attachment and the pressure exerted on it, in comparison with other microphones available (Baxendale and Yao 1991, Smith & Stokes 1993). It is not sensitive to air-bound sounds.

Quadriceps muscle were chosen for AMG investigation in these

experiments for the following reasons: 1) the relative large muscle mass and surface offered many recording sites. 2) the radius of curvature of the muscle changes relatively little during contractions and so there are fewer problems with displacement of the accelerometer. 3) quadriceps is used during functional electrical stimulation to aid walking and standing in handicapped humans.

The voluntary force recorded from quadriceps is the summation of forces generated in the whole of the quadriceps group: rectus femoris, vastus medialis, vastus lateralis and vastus intermedius muscle. This group of muscles contract and bring about the extension of knee. In addition, the rectus femoris flexes the hip. The force recorded is the sum of the whole muscle group but the AMG signal may be that of the muscle immediately under the recording site. However, there is evidence that rectus femoris is the major force contributor in extension between 90° to 140° (Basmajian and De Luca 1985). The joint angle was fixed at 90° in this study. Thus, rectus femoris may be the major force contributor and the AMG recorded in association with contraction was dominated by the sound coming from rectus femoris. Furthermore, the fixed angle of knee joint and the standardised posture should eliminate changes in muscle activation strategy.

The experimental set-up used to record forces in this experiment does permit certain amount of movement at the start and end of the contractions. This is evident in the rather large AMG signal at the start and end of each contraction, as seen in figure 32 and 35. But the artifacts in the AMG signal were more prominent in isometric holding contractions, where AMG and force signal being analysed were those of steady state phase. Thus, the artifact interfered very little with result obtained. It seemed rather possible that the "jerking" signal at both end of contraction was the result of muscle tendon taking up tension from a slack state and vibrating like a piece of rubber band being stretched. The speed of that stretching determined the amplitude of the vibrating signal. This theory could also explain the smaller artifacts observed in contractions with slow rate of force change.

In this study, AMG signals were analysed for their frequency content by the Fast Fourier Transform method (FFT). This gives a detailed representation of signal in terms of the frequencies contained and the power associated with each frequency (Diemont, Figini, Orizio, Perini & Veicsterinas 1991). However, there are drawbacks of FFT method. The most significant is the assumption of a constant signal frequency content during the transform interval (Wood, Buda & Barry 1992). This can be minimised by keeping the data samples short.

2.4.2 RECORDING SITES:

As stated in the introduction, the quality and possibly the quantity of the sound produced by muscle contraction can vary with acoustic sensor employed. Therefore the discrepancy in reported AMG-force relationships obtained by different groups of workers (see Table I), may be due to the variety of recording devices used. One possibility is that the location of the source of sound produced may be changing with time or degree of exertion. Consequently, the first priority, in this study was to find the optimal site for recording AMG.

Data from this study shown that 1) There are best sites for recording optimal AMG signals at any given force in different subjects. 2) The best recording site changes with varied exertion, and with no visible trend in individual subjects. 3) The best recording site differ between different subjects and different degree of exertion. This is summarised in figure 26. The movement of the optimal AMG signal intensity position did not follow a specific pattern. The inter-subject variations were great. So it appears there is no single common optimal site for AMG recording in any individual studied. It thus become impractical to find on common position best for recording AMG signal, if AMG intensity is the parameter of consideration in an investigation.

There was very little AMG signal at the boundary of the muscle, which is to be expected as the AMG signal recorded was the summation of those from all muscle fibres activated and there are fewer muscle fibres near the muscle boundary. The signal from other muscle fibres in the centre would be weakened on reaching the boundary. The location of higher AMG intensity tends to lie within the upper and lateral region of the thigh. This trend of higher AMG in lateral site of rectus femoris is consistent with the anatomical arrangement of the quadriceps, with the sartorius muscle covering proximal medial portion of Rectus Femoris. Although rectus femoris is the major force contributor of quadriceps group at knee joint angle of 90 degree (Basmajian & De Luca 1985), the more superficial location of sartorius muscle enable its AMG signal to transmit to the surface of skin through less boundaries, with less dampening and higher intensity. The relatively low AMG intensity at the distal end of rectus femoris is also consistent with the anatomical arrangements, where the fibre ends and inserts into the muscle tendon with higher stiffness and lower vibration amplitude. This notion is supported by frequency content analysis on maps of the thigh at different force levels, as seen in figure 30, demonstrating the low AMG power at distal end of the thigh with a dominant frequency at about 6 to 12Hz. It is noted that the AMG intensity and peak frequency were lower at the site of patella. This is consistent with the theory of AMG being vibration of the whole muscle in the transverse direction and the patella tendon will experience more force in longitudinal direction as muscle contracts than transverse direction.

The power spectra at the lateral sites agree well with AMG intensity recorded and the frequency power spectral range increased up to 60Hz with multiple peaks varying between sites and contraction levels. The increase in frequency range and the number of peaks are in good agreement with the present knowledge of muscle motor unit behaviour: recruitment and decruitment of different types of motor unit. Each has different contractile strength and response to different frequencies of neuronal input. This motor behaviour together with motor unit territory could account at least in part, for the varied AMG frequencies content at different sites.

This finding reinforce result of preliminary study of Zhang et al (1991). They investigated the AMG signal from quadriceps, during isometric contractions and showed the dependence of AMG power, frequency content on different positions. However, they only worked with four positions of recording on the rectus femoris and the report was not detailed, which made further comparison difficult, even though they employed accelerometer as the case in this investigation. The other report on AMG signals intensity recorded over different positions on a muscle were Wee & Ashley (1991). They reported AMG signal at the point of stimulation was double that of a distant site in human biceps and triceps. Their results differ from the present finding. The difference may be in

three area: 1) the recording device: uses of an accelerometer in this study, as opposed to piezoelectric transducer taped to the skin. The tape exerting forces on transducer could influence signal intensity, especially with large change in muscle radius. 2) the types of contractions: voluntary contractions verses electrical stimulation, which capable of synchronising muscle fibre activities. 3) different muscle studied: the biceps is smaller compared with quadriceps and the change in muscle radius would be different between the two muscles. Recruitment strategy and threshold force for motor unit activation will differ between these two muscles and contribute to discrepancy of results between studies.

Based on data presented in figure 26, 27 and 28, general high AMG signal were recorded from the mid-proximal region of the left thigh. In addition, the change in AMG amplitude with force was linear at these positions and have higher rate of changing AMG with force in this region (table 3). The presence of a high AMG intensity region at the mid and lateral sites is consistent with the theory that AMG represents the mechanical event of muscle contraction. With majority of the fibres bundled together in the muscle belly and the fact that highest vibration intensity signal would be theoretically midway between the two poles in an vibrating string model, AMG would be highest at the mid point of the muscle. Furthermore, if AMG signal is spreading out like a wave from the active fibres to the surface of the thigh, a good position for detecting and recording those signals with higher amplitude should be around the middle of the muscle. This is confirmed in figure 24, where AMG intensity recorded from position 3(0) is high in all force level tested. Position p3(0)thus became the convenient site for subsequent AMG recording.

2.4.3 AMG-FORCE RELATIONSHIP DURING ISOMETRIC CONTRACTIONS:

Data obtained during isometric force holding and varying contractions, show a statistical significant linear relationship between force production and the accompanying AMG signal processed by both RI and RMS methods. This linearity existed in whole range of force tested between 0 and 100%MVC, as shown in figure 33, 34, 36 and 37. Furthermore, the linearity was found to exist in data recorded from different positions on the surface of the thigh, as displayed in figure 27, 28 and table 3. The same relationship was also found to exist within one force varying isometric contraction.

The linearity between force and AMG signal intensity, is in agreement with the previously reports on the human quadriceps (Stokes & Dalton 1991, Smith & Stokes 1993, Zhang et al. 1991), on adductor pollicis muscle (Stokes & Cooper 1992), biceps brachii muscle (Oster & Jaffe 1980; Barry 1985; Zwarts & Keidel 1991) and tricep brachii (Rouse & Baxendale 1991) (see table 1 for detail). The force increment in a fresh muscle is known to be determined by two factors: the recruitment of more motor units and increasing firing rate of the early recruited motor units. The origin of AMG is thought to be vibration of the whole muscle (Barry & Cole 1988b). The shortening of sarcomeres in a contraction will pull at both end of the muscle and the change of muscle dimension will result in the vibration of the whole muscle. The amplitude of this vibration depends on the degree of contraction force. Large muscles generally have more motor units and require less fine control over force. They rely on recruitment more than change in firing rate for force production, as reviewed by Basmajian & De Luca (1985). A large muscle, such as biceps, is known to have recruitment up to high force levels such as 80%MVC (Kukulak and Clamann 1981). Thus, this linearity between AMG and force found in this study could be explained base on recruitment strategy. Increase in firing rate of active motor units would be expected to contribute to higher frequency components of the AMG signal, certainly at the higher force range. Increase power in this higher component will surely shift the median frequency of AMG to higher value. This shift was not observed during isometric contractions in this study. Therefore, increase in firing rate of active motor units contributes little in the quadriceps force at higher force range.

The results obtained from this series of experiments are inconsistent with other reports of a linear relationship only up to 80%MVC and then a sharp decline, by Orizio, Perini & Veicsteinas (1989a) on the biceps brachii. The results also differ from a non-linear relationship as reported by Stokes, Moffroid, Rush & Haugh (1988), Barry (1990), Maton, Petitjean & Cnockaert (1990), Stile & Pham (1991) and Smith & Stokes (1993). (see table 1). The different AMG and force relationships reported may be due to the different muscle architecture and functional difference in terms of fibre composition, recruitment strategy and distribution of fibre type in a muscle. It may be possible that these differences can change the sound origin and sound amplitude at different force levels. This will be a particular problem if the recording site is fixed at one point. As mentioned above (in section 2.3.2), AMG recordings made on different sites over the surface of thigh did not indicate movements of underlining AMG source as the force changes. Therefore, change in muscle dimension during a contraction and the subsequent displacement of microphone can not be accounted for the discrepancy between the AMG force relationships reported by Orizio et al (1989a), Barry (1985), Maton et al (1990) and Stile & Pham (1991); and that obtained in this series of study.

The comparison is further complicated due to the different methods and material used in the above mentioned studies. The use of different devices can lead to quite different results. Bolton, Parkes, Thompson, Clarke & Sterne (1989) and Smith & Stokes (1993) found that the pressure exerted on a microphone can affect the AMG intensity being recorded. Orizio and co-worker(1989a) used a contact microphone, which is sensitive to pressure rather than acceleration or vibrations, which will affect results. The accelerometer used in studies reported here, is a light and sensitive device, which only response to vibration and acceleration with little other influence from the surrounding. In addition, the accelerometer is very insensitive to pressure and only responses to vibration on one plane. These characteristics enables more accurate recording of signal. An other major factor is the different muscles studied. There are only three reports of AMG-force relationships from human muscles in experiments using accelerometer as recording device for isometric contractions (table 1). In these three reports, two linear AMG-force relationships were found on quadriceps. The other reported a S-shaped AMG-force relationship in bicep brachii. It thus implies that AMG may be a better indicator for force in a large muscle.

Within force varying isometric contractions, the relationship between AMG signal intensity and force exertion is still linear in nature during contracting and relaxing phases, as illustrated in figure 38, 39 and 40. A significant difference existed between the rate of AMG intensity rise with force during contracting and relaxing phases in each individual subject. The summary data from figure 41 reveals that the rate of AMG intensity change with force increment is not significantly (p>0.05) altered at different peak force during relaxing phase and contracting phase. This indicates a close linkage between AMG signal amplitude and force production within isometric force varying contraction, and this strong link is not broken with change in peak force and hence rate of force increase. 「「「「「「「」」」を見ているというできた。「「」」というできた。

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This phenomena, to the author's knowledge, is not reported of isometric contractions in the literature. Most of which concentrated on the relationship between AMG signal intensity and the associated force production rather than on the rate of that change. The only report on rate or gradient of linear AMG-force relationship was that by Dalton & Stokes (1991) on dynamic concentric and eccentric contraction in human biceps brachii. They reported a lower rate of AMG intensity change with increasing force in eccentric contractions, in comparison to concentric contractions. It seemed that the AMG signal amplitude is closely linked to force and unaffected by rate of that force change in voluntary contractions. But initial muscle length might affect the AMG-force ratio with increasing peak force. This requires further investigation.

2.4.4 VOLUNTARY FATIGUING CONTRACTIONS:

This set of experiments were carried out with maximal effort or near maximal voluntary contractions, which will activate all motor units in the whole muscle and thus eliminate variation in motor unit numbers during a single contraction. In addition, the relatively short time course taken to fatigue muscle is preferred for experimental convenience and easy analysis.

Force production decreased with time in both test conditions, but the force decline was more rapid in the sustained contractions than in the intermittent contractions. It was noted that the subjects sustained continuous isometric contraction longer in this study than reported values of one to two minutes. This may be due to the fact that the subjects chosen were of athletic inclination and they were well able to tolerate pain. This notion is supported by Prof. N. Spurway's work in Glasgow university UK (private communication). In addition, there might be some metabolic adaptation in these semi-professional athletes, which enable them to tolerate blood occlusion during contraction.

The difference in fatiguing time course of intermittent and sustained contractions, may stem from the different types of fatigue mechanism. In the sustained contraction till fatigue, the blood is occluded and there is a greater tendency to develop peripheral fatigue. But contribution from central fatigue could not be ruled out. In the intermittent fatiguing protocol, central fatigue may be the main cause of force decline.

During continuous fatiguing test, as seen in figure 47, the force decreases with time and AMG amplitude change varied between subjects. The AMG-force ratio, as in figure 48, appears to be constant at least until 175s. Beyond this the ratio increased, but the data was only from one subject, and the duration of the contraction suggest that they may not be the truly maximal in the early stages. Alternatively, some other form of fatigue may have developed in these long duration contractions. One possibility is that fatigue induced tremor contributes to the AMG later in the contraction. This can be discounted, at least up 175s, since the constant AMG-force ratio would not allow for additional tremor. The

force oscillations seen in figure 43-46 are too low frequency to be considered as tremor. In addition the collective AMG frequency spectrum does not show any significant change in median frequency during contractions (figure 53). This would have been a sensitive test for the development of termor induced AMG components.

Data from the intermittent isometric fatiguing test show a different trend, as in figure 49. With force decline, the AMG intensity was observed to increase, decrease or remain constant in different subjects. The summary graph in figure 50 displayed increase in AMG-force ratio throughout the time course of the test. A line could be fitted to the data and the positive slope of it was found to be significantly different from zero. This indicates a dissociation between force and AMG fatigue by the intermittent contraction and this dissociation increases with progressive fatigue. It imply a different form of fatigue developed during continuous and intermittent contractions.

This tight link between AMG and force during continuous fatiguing contractions agrees well with data of continuous fatiguing test from biceps of 75%MVC (Barry et al 1985) and biceps brachii of MVC (Zwart & Keidel 1991). The short duration and the high initial force in this fatiguing experiment, ensure activation of most of the motor units in quadriceps. In muscle fatigue by sustained contraction, force loss was thought to be due to the impairment of maximal shortening velocity and slowing of relaxation in the active fibres (Bigland-Ritchie and Wood 1984) as well as decline in mechanical output of motor units. The decline in motor unit twitch force will result in fall of AMG amplitude and the lengthening of relaxation. These will shift the AMG median frequency to lower values. This shift is only demonstrated in some of the subjects in figure 53.

Furthermore, the increase in intramuscular pressure and in passive muscular stiffness occurring during fatigue could reduce fibre dimensional change and thus AMG amplitude

However, the results reported here does not agree with AMG generated from biceps brachii muscle contractions of 20, 40, 60%MVC sustained till fatigue (Orizio et al 1989b). In the same report, they shown an decreasing trend of AMG signal intensity with time during sustained 80%MVC, but did not calculate its correlation with force.

A dissociation between AMG signal amplitude and force was observed in fatigue by intermittent contraction study and shown in figure 49 & 50. In these figures there is only one case of AMG signal decline with fall of force. This is not in agreement with reports of fatigue by intermittent contractions in first dorsal interosseous (Barry, Hill & Dukjin 1992) and quadriceps (Dalton, Comerford & Stokes 1992). There are lesser peripheral fatigue and more fatigue from central nervous system in intermittent fatiguing test of a near maximal force. The change in muscle force production in central fatigue is likely to associate with change in the number of active motor units or the firing rate of active motor units or both. The degree of contribution from each of these elements require further study. Physiological tremor as a major contributor to AMG signal change could not account for all observations. Because change in physiological tremor with low frequency content would be expected to shift the AMG median frequency to lower values. Again, the shift was only observed in 3 subjects.

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2.4.5 AMG FREQUENCY CONTENT:

The results from this study shown the AMG frequency spectrum

lies between 5 to 35Hz, with very little activity beyond 60Hz. This is illustrated in figure 29 & 30. It is consistent with other reported value of Barry (1988), Zhang et al (1992) & Stile and Pham (1991). The frequency spectra are similar as forces increase and the median frequency are unaltered as shown in figure 31. This is in agreement with Diemont, Figini, Orizio, Perini & Veicsteinas (1988), Wee & Ashly (1987), Rouse & Baxendale (1991). The frequency spectra during force-varying contractions are more complex than those recorded during constant force contraction. This probably reflects the more complex mechanical events during the force-varying contraction. The median frequency of AMG signal remain constant with changing force level and changing position of recording, as shown in figure 31.

Figure 30 shows the range of AMG frequencies present at different points over the surface of quadriceps. The most intense and complex spectra are seen near the mid-line towards the proximal end of the muscle. Much simpler spectra are seen at the margins of the muscle suggesting either simpler AMG generation processes or a degree of mechanical filtering of the AMG. It is interesting to note that the spectra recorded at the patellar tendon shows considerable similarities to the tremor spectrum and remains unchanged with changing force. This is probably due to the high stiffness of the region, the AMG recorded is more of a transverse vibration than a longitudinal one and the tendon will have more vibration on the longitudinal direction with muscle contraction. There were observed intra-subject differences in AMG median frequency change with progressive fatigue, as shown in figure 53. This is unlike reports of Zwarts & Keidel (1991) who found only unchanged median frequency, Diemont et al (1988) and Maton et al (1990) discovered change in median frequency of AMG in fatigue.

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CHAPTER 3

EXPERIMENT ON RABBIT ANTERIOR TIBIALIS

3.1 INTRODUCTION

It is often easier to work with the experimental animals due to the relatively controlled conditions of the investigation and the possibility of isolating single factors in complex events. This is not easily done in human subjects. With experiments on animals, denervation techniques can avoid reflex responses elicited by stimulation. In addition, stimulation which may cause pain sensations in normal human subjects, can be employed in anaesthetized animal preparations.

3.1.1 THE NATURE OF AMG:

The nature of AMG is thought to represent the mechanical components of contraction. Oster (1984), who claimed that the action of single fibre during a contraction is the cause of AMG and the waveform of AMG varies with muscle length, peak twitch force and temperature. The second theory of the nature of the sound is that a change in the radial dimension of the fibre during a contraction (Gordon & Holbourn 1984, Brozovich & Pollack 1983). Physiological tremor has also been considered as the source of AMG signal by Oster & Jaffe (1980). Rhatigan, Mylrea, Lonsdale & Stern (1986) believed the oscillation due to pulling of elastic elements at each steps of contraction is the cause of muscle sound. Perhaps the most well supported theory regarding the origin of muscle sound is "lateral movement " of a whole muscle (Barry 1987, Frangioni, Kwan-Gett, Dobrunz & McMahon 1987 & Barry and Cole 1988). They shown that the intensity of the sound is linearly related to lateral acceleration and inversely proportioned to distance from the muscle and cosinusoidally dependent on the angle from the major plane of lateral movement.

3.1.2 THE FREQUENCY COMPONENTS OF AMG:

It is rather convenient for investigators to use animal muscles in the study of AMG produced by electrical stimulation. Cole and Barry (1991) set out to test the possibility of using frequency content of AMG to track force level. They constructed a mechanical model of frog semitendinosus muscle, using a string with distributed mass. This model simulates properties related to the muscle such as mass, length, elastic modulus, tension loading, fluid medium. This model combined with resonant frequency determined from AMG data to predict force. It showed a near perfect match between the actual recorded forces from frog muscle and those recorded from the mechanical model. In addition, it was demonstrated that the amplitude of AMG is larger at the optimal length for force production than that at a longer length. The time course of the resonant frequency rise is more rapid at the optimal length than it counterpart at longer than optimal length. The authors suggested that the time course of tension change was the dominant factor in changing the resonant frequency of this muscle during isometric contraction, and the effect of elastic modulus may be important in a muscle with larger radius -length ratio.

3.1.3 AIM OF EXPERIMENT:

Normal movements are associated with changes in muscle length, force and velocity. The following experimental protocols were an attempt to answer a few questions:

1) How does change of muscle length effect the production of AMG signal ?

2) How does the stimulation frequency affect the AMG

characteristics ?

3) How does fatigue developed during electrical stimulation affect the AMG signal ?

4) How do stimulation frequency and fatigue affect AMG signal ?

3.2 METHODS

3.2.1 ANIMALS:

Male New Zealand White rabbits, purchased from the government approved commercial sources, were used in the animal experiments. The choice in using these animals are: 1) relative homogeneous muscle fibre types in Anterior Tibialis. It has about 87% fast twitch muscle fibres and 13% slow fibres as stated by Lieber, Ferro & Hargens (1991); 2) its easy access and fixation of muscle on the experimental setup. The big body size of the rabbit allows operations to be carried out with relative ease, and also reduced the risk of hypothermia and heat loss.

3.2.2 ETHICAL CONSIDERATIONS

Ethical approval to the experiment design was given with Home office granted licences (project licence number 60/01063 and personal licence number 60/03992), and all experiments comply with requirements set out in Home Office Animal (scientific procedure) Act 1986.

3.2.3 COMMON PROCEDURES:

Experimental set-up: Initial anaesthesia of the rabbits was achieved

by inhalation of up to 4% Halothane in 80% N_2O and 20% O_2 gases. Gases were continuously supplied through a face mask before cannulation of the trachea. The state of anaesthesia of the rabbit were checked frequently by examine reflex response to painful stimulus such as toe pitching and touch of cornea. Attention was also drawn to the respiratory rate, depth and colour of mucus membrane to determine the effect of anaesthetic on depression of respiration. Muscle temperature was constantly examined during experimental procedure with a thermometer placed on and paraffin pool. All operations were performed with the rabbit on a thermal controlled blanket and overhead heat lamps were used to regulate ambient temperature.

Surgical procedures:

a) Tracheostomy: With the rabbit in prone position and under deep anaesthesia, a classical tracheostomy was performed. A medial incision in the anterior region of the hyper-extended neck exposed the trachea. After cauterising any possible bleeding, the trachea was incised partially between two cartilage rings and a glass tracheal canula was inserted and carefully fixed on place with string. After checking for airway clearance and satisfactory ventilation ensured, the skin incision was then closed and the neck extension released. b) Peroneal Nerve exposure and TA isolation: The whole leg of the animal was shaved and incision was made to expose the peroneal nerve near the knee joint. Nerves to other muscles in the anterior compartment were cut. Common peroneal nerve exposed at lateral malleolus of tibial was freed from connective tissue and placed over stimulating electrodes (see details in section 3.2.4). The nerve was crushed to eliminate reflex effects. The superficial muscle of Tibialis Anterior (TA) was then isolated and freed. The knee and ankle of the rabbit were rigidly clamped after injection of local anaesthetic (Oxycaine 1.5 - 2mg/kg body weight). Tendon of TA was sewn and tied with ligature, then freed from ankle joints and attached to a strain gauge positioned to preserve muscle line. The accelerometer was stitched on the surface of TA belly. Skin was sewn on to a metal frame to create a paraffin pool maintained at temperature range of 35°C-38°C, and light plastic film was used to minimise loose of moisture from the muscle. This set-up is shown in figure 54.

3.2.4 STIMULATIONS

Bipolar silver wire electrodes were employed in these experiments for electrical stimulation of the peroneal nerve supplying Tibialis Anterior. The stimulation pulse width was kept constant at 100 micro seconds and stimulation intensity was gradually increased till a twitch could be observed in Tibialis Anterior muscle. The intensity of the current used in the later experiments were 10 times above the current threshold for visible contractions. The stimulation frequency and the duration of stimulation varies with individual experimental protocols, which will be detailed in the following sections. The signal capture, storage, processing and analysis were carried out with the same methods stated in chapter 2

3.2.5. SIGNAL PROCESSING

Both the AMG signal and Force were recorded, store and retrieved by the same method as stated in Chapter 2, section 2.2.2 "Signal recording"; section 2.2.3 "Data recording and storage" and section 2.2.4 "signal processing and analysis". Figure 54. The experimental setup used in animal tests. The rabbit hind limb were immobilised with metal clamps at knee joint and ankle joint level (denoted by arrows A). Anterior Tibialis was isolated and the nerve innervating it was stimulated by bipolar electrode (arrow B). Reflexive action was prevented by crushing the nerve proximal. An accelerometer was stitched onto the muscle belly (arrow C) and force production were recorded from strain gauge attached to ligament (arrow D) sewn and tied on to tendon of Anterior Tibialis.


AMG: The AMG signal was captured from tape with 512 points. The AMG intensity value were measured both at the begin of a contractions and as 5 to 10 average of the peak to peak values of the very last few AMG signal waveforms. Only-rectifying-integrating (RI) method was used for AMG signal processing. The AMG sensor used in these tests were the same Entran Accelerometer employed in the Human experiments (see Chapter 2 section 2.2.2)

Force: The force transducer deployed in these tests was that of RS 20Kg Load cell (Supplied by R.S. Components, UK), which operates within the temperature range of -30 to 70 degree C and has maximum load of 20Kg with accuracy being 0.05%, as shown in figure 55.

3.2.6 STATISTICAL METHODS.

In most cases, data are presented as characteristics of individual muscles. In the case of calculated collective data, comparison of standard deviation or standard errors of group data was achieved by student t-test or rank test.

3.2.7. SPECIFIC EXPERIMENTAL PROTOCOLS:

A) THE EFFECT OF MUSCLE LENGTH ON FORCE GENERATION AND AMG INTENSITY:

This series of experiments were performed under two sets of conditions,:

1) The electrical stimulation frequency being that of a single pulse producing twitches. The rabbits were stimulated at 1Hz and the intensity



Figure 55. The calibration curve of strain gauge used during all animal experiments.

of the stimulation was 10 times the threshold for a visible twitch. The stimulation train lasted 5 to 10 seconds.

2) The electrical stimulation were delivered in short trains of 1 to 5 seconds. The frequency of stimulation was gradually increased till a full tetani was produced in Tibialis Anterior, which generally occurs at 75Hz. A supra-tetanic stimulation frequency (about 100Hz), was then chosen to stimulate the muscle to ensure full tetani.

The muscle length was controlled by the precise movement of the force strain gauge away or towards the rabbit. The optimal muscle length for force production was found by repeated stimulation across a range of lengths. The muscle length increased and decreased in steps of 2mm (from -12mm till +16mm) around the optimal length. Details for the set-up are illustrated in figure 54. Both force and the accompanying AMG signal were recorded at each length. The fatigue state was assessed by comparison of single twitches at the beginning and end of each subset of tests.

B) THE EFFECT OF STIMULATION FREQUENCY ON FORCE GENERATION AND AMG INTENSITY:

The effects of different stimulation frequencies on the force production in the muscle and the accompanying AMG signal were also investigated. Stimulation of Tibialis Anterior was carried out with increasing stimulation frequency in range of 1 to 200Hz. The stimulation intensity was still 10 times above the threshold value for a visible twitch.

C) THE EFFECT OF FATIGUE BY ELECTRICAL STIMULATION ON FORCE GENERATION AND AMG INTENSITY: Fatigue, due to electrical stimulation, was studied under two sets of conditions:

1) During continuous stimulation with frequencies between 1 to 200Hz. The duration of these tests varied with the frequency. At 1Hz the run lasted about 20 Minutes, but at 50Hz, eliciting partly fused contractions, they lasted 20 to 45 seconds.

2) The intermittent form of fatiguing stimulation, in which a burst of electrical stimulation lasting for one second is followed by a period of one second. The frequencies used were those capable of eliciting partly fused tetanus in the muscle, such as 50Hz.

D) THE EFFECT OF FATIGUE ON THE FORCE -STIMULATION FREQUENCY RELATIONSHIP AND THE AMG -STIMULATION FREQUENCY RELATIONSHIP. Electrical stimulation on fatigued muscle: The Tibialis Anterior muscle was fatigued with continuous or intermittent stimulation and then subjected to the same stimulation protocol with changing frequency as in protocol b). The stimulation intensity was 10 times above threshold value for visible contraction and the muscle was stimulated with increasing frequency between 1 to 200Hz.

3.3 RESULTS

3.3.1 THE EFFECT OF MUSCLE LENGTH ON AMG INTENSITY: These series of experiments were performed under two sets of

conditions: 1) with single twitches and 2) with brief periods of tetanic stimulation.

SINGLE SHOCK ELECTRICAL STIMULATION AT DIFFERENT MUSCLE LENGTH:

Rabbit Tibialis Anterior muscle has a twitch rise time of about 25 ms, consequently, it is a fast contracting muscle. One example of this is illustrated in figure 56. The AMG signal begins before any external force was be recorded. The interval between onset of the AMG and onset of the force increase was about 5ms. The force rise time, in this case, was 20 ms and the half relaxation time was 22.5ms.

Twitches were elicited over a range of about 12mm. There was a clear variation in the magnitude of the force development. This allowed easy identification of L_{∞} the optimal length at which the muscle develops the maximal force. Figure 57 show data from 6 experiments. In each case the length is expressed relative to L_{0} . Force generation exhibits a well known bell-shaped or near bell-shaped relationship with variations in muscle length. The twitch magnitude was reduced to about 50% with changes of 6mm in either direction. All the muscle tested demonstrated this relationship very clearly, though there are small characteristic differences between individual muscles.

The AMG amplitude did not display a simple relationship with the force at all muscle lengths. Figure 57a shows an AMG signal intensity closely associated with force production. Whilst figure 57b shown the opposite effect. In this case the AMG amplitude varied in an inverse bellshaped fashion, and the minimal AMG signal intensity occurred at the optimal length for force production. The other four muscles tested, figure Figure 56. Force and AMG waveforms recorded from rabbit anterior tibialis muscle stimulated by a single electrical pulses.



Figure 57. Force (circles) and AMG signal intensity (squares) generated from 6 (a to f) rabbit Anterior Tibialis muscle by single electrical pulses at intensity 10 times of the threshold at a series of muscle lengths. The muscle length, from which maximal force output was recorded, were set as length 0mm.



Figure 58. Summery data of force and the accompanying AMG signal generated from 6 (a to f) rabbit Anterior Tibialis muscles at a series of muscle lengths. The muscle length, from which maximal force output was recorded, were taken as length 0mm. The bottom graph shown AMG-force ratio and the values displayed in the top two graphs were all normalised to the maximal value as 100%.



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56c to f, were found to have AMG amplitudes which changed with muscle length in a linear or near linear manner. There was great variation in the gradient. All these data were collected and the result displayed in figure 58. Despite the dissimilarity in AMG intensity change with muscle length, 5 out of 6 AMG-force ratios obtained had displayed a relationship with change in muscle length, well fitted with an inversed dome shape curve. The highest value situated at or close to the optimal muscle length for force production.

ELECTRICAL STIMULATION WITH FREQUENCIES PRODUCING TETANUS AT DIFFERENT MUSCLE LENGTHS:

Broadly similar results were obtained with tetanic stimulation at about 50Hz. Figure 59 shows a typical period of stimulation. The force generated is measured from the base line to the average voltage of the peak force ripples. The AMG signal intensity was that of the average of last 10 peak to peak values of AMG waveforms. Figure 60 shows results from 6 muscles under tetanic stimulation. A clear L_0 can be identified by tetanic simulation, though interestingly this does not always coincide with the L_0 identified during single twitch stimulation. Similar to contractions elicited by the single electrical pulse, the tetanic force production shows a characteristic bell-shaped relationship with changing muscle length. There are clear optimal lengths for force production, and the force decreases with variation in muscle length around the optimal length. The rate of force change with length change in muscle differ between muscles. Again the relationship between AMG and length does not seem simple.

Data from these six muscle are collected together and illustrated in

Figure 59. An example of the force and AMG signal waveforms recorded from rabbit anterior tibialis muscle, when stimulated by 50Hz electrical pulses with 10 times the threshold voltage for a visible muscle twitch.

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Figure 60. Force (circles) and the accompanying AMG signal intensity (squares) generated from 5 (a to e) rabbit Anterior Tibialis muscle by periods of 50Hz stimulation at a series of muscle lengths. The muscle length, from which maximal force output was recorded, were set as length 0mm.



Figure 61. Summary data of force and AMG intensity generated from 6 rabbit Anterior Tibialis muscles by periods of 50Hz stimulation at a series of muscle lengths. The muscle length, from which maximal force output was recorded, were set as length 0mm. The bottom graph shown AMG-force ratio and the values displayed in the top two graphs were all normalised to the maximal value as 100%.

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figure 61. AMG-Force ratio of the data is represented by the bottom graph in the figure and it indicates that majority of muscles (5 out of 6) have the ratio change with muscle length fitted well to a shallow inverse dome shape curve.

3.3.2 STIMULATION FREQUENCY & FORCE-AMG RELATIONSHIPS:

The force developed by the muscle and the accompanying AMG during stimulation at a range of frequencies were investigated.

At a fixed muscle length: Typical force and AMG signals from one muscle are displayed in figure 62. In each case the stimulation frequency was too low to tetanise the muscle fully. Thus each stimulus pulse causes a modulation of force and an associated AMG wave. At the lower stimulation frequencies, figure 62a, the AMG event is triphasic. The waveform of the AMG event changes at higher frequencies. The peak to peak amplitude decreases as the force rises at higher frequencies. In addition, the individual event becomes more biphasic. At the higher stimulation frequencies the first AMG event in the train is substantially greater in amplitude than the subsequent events. Data of 7 experiments are summarised in figure 63. The results from these experiments are consistent. They showed the force-frequency relationship for the muscles are similar and that there were steady force output below 25Hz and big force increment with increasing stimulation frequency up 75hz. There is relatively little increase in force as stimulation frequencies rise above 100Hz. The AMG has almost exactly the inverse relationship. Its amplitude falls between 25 and 50Hz, so that as the tetani becomes more complete the AMG intensity is reduced greatly. Ultimately, with Figure 62. Traces of 10 seconds of force (top trace) and AMG (bottom trace) signals during electrically stimulated contractions at a)21Hz b)26Hz c)40Hz d)51Hz and e)100Hz. The records were made at L_0 .



Figure 63. Force and AMG recorded from 7 rabbit Anterior Tibialis muscles, when stimulated at a range of frequencies. Data were normalised to the maximal value obtained. The relationship between AMG-Force ratio and the stimulation frequency is displayed at the bottom graph. The curve fitted to data were of secondary regression curves. The regression coefficients are displayed in the graphs.



stimulation at 200Hz, which fully fuses the contraction, the maximal force is developed but the AMG is absent.

When the ratio of AMG-force were calculated and expressed in graphical form in figure 63 bottom graph, it exhibited a similar relationship to stimulation frequency to that of AMG signal intensity. It falls into a minimum at higher stimulation frequency and the maximal ratio were obtained below 25Hz. Further analysis of force and AMG signal, as well as AMG-force ratio, obtained from muscles stimulated by pulse frequency between 20 and 60Hz is shown in figure 64. This figure shows a linear relationship between force, AMG, AMG-force ratio and stimulation frequency, which could be fitted by first order regression lines: Y=1.278X-2.8186 ($r^2=0.713$) for force-frequency; Y=-2.18X+144.04 ($r^2=0.6552$) for AMG-frequency and Y=-0.079X+4.667 ($r^2=0.654$) for AMG-force relationship with stimulation frequency.

Effects of changes in muscle length: The same force-frequency curve were observed when muscle length change. With change in the muscle length away from L_{o} , the force-frequency curve obtained shifted downwards, indicating a decline in force production with changes in muscle length away from the optimal at all frequency of stimulation. The AMG signal intensity also changes with frequency of stimulation at different muscle length, in a less systematic way.

When the data are expressed in terms of ratio between AMG signals intensity and force production, as in figure 65, a clear trend emerges. At all muscle lengths tested, the AMG-force ratio has a peak with the lower frequency stimulation and declines as the frequency increases. This effect is strongest at the shortest muscle length tested and becomes weaker as muscle length was reduced.

Figure 64. Extracted data from figure 63, showing the force and AMG recrded from 7 rabbit anterior Tibialis muscles, when stimulated betwenn 20-60Hz. Data were normalised to the maximal value obtained. The relationship between AMG-force ratio is displayed at the bottom graph. The data could be fitted by straight lines.



Stimulation Frequency (Hz)

muscle lengths.

Figure 65. The changes in AMG-Force ratio with stimulation frequency from rabbit Anterior Tibialis muscle at different

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3.3.3 FATIGUING TEST:

As mentioned in the introduction section, part of the aim of this study was to clarify the AMG signal characteristics and the effect of fatigue.

THE IDENTIFICATION OF FATIGUE: TWITCH RISE TIME AND RELAXATION RATE.

The measurement of twitch rise times and half relaxation times was performed to confirm the development of muscle fatigue. Twitches were studied during both continuous and intermittent stimulation till exhaustion. A graphical display was shown in figure 66, which is the average value of force generated and the associated AMG of 15 contractions, induced with single electrical stimulation pulses. There is a large reduction in the twitch force developed. The mean twitch rise time during control conditions before fatiguing test was 22.5mS, and the mean half relaxation time was 47.5mS. After the fatiguing stimulation, the mean twitch rise time was prolonged to 25mS, while the mean half relaxation time was prolonged to 400 the force rise time were similar in value, the much prolonged relaxation time suggest fatigue. The AMG signal changes much less than the force. The AMG does fall in amplitude and each component occurs a little later.

FATIGUE TESTS USING CONTINUOUS AND INTERMITTENT STIMULATION:

The characteristics of AMG during fatiguing contractions were studied during stimulation at 50Hz at about Lo. Two stimulation patterns were used: 1) continuous stimulation; 2) intermittent stimulation at 50Hz for one second followed by one second rest. The changes in the force frequency and AMG frequency curves were also compared in fresh muscle and after periods of fatigue had been induced by sustained muscle activity.

Figure 67 shown data from continuous 50Hz stimulation till fatigue of

Figure 66. The twitch response to a single supra-threshold electrical pulse before and after fatigue in a rabbit anterior tibialis muscle.



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Figure 67. Force generation (open circles) and the accompanying AMG signal production (closed circles) recorded from five rabbit Anterior Tibialis muscles during continuous 50Hz stimulation till fatigue. Data were normalised to the initial values as 100%.



Time (seconds)

five muscles, open circles denote force production and closed circles AMG production. All data were normalised to the initial value at the start of experiment. There was a degree of force potentiation during the course of stimulation in 4 out of 5 muscles and it reached its peak at various times after the start of stimulation. The AMG signal intensity, however, did not always follow the change in force production. There were initial increases in AMG production at the first few seconds and then declined the reached a minimum about 20 seconds into the contraction for all muscles tested. There were two cases of close association between force and AMG, but in the three other cases there was no obvious link between AMG and force.

However, the AMG-force ratios change with time in a similar fashion as AMG signal. This is displayed in figure 68. In this figure, the AMG-force ratios increased in the first seconds, reaching a peak between 2-5 seconds. They then fell rapidly till a steady phase is reached after 20 seconds into the contraction. The data from intermittent fatiguing test are illustrated in figure 69, where all values were normalised to the value obtained at the start of experiment. It is evident that it would take longer to fatigue a muscle with intermittent fatiguing protocol (70-100s) than the sustained stimulation protocol (25-50s). Similar to data from sustained stimulation protocol, there were degree of force potentiation during the course of experiment. This is sometime accompanied by corresponding AMG potentiation.

The degree of association between AMG and force is illustrated in figure 70, in the form of AMG-force ratio. In this figure, the AMG-force ratio of all 4 muscles tested shown an initial decline during first 8 to 15 contractions, then remain constant thereafter.

Figure 68. AMG-force ratio recorded from five rabbit anterior tibialis muscles during continuous 50Hz stimulation till fatigue. Data were normalised to the initial values as 100%.




Figure 69. Force generation (open circles) and the accompanying AMG signal production (closed circles) recorded from five rabbit Anterior Tibialis muscles during intermittent 50Hz stimulation till fatigue. Data were normalised to the initial values as 100%.

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Figure 70. AMG-force ratio recorded from five rabbit Anterior Tibialis muscles during intermittent 50Hz stimulation till fatigue. All data are nonnalised to the initial values as 100%.

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The effects of fatigue on force production and the accompanying AMG signal were investigated in this experiment. Both continuous and intermittent stimulation protocols were used. Different conditions were deployed: 1) Electrical stimulation on fresh muscles and 2) Electrical stimulation on fatigued muscle. The result obtained from 5 individual muscle tested is graphically demonstrated in figure 71, 72 and 73. The maximal signal of force and AMG were taken from the highest value at fresh state for any given muscle.

The relationship between force production and frequency of stimulation in a fatigued muscle, as shown in figure 71, is very similar to those obtained from fresh muscles, as in figure 63 & 64. The force increase was small in low stimulation frequency range, but increased sharply with higher stimulation frequency eventually reaching a plateau. Fatigue caused a decline in force production at all stimulation frequencies.

AMG signal intensity, as shown in figure 72, related to stimulation frequency in a general exponential decline, both at fresh or fatigued state. This decline slows down greatly, when stimulation frequency increases beyond 100Hz. When the muscles are fatigued, AMG signal intensity was reduced at frequency range below 100Hz. Beyond 100Hz, the AMG signal intensity is very similar in value between fresh and fatigued state.

The AMG-force ratio and stimulation frequency relationship is shown in figure 73. This figure displayed visual similarity to AMGfrequency relationship in figure 72. However, the exponential decline of the ratio slows greatly at a lower stimulation frequency than shown in figure 72. The turning point for individual muscle tested was different in Figure 71. The force recorded from five rabbit Anterior Tibialis muscles before (solid symbols) and after (open symbols) fatigue. The force developed at a range of stimulation frequencies is shown.



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Figure 72. The AMG signal intensity recorded from five rabbit Anterior Tibialis muscles, before (solid symbols) and after (open symbols) fatigue. The AMG elicited at a range of stimulation frequencies is shown.

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Figure 73. The Force-AMG ratio obtained from data plotted in figures 67, 68. Solid symbols indicate data at fresh and open symbols denote data at fatigue state.



value.

3.4 DISCUSSION

Experiments were carried out on anaesthetized rabbits, to further investigate the AMG characteristics. This allowed greater mechanical isolation of the muscle and fuller control of the stimulation. In addition, tremor and reflex changes in muscle force seen in voluntary contractions could be reduced. The findings are presented in the following sections, following the same order of experimental protocol as in the method section of this chapter.

3.4.1 AMG AND CHANGE IN MUSCLE LENGTH:

During single twitch stimulations: The AMG signal appears before force and the highest AMG intensity occurs during the rising phase of the twitch, as shown in figure 56. The force shown a well known bell-shaped relationship with changing lengths of muscle. The corresponding AMG intensity changes with muscle lengths in non-systematic way with a displaced maximal away from L_0 length. These are shown in figure 57 & 58. The AMG-force ratio change with muscle lengths with a relationship closely assembles the force-length curve, as seen in figure 58.

The origin of AMG is still under investigation. The early onset of AMG signal before force indicates that AMG signal must be related to events early in electro-contraction coupling.

During tetanic stimulation: Similar to twitch stimulation, the AMG signal has highest intensity during force rise phase with earlier onset. As

the force approaching tetani, burst of high AMG activity associates with stimulus gradually diminishes. This is visible in figure 59. The same forcelength relationship is observed as in twitch stimulations. Again, the AMG signal shown no systematic relationship with length change, as displayed in figure 60 & 61. However, the AMG-force ratio seems more regular but the inverted bell-shaped curve strongly resembles the inverse of the clear force-length relationship.

3.4.2 AMG AND STIMULATION FREQUENCY:

The twitch duration from figure 56 is about 50ms. The frequency needed to produce unfused tetanic contraction must be above 20Hz. This is confirmed in figure 62, where AMG signal during steady force diminishes with increasing frequency of stimulation. The relationship between force and stimulation frequency is a well known one and fitted well with a S-shaped curve. The AMG signal change with stimulation frequency was that of the inverse of force-frequency curve. The AMGforce ratio change with stimulation frequency in a manner closely resembles AMG-frequency relationship. These are shown in figure 63, Based on the fact that it requires over 20Hz stimulation frequency to produce unfused contractions and the near zero AMG amplitude beyond 55Hz. An extract of data from figure 63 is shown in figure 64. In this figure, the force, AMG and AMG-force ratio change with stimulation frequencies in a linear fashion. The force increases significantly with increasing stimulation frequency, while and AMG and AMG-force ratio decrease significantly. This AMG-force ratio fall with increasing stimulation frequency is still evident when muscle lengths changes. However, the fall seemed to be steeper at short muscle lengths, as

exhibited in figure 65.

3.4.3 AMG AND FATIGUE:

As muscle fatigues, both force and AMG decreased. This is evident in twitch stimulation in figure 66, continuous and intermittent stimulation in figure 67 & 69, and fatiguing effect on force and AMG produced by changing stimulation frequencies in figure 71 & 72. The AMG-force ratio rises through the first 5 seconds of continuous stimulation then falls progressively to almost zero, as shown in figure 68. The AMG-force ratio during the intermittent stimulation fell immediately, and was almost complete by 20 stimulation, as seen in figure 70. These suggest different underlying mechanism of fatigue exists between continuous and intermittent stimulation. In addition, they indicate that the AMG falls significantly faster than muscle force as fatigue develops. However, the summary AMG-force ratio obtained from changing stimulation frequencies did not show significant change with fatigue.

3.4.4 SUMMARY RESULTS:

The overall results shown that there is no clear relationship existed between AMG signal and force production in a stimulated contraction. However, it is clear that forces produced by high frequency stimulation is associated with little or no AMG signal. This is also apparent in later part of fatigue when force falls. This dissociation between AMG and force could be attributed to the increase stiffness and intramuscular pressure and the diminished muscle movement on reaching tetani and fatigue.

3.4.5 COMPARISON WITH REPORTED DATA:

A > Effects of changing muscle length: The AMG intensity showed a less direct relationship with change in muscle length, as shown in figure 57 and 60. This observation is similar to reported results in Barry (1987) and Frangioni et al (1987) recorded from twitches. Though they only examine AMG accompanying twitches, they did report a bell-shaped AMG intensity relationship with changing length and the AMG-length curves were out of phase with the force-length curve peaking at shorter muscle length at about 90% L₀. Results from this series of experiment shown no systematic relationship and the peak AMG intensity were found for muscle length both longer and shorter than the optimal length.

It is possible that the discrepancy lies: 1) the different AMG recording device used ie. hydrophone vs. accelerometer. The hydrophone is sensitive to wave, including ambiant noise; while the accelerometer used in this experiment is sensitive to movement in only one direction, 2) The controlled environment of the experiments; Barry (1987) performed experiment in temperature range of 7-20°C, Frangioni et al (1987) studied at 20°C. Experiments reported here was under temperature of 35-37°C. Temperature change will affect twitch force. If AMG is in anyway linked to force, changes in temperature will certainly affect AMG signal in amplitude (Barry 1987) or waveform or both. 3) the difference between muscles: the AMG recorded is thought to be due to the lateral movement of muscle (Barry & Cole 1988), and the muscle vibrate at resonant frequency. The resonant frequency of muscle is dependent on the muscle geometry such as size, stiffness, length, mass and viscosity, these factors are different in different muscles and would be different even in the same muscles from different animal species.

B> Effect of change in frequency of stimulation: In this study,

AMG amplitude was found to decrease with increasing force during contractions induced by a series of frequencies of the sub-tetanic range (figure 63), and there was a significant decline in AMG-force ratio, as shown in figure 63 & 64. This suggest a dissociation between AMG and force in stimulated contractions. This data is consistent with results from reports of cat soleus muscle by Zhang, Herzog and Vaz (1994) and Vaz, Herzog, Zhang & Zhao (1994). Though the fibre composition and geometry are quite different between cat soleus and rabbit anterior tibialis, they both exhibited this dissociation of AMG amplitude and force.

The same dissociation between AMG and force with changing stimulation frequency was observed at a series of muscle lengths. The dissociation is more pronounced at short muscle lengths. This agrees well with part of data from Vaz et al (1994), though they only attempted stimulation at two lengths and cat soleus only works in the ascending limb of force-length relationship.

C> Effect of fatigue and AMG signal:

There was a general trend of decrease in AMG intensity with progressive fatigue, corresponding to the fall in force production. This is readily observable in figure 67 and 69. The decline in AMG signal intensity might be due to decreased peak twitch of motor units with fatigued rather than decruitment of motor unit. This decline in peak twitch force excerts less driving force on muscle to vibrate as a whole. This is consistent with the absence of median frequency shift in AMG signal spectrum and the diminished amplitude. Again, there are no simple formulae for describing AMG-force relationship. The clear dissociation between AMG and force imply that AMG depends less on the force producing mechanism in fatigue and AMG intensity alone will not be a

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GENERAL DISCUSSION

It was the original aim of this study to identify the optimal positions or conditions for recording AMG; investigate AMG characteristics and its relation to force of contraction; assess the possibility of AMG as an indicator of muscle fatigue and construct an animal model for study of electrically elicited muscle contractions.

4.1 SUMMARY OF DATA FROM THIS INVESTIGATION

The overall data indicates that there is no optimal position for recording AMG in an active human muscle. The AMG signal amplitude remain closely linked to force during voluntary isometric contractions. A tight association between AMG amplitude and force output during fatiguing contractions was only observed in sustained contractions but not the intermittent form. On the contrary, AMG amplitude does not have a simple relationship with force output in all form of stimulated contractions of rabbit anterior tibialis. In both data obtained from human and rabbit experiments, the AMG frequency spectra were complicated. The median frequency did not significantly change with force and fatigue in voluntary contractions. The AMG spectra were swamped with high energy signal at a frequency corresponding to the stimulation frequencies. These data lead to the conclusion that the AMG amplitude could be a force indicator in voluntary contractions and sustained contraction till fatigue, but not in the stimulated contractions. In addition, the median frequency of AMG signal could not be useful in predicting force output in both voluntary and stimulated contractions.

The different AMG signal behaviour with changing force during voluntary and stimulated contractions could, at least in part be attributed to:

1) The different muscle fibre type composition. The quadriceps were of mixed fibre types and rabbit anterior tibialis has predominant fast muscle fibres. The fast muscle fibres would have higher twitch force, and thus higher driving force for muscle movement (Marchetti, Felici, Bernardi, Minasi & Filippo 1991). Hence, the AMG signal amplitude would be higher. Furthermore, the motor unit fibre territory will not be the same in these two muscles.

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2) The different recruitment strategy used: Voluntary force is controlled by motor unit recruitment / decruitment and firing rate. If AMG is linked to force, then its' amplitude would reflect the number of motor unit active and their change in firing rate. The unchanging AMG median frequency and changing AMG amplitude observed in human experiments agree well with the fact that force generation in quadriceps is largely governed by motor unit recruitment. The possible reflexive influence on force and AMG could not be over looked. The stimulated muscle, on the contrary, had all motor units activated and the activation frequency is controlled by the stimulation pulses. As shown in figure 62 & 63, there are non-linear changes in AMG signal amplitude with changing stimulation frequency, which may reflect the degree of synchronization.

3) The degree of filtering and signal distortion: The AMG signal recording were made on the surface of the skin during voluntary contractions and directly on the muscle surface during stimulated contractions. The filtering effect will be higher in the voluntary contractions. This will be further complicated by the relative depth in location of slow motor unit fibres and faster motor unit fibres.

4) Difference in muscle properties: The rabbit anterior tibialis and human quadriceps differ in size, mass, length, stiffness, muscle pennation and elastic elements. These differences will determine the resonant frequency stiffness of the muscle (Barry & Cole 1988a, 1988b) and thus affect AMG amplitude.

4.2. AMG CHARACTERISTIC COMMON TO BOTH HUMAN AND ANIMAL DATA

The AMG signal spectrum has power concentrated in a range between 0-45Hz, with very little frequency power above 60Hz. This is clearly shown in figure 28, 29 and 51.

The median frequency of AMG signal was found to be dissociated from force production in all situations examined. This indicates the lack of shift in AMG power frequency. If physiological tremor had increased, one would expect a downward shift in the AMG median frequency. If force was increased by way of increasing firing rate of motor units leading to summation of fibre twitch, there would be an increase in AMG median frequency. Neither of this is observable in collective data. Furthermore, the AMG signal spectrum was greatly influenced by the stimulation frequency. This observation only agrees with reports from stimulated contractions (Stokes & Cooper 1992) and voluntary eccentric contractions (Dalton & Stokes 1993). Data from this experiment did not agree with reports of changing AMG signal frequencies: in voluntary contraction by Zhang, Frank, Rangayyan & Bell (1992), Maton et al (1990), Orizio, perini & Diemont (1990); change in median AMG frequency with stimulation rate and fatigue (Zhang, Frank, Ragayyan & Bell 1993); The frequency of AMG signal was found dependent on the length of the muscle (Frangioni et al 1987).

The AMG signal is thought to be generated at the resonant frequency of muscles (Barry & Cole 1990). The resonant frequency is dependent on the properties of muscle and test conditions, such as the size, mass, length, elasticity, tension loading and the fluid medium. All these factors would be very different from muscle to muscle, and thus account for part of the difference in results. In addition, the task dependent nature of muscle activation and muscle contributing to more than one joint movements might affect the AMG signal amplitude in different conditions.

4.3 THE NEED OF FEEDBACK IN FES

The functional electrical stimulation of paralysed muscles for regaining normal movements has little success due to the problem of fatigue and variability of the stimulation, which results in poor reproducibility of movements. This calls for a good feedback control system, which will increase reproducibility and at the same time reduce fatigue caused by stimulation protocols. This feedback system relies on quantity and quality of feedback data, the sensor making accurate recording and the speed and effective analysis of those data.

4.4 SENSORS AVAILABLE AND THEIR SUITABILITY IN FES APPLICATIONS

The sensors, for force monitoring in a FES application, are currently under investigation and in clinical application, such as gait analysis, lower level prothosis control. They are EMG signal (Graupe & Kohn 1988), AMG signal (Barry 1985, Frangioni et al 1987) and intramuscular pressure (Sejersted & Hargens 1995). The quality of a sensor should be that of: good accuracy, good interrelation with control strategy, wide range of measurement, adequate band width of response and ease of mounting.

As mentioned in the beginning of chapter 1, the EMG has an advantage of easy mounting and online analysis. But it is not a good force indicator during fatiguing contractions. EMG can be contaminated by electrical stimulation and movement of electrodes. The frequency content of EMG signal changes with fatigue more as a consequences of muscular activity. These characteristic reduced the potential for EMG being a good and convenient force indicator.

The intramuscular pressure (IMP) had shown promise due to strong linkage with force irrespective of the mode and speed of contraction. It is thought a better predictor of force than EMG. The signal is determined by the tension of the muscle fibres, recording depth and the fibre geometry, such as fibre curvature or pennation angle (Sejersted & Hargens 1995). However, there is not much data available on the IMP changes during intermittent or dynamic contractions lasting more than a few minutes, even less with fatigue.

The investigation into AMG signal has a longer history than IMP. The suitability of AMG signal as a force indicator will be discussed in the following section, relating to data from this experiment and the literature.

4.5 THE POTENTIAL OF AMG SIGNAL AS A FORCE INDICATOR IN FES APPLICATIONS.

In FES application, such as restoration of movement, there are needs to control over the speed and range of movement by regulating force. This regulation is achieved by changes in stimulation frequency to bring about changes in force and joint angle involving muscle length change. The state of muscle should be monitored to avoid fatigue and reduce muscle damage. If AMG could be an monitor for force, then it must show link with force under these conditions.

4.5.1 THE AMG SIGNAL AND STIMULATION FREQUENCY

The AMG signal amplitude change with increasing stimulation frequencies in stimulated contractions and the changes mirroring the forcefrequency curve (figure 62 & 63). This is in agreement with Stokes & Cooper (1992) observation in human adductor pollicis. Furthermore, the AMG-force ratio changes with stimulation frequency in a fashion closely resembling AMG-frequency curve (figure 62 & 63). This imply that the AMG signal amplitude depends very little on the force production in stimulated rabbit tibialis anterior, and the AMG signal amplitude could not be used as a good indication of force when the stimulation frequency changes. The AMG signal might depend more on motor control rather than the intrinsic contractile process. However, both this study and the report of Stokes and Cooper (1992) examined small muscles with relatively small numbers of motor units. In addition, the frequency of stimulation used in FES applications are typically between 20-40Hz, which might be producing twitches instead of tetani in a muscle with mixed or slow fibres. Further investigation is required to assess AMG response to stimulation in a larger muscle with mixed fibre types, such as quadriceps.

4.5.2 THE AMG SIGNAL IN CONTRACTIONS WITH CHANGING MUSCLE LENGTHS

The AMG signal amplitude has diverse relationship with force when muscle length changes during stimulated contractions. This is evident in figure 56, 57, 59 & 60. A dissociation between summary data of AMG amplitude and force is shown in both figure 57 & 60. This is not in agreement with reports in the literature, which shown a bell-shaped curve of AMG-length relationship with a displaced maxima (Barry & Cole 1987, Frangioni et al 1987 and Dobrunz, Pelletien & McMahon 1990). The discrepancy between results from frog gastrocnemius (Barry & Cole 1987, Frangioni et al 1987) and those reported here on rabbit anterior tibialis, could not be totally accounted for by the fibre composition difference.

The data from this experiment, shown in figure 56 & 59, demonstrated that AMG relationship with force remained as a property of individual muscle. This indicate that AMG amplitude might not be useful as a force indicator when muscle length are changing in a population, but could be useful in some muscles. On the other hand, the length change in quadriceps muscle to bring about knee extension in FES could only change muscle length at the most about 5% of muscle length at full extension. The data presented AMG signal change with muscle length in the range of 12-16% resting length. The AMG-force ratio change with 5% of L₀ will not affect the AMG signal to a great extent. Further work is required to examine closely the discrepancy between data reported before AMG signal could be discarded as an indicator of force.

4.5.3 THE SENSITIVITY OF AMG IN FATIGUE BY STIMULATION

Prolonged force rise phase and relaxation phase in stimulated twitch was observed after fatigue, and the AMG shown a corresponding decrease

in peak-peak amplitude and signal cycles (figure 67). The force changes are thought to be brought about by decreased contractile speed and relaxation rate, changes in recruitment strategy of the muscle, changing firing rate of active motor unit as well as lowering firing threshold of fast motor unit in voluntary contractions (as reviewed by Edwards 1985). There are possible contribution from decruitment of fast-twitch fibres. leading to a broadened twitch width (Fang and Montimer 1991), though it might only be relevant in a small muscle which rely on changes in both recruitment and firing rate as mechanism of force generation starting from a low force level. Furthermore the slow contractile speed allows a decrease in motor units firing rate for the same force output. All these changes could diminish the AMG amplitude as the driving force decreases for whole muscle lateral movement reduces. This is in agreement with observation by Barry & Cole (1991). The decrease in AMG amplitude observed during fatigue induced by voluntary contractions (figure 46 & 48), could be due largely to the decrease in amplitude and slowing of twitch, when the change in AMG median frequency is absent, though the precise relationship between AMG and force is more complicated and depends on other factors and vary between individuals.

The stimulated contractions, as stated previously, had activated all motor units with predetermined frequency. The decrease in AMG signal amplitude with progressive loss in force, shown in figure 65 & 67, could only be accounted for by the decrease in twitch force of individual motor units. There might be contribution from increased asynchrony of the motor fibres as different type of motor units has different fatiguability.

The AMG signal amplitudes were so varied that there is no simple relationship between summary data of AMG and force in fatiguing contractions. The AMG signal remained a property of individual muscles. This imply that AMG signal amplitude is dependent less on force and more on other factors, such may be different strategy of force generation and the subsequent changes in muscle properties. This leads to the conclusion that if AMG signal is useful for force indication in fatigue, alternative parameter must be sought other than the amplitude of the signal. These might be in the power changes in certain frequency range, but certainly not the median frequency.

4.6 ENVIRONMENTAL INFLUENCE ON AMG SIGNAL PRODUCTION

Not unlike other scientific studies, all observations associated with AMG are susceptible to contamination by artifacts and any feedback signal in FES must be relatively pure to be useful. The influence by subcutaneous blood flow is ruled out by Oster and Jaffe (1980). But their claim that the AMG signal is independent from muscle temperature is contested by Barry (1987).

Specifications of equipments, such as strapping microphone around the muscle under study (Orizio et al 1989a, 1989b), could potentially introduce artifacts in AMG signal. With muscle bulk movement during a contraction (Barry & Cole 1988b), the pressure exerted on the microphone by the strap may cause distortion of AMG signal and further induce fiction between skin and microphone. This problem may be overcome by the use of accelerometer, which is not sensitive to pressure but movement.

Some influence of cardio-respiratory system may be recorded in the raw AMG signal (Lakie, Walsh & Wright 1982). This may be compensated by employing filters. Again the use of accelerometer with sensitivity in one direction will eliminate the artifacts and ambient vibration.

One element of influence may not be easily eliminated by external manipulation, that is of physiological tremor as noted by Wollaston (1810): "In attempting to lessen the number of vibrations, there appears to be a degree of unsteadiness which prevents any accurate measurement of the real number". The physiological tremor could be reduced by cooling the muscle under investigation and occlusion of blood flow (Oster & Jaffe 1984). The physiological tremor is a particular problem in sustained contractions of high forces Barry et al (1985). The interference of physiological tremor can be eliminated at the expanse of loosing some AMG signal, by filtering the signal with a 12Hz high-pass filter.

Wee and Ashley (1989) pointed to another source of interference: the sound produced by the distant contracting muscle. Especially when a group of muscle is contracting, sound from synergic muscles and that from muscle under investigation is rather difficult to separate. This factor needs to be considered when study is carried out on a large muscle group such as quadriceps. Accelerometer with high sensitivity in one plane could ensure the recording signal has predominant energy coming from area directly under fixation point.

4.7 CONCLUSION:

Skeletal muscle contractions can generate transverse mechanical waves, which is measurable by a sensor, like accelerometer. This wave is biphasic in nature and has an early onset than external force.

The data presented in this report corroborate a linear relationship

between AMG and force level in isometric contractions of human quadriceps muscle. This close link between AMG signal intensity and force production is also present in fatigue induced by sustained voluntary contractions but not by intermittent contractions.

The data obtained from rabbit tibialis anterior muscle showed less systematic relationships between AMG and force production. AMG signal intensity did not display a close relationship with force under condition of changing muscle length, stimulation frequency and fatigue. There were no significant change in AMG signal median frequency during voluntary contractions. The AMG signal were dominated by stimulation frequency.

The good correlation between AMG and force in voluntary contractions, shown promise as an indicator of voluntary force from isometric contractions and fatigue by sustained contractions. The possible clinical use of AMG could be in the area of assessing force output from muscles with limited access, such as paraspinal muscle and facial muscles. It could also be used in conjunction with EMG to assess the state of the muscle function in health and disease and muscle mechanic in training. The use of AMG as force indicator in stimulated contractions, such as FES applications, requires further investigation. The AMG signal did not appear to related strongly to force output under the conditions investigated. Alternative properties of AMG signal, such as power content in specific frequency ranges, should be investigated further for FES applications. The AMG median frequency certainly is not a good force indicator under all conditions tested in the experiment.

4.8 SOME SUGGESTION FOR FURTHER WORK:

This study had contributed further to the knowledge of AMG and

its relationship in both voluntary isometric contractions and electrically stimulated contractions. However, isometric contractions were not the only type of contractions involved in everyday life activities and it thus become important to carry this work further in the area of investigating AMG characteristics in dynamic movements, where muscles might be involved in two action: movement and rotation of the joints, and the subsequent fatigue state.

During fatiguing contractions, the AMG signal behaviour was not tightly linked to force output, but does depend on the initial force in fatigue. Further study required to clarify AMG signal characteristics in fatiguing by different initial force with sustain, intermittent and dynamic contractions. This could be an important area to be investigated and both AMG and EMG signal might contribute to further understanding of muscle activation strategy. This study consistently demonstrated that AMG signal median frequency is not a good indicator of force, but the relative power content of AMG signal in specific ranges should be investigated before AMG signal could be discarded as force indicator.

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